













SOCRATIC LECTURES

12 TH INTERNATIONAL SYMPOSIUM LJUBLJANA, JANUARY 11, 2025

PEER REVIEWED PROCEEDINGS PART II

EDITED BY:

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Z-STEAM







Socratic Lectures

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Program of the Symposium Socratic Lectures, January 11, 2025, 10:00 - 17:00 (Ljubljana time)

- 10.00 Welcome to participants (Veronika Kralj-Iglič, University of Ljubljana)
- 10.05 10.45 Plenary lecture: Saara Laitinen: Extracellular vesicles from blood cells and in blood. The avenue of future therapies

10.45 Break

11.00 Scientific sections

Section 1: Medicine (Chairs: A. Nemec Svete, K. Troha)

- 11.00 11.20 G. Omejec: Ultrasonographic assessment of diaphragm function
- 11.20 11.40 A. Troha: Advancements and future prospectives of platelet-rich products and extracellular vesicles in otology
- 11.40 12.00 A. Čuček: Mammary gland anatomy, histology, pathology, and physiotherapist management post surgery
- 12.00 12.20 K. Pahovnik: A case of autosomal dominant osteopetrosis with a novel mutation in CLCN7
- 12.20 12.40 L. Dolinar: Destigmatization of erectile dysfunction
- 12.40 13.00 D. Dolinar, I. Potparič: Premature fracture of a modular femoral neck after total hip arthroplasty

Section 2: Veterinary Medicine (Chairs: V. Erjavec, M. Šimundić)

- 11.00 11.20 L. Jarnovič: Brachicephaly in cats. A silent problem in feline health
- 11.20 11.40 B. Perc: Hematology-derived inflammatory markers in dogs with BOAC
- 11.40 12.00 I. Premrl: Foreign body removal from the canine digestive tract: findings and outcomes
- 12.00 12.20 M. Šimundić: Approach to the patient with polyuria/polydipsia
- 12.20 12.40 B. Dučić: Retained tooth roots in cats
- 12.40 13.00 M. Arko: Extracellular vesicles in bovine colostrum and milk

Section 3: Physiotherapy (Chairs: R. Vauhnik, A. Jakopič)

- 11.00 11.20 Bec: Effects of transcutaneous electrical nerve stimulation in patients with fibromyalgia syndrome
- 11.20 11.40 K. Breznik: Pain relief treatment of fresh sacral fracture for a professional snowboarder one month before participation in the Winter Olympic Games: a case report
- 11.40 12.00 A. Jakopič: Effects of hydrotherapy on functional capacity of patients after stroke
- 12.00 12.20 M. Jelen: The effects of passive loading of the knee joint after anterior cruciate ligament reconstruction on knee stability
- 12.20 12.40 E. Strle, L. Uhan: Surgical treatment of chronic post-traumatic hamstring origin pain with tendon transfer from the unfused ischial tuberosity apophysis to the ramus of the ischial bone: a case report
- 12.40 13.00 M. Zoroja: Ring shaped lateral meniscus Raise awareness and avoid unnecessary surgery

Section 4: Green transition (Chairs: T. Griessler Bulc, M. Bavcon Kralj)

- 11.00 11.30 A. Drozd-Rzoska: Physics & Society: High Pressures for innovative pro-health foods (keynote lecture)
- 11.30 11.45 A. Kranjc Požar: Microalgal technologies: Treatment strategy for contaminants of emerging concern
- 11.45 12.00 E. Andreasidou: Uptake of contaminants of emerging concern in tomato plants irrigated with treated wastewater
- 12.00 12.15 L. Klemenčič: Harvesting of algae biomass by electrocoagulation







- 12.15 12.30 J. Danilović Luković: Impact of ionizing radiation on microalgae: Enhancing biotechnology potential
- 12.30 12.45 U. Šunta: Surface properties of algal biomass and microplastics: An exploration of point of zero charge and contact angle
- 12.45 13.00 T. Zrnec Drobnjak: Production of plant biostimulants from microalgae grown on the biogas digestat

Section 5: Nanostructurome pipeline (Chair: P. Trebše, U. Lavrenčič Štangar)

- 11.00 11.10 I. Jerman: Infrared and Raman spectroscopy
- 11.10 11.20 J. Jakše: Next generation sequencing
- 11.20 11.30 J. Hočevar: NMR Spectroscopy
- 11.30 11.40 R. Cerc Korošec: Thermoanalytical techniques
- 11.40 11.50 M. Novinec: Mass photometry for molecular mass determination
- 11.50 12.00 G. Schlosser: LC-MS lipidomics, metabolomics
- 12.00 12.10 E. Heath: LC-MS/MS methods
- 12.10 12.20 P. Hansson: SAXS and investigation of diffusion and transport of drug discovery systems in extracellular matrix models
- 12.20 12.30 V. Kralj-Iglič: Scanning electron microscopy
- 12.30 12.40 O. Vanderpoorten: Nanospacer: Nanofluidic sizing of biomolecules and EVs
- 12.40 12.50 G. Liguori: in vitro cell assays
- 12.50 13.00 S. Michelini: ELISA assays
- 12.50 13.00 All partners: Discussion on sample preparation and characterization

Section 6: FarmEVs: Different sourced extracellular vesicles and their potential (Chairs: G. Pocsfalvi, A. Di Loria)

- 11.00 11.20 G. Guerriero: Plant extracellular vesicles
- 11.20 11.40 I. Schabussova: Bacterial extracellular vesicles
- 11.40 12.00 S. Vainio: From sustainable soil to lifelong health speculations from ecological nanointeractomics
- 12.00 12.10 R. Mammadova: Enhancing therapeutic potential: Loading strategies for plant-derived nanovesicles
- 12.10 12.20 Glamoclija: Trichinella spiralis extracellular vesicles alleviate OVA-induced allergic inflammation in BALB/c mice
- 12.20 12.30 P. Devarkanda: Bean PDNVs isolation and characterization from the dwarf organic Borlotti bean Lingua di Fuoco
- 12.30 12.40 E. Cepec: Biostimulants from algae biomass uncovered: phytohormones determination and plant growth stimulation
- 12.40 12.50 B. Korenjak: Interferometric light microscopy of diluted blood in different species

Section 7: Biophysics (Chair: A. Iglič)

- 11.00 11.20 S. Rzoska: Unique properties of high-pressure & high-temperature formed glasses: The application for new-generation batteries (Keynote lecture)
- 11.20 11.40 T. Beke Somfai: Peptide-based antibiotic supramolecules (Keynote lecture)
- 11.40 12.00 L. Mesarec: Equilibrium membrane shapes influenced by different concentrations of orientationally ordered curved rod-like membrane protein
- 12.00 12.20 M. Mussel: On spikes and sound: Debating the physical nature of action potentials
- 12.20 12.40 L. Bar: Impact of inclusions on the organization and phase behavior of lipid membrane models
- 12.40 13.00 M. Drab: Monte Carlo studies of vesicle shapes with anisotropic membrane inclusions and volume constraints

Section 8: Physics (Chair: S. Kralj)

- 11.00 11.30 A. Jelen: Microscopic patterns and complexity (Keynote lecture)
- 11.30 11.45 A.A. Sojecka: Doomsday criticality for the global society
- 11.45 12.00 T. Blazevic: Maximal entropy production
- 12.00 12.15 D. Dovnik: Fractional topological charges
- 12.15 12.30 J. Sgerm, A. Ribas, L.G. Fugina: Universality of physics of phonons
- 12.30 12.45 G. Goričan: Kibble-Zurek mechanism and applications
- 12.45 13.00 M. Župec, M. Kodrin, R. Rojko, M. Štorman: Crystal lattices in nature







Section 9: Liquid Crystals (Chairs: A. Jelen, B. Švajger)

- 11.00 11.20 T. Javornik: Importance of the critical point in thermotropic nematic liquid crystals in terms of their sensitivity
- 11.20 11.40 A. Hoelbl: Localised excitations in liquid crystals as particle analogues
- 11.40 12.00 E. Čokor: Imry-Ma patterns in confined nematic liquid crystals
- 12.00 12.20 M. Potrč: Lyotropic liquid crystalline phases
- 12.20 12.40 B. Švajger: Volume and surface phase transitions in confined nematic liquid crystals
- 12.40 13.00 M. Zid: Mode coupling and memory effects

Section 10: Education (Chair: G. Torkar)

- 11.00 11.20 G. Torkar: Environmental education for behaviour change
- 11.20 11.40 I. Devetak: Pre-service primary school teachers' understanding of biogeochemical cycling
- 11.40 12.00 L. Vinko: Environmental literacy of chemistry teachers
- 12.00 12.20 S. Beslagic: Robotics in high school
- 12.20 12.40 T. Plešnik: Undergraduate physiotherapy student perceptions of teaching and learning activities associated with clinical education
- 12.40 13.00 Ž. Rode: Pre-service teachers as citizen scientists

Section 11: Bridging Science, Health and Arts I (Chair: Y. Istileulova)

- 11.00 11.20 N. Gomes: Transformative educational strategies with artificial intelligence (Keynote lecture)
- 11.20 11.40 S. De Lasala Porta: Truth, beauty, and ethics in art, science, and health: Interdisciplinary reflections and philosophical perspectives
- 11.40 12.00 F. Banabed: Exploring health humanities from a global south perspective
- 12.00 12.20 E. Rosakebia: Balancing care and creation: The role of poetry in caregiving and emotional resilience
- 12.20 12.40 F. Dalpane: Making art as bravery: A virtue-ethics perspective
- 12.40 13.00 Y. Istileulova: Planetary health through arts: De rerum natura and Earth's first music in cosmic harmony

Section 12: Bridging Science, Health and Arts II (Chair: V. Vidrih Perko)

- 11.00 11.20 R.S. Thomas: Combining an art therapy-trauma protocol and sensory motor art therapy to support a woman with breast cancer
- 11.20 11.40 E. Hribernik: Man on the stage of life introduction to an Italian baroque opera
- 11.40 12.00 A.L. Mastruzzo: The performer as a multi-expressive artist in Argentina's current flute experimentation
- 12.00 12.20 A. Karboski: Vladimir Stiftar and his views on the formation of the architectural and cultural landscape of the Eupatoria resort
- 12.20 12.40 N. Paliska: Mozart iconography
- 12.40 13.00 V. Vidrih Perko: Learning the immortal soul of humanism (Keynote lecture)

13.00 Break

14.00 Plenary lectures

- 14.00 14.30 V. Hlavackova Pospichalova: Extracellular vesicles in ovarian cancer
- 14.30 15.00 N. Gov: Magnets, Ants and Humans. The physics of collective transport by ants
- 15.00 15.30 H. Murto: Organic farming
- 15.30 16.00 E. Mihajlović: Claudio Monteverdi and so called seconda pratica or how he brings real human feelings into music

16.00 Student zone

17.00 Closing of the symposium







Satellite event: Concert at the Betteto Hall, Academy of Music, Casino Building 10.1.2025 at 18.00 Tentative Program:

Classical music:

C. Franck: Petit offertoire. Organ: Yelena Istileulova

C. Monteverdi/G.F. Busenello: Prologue from opera Coronation of Poppaea. Sopranos: Fortuna: Ronja Prapotnik, Virtu: Eva Kokot, Amore: Brina Vukovič, Harpsichord: E. Mihajlović, Organ positive: Branko Rezić

A. Aljabjev A: Nightingale. Flute: Anita Prelovšek, Piano: Elena Startseva Somun

S. Rachmaninov: Vocalise. Violin: Vasilij Meljnikov, Piano: Lara Oprešnik

C. Monteverdi/G.F. Busenello: Regina disprezzata from opera Coronation of Poppaea. Soprano: Ottavia: AlessandraTessaro, Harpsichord: E. Mihajlović, Organ positive: Branko Rezić

S. Rachmaninov: Etude Tableaux Op 33 No 3. Piano: Lara Oprešnik

G. Brun: Romance. Flute: Anita Prelovšek, Piano: Elena Startseva Somun

C. Monteverdi/G.F. Busenello: Duetto Demigella and Valetto from opera Coronation of Poppaea. Sopranos: Demigella: Nives Hadžić, Valetto: Eva Kokot, Harpsichord: E. Mihajlović, Organ positive: Branko Rezić

W.A. Mozart: Alla turca from Sonate in A major. Piano: Denis Luin

F. Chopin: Polonaise G sharp minor. Piano: Matic Bogataj

G. Ipavec/A. Čopi - Simon Gregorčič: Mountain flower: Chorus Studenec Poetry by Ifigenia Simonović

Ifigenija Simonović is writing poetry, essays, book reviews, translating, and painting various objects for more than fifty years. She is the author of ten poetry collections, three books of essays, three books for children, and is also known as a publisher of eight books of poetry by Vitomil Zupan. In 2009, she received the Rožanec award. Between 2017 and 2021, she was the president of the Slovenian PEN Center. Her poetry is often dark, but love persistently shines through the cracks that are drawn on her path by real-time experiences.

H. Lavrenčič/Anonymus: The coque has sung: Chorus Studenec

E. Adamič/O. Župančič: Evening song: Chorus Studenec

Chorus Studenec Pivka, led by Irena Rep, is composed of experienced singers. In its 22 seasons, various genres were performed but the singers prefer to sing arrangements of folk songs. They have around 30 concerts annually, home and abroad, most importantly, the visits to Slovenian societies throughout Europe.

Contemporary music

A. von Sultanova/Titus Lucrecius: De rerum natura. Piano, Voice: Aleona von Sultanova

S Kralj: Topology. Piano: Samo Kralj

H. Mancini: Pink Panther. Trombone: Emil Somun, Piano: Elena Startseva Somun

A. von Sultanova/A. von Sultanova: For Samuel Gmelin - water, water. Piano, Voice: Aleona von Sultanova

J. Rae: Sonatine (Aquarelle, Notturne, Firedance). Flute: Anita Prelovšek, Piano: Elena Startseva Somun

A. Schnittke: Suite old style. Violin: Branko Brezavšček, Piano: Elena Startseva Somun

L. Oprešnik: Fugue. Piano: Lara Oprešnik

B. Kobal: Dic verbo. Soprano: Kaya Tokuhisa, Organ: Jana Jamšek

D. Zupanič Turković: Cantique de Baruch Spinoza from Mass in E minor. Organ: Aleona von Sultanova, Percussion: Bojan Ilievski, Piano: Lara Oprešnik, Flute: Anita Prelovšek, Voice: Veronika Kralj-Iglič

Satellite event: Recital of the organist Roberta Schmid at the Church of Assumption, Tromostovje 14.1.2025 at 20.00.

Program:

D. Buxtehude: Passacaglia in D minor BuxWV 161

J. S. Bach: Choral Das alte Jahr vergangen ist BWV 614

J. S. Bach: Choral Erbarme dich mein Gott BWV 721

A.G. Ritter: Sonate no. 2 in E minor op.19

G. Mushel: Toccata

J. Rheinberger: Passacaglia

L. Vierne: Carillon de Westminster







Editorial

12th Socratic Lectures symposium took place online on January 11, 2025. It featured five plenary lectures, 12 scientific sessions, a poster session and a newly introduced session called "The student zone".

As we received more than 40 papers, The Proceedings of the 12th Socratic Lectures is organized in three parts. Part I contains mainly papers from the medical fields, Part II contains mainly papers from the natural and social sciences and Part III contains standard operating procedures of a pipeline for assessment of extracellular particles. The pipeline was designed within the project Nanostructurome. Socratic lectures thus present an important meeting point for the participants of various projects and for the publication of scientific results, opinions, repositories, and any kind of documents that are useful in the management of the projects. But in the first place, the 12th Socratic lectures were a meeting point for scientists, artists and friends from many different fields. We were able to enjoy the fruit of the fields which were kindly provided to us by the participants of the events for which we remain forever thankful.

With the participation of world top scientists from the fields and integration of science in international artistic production, Socratic lectures strive to present scientific and artistic excellence to the students and involve them in the creation of science and art. We conclude with a big thanks to all the participants who in the spirit of Socrates donated their contributions and to all those who made the events possible. Kindly invited to the next Socratic lectures.

Veronika Kralj-Iglič, Anna Romolo and Yelena Istileulova







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Reflection/Review

Microalgal Technologies: Treatment Strategy for Contaminants of Emerging Concern

Kranjc Požar Ajda^{1,*}, Atanasova Nataša¹, Griessler Bulc Tjaša², Istenič Darja^{1,2}

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Abstract:

The increasing presence of contaminants of emerging concern (CEC) in aquatic environments poses significant risks to ecosystems and human health. Traditional wastewater treatment plants (WWTPs) are not equipped to efficiently remove these pollutants, necessitating the development of advanced treatment methods. This article explores the potential of microalgal technologies as a sustainable and cost-effective strategy for CEC removal. Microalgae, such as Chlorella sp., Chlamydomonas sp., and Scenedesmus sp., demonstrate resilience to high concentrations of CEC and offer multiple mechanisms for pollutant removal, including biosorption, bioaccumulation, biodegradation, photodegradation, and volatilization. The article discusses various cultivation methods, including planktonic, immobilized, and mixed biofilm cultures, and highlights the advantages and challenges associated with each. Further research is needed to optimize microalgal cultivation conditions and integration into wastewater treatment processes in order to achieve high CEC removal efficiency. By addressing these challenges, microalgal technologies can enhance the efficiency and sustainability of wastewater treatment systems, contributing to improved water quality and environmental protection.

Keywords: Microalgae; wastewater treatment plants; quaternary treatment; *Chlorella* sp.; cultivation methods





1. Introduction

Traditionally, water quality management has concentrated on addressing key pollutants such as nutrients, organic matter, suspended solids, heavy metals, and human pathogens (Pal et al., 2014); however, in recent years, growing attention has been directed toward contaminants of emerging concern (CEC). These contaminants pose significant risks to aquatic ecosystems and human health, sparking widespread concern and prompting further research (Tran et al., 2018).

CEC encompass both naturally occurring and artificially synthesized chemicals that are associated with known or uncertain adverse effects on human health and the environment (Ahmed et al., 2021). These substances are characterized by their relative polarity, resistance to natural biodegradation, and potential for bioaccumulation (Kumar et al., 2022). CEC include a wide array of compounds, such as pharmaceuticals and personal care products, endocrine-disrupting compounds, perfluorinated compounds, surfactants, gasoline additives, disinfection by-products, algal and cyanobacterial toxins, organometallic compounds, brominated and organophosphate flame retardants, plasticizers, and nanoparticles.

Most CEC enter waterways through human-mediated routes, including the direct discharge of raw or treated municipal wastewater, landfill leachate, urban and rural runoff, groundwater infiltration, and industrial waste discharge (Tran et al., 2018). Among these, municipal wastewater treatment plants (WWTPs) are the most significant contributors to the continuous discharge of CEC into aquatic environments. The number of specific CEC detected in wastewater discharge ranges from 25 to 200 compounds, a figure likely to increase with advancements in analytical techniques and the industrial use of newly synthesized compounds (Norvill et al., 2016; Tran et al., 2018; Villar-Navarro et al., 2018; Wang et al., 2016).

Conventional WWTPs were originally designed to remove nutrients and organic matter and are not equipped to efficiently eliminate CEC (Lopez et al., 2022). These traditional methods, including processes like waste stabilization ponds, up-flow anaerobic sludge blankets, activated sludge anoxic-aerobic process and sequencing batch reactors, achieve CEC removal rates of only 40–68% (Dubey et al., 2023).

To address these limitations, several advanced technologies have been introduced to enhance the removal of CEC in WWTP effluents, such as chemical precipitation, advanced oxidation process, reverse osmosis, ion exchange, adsorption-based technologies and electroflocculation. However, these methods often incur high costs due to the need for specialized materials like photocatalysts and synthesized adsorbents, as well as fluctuating factors such as material availability, labor, electricity, and recycling costs (Patel et al., 2019). Given these challenges, the development of cost-effective and sustainable treatment methods for CEC removal is imperative. Integrating microalgae into wastewater treatment systems presents a promising, eco-friendly approach (Kumar et al., 2022), with high potential of resources recovery and reuse (e.g. algal biomass) The goal of this paper is to provide a critical review of algal-based technologies for removal of CEC in order to assess their potential and indicate the direction of further research to address their challenges.

2. Advantages of microalgal - based bioremediation

The algal-based technologies have emerged as a promising approach for wastewater treatment, offering numerous advantages. Research has shown that various microalgae species, such as *Chlorella* sp., *Chlamydomonas* sp., *Scenedesmus* sp., and *Picocystis* sp., exhibit resistance to a wide range of CEC even at high concentrations. This resilience makes them valuable candidates for removing pollutants from wastewater with elevated contamination levels (Hom-Diaz et al., 2015; Kumar et al., 2022; Prosenc et al., 2021; Quan et al., 2023). Microalgae can efficiently assimilate nutrients present in wastewater, performing bioremediation while significantly reducing the expenses associated with chemical inputs (Kumar et al., 2022). Additionally, this process is environmentally friendly, enhancing CO₂ sequestration and increasing dissolved oxygen (DO) levels in water through the photosynthetic activity of microalgal cells (Qi et al., 2021). Furthermore, microalgae inhibit the growth of harmful bacteria and bloom-forming organisms, thereby reducing microbial loads in aquatic systems (Zhang et al., 2022).







Beyond improving wastewater treatment efficiency, this strategy also supports wastewater valorization by generating valuable biomass. This biomass can be utilized to produce eco-friendly bioproducts, contributing to the circular bioeconomy and promoting sustainable resource management (Kumar et al., 2022).

3. Microalgal mechanisms for mitigating CEC

3.1 Biosorption/bioadsorption

Bioadsorption by microalgal cells occurs when contaminants are adsorbed either onto the cell wall components or onto organic substances, such as extracellular polysaccharides (EPS), excreted by the cells into the surrounding environment (Saavedra et al., 2018). This process is non-selective and reversible, allowing various types of CEC to be simultaneously captured on the microalgal cell surfaces (Prosenc et al., 2021). As a passive, non-metabolic interaction, bioadsorption involves the binding of CEC to the negatively charged cell surfaces, which include both the cell wall and secreted substances (collectively termed cell surfaces). These surfaces exhibit a chemical affinity for positively charged contaminants (Xiong et al., 2017).

The ability of CEC to adsorb onto microalgal surfaces depends on its chemical structure. Hydrophobic, cationic contaminants are drawn to the microalgal surface through electrostatic interactions, while hydrophilic CEC tend to be repelled (Xiong et al., 2017). Once a CEC reaches the cell surface, the extent of adsorption is determined by the surface area and chemical composition of the microalgal cells (Norvill et al., 2016). Microalgal cell surfaces contain a variety of functional groups, including carboxyl, hydroxyl and sulfate groups, each with varying affinities for organic and inorganic compounds (Çetinkaya Dönmez et al., 1999).

Bioadsorption involves several chemical processes at the cell surface, including adsorption reactions, ion exchange, surface complexation reactions, chelation, and microprecipitation (Çetinkaya Dönmez et al., 1999). The rate and thermodynamics of the adsorption process are influenced by the physico-chemical properties of the surrounding environment, such as temperature, redox potential and pH.

As bioadsorption is a non-metabolic process, contaminants can bind to both living and non-living microalgal cells, as most cell receptors for CEC remain functional even after the cell has died (Choi & Lee, 2015).

3.2 Bioaccumulation

Bioaccumulation is an active, energy-driven process by which CEC are stored in their original form within microalgal cells. This occurs through the crossing of the cell membrane, where CEC bind to intracellular proteins and other compounds. Unlike bioadsorption, bioaccumulation is slower and occurs exclusively in living cells (Xiong et al., 2018).

Microalgal cells can uptake CEC through three primary mechanisms: passive diffusion, passive-facilitated diffusion and energy-dependent/active uptake across the cell membrane.

In passive diffusion, CEC move across the membrane from areas of high external concentration to areas of low internal concentration without requiring energy expenditure from the cell. This process is facilitated by the hydrophobic nature of the cell membrane, which allows low molecular weight, non-polar, lipid-soluble contaminants to diffuse through. However, polar molecules, high molecular weight compounds, and ions cannot pass through the membrane passively (Xiong et al., 2018).

Passive-facilitated diffusion refers to the process by which CEC cross the cell membrane with the assistance of transporter proteins. These proteins facilitate the influx of polar molecules into the cell (Wilde & Benemann, 1993).

The final mechanism, active transport, involves the movement of CEC across the cell membrane, a process that requires energy expenditure by the cell. In active transport, the compound often moves against a concentration gradient, though this is not always the case Regardless of the mechanism, bio-uptake is influenced by various factors in the surrounding environment. These include the physico-chemical conditions, such as temperature and





pH, the metabolic state and health of the cell, and the presence of any metabolic inhibitors (Wilde & Benemann, 1993).

3.3 Biodegradation (biotransformation)

Biodegradation of CEC by microalgae is one of the most promising pathways for the remediation of these pollutants. Unlike bioadsorption or bioaccumulation, which function primarily as biological filters to concentrate and remove CEC from the surrounding aqueous solution, biodegradation involves the metabolic transformation of complex compounds into simpler, less harmful molecules. This process offers an advantage over bioadsorption and bioaccumulation by addressing the issue of managing CEC-laden microalgal biomass generated during these treatments (Tiwari et al., 2017).

Microalgal biodegradation can occur through two primary mechanisms: metabolic degradation and co-metabolism. In metabolic degradation, the CEC serves as a carbon source or electron donor/acceptor for the microalga, enabling its breakdown, while in co-metabolism, the CEC is degraded by enzymes that catalyze the degradation of other substrates present in the environment (Tiwari et al., 2017).

The process of biodegradation can occur either intracellularly, extracellularly, or through a combination of both. In the intracellular process, the CEC is bio-uptaken by the microalga. Extracellular degradation, on the other hand, occurs when microalgae secrete enzymes into the extracellular polymeric substances (EPS), which act as an external digestive system. The EPS can also serve as a surfactant and emulsifier, increasing the bioavailability of CEC and facilitating their subsequent uptake by the cell. In combination process, initial degradation typically occurs outside the cell, with the breakdown products being further degraded inside the cell (Tiwari et al., 2017; Xiong et al., 2018).

3.4 Photodegradation and volatilization

Even if a CEC cannot be bioremediated by microalgae through bioadsorption, bioaccumulation, or biodegradation, microalgae may still contribute to its successful remediation. Two processes—photodegradation and volatilization—can be enhanced by the presence of microalgae or the microalgal treatment system itself (Abo et al., 2016).

Photodegradation of a CEC can occur through two mechanisms: photolysis and photooxidative degradation. Photolysis happens when a contaminant absorbs light, causing direct chemical alteration and subsequent degradation of the compound. In contrast, photooxidative degradation involves the breakdown of the contaminant through interactions with hydroxyl radicals or other oxidants, which are formed from reactions with dissolved organic molecules or nitrate under light exposure (Abo et al., 2016).

Light exposure is essential for photodegradation, but in microalgal treatment systems, light is often absorbed or scattered by the cells, reducing its availability. This can slow down photodegradation. However, the design and operation of these systems can be adjusted to improve light penetration. Microalgae release dissolved organic molecules (DOM) into the water, which can enhance photodegradation. DOM includes compounds like hydrophilic organic acids and humic substances. These molecules can facilitate photodegradation through various mechanisms, such as producing hydroxyl radicals or participating in redox reactions. This process helps break down CEC. (Norvill et al., 2016). For example, studies have shown that microalgal DOM can aid in the removal of pharmaceuticals like ibuprofen through indirect photodegradation (de Wilt et al., 2016).

Volatilization refers to the loss of volatile organic compounds from the liquid phase into the atmosphere. This process depends on the physico-chemical properties of the CEC, such as the Henry's law constant, and the operating conditions of the treatment system, including factors like aeration or agitation rates, temperature, and atmospheric pressure (Tran et al., 2018). In microalgal-based treatment systems, high aeration rates provided by mixing devices (such as paddlewheels, bubble lift columns, and stirrers), along with increased sunlight and higher temperatures compared to conventional wastewater treatment systems, can enhance the removal of volatile CEC (Matamoros et al., 2015).





4. Efficiency of microalgal CEC removal

Several species of microalgae have been tested for their ability to remove CEC, and the choice of species can significantly impact the efficiency of the bioremediation process (Wang et al., 2016).

Microalgae-based systems with optimized conditions, such as high aeration rates, ample sunlight exposure and suitable operating temperatures (whether during warm or cold seasons), can significantly enhance the removal of volatile CEC, including musk fragrances like tonalide and galaxolide (Matamoros et al., 2015).

Among the most frequently studied species for CEC removal are *Chlorella* sp., *Chlamydomonas* sp., and *Scenedesmus* sp., which have been extensively tested in proof-of-concept studies. These species are particularly valued for their robustness and adaptability under stressful environmental conditions. Despite the high diversity of microalgae species, only a few have been sufficiently studied for their potential in bioremediating CEC (Maryjoseph & Ketheesan, 2020). Appendix 1 presents selected CEC and microalgal cultures utilized in various studies, along with their transfer pathways.

Microalgae can be cultivated either individually or combined in both open and closed systems. These cultivation methods are gaining recognition for their potential in secondary and/or tertiary treatment in wastewater treatment plants (Norvill et al., 2016) and for the removal of CEC (Chi et al., 2019; Kurade et al., 2016). The following sections describe three main cultivation methods: planktonic cultures, immobilized cultures and mixed biofilm cultures.

4.1 Planktonic cultures

Open systems are commonly categorized as stabilization ponds and high-rate algal ponds (HRAPs) (Norvill et al., 2016). The main advantages of open systems include their simplicity in construction and operation, along with low operating costs. However, these systems face several challenges, such as susceptibility to contamination by other microorganisms, loss of CO₂ to the atmosphere, and the requirement for large land areas (De Godos et al., 2012). Additionally, their reliance on environmental factors—such as temperature and light—can limit the efficiency of the cultivation process (Matamoros et al., 2016; Matamoros et al., 2015).

Closed photobioreactors (PBRs) offer an alternative to open ponds and come in various configurations, such as tubular, bubble column, air-lift, and flat panel (Torgal, 2016). Unlike open systems, PBRs reduce microbial contamination and allow for the control of variables like pH, temperature, light, and CO₂ concentration (Huang et al., 2017). However, in both open and closed systems, nutrient composition and biological contamination can impact microalgal biomass production and the removal of contaminants of emerging concern (CEC) (Procházková et al., 2014).

4.2 Immobilized cultures

Cell immobilization refers to the restriction of cell mobility through entrapment or attachment to an organic or inorganic water-insoluble solid support (Bouabidi et al., 2019). Common immobilization methods include adsorption onto surfaces, aggregation, encapsulation in semi-permeable membranes, covalent bonding, and entrapment within porous or fibrous polymers. The inherent ability of microorganisms to attach to surfaces often leads to the formation of biofilms, contributing to the immobilization process (Gonçalves et al., 2017).

Immobilized systems offer several advantages, such as maintaining high microalgal concentrations and activity, improving the efficiency of CEC removal, simplifying harvesting, and enhancing resilience to stress, including exposure to toxic contaminants (Zhang et al., 2016). Immobilized microalgae have been successfully used to remove nitrogen, phosphate, and heavy metals from wastewater (Shen et al., 2009). More recently, they have also been employed to remove CEC from wastewater.

4.3 Mixed cultures

Consortia of microorganisms can exhibit greater robustness and stability in response to sudden environmental conditions, as well as protection against invasion by predators and competitor species, compared to individual species (Subashchandrabose et al., 2011). The







presence of bacteria can benefit microalgae growth by removing oxygen and supplying carbon dioxide, altering the surrounding environment and providing essential nutrients and growth factors, such as chelators and phytohormones (Wang et al., 2016).

Several studies have demonstrated enhanced microalgal growth when associated with bacteria, with mutual benefits arising from the excretion of bacterial growth-promoting factors, including microalgal extracellular polymeric substances (EPS) (Cho et al., 2015; Hernandez et al., 2009).

5. Challenges and prospects of microalgal-based removal of CEC

Recent advances in microalgal-based bioremediation of CEC present promising, sustainable alternatives to traditional physicochemical processes in wastewater treatment. However, several significant challenges must be addressed before microalgae can be fully implemented as an eco-friendly and effective bioremediation platform for CEC in wastewater. One of the key obstacles is the relatively low performance of microalgae, as different species exhibit varying levels of adaptability and tolerance to CEC toxicity. Future research should focus on isolating and selecting microalgae strains with high growth rates and improved contaminant removal efficiency, as well as enhancing strain performance through methods such as random mutagenesis, genetic engineering, and adaptive laboratory evolution (Arora & Philippidis, 2021).

Another challenge lies in the potential use of microalgal biomass after CEC treatment for food, feed, or biofertilizer applications. Bioaccumulation of CEC in microalgae may hinder such uses, as contaminants could re-enter the food chain and affect the environment (Hena et al., 2020). Therefore, further studies are needed to ensure the safe and sustainable use of microalgal biomass following CEC treatment. Recent research has shown that laccase enzymes can effectively degrade contaminants like Bisphenol A from growth media (Clark et al., 2022). Toxicity in wastewater can also hinder the growth of certain microalgal species, particularly in large-scale real-world applications. The toxicity of wastewater depends on its source and composition (Pittman et al., 2011). Acclimation or adaptation of microalgae to these conditions is a key area of study. Genetic adaptation has been shown to enable microalgae to tolerate high doses of contaminants such as antibiotics, herbicides, and mine waste (García-Balboa et al., 2013). Additionally, microalgae have demonstrated the ability to acclimate to sub-lethal stresses like heavy metals, singlet oxygen, salinity, and high light conditions, often leading to the production of toxic-degrading enzymes (Osundeko et al., 2014; Singh et al., 2020).

A major bottleneck in the application of microalgae for CEC treatment is the lack of suitable cultivation facilities. Conventional systems like open ponds, raceway systems, and high-rate algal ponds are cost-effective and scalable but face limitations, including inefficient light utilization, contamination from predators and heterotrophs, and the need for large land areas (Cheng et al., 2021). To overcome these limitations, closed photobioreactors such as vertical column, tubular, flat plate, membrane, and biofilm-based systems have been developed. However, these systems are more costly to install and maintain, making them less suitable for large-scale, cost-effective wastewater treatment (Sathinathan et al., 2023).

Therefore, an engineering breakthrough is needed to develop economical, efficient, and practical in situ treatment systems for microalgal-based CEC wastewater treatment. Furthermore, most studies on microalgal-based CEC bioremediation focus on synthetic water or culture media under laboratory conditions, with only a few studies addressing natural wastewater or surface water. To ensure the real-world applicability of this technology, further research is required to explore the practical use of microalgae for CEC removal in industrial-scale wastewater treatment systems (Kumar & Shukla, 2023).

Also, further research is essential to determine the most effective stage of wastewater treatment for integrating microalgal cultures. Specifically, studies should focus on identifying whether microalgal cultures are most efficient in the secondary, tertiary, or quaternary stages of treatment. Additionally, research should explore the optimal forms and configurations of microalgal cultures, such as planktonic, immobilized, or mixed biofilm cultures, to maximize the removal of CEC. Understanding these factors will help optimize the design and operation of wastewater treatment systems, enhancing their efficiency and sustainability.





6. Conclusions

In conclusion, microalgal technologies present a promising and sustainable approach for the treatment of CEC in wastewater. The integration of microalgae into wastewater treatment systems offers numerous advantages, including bio- or photo- degradation of CEC as opposed to just displacement from water, cost-effectiveness, environmental benefits, and the potential for wastewater valorization. Microalgae can effectively remove a wide range of CEC through mechanisms such as biosorption, bioaccumulation, biodegradation, photodegradation, and volatilization.

However, several challenges must be addressed to fully realize the potential of microalgal-based bioremediation. These include optimizing the selection and enhancement of microalgal strains, efficient integration into existing wastewater treatment systems, ensuring the safe use of microalgal biomass post-treatment, and developing economical and efficient cultivation systems. Further research is essential to determine the most effective stage of wastewater treatment for integrating microalgal cultures and to explore the optimal forms and configurations of these cultures.

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Appendix A Table A1. Selected CEC and microalgal cultures with removal efficiency and their transfer pathways.

Appendix 1

Table 1A. Selected CECs and microalgal cultures with removal efficiency and their transfer pathways

	O .		•	1 ,
Contaminants	Microalgal species	Removal efficiency [%]	Mechanisms (pathways)	Reference
17 α-Boldenone	Mixed consortia	82	Not determined	(Zhou et al., 2014)
17 α -Estradiol	Scenedesmus dimorphus	85	Biodegradation	(Zhang et al., 2014)
17α - Ethynylestradiol	Chlamydomonas reinhardtii	100	Bioadsorption, bio-uptake,	(Hom-Diaz et al., 2015)
	Selenastrum capricornutum	60-95	biodegradation	
	Nannochloris sp.	60		(Bai & Acharya, 2019)
	Desmodesmus subspicatus	68		(Maes et al. <i>,</i> 2014)
17 β-Boldenone	Mixed consortia	75-86	Not determined	(Zhou et al., 2014)
17 β-Estradiol	Chlamydomonas reinhardtii	100	Bioaccumulatio n,	(Hom-Diaz et al., 2015)







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	Desmodesmus subspicatus	95	bioadsorption, biodegradation	
	Selenastrum capricornutum	88		
	Nannochloris sp.	60		(Bai & Acharya, 2019)
	Chlorella	>92		(Huang et al., 2016)
7-amino	Chlorella sp.	100	Photodegradati	(Guo et al.,
cephalosporanie acid	Chlamydomonas sp.	100	on, bioadsorption	2016)
	Mychonastes sp.	100		
Acetaminophen	Mixed consortia	99	Not determined	(Matamoros et al., 2016)
Alprazolam	Mixed consortia	87	Not determined	(Hom-Diaz et al., 2017a, 2017b)
Amitriptyline	Chlorella sorokiniana	68	Bioadsorption	(Gojkovic et al., 2019)
	Chlorella vulgaris	42		
	Chlorella saccharophila	92		
	Coelastrella sp.	60		
	Coelastrum astroideum	68		
	Desmodesmus sp.	37-47		
	Scenedesmus sp.	85		
	Scenedesmus obliquus	69		
Amoxicillin	Chlorella pyrenoidosa	77	Not determined	(Li et al., 2015)
	Microcystis aeruginosa	18-31		(Liu et al., 2015)
Atenolol	Mixed consortia	85-98	Not determined	(Hom-Diaz et al., 2017a, 2017b)
Azithromycin	Mixed consortia	89	Not determined	(Hom-Diaz et al., 2017a, 2017b)







Biperiden	Chlorella	35	Bioadsorption	(Gojkovic et al.,
Diperiden	sorokiniana	33	Diodasorption	2019)
	Chlorella vulgaris	93		,
	Chlorella	89		
	saccharophila			
	Coelastrum	9		
	astroideum			
	Desmodesmus	41-71		
	sp.			
	Scenedesmus sp.	53		
	Scenedesmus	48		
	obliquus			
Bisphenol A	Chlorella	43	Bio-uptake,	(Fu et al., 2023)
	pyrenoidosa		biodegradation	
	Desmodesmus	26		(Wang et al.,
	sp. WR1			2017)
	Chlamydomonas	24		(Ji et al., 2014)
	mexicana			
	Chlorella vulgaris	24		
Bupropion	Chlorella	60	Bioadsorption	(Gojkovic et al.,
	sorokiniana			2019)
	Chlorella vulgaris	82		
	Chlorella	88		
	saccharophila			
	Coelastrella sp.	89		
	Coelastrum	94		
	astroideum			
	Desmodesmus	86-90		
	sp.			
	Scenedesmus sp.	70		
	Scenedesmus	95		
	obliquus			
Caffeine	Mixed consortia	99	Biodegradation	(Matamoros et
				al., 2016)
	Mixed consortia	26-81		(Gojkovic et al.,
				2019)
Carbamazepine	Mixed consortia	4-15	Bioadsorption,	(Zhou et al.,
			biodegradation	2014)
	Mixed consortia	20		(Matamoros et
				al., 2016)







	Chlamydomonas mexicana	35		(Xiong et al., 2017b)
	Scenedesmus obliquus	35		,
	Chlorella	10-30		(de Wilt et al.,
	sorokiniana	10 00		2016)
	Nannochloris sp.	20		(Bai & Acharya, 2016)
	Desmodesmus sp.	71		(Gojkovic et al., 2019)
Carbendazim	Mixed consortia	14-30	Not determined	(Zhou et al., 2014)
Cefradine	Chlorella pyrenoidosa	23	Not determined	(Li et al., 2015)
Cefradine	Chlorella pyrenoidosa	76	Not determined	(Chen et al., 2015)
Ciprofloxacin	Chlorella vulgaris	89,9	Biodegradation, bioadsorption, photodegradati on	(Al-Mashhadani and Al- Mashhadani, 2023)
	Chlamydomonas sp.Tai-03	100		(Xie et al., 2020)
	Mixed consortia	74-79		(Zhou et al., 2014)
	Mixed consortia	20-30		(Hom-Diaz et al., 2017a, 2017b)
	Chlamydomonas mexicana	13-56		(Xiong et al., 2017b)
	Nannochloris sp.	100		(Bai & Acharya, 2016)
	Dictyosphaerium sp.	11		(Gentili & Fick, 2017)
Clarithromycin	Mixed consortia	100	Not determined	(Zhou et al., 2014)
Climbazole	Mixed consortia	30-70	Biodegradation	(Zhou et al., 2014)
	Scenedesmus obliquus	88		(Pan et al., 2018)
Clofibric acid	Mixed consortia	0-30	Not determined	(Zhou et al., 2014)







Clomipramine	Chlorella sorokiniana	96	Bioadsorption	(Gojkovic et al., 2019)
	Chlorella vulgaris	100		2017)
	Chlorella	100		
	saccharophila			
	Coelastrella sp.	34		
	Desmodesmus	29-42		
	sp.			
	Scenedesmus sp.	73		
	Scenedesmus obliquus	78		
Codeine	Chlorella sorokiniana	50	Biodegradation, photodegradati	(Gojkovic et al., 2019)
	Chlorella vulgaris	57	on	
	Chlorella saccharophila	42		
	Coelastrella sp.	46		
	Coelastrum astroideum	72		
	Desmodesmus sp.	37-80		
	Scenedesmus sp.	33		
	Scenedesmus obliquus	59		
Diazinon	Chlorella vulgaris	94	Biodegradation	(Kurade et al., 2016)
Diclofenac	Picocystis sp. CINS 23	>90	Bioadsorption, bioaccumulatio	(Ali et al., 2022)
	Mixed consortia	92	n, biodegradation	(Matamoros et al., 2016)
	Chlorella sorokiniana	40-60		(de Wilt et al., 2016)
	Chlorella sorokiniana	30		(Escapa et al., 2015)
	Chlorella vulgaris	21		,
	Mixed consortia	55		(Villar-Navarro et al., 2018)
Diltiazem	Mixed consortia	72-77	Not determined	(Hom-Diaz et al., 2017a, 2017b)







Di-n-butyl Chaetoceros 22,5 Biodegradation (Chi et al., 2019) phthalate muelleri Cylindrotheca 91,4
Cylindrotheca 91,4
closterium
Diphenhydrami Chlorella 73 Biodegradation (Gojkovic et al
ne sorokiniana 2019)
Chlorella vulgaris 98
Chlorella 93
saccharophila
Coelastrella sp. 87
Coelastrum 87
astroideum
Desmodesmus 88-92
sp.
Scenedesmus sp. 86
Scenedesmus 85
obliquus
Enrofloxacin Mixed consortia 75-77 Not determined (Zhou et al.,
2014)
Erythromycin Mixed consortia 63-86 Not determined (Zhou et al.,
2014)
Mixed consortia 85 (Hom-Diaz et
al., 2017a,
2017b)
Estriol Scenedesmus 85 Biodegradation (Zhang et al.,
dimorphus 2014)
Estrone Mixed consortia 85 Biodegradation (Zhou et al.,
Scenedesmus 85 2014)
dimorphus
Flecainide Chlorella 71 Photodegradati (Gojkovic et al
sorokiniana on 2019)
Chlorella vulgaris 100
Chlorella 100
saccharophila
Coelastrella sp. 52
Coelastrum 66
astroideum
Desmodesmus 72-96
sp.
Scenedesmus sp. 40





	Scenedesmus obliquus	93		
Fluconazol	Desmodesmus sp.	33	Bioadsorption	(Gojkovic et al., 2019)
Fluoxastrobin	Synechococcus sp.	Not determine d	Bioadsorption	(Stravs et al., 2017)
Fluxonazole	Mixed consortia	25	Not determined	(Zhou et al., 2014)
Hydrochlorothi azide	Mixed consortia	44-84	Not determined	(Hom-Diaz et al., 2017a, 2017b)
Hydroxyzine	Chlorella sorokiniana	76	Biodegradation	(Gojkovic et al., 2019)
	Chlorella vulgaris	93		
	Chlorella saccharophila	93		
	Coelastrella sp.	80		
	Coelastrum astroideum	96		
	Desmodesmus sp.	87-100		
	Scenedesmus sp.	73		
	Scenedesmus obliquus	95		
Ibuprofen	Mixed consortia	99	Bio-uptake, biodegradation	(Matamoros et al., 2016)
	Mixed consortia	98		(Hom-Diaz et al., 2017a, 2017b)
	Chlorella sorokiniana	100		(de Wilt et al., 2016)
	Nannochloris sp.	40		(Bai and Acharya, 2016)
	Navicula sp.	60		(Ding et al., 2017)
Ketoprofen	Mixed consortia	36-85	Not determined	(Hom-Diaz et al., 2017a, 2017b)
Kresoxim- methyl	Mixed consortia	Not determine d	Not determined	(Stravs et al., 2017)







Levofloxacin	Scenedesmus	93,4	Biodegradation,	(Xiong et al.,
	obliquus	10.00	bioaccumulatio	2017a)
	Chlorella vulgaris	10-90	n, bioadsorption	
Lincomycin	Mixed consortia	80	Not determined	(Zhou et al., 2014)
Lorazepam	Mixed consortia	30-60	Not determined	(de Wilt et al., 2016)
Memantine	Chlorella sorokiniana	87	Bioadsorption, biodegradation	(Gojkovic et al., 2019)
	Chlorella vulgaris	100		,
	Chlorella	100		
	saccharophila			
	Coelastrella sp.	78		
	Coelastrum astroideum	73		
	Desmodesmus sp.	44-86		
	Scenedesmus sp.	92		
	Scenedesmus obliquus	86		
Metoprolol	Chlorella	100	Biodegradation	(de Wilt et al.,
1	sorokiniana			2016)
	Dictyosphaerium	99		(Gentili & Fick,
	sp.			2017)
	Chlamydomonas	99		(Stravs et al.,
	reinhardtii			2017)
Metronidazole	Chlorella vulgaris	100	bioadsorption	(Hena et al., 2020)
Mitrazapine	Chlorella sorokiniana	63	Bioadsorption, biodegradation	(Gojkovic et al., 2019)
	Chlorella vulgaris	69		,
	Chlorella	80		
	saccharophila			
	Coelastrella sp.	70		
	Coelastrum	67		
	astroideum			
	Desmodesmus	55-85		
	sp.			
	Scenedesmus sp.	77		
	Scenedesmus obliquus	62		







Naproxen	Mixed consortia	89	Biodegradation	(Matamoros et
				al., 2016)
	Mixed consortia	10-70		(Hom-Diaz et
				al., 2017a,
				2017b)
Nonylphenol	Chlorella	48	biodegradation	(Feng et al.,
	pyrenoidosa			2022)
Norfloxacin	Mixed consortia	41-53	Not determined	(Zhou et al.,
				2014)
Norgestrel	Chlorella	60	Biodegradation	(Peng et al.,
	pyrenoidosa			2014)
	Scenedesmus	95		,
	obliquus			
Ofloxacin	Mixed consortia	43-52	Not determined	(Zhou et al.,
		10 02		2014)
	Mixed consortia	66		(Hom-Diaz et
	TVIIXEd CONSOTTIA	00		al., 2017a,
				2017b)
Orphenadrine	Chlorella	82	Bioadsorption	(Gojkovic et al.,
Orphenaumic	sorokiniana	02	bloadsorption	2019)
	Chlorella vulgaris	100		2017)
	Chlorella	98		
		98		
	saccharophila	70		
	Coelastrella sp.	78		
	Coelastrum	66		
	astroideum			
	Desmodesmus	75-82		
	sp.			
	Scenedesmus sp.	79		
	Scenedesmus	95		
	obliquus			
Oxytetracycline	Picocystis sp.	100	Bioadsorption,	(Ali et al., 2022)
	CINS 23		bioaccumulatio	
			n,	
			biodegradation	
Paracetamol	Mixed consortia	88-94	Biodegradation,	(Zhou et al.,
			photodegradati	2014)
	Chlorella	100	on	(de Wilt et al.,
	sorokiniana			2016)
	Chlorella	41-69		(Escapa et al.,
	sorokiniana			2015)
				,







Paroxetine	Mixed consortia	99	Not determined	(Hom-Diaz et al., 2017a, 2017b)
Phthalic acid esters	Chaetoceros muelleri	95,5	Biodegradation	(Chi et al., 2019)
	Cylindrotheca closterium	97,7		
Progesterone	Mixed consortia	83-87	Biodegradation	(Zhou et al., 2014)
	Chlorella pyrenoidosa	95		(Peng et al., 2014)
	Scenedesmus obliquus	95		
Roxithromycin	Mixed consortia	87-94	Not determined	(Zhou et al., 2014)
Salicylic acid	Mixed consortia	97	Bio-uptake, biodegradation	(Zhou et al., 2014)
	Nannochloris sp.	60		(Bai & Acharya, 2019)
	Mixed consortia	33		(Hom-Diaz et al., 2017a, 2017b)
	Chlorella sorokiniana	73		(Escapa et al., 2015)
	Mixed consortia	90		(Villar-Navarro et al., 2018)
	Chlorella sorokiniana	93-98		(Escapa et al., 2015)
	Chlorella vulgaris	25		(Escapa et al.,
	Scenedesmus obliquus	93		2017)
Salinomycin	Mixed consortia	71-79	Not determined	(Zhou et al., 2014)
Sulfadiazine	Mixed consortia	52-75	Not determined	(Zhou et al., 2014)
Sulfadimethoxin e	Mixed consortia	56-78	Not determined	(Zhou et al., 2014)
Sulfamethazine	Mixed consortia	18-48	Not determined	(Zhou et al., 2014)
Sulfamethoxazo le	Nannochloris sp.	32	Bioadsorption, biodegradation,	(Bai & Acharya, 2019)







	Nannochloris sp.	40	photodegradati on	(Bai & Acharya, 2016)
	Mixed consortia	0-18		(Stravs et al., 2017; Zhou et al., 2014)
Sulfapyridine	Mixed consortia	98	Not determined	(Zhou et al., 2014)
Testosterone	Mixed consortia	100	Not determined	(Zhou et al., 2014)
Tetrabromobisp henol A	Chlorella sphaericum & Scenedesmus quadricauda	98	Biodegradation	(Peng et al., 2014)
Tetracycline	Chlorella sp.	68	Bioadsorption, photodegradati on	(Suárez- Martínez et al., 2022)
	Scenedesmus quadricauda	48,84		(Daneshvar et al., 2018)
	Chlorella vulgaris	69		(De Godos et al., 2012)
Thiamethoxam	Chlorella sp. TXH	97,5	Biodegradation, bioadsorption, bioaccumulatio	(Quan et al., 2023)
Thiamphenicol	Chlorella sp. L38	77,7	Biodegradation,	(Song et al.,
•	Chlorella sp. UTEX1602	87,3	bioaccumulatio n, and biosorption	2020)
Tramadol	Dictyosphaerium sp.	57	Bio-uptake, biodegradation	(Gentili & Fick, 2017)
	Scenedesmus obliquus	91		(Ali et al., 2018)
	Chlorella vulgaris	51		(Gojkovic et al.,
	Desmodesmus sp.	14-45		2019)
Trichlorfon	Chlamydomonas reinhardtii	100	Biodegradation	(Wan et al., 2020)
Triclocarban	Mixed consortia	81-99	Not determined	(Zhou et al., 2014)
Triclosan	Mixed consortia	31-58		(Zhou et al., 2014)







Mixed consortia	95	Biodegradation,	(Matamoros et
		photodegradati	al., 2016)
Nannochloris sp.	100	on	(Xiong et al.,
			2017b)
Nannochloris sp.	72		(Bai & Acharya,
			2016)
Chlorella	77		(Wang et al.,
pyrenoidosa			2013)
Microcystis	46		(Wang et al.,
aeruginosa			2016)
_	100		(Rühmland et
1			al., 2015)
Chlorella	40	Bioadsorption	(Gojkovic et al.,
sorokiniana		1	2019)
Chlorella vulgaris	100		,
Chlorella	95		
saccharophila			
Coelastrum	54		
astroideum			
Desmodesmus	63-73		
-	49		
-	60		
•	0-37	Not determined	(Zhou et al.,
			2014)
Chlorella	40-60		(de Wilt et al.,
sorokiniana			2016)
	<4		(Gentili & Fick,
, · ·			2017)
	60		(Escapa et al.,
sorokiniana	- -		2015)
	75	Not determined	(Zhou et al.,
			2014)
	Nannochloris sp. Chlorella pyrenoidosa Microcystis aeruginosa Nannochloris sp. Chlorella sorokiniana Chlorella vulgaris Chlorella saccharophila Coelastrum astroideum Desmodesmus sp. Scenedesmus sp. Scenedesmus obliquus Mixed consortia Chlorella sorokiniana Chlorella	Nannochloris sp. 100 Nannochloris sp. 72 Chlorella 77 pyrenoidosa Microcystis 46 aeruginosa Nannochloris sp. 100 Chlorella 40 sorokiniana Chlorella yulgaris 100 Chlorella 95 saccharophila Coelastrum 54 astroideum Desmodesmus 63-73 sp. Scenedesmus sp. 49 Scenedesmus sp. 49 Scenedesmus 60 obliquus Mixed consortia 0-37 Chlorella 40-60 sorokiniana Dictyosphaerium <4 sp. Chlorella 60 sorokiniana	Nannochloris sp. 100 Nannochloris sp. 72 Chlorella 77 pyrenoidosa 46 aeruginosa 100 Chlorella 40 sorokiniana Chlorella 95 saccharophila Coelastrum astroideum Desmodesmus 63-73 sp. Scenedesmus 60 obliquus Mixed consortia 0-37 Chlorella 40-60 sorokiniana Dictyosphaerium 54 sp. Chlorella 60 sorokiniana







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Research

Surface Properties of Microalgal Biomass and Microplastics: Exploring Point of Zero-Charge and Contact Angle

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Abstract:

The surface properties of microalgal biomass and microplastics, focusing on the point of zero charge (PZC) and water contact angle (CA), were investigated. Microalgae, particularly *Chlorella vulgaris* and mixed microalgal consortia, were cultivated and analysed for their surface characteristics. Oven-dried and freeze-dried samples exhibited varying degrees of hydrophilicity, with freeze-dried microalgal consortia exhibiting more hydrophilic surface. The PZC values indicated a higher density of negative charges on the surface of *C. vulgaris* compared to the microalgal consortia biomass. Microplastics (MPs) from agricultural mulch films, including biodegradable and non-degradable types, were also examined. Results showed that naturally aged MPs exhibited more hydrophilic surfaces compared to their pristine counterparts. The PZC values of microplastics varied, with some showing neutral to slightly negative charges at environmental pH levels. The findings underscore the importance of surface characterization in understanding the sorption mechanisms of contaminants.

Keywords: Microplastics; Microalgal biomass; Surface properties; Point of zero-charge; Water contact angle; Sorption





1. Introduction

Sustainable development, as outlined by the United Nations (UN, 2015), can be enhanced with resource recovery and reuse in agricultural practices as well as reducing pollution in the environment due to insufficiently treated wastewater (WW). Nature-based solutions for wastewater treatment using microalgae are considered an efficient technology for the removal of contaminants of emerging concern (CECs) with simultaneous removal of nutrients from WW and incorporation into microalgal biomass (Maryjoseph & Ketheesan, 2020; Prosenc et al., 2021). Microalgae-based WW treatment process can take place in open ponds such as high-rate algal ponds (HRAP) or closed systems such as photobioreactors (PBRs). Either way, the end-products are treated WW and microalgal biomass. Treated WW can be used for irrigation, while microalgal biomass can be used to obtain high-value products: biostimulants to be used in agriculture, source of biopolymers, nutraceuticals (fatty acids, sterols, vitamins, minerals) and pigments (chlorophyll, phycocyanin, carotenoids) (Abdelfattah et al., 2023). Removed CECs can be either degraded or retained by microalgal biomass, including microplastics (MPs).

Increased yield in agricultural fields relies on the use of fertilizers (organic or mineral), CECs like pesticides and various materials, applied to the surface of the soil uncovered or covered with mulch (Alvarez et al., 2021; Bhuvaneswari et al., 2022; Hofmann et al., 2023). The most widely used material for mulch is non-biodegradable polyethylene (PE), which is lately being replaced with biodegradable and biobased polymers such as polybutylene co-adipate co-terephthalate (PBAT), thermoplastic starch (TPS), polylactic acid (PLA), polyhydroxy alkanoate (PHA) and their blends (Sintim et al., 2020). Mulch films, like all other plastics, can degrade in the environment when exposed to physical, chemical, and biological factors, forming MPs (i.e. particles in the size from 1 µm to 5 mm (ISO, 2023)). MPs are known contaminants in the soil, the source of which are overused agricultural materials (mulch, greenhouses), littering, wind deposition, irrigation with WW, composting, and fertilisation with organic fertilisers (Sa'adu & Farsang, 2023). MPs can alter the physical, chemical and microbial properties of soil and can be translocated between the environmental compartments (Dissanayake et al., 2022, Yadav et al., 2022).

Additionally, MPs were found to be an adsorbent and can act as a vector for other CECs such as antibiotics and pesticides, especially hydrophobic ones, and can enhance their persistence in soil (Šunta et al., 2020; Zhang et al., 2021; Wu et al., 2022). The sorption behaviour of CECs in the soil is dependent on the characteristics of the sorbate as well as on the characteristics of sorbents - in this case MPs and microalgal biomass (García-Delgado et al., 2020). There are many mechanisms involved in the sorption of contaminants on the surface of sorbents. Two of them are hydrophobic and electrostatic interactions that can be explained by the determination of the contact angle and point of zero charge of the sorbent, respectively.

The Contact angle (CA) is a measure of the wettability of a solid surface by a liquid, the most commonly used is water. CA is defined as the interface angle, formed at the three-phase boundary, that is generated after the application of a drop of liquid on the solid surface, between the gas-solid surface, gas -liquid surface and solid-liquid surface. The CA is a result of the balanced forces due to the interfacial tensions of solid surface, liquid and air. According to the Young-Laplace's equation, the CA (θ) can be expressed as in Eq. (1):

$$\cos\theta_{\gamma} = \frac{\gamma_{SV} - \gamma_{SL}}{\gamma_{LV}} \tag{1}$$

where θ_{γ} represents the contact angle, γ_{SV} represents the surface energy of the solid-air interface, γ_{SL} represents the surface energy of the solid-liquid interface, and γ_{LV} represents the surface energy at the liquid-air interface. The water CA values of less than 90° indicate the surface is hydrophilic, while the values of more than 90°C indicate that the surface is hydrophobic (Guo & Zhao, 2024; Kholodov et al., 2015).

The point of zero charge (PZC) is a pH value at which the surface density of positive charges is equal to that of negative charges under given analytical parameters (temperature conditions, composition of aqueous solutions) (Rey et al., 2011). PZC is determined using the salt addition method. The method is based on measuring the differences between the initial and final pH values in the salt solutions (from pH 2-11) over a certain period. Based on the measured differences, the surface can exhibit a negative charge in case of





lower final pH values and a positive charge in case of higher final pH values as can be seen in **Figure 1** (Bakatula et al., 2018; Zia-Ur-Rehman et al., 2020).

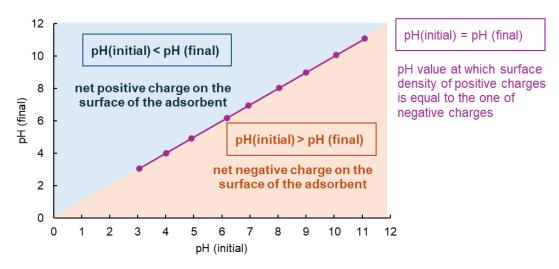


Figure 1. The surface charge of the adsorbent concerning the differences in pH values, determined in salt solution with the salt solution method for determination of the point of zero charge (adopted from Zia-Ur-Rehman et al., 2020).

The overall aim of this research was to determine the surface properties (PZC and CA) of MPs and dry microalgal biomass as sorbents of CECs, and to investigate how the drying process affects the surface characteristics of microalgal biomass. This will help predict the behavior of different combinations of MPs, CECs, and microalgal biomass when added to soil.

2. Material and Methods

2.1. Microplastics

Surface properties of microplastic particles (MPs) from agricultural mulch were assessed, namely of two biodegradable and one non-degradable film. Polymer types of plastic mulch used were co-polymer of polybutylene adipate terephthalate and polylactic acid (PBAT/PLA), TPS, and high-density PE (PE_black). Obtained mulch of each polymer type was manually cut into smaller particles to obtain large MPs in the size between 1 mm and 5 mm.

2.2. Microalgal biomass

Microalgal biomass analysed consisted of a monoculture *Chlorella vulgaris* (*C. vulgaris*), cultivated in the laboratory, and a mixed microalgal consortia, cultivated during the WW treatment process in HRAP.

Monoculture of *C. vulgaris*, acquired from AlgEn (Algal Technology Centre, Slovenia), was cultivated in a sterile Bold's Basal Medium (BBM; PhytoTech Labs, USA). *C. vulgaris* was cultivated in sterile 250 ml Erlenmeyer flasks, which were sealed with foam caps to enhance gas exchange. The inoculated flasks were continuously agitated at 150 rpm using an orbital shaker (RS-OS-20, Phoenix Instrument, Germany) within a custom-built growth chamber. Cultivation conditions were as follows: 25 °C, artificial illumination provided by FLUORA lamps (OSRAM, Germany) with an average illumination intensity of 1380 lx. The obtained biomass of *C. vulgaris* was harvested using a UNIVERSAL 320 centrifuge (Hettich Zentrifugen, Germany) at 8000 rpm for 5 min.

Mixed microalgal consortia was harvested from the HRAP pilot plant (45° 52.5' N, 13° 54.3' E), used for treating effluent from primary treatment of WW at the Central Wastewater Treatment Plant Ajdovščina (CWWTP Ajdovščina). 1 L of WW containing microalgal biomass from HRAP was concentrated with centrifugation at 8000 rpm for 5 min (UNIVERSAL 320, Hettich Zentrifugen, Germany).





2.3. Measurements of contact angle

CA of MPs and microalgal biomass were measured with tensiometer Theta Attention (Biolin Scientific, Sweden) using the sessile drop method (Kholodov et al., 2015; Cramer et al., 2022). MPs and microalgal biomass were mounted onto the microscopic object glass slides using double-sided adhesive tape (4965, Tesa, Germany). A droplet of water (5 μL) was extruded from the syringe onto the sample, while simultaneously the magnifying camera on the tensiometer recorded (OneAttension software) the droplet formation on the sample (repeatability n=9). The geometry of each droplet was evaluated by the Young-Laplace's equation and CA was measured at the three-phase contact point between the surface of the sample, the water droplet and the air.

2.4. Measurements of point of zero charge

The salt addition method was used to determine the point of zero charge (PZC) of MPs and dry microalgal biomass according to the modified method by Santaeufemia et al. (2021). Briefly, charges in the pH were measured with a Multi 3620 IDS meter and Sentinx 940 probe (WTW, Germany) in nine pH solutions (3-11) containing 40 mL of 0.01 M CaCl₂. In each solution, the pH value was adjusted to the initial pH of the solution ($pH_{initial}$) using 0.1 M NaOH and/or 0.1 M HCl. To each of the solutions with adjusted pH, 40 mg of MPs or microalgal biomass was added and left to stir on magnetic stirrers (Velp Scientifica, Italy) at approx. 300 rpm. After 1 h, the pH was measured and denoted as pH_{final} . Difference in the pH (ΔpH) for each of the solutions was calculated according to the Eq. (2) and the PZC was obtained from the intersection of ΔpH with the x-axis on the graph ΔpH vs. $pH_{initial}$.

$$\Delta pH = pH_{initial} - pH_{final} \tag{2}$$

3. Results and Discussion

3.1. Surface properties of dry microalgal biomass

Microalgae are unicellular organisms mainly present in aquatic environments. Based on the previous research by Ozkan & Berberoglu (2013), it was expected for the surface of the tested microalgal biomass to exhibit a hydrophilic nature, regardless of different sample preparations. CA of laboratory culture of *C. vulgaris* was determined to be similar in case of direct (oven) drying of the biomass (86.1±9.0°) and freeze-dried biomass (82.8±6.3°). Contrary, the surface of microalgal consortia was more hydrophilic when biomass was freeze-dried (65.0±6.6°) compared to when it was oven (direct) dried (93.1±4.4°). The drying method can affect the hydrophobicity/hydrophilicity of microalgal biomass due to the structural changes in the cell wall. Ultimately, the breakage of the cell wall can expose more hydrophobic intracellular components therefore, making the surface more hydrophobic (Machado et al., 2022).

The surface charge of microalgal biomass was found to be similar for both tested microalgal biomasses, regardless of the used drying method. PZC of the culture of *C. vulgaris* was slightly acidic, determined at pH of 5.7 and 5.6, for direct and freeze drying, respectively (**Figure 2**). Microalgal consortia biomass exhibited slightly higher PZC at pH values of 6.2 and 6.7, for direct and freeze drying, respectively. Obtained PZC values are in the range of previously reported PZC values for dry microalgal biomass, tested in different salt solutions (Mohammed et al., 2019; Bakatula et al., 2018). Therefore, at environmental pH values (7-8), the surface of *C. vulgaris* had a higher density of negative charge compared to the microalgal consortia. Negative surface charge in microalgal biomass is attributed to the presence of carboxylic and amino functional groups on the membrane of the microalgal cells (Li et al., 2022).





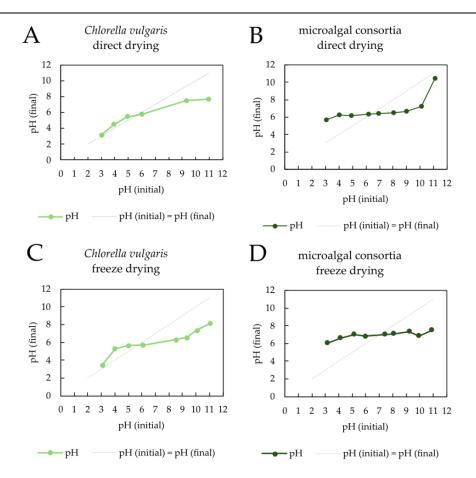


Figure 2. Initial (pH (initial)) vs. final (pH (final)) pH measurements of salt solution containing microalgal biomass, dried in two different ways: direct oven drying (*Chlorella vulgaris* (A) and microalgal consortia (B)) and freeze-drying (*Chlorella vulgaris* (C) and microalgal consortia (D)). The point of zero charge (PZC) of each microalgal biomass is characterised by the crossing of the pH (final) curve at different pH (initial) values with the linear curve of equal pH (initial) and pH (final) in salt solution (pH (initial) = pH (final)).

3.2. Surface properties of microplastics

MPs, in general, are reported to primarily have a hydrophobic nature, since most of the structural polymers are hydrophobic. The wettability of the tested mulch MPs in this study based on the polymer types was as follows: PBAT/PLA (84.7±5.9°) > TPS (76.5±4.5°) > PE_black (73.0±7.8°). For pristine MPs, the obtained water CA for PLA and PBAT are in accordance with the literature, 75-85° and 82-92°, respectively (Tümer et al., 2022; Pan et al., 2024). Contrary, TPS film exhibited more hydrophobic surface compared to the reported CA values in the range from 53° to 62° (Jantanasakulwong et al., 2016; Zhong et al., 2022), and PE mulch exhibited more hydrophilic surface compared to the reported values for PE from 89° to 99° (Aktas et al., 2023; Accu Dyne Test, 2025). The discrepancy with the literature could be due to the possible presence of additives in mulch films that alter the hydrophobicity/hydrophilicity of the material to achieve the desirable characteristics of mulch (moisture control and funnelling the excess rainfall away from the roots, heating properties of plastic to regulate soil temperature) (Kasirajan & Ngouajio, 2012).

Exposure to environmental factors (physical, chemical, and biological) can cause ageing and degradation of MPs. Thus, the surface of MPs can become hydrophilic (Harraq & Bharti, 2021). This was observed also in our case, where all tested polymer types exhibited more hydrophilic surface of the naturally aged particles compared to their pristine counterparts. The CA of naturally aged MPs followed

PE black $(67.6\pm7.3^{\circ}) \approx PBAT/PLA (65.5\pm5.0^{\circ}) > TPS (57.3\pm3.5^{\circ}).$





The surface of tested MPs exhibited neutral to slightly negative charge (**Figure 3**). Neutral charge at environmental pH values was observed for pristine PBAT/PLA and TPS MPs, and naturally aged PE_black (all PZC 7.3) and PBAT/PLA (PZC 6.8). A slightly acidic and therefore more negatively charged surface was determined in the case of pristine PE_black (PZC 6.3) and naturally aged TPS (PZC 6.1).

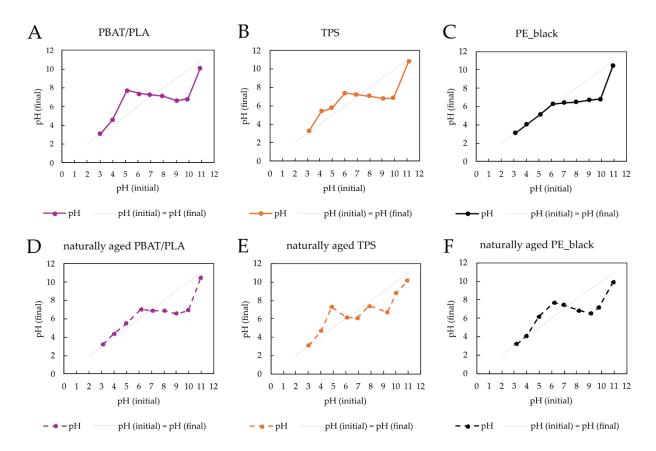


Figure 3. Point of zero charge of pristine microplastics (polybutylene adipate terephthalate (PBAT)/polylactic acid (PLA) – A, thermoplastic starch (TPS) – B, and polyethylene (PE_black) – C) and naturally aged microplastics (PBAT/PLA – D, TPS – E, and PE_black – F). The point of zero charge (PZC) of each type of microplastics is characterised at the crossing of pH $_{\rm f}$ curve at different pH $_{\rm f}$ values with the linear curve of equal pH $_{\rm f}$ and pH $_{\rm f}$ in salt solution (pH (initial) = pH (final)).

5. Conclusions

Surface characterization of sorbents is important in order to explain the sorption mechanisms that drive the sorption process of certain organic contaminants. Two methods for characterization of surfaces – point of zero charge (PZC) and water contact angle (CA) – were used to characterize the surface of microalgal biomass and microplastic particles (MPs) as sorbents. The drying method of microalgal biomass can modify the surface characteristic, especially the polarity of the surface due modifications of the cell wall and elimination of intracellular hydrophobic substances. The effects of used drying method can vary, based on the composition of the used microalgal biomass. On the other hand, ageing of MPs can change the surface charge and wettability (CA), therefore affecting the sorption process of organic contaminants in the aquatic and terrestrial environment.

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Conflicts of Interest: The authors declare no conflict of interest.





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Research

Biostimulants from Algae Biomass Uncovered: Phytohormones Determination and Plant Growth Stimulation

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mones Detection and Plant Growth

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Abstract:

Microalgae are increasingly recognized as a valuable source of bioactive compounds, including phytohormones, which play crucial roles in regulating plant growth and development. This study investigates the potential use of microalgae biomass as natural biostimulant for sustainable agriculture, where focus on boosting crop resilience and productivity is needed. While most previous studies of the microalgae phytohormones production have been limited to laboratory-scale experiments using expensive synthetic growth media, this research advances the field by utilizing pilot-scale systems for algae biomass cultivation in real municipal wastewater (WW), which represents an alternative, cost-effective growth medium. Microalgae cultures were grown under controlled laboratory conditions and in a pilot high-rate algae pond (HRAP), and specific phytohormones in selected microalgae cultures were quantified. Biomass samples were prepared using various methods (fresh utilisation, freezing, drying), with lyophilization being identified as more effective for preserving biostimulative activity, compared to frozen and thawed biomass, which was found to be less suitable. Preliminary tests, performed with commercial phytotoxkit liquid samples, confirmed biostimulatory effects of algae biomass on roots growth of white mustard (Sinapis alba). Phytohormone profiling, conducted using LC-MS/MS, revealed up to 30-fold higher levels of auxin indole-3-acetic acid (IAA) in pilot-scale cultures compared to laboratory-grown biomass. These findings highlight the potential for scaling up microalgal cultivation to produce phytohormoneenriched biomass. Understanding how different growth conditions influence phytohormone production in microalgae is essential for developing large-scale application guidelines. Variability in biomass composition due to fluctuating wastewater characteristics necessitates further research, as well as the development of optimized application strategies to enhance biostimulatory efficacy.

Keywords: High-rate Algae Pond; Microalgae Cultivation; Phytohormones Screening; Plant Growth Promoters





1. Introduction

1.1. Microalgae as a source of phytohormones

Microalgae are a diverse group of photosynthetic microorganisms, including both eukaryotic and prokaryotic organisms, cyanobacteria (Dineshkumar et al., 2017; Singh & Saxena, 2015). Algal biomass is gaining attention due to its many beneficial properties and its importance across various industries, including cosmetics, food (nutritional supplements, animal feed), as well as to produce pigments and other bioactive compounds which are valuable in medicine and the pharmaceutical industry (Basheer et al., 2020; Castro et al., 2023; Eze et al., 2023). Some species of microalgae have ability to produce polysaccharides, antimicrobial compounds and other metabolites, such as phytohormones, which are well known for their biostimulatory properties and can positively influence plant growth, development, and resilience under stress conditions (Gonçalves, 2021; Guo et al., 2020; Haak et al., 2017; Liu & Ruan, 2022). Auxins and cytokinins play a critical role in plant cell division and elongation, while gibberellins regulate seed germination and plant growth, and abscisic acid acts as a stress-related hormone under adverse conditions (Stirk et al., 2014; Wang et al., 2021). The use of phytohormones derived from microalgae offers a promising sustainable solution in agriculture by reducing the need for synthetic fertilizers, thereby contributing to a decrease in their negative environmental impact (Kapoore et al., 2021; Miranda et al., 2024; Oancea et al., 2013).

Microalgae require specific growth conditions, including appropriate light, temperature, pH, nutrients concentration, and the presence of CO₂, which increases production costs, cultivation, harvesting, and processing costs, particularly for biomass with high valuable compounds that often necessitates optimized growth conditions, expensive synthetic media, or complex bioreactors (Amaro et al., 2023; La Bella et al., 2022; Menegazzo & Fonseca, 2019). This requires the development of innovative, energy-efficient, and cost-effective technologies to enable sustainable production of high-quality algae biomass and its easy integration into agricultural practices (Barsanti & Gualtieri, 2018; Do et al., 2021; Parmar et al., 2023).

Microalgae are capable of thriving in diverse environments, including mixotrophically in wastewater systems (Najdenski et al., 2013; Orfield et al., 2014; Singh & Saxena, 2015). They can be cultivated auto/hetero/mixotrophically either in closed (photo/fermentation) bioreactors or open systems, such as high-rate algae ponds (HRAPs) (Borowiak & Krzywonos, 2022; Muhammad et al., 2020; Tran et al., 2020). Their ability to assimilate nutrients from wastewater (WW) while producing valuable biostimulatory compounds underscores potential for agricultural application and environmental remediation, increasing sustainability (Chew et al., 2017; Kumar et al., 2024). While WW has shown potential for microalgae producing biostimulants, the variability in its composition and specific microbial consortium could affect consistency in phytohormones production (Chai et al., 2024; Li et al., 2024; Oruganti et al., 2022). Production of phytohormones (types and levels) varies among different microalgal and bacterial strains as well as on factors in microorganisms, such as cultivation method, light, and stress factors (Mujtaba & Lee, 2016; Wang et al., 2021). Stirk & van Staden (2020) demonstrated that phytohormone production is closely linked to algal growth and the cell cycle, with different types and concentrations of hormones produced at various growth stages. In actively growing cells, auxins and cytokinins appear to regulate the cell cycle, gibberellins are associated with cell elongation, and abscisic acid functions as a stress hormone (Stirk et al., 2014). Therefore, standardize cultivation parameters are needed and further research focuses should include exploring the production of phytohormones by selected microalgae strains under varying culture conditions, with an emphasis on their application in agriculture (Liang et al., 2009; Magalhães et al., 2024; Mohsenpour et al., 2021).

1.2. Phytohormones and biostimulatory screening of microalgae

Specific methods are required for effective extraction, purification and quantification of target phytohormones in microalgae (Do et al., 2021). Unlike other intracellular active substances, phytohormones encompass many different types with varying structural charac-





teristics, numerous derivatives or metabolites. Additionally, phytohormones are not stable, and they are present in low concentrations in microalgae, therefore sensitive, simple, and rapid processes are needed for determination of phytohormones (Wang et al., 2021). Appropriate techniques for determining endogenous phytohormones are gas chromatography (GC) or high-performance liquid chromatography (HPLC), and for more complex samples, liquid chromatography coupled with mass spectrometry (LC-MS) has been determined as suitable (Zhou et al., 2003; Hoyerová et al., 2006). Form of LC, coupled with tandem mass spectrometry (LC-MS/MS), is an advanced form of LC-MS analysis and is extensively used in targeted analysis, where specific compounds are selected for quantification or identification. This makes it ideal for comprehensive coverage of nearly all known plant hormones and meeting the high accuracy demands of researchers (Creative Proteomics). GC is a commonly used method offering even greater precision and reliability (Birkemeyer et al., 2003; Wang et al., 2021).

Quantification of phytohormones in microalgae samples with LC-MS/MS, along with potential biostimulatory screening of biomass, could provide a comprehensive approach to understanding phytohormone production in microalgae for process optimizations, enabling scalable applications in sustainable agriculture (Nephali et al., 2020; Stirk & van Staden, 2020; Wang et al., 2021). Seedling growth tests are widely used to assess the biological activity of various substances, which include bioactive substances in microalgae that could influence plant growth and development (Godlewska et al., 2016; Rogovska et al., 2012). White mustard (*Sinapis alba*) seeds are common in such tests because of their fast germination, high sensitivity and uniformity (Pannacci et al., 2013).

Most research of microalgae producing phytohormones remains confined to laboratory-scale studies using synthetic media, leaving critical gaps in understanding their performance in real-world conditions (Abdelfattah et al., 2023; Chakraborty et al., 2023; Villaró-Cos et al., 2024). The research aim of this study was to examine production of phytohormones and biostimulatory potential of microalgae, cultivated under controlled laboratory conditions, either in sterile or semi-sterile growth medium, and on pilot scale in HRAP; an open system demonstrating secondary WW treatment. We hypothesize that from microalgae, grown on HRAP, higher concentration of bi-ostimulants can be gained compared to the lab scale.

Therefore, the biostimulatory effects of microalgae biomass prepared in various ways were investigated, and a direct effect on seed germination and early growth of *S. alba* was observed. Different samples of biomass were lyophilized (freeze-dried) for phytohormones quantification conducting LC-MS/MS, as described below.

2. Material and Methods

2.1. Microalgae culturing, harvesting and biomass processing

Chlorella vulgaris cultures were grown under controlled laboratory conditions, at the room temperature, under fluorescent 4000 K (FLUORA, Osram, Germany) and LED lights (65 µmol/m²s). Cultivation was performed in sterile 2 L Erlenmeyer flasks placed on a magnetic stirrer, containing 1.2 L of Bold's Basal Medium (BBM; Phytotechlab) (**Figure 1a**), and in a 30 L photobioreactor (PBR) operating under semi-sterile conditions, with stirrer and CO₂ addition, containing supplemented BBM with 0.34 g/L NaNO₃. The PBR maintained a working volume of 27 L (**Figure 1b**).

Mature cultures were harvested by centrifugation, washed with sterile water to remove residual medium, and processed either by oven-drying at 45 °C or lyophilization. The dried biomass was stored in airtight, light-protected containers at room temperature until further analysis. For quantitative phytohormone analysis, approximately 100 mg of lyophilized biomass was needed. Samples were labeled "F, W, D, L" to indicate the preparation steps: fresh, washed, dried, and lyophilized biomass, respectively.

Preliminary tests were performed to confirm biostimulatory effect of algae biomass on *Sinapis alba* (*S. alba*) seeds; various samples from PBR, including fresh biomass, dried, lyophilized or few weeks old from the fridge (labeled "O") were analyzed (**Figure 1**).





Mixed culture of *Chlorella, Scenedesmus*, and *Pseudopediastrum* species (**Figure 1c**) was obtained from the pilot HRAP in the beginning of September 2024, with real municipal WW (following primary treatment) used as the growth medium.

Biomass was harvested from the sedimentation tank, without washing, and either directly lyophilized or subjected to a freeze-thaw cycle before lyophilization. The processed samples were stored at room temperature in airtight, light-protected containers until further analysis. Samples were labeled "F, FR+T, L" to indicate the preparation steps: fresh, previously freeze-thawed, and lyophilized, respectively (**Figure 1**). The biostimulatory effect of selected algae biomass from the pilot HRAP on *S. alba* root growth has not yet been confirmed.

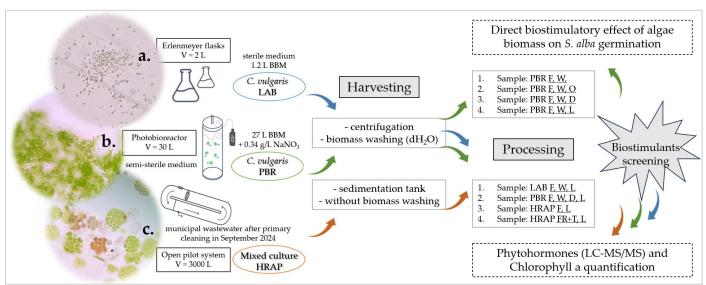


Figure 1. Microalgae culturing, harvesting and biomass processing; **a.** *C. vulgaris* grown in sterile laboratory conditions (LAB), **b.** *C. vulgaris* grown in semi-sterile laboratory conditions (PBR), **c.** mixed algae-bacteria culture grown in open pilot system (HRAP). **F** = fresh, **W** = washed, **O** = few weeks in the fridge (old), **D** = dried at 45 °C, **L** = lyophilized, **FR+T** = freezed and thawed.

2.2. Phytohormones quantification in microalgae and biostimulatory potential of biomass Selected 4 microalgal samples were analyzed for quantification of specific phytohormones using LC-MS/MS (AB Sciex QTRAP 6500, Waters Xevo TQ-S) at the Creative Proteomics (USA). For quantification of phytohormones, approximately 100 mg of lyophilized biomass was needed. The obtained data were normalized against the company's internal standards to account for experimental variation and differences in hormone extraction and ionization efficiency.

Chlorophyll A (Chl A) content (mg/g) was determined using a spectroscopic method (UV-VIS) according to the modified method by Lichtenthaler & Buschmann (2001). Briefly, a 0.1% suspension of lyophilized biomass sample (w/w), mixed with water (dH2O) was prepared. Chl A was extracted by the following procedure: centrifugation at 6500 rpm for 25 min, discarding the supernatant, addition of 95% ethanol (8 mL), heating at 60°C for 1 hour, centrifugation at 4500 rpm for 10 min, measurement of absorbance at 649 nm, 664 nm and 750 nm. Concentration of Chl A in mg/g was calculated according to Eq. (1),

$$Chl\ A = (13.36 \times (A_{664} - A_{750}) - 5.19 \times (A_{649} - A_{750})) \times (V_{EtOH}/(V_{sample} \times l_{cuvette})) \qquad . \tag{1}$$

Eq.(1) incorporates coefficients (13.36 and 5.19) to accurately quantify chlorophyll A (Chl A) in mg/g. Additionally, the calculation adjusts for the volume of solvent (V_{EIOH}), the sample volume (V_{sample}), and the cuvette path length (l) to standardize the result.

Phytotoxkit liquid bioassays (Microbiotests) were conducted using *Sinapis alba* seeds to evaluate fresh (F), aged (O), dried (D), and lyophilized (L) samples from PBR. A 10% dry matter buffer mixture was prepared, then algae solutions were applied to test plates at 0.5% and 5% concentration prepared in final 20 mL, where 6 seeds were placed per half





plate, and distilled water was used as the control. After 3 days, root growth was determined with ImageJ tool (available for downloading online), and the germination index (GI) was calculated (Eq. (2)). GI of >120% indicated biostimulation, while GI <80% suggested phytotoxicity.

$$GI [\%] = ((RL \times GR \text{ of sample})/(RL \times GR \text{ of control})) \times 10$$
 . (2)

Eq.(2) accounts for average root elongation (RL) and germination rate (GR), providing a comprehensive determination of the biostimulatory or phytotoxic potential of the tested biomass. Germination rate (GR) was determined as the percentage of germinated seeds relative to the total number of tested seeds.

3. Results

Among detected phytohormones were auxins, cytokinins and gibberellins (**Table 1**). Phytohormones' levels were varying in samples; algae biomass grown in HRAP exhibited up to 30-fold higher levels of indole-3-acetic acid (IAA) compared to laboratory-grown biomass. However, these elevated levels may partially resulted from exogenous sources present in directly used biomass, and some bacteria could secrete IAA to enhance the interaction between algae and bacteria which could be the reason for the lowest content in biomass cultured under sterile laboratory conditions (Chen et al., 2019).

Drying (lyophilization) was identified as a suitable method for preserving phytohormones and biostimulant activity, but freezing and thawing affected the content of phytohormone, as well as Chl A (**Figure 2**).

Table 1. Phytohormones quantification (ng/g) conducting LC-MS/MS (Creative Proteomics, USA) in lyophilized biomass.

Phytohormones (ng/g)	Cytokinins		Gibberelins		Auxins				
/ Sample (preparation)	cZ	cZR t	ZR	GA9	GA4	GA29	IAA	MethlyIAA	IAA-Trp
Chlorella vulgaris LAB (F, W, L)	0.3	1.2	0.1	/	7.3	/	7.7	0.8	/
Chlorella vulgaris PBR (F, W, D, L)	0.2	0.8	/	/	/	/	119.0	1.4	/
Mixed culture HRAP (F, L)	0.6	0.9	0.1	5.6	/	1.5	3083.2	3.6	0.4
Mixed culture HRAP (FR+T, L)	7.0	21.6	1.2	/	/	/	1356.9	24.8	/

F = fresh, W = washed, D = dried, L = lyophilized, FR+T = freezed and thawed, cZ = c-Zeatin. cZR = c-ZRiboside, tZR = t-ZRiboside, GAs = gibberelins, IAA = indole-3-acetic acid, IAA-Trp = IAA-tryptophan. Abscisic acid (ABA) was not detected.

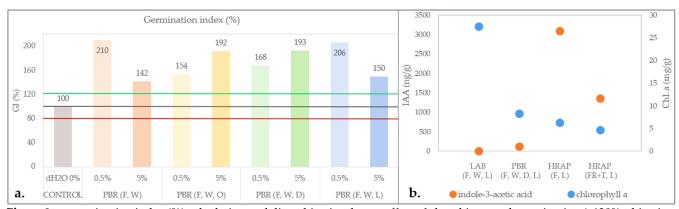


Figure 2. a. germination index (%) calculation and direct biostimulatory effect of algae biomass determination (>120% = biostimulatory effect, <80 = phytotoxic effect, 80-120% = no effect), **b.** indole-3-acetic acid (IAA, ng/g) and chlorophyll a (Chl. a, mg/g) content in lyophilized biomass. **F** = fresh, **W** = washed, **O** = few weeks in the fridge (old), **D** = dried at 45 °C, **L** = lyophilized, **FR+T** = freezed and thawed.





4. Discussion

We have previously reported that freeze-thawed algal biomass was not effective as a biostimulant for *S. alba* seeds (Cepec et al., 2024). One possible explanation is the damage of the cell wall during thawing at room temperature, which could leads to the release and likely degradation of intracellular contents. The freeze-thaw principle for cell disrupting has been described, where the efficiency of cell rupture using this method depends on factors such as the freezing rate, thawing conditions, extraction solution, and algal species (Chittapun et al., 2025). Recent studies have shown that an increase in thawing temperature and duration can exacerbate the loss of photosynthetic pigments (Chen et al., 2024).

Light intensity is a major factor influencing chlorophyll content; however, culture concentration, CO₂ availability, and nutrient conditions can also play significant roles. Self-shading likely reduces the average light available to cultures in PBRs, whereas in HRAPs, chlorophyll content tends to increase with decreasing water depth. Seasonal variations and the presence of environmental pollutants further influence chlorophyll levels in microalgae (da Silva Ferreira & Sant'Anna, 2017; Kim et al., 2018; Perin et al., 2022; Sutherland et al., 2014).

Our results match others, as IAA being the most prevalent phytohormone in actively growing microalgal strains, where concentrations of cZ>tZ-type cytokinins were reported (Stirk et al., 2009; Stirk et al., 2013). Stress-responses can explain lower content of active forms and increase of precursors in freeze-thawed sample, additionally, bioactive compounds are susceptible to degradation over time, as exposing the secondary metabolites to oxygen is quickening their degradation (Stirk & van Staden, 2020; Stirk et al., 2021). Other studies have similarly indicated that growth conditions and biomass processing significantly affect the phytohormone content and biostimulant potential of biomass (Ranglová et al., 2021). For example, the phytohormone content in Chorella sorokiniana is highly influenced by culture conditions, however, our findings differ slightly, suggesting that phytohormone production in microalgae is both growth- and species-specific (Stirk et al., 2024). In contrast, lower auxin concentrations were detected in C. sorokiniana grown phototrophically in outdoor raceway ponds with synthetic growth medium compared to laboratorygrown cultures (Do et al., 2020). On the other hand, microalgal biomass cultivated in synthetic medium combined with municipal WW exhibited higher auxin and cytokinin concentrations compared to biomass grown in synthetic medium alone (Elakbawy et al., 2022).

Microalgae produce bioactive compounds under specific conditions, but the precise mechanisms and environmental factors regulating their synthesis remain insufficiently understood (Do et al., 2020; Senousy et al., 2023). Based on our results, we conclude that further research is essential to fully understand the complex interplay of factors influencing the production and stability of bioactive compounds in microalgae. Special emphasis should be placed on optimizing mixotrophic cultivation at larger scales while reducing production costs.

5. Conclusions

The findings of this study suggest that HRAP systems can be optimized for large-scale production of phytohormone-enriched biomass and freeze-drying is a reliable method for preserving the biostimulatory potential of microalgae biomass. Therefore, in further research it is necessary to standardize cultivation parameters and optimize production systems. Future studies should explore how varying culture conditions influence phytohormone synthesis in selected microalgae cultures. The focus should remain on enhancing their application as biostimulants to support plant growth and improve stress resistance, conducting pilot-scale experiments using real municipal wastewater. This approach not only offers a natural alternative to synthetic chemicals but also contributes to environmental sustainability by utilizing waste resources in biomass production. Understanding influence of growth conditions and processing methods on the production and activity of phytohormones in microalgae can guide the development of high-quality biomass for agricultural use.







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Research

Electrochemical Harvesting of Microalgae

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Abstract:

The rapid growth of the global population is driving increased energy and resource consumption, necessitating alternative feedstocks. Microalgae, with their high growth rates and valuable natural compounds, offer a sustainable solution for producing biofuels, pharmaceuticals, cosmetics, fertilizers, and animal feed. However, harvesting microalgae remains a significant challenge, accounting for 20-30 % of production costs due to factors such as small cell size, low density of microalgae solution, and negative surface charge of microalgae cell. Conventional methods, including centrifugation and filtration, are also high energy consuming. This study explores electrochemical processes as an alternative harvesting technique, focusing on electrocoagulation-flotation with aluminium and aluminium-graphite electrodes and on electroflotation with graphite electrodes. The additional focus of the research was to explore the harvesting efficiency with graphite electrodes to reduce environmental impact by the usage of aluminium. Experiments were conducted at voltages of 5, 8, and 10 V with harvesting durations of 4 and 8 minutes. Two types of microalgae samples were tested: lab-scale cultivated, and pilot-plant cultivated microalgae from high-rate algal pond at the central wastewater treatment plant Ajdovščina. Harvesting efficiency was determined by optical density at 680 nm. Results showed that higher voltages and extended durations improve efficiency, with a maximum at 98.2 % using aluminium electrodes. Combined aluminium-graphite electrodes achieved a comparable efficiency of 96.8 %, reducing aluminium usage by 50 %. Graphite electrodes alone achieved an efficiency of 66.5 %. Electroconductivity also plays a crucial role in the efficiency of electrochemical processes.

Keywords: Electrochemical process; Electroflotation; Electrocoagulation; Graphite electrodes; Aluminium electrodes; High-rate algal pond





1. Introduction

The rapid population growth may lead to a rise in global energy and resource consumption. This growing demand can be addressed using alternative sources. Microalgae, for instance, contain natural compounds that serve as a valuable source of alternative raw materials for producing biofuels, pharmaceuticals, cosmetics, fertilizers, and animal feed. Additionally, microalgae are resistant to environmental conditions and can be cultivated in areas unsuitable for traditional plant farming. Microalgae are single-cell, photosynthetic microorganisms that grow very fast under the three essential elements: a light source, nutrients (primarily nitrogen, phosphorus, and trace metals), and a carbon source (provided in the form of CO₂) (Roy & Mohanty, 2019).

A bottleneck in microalgae production is harvesting, which involves separating algal biomass from the liquid medium in which it is suspended. According to some estimates, this process accounts for 20–30 % of total production costs (Suparmaniam et al., 2022). The high costs and low efficiency of certain harvesting processes are primarily due to: 1) the small size of cells (5-50 μm), 2) the negative charge on cell surfaces, 3) the cell density being similar to that of water (1.08–1.13 g/mL), 4) the relatively low concentration of microalgae in the liquid medium (0.5–5 g/L) (Young et al., 2021). Conventional harvesting techniques, such as centrifugation, filtration, flocculation, and gravitational sedimentation, have some drawbacks in terms of efficiency, toxicity, costs, and sustainability. To address these limitations, new harvesting methods must be explored, including bioflocculation, electroflotation-coagulation, ultrasonic aggregation, magnetic separation, and phototaxis (Zhu et al., 2024). Electrochemical (EC) microalgae harvesting is based on the application of a direct electrical current through electrodes into a culture broth. During this process, electrolysis occurs between the anode and cathode. Electrolysis can take place using either metallic or non-metallic electrodes. Metallic electrodes are sacrificial electrodes and thus releasing metal ions that can contaminate the final biomass and liquid, making the final products unusable. The objective is to use non-metallic, non-sacrificial electrodes that remain inert during electrolysis and do not react with the solution or biomass, such material like graphite (Coons et al., 2014).

Microalgae can thrive in diverse environments, including in nature-based solutions for wastewater treatment such as high-rate algal ponds (HRAPs). HRAPs are shallow, open ponds made for the treatment of wastewater where the wastewater represents a source of the nutrients for microalgal growth (Magalhães et al., 2024). The aim of this study is to separate microalgal biomass from wastewater originating from the HRAP system. The concentrated microalgal biomass holds potential as biofertilizer and biostimulant in agriculture, while the treated water can be repurposed for irrigation. Therefore, it is important to address potential environmental and health risks in compliance with Directive (EU) 2024/3019 on urban wastewater treatment, as well as Regulation (EU) 2020/741 on minimum requirements for water reuse.

EC processes for harvesting microalgae has been reported to be effective by some researches. Liu et al. (2018) investigated the efficiency of graphite and aluminium electrodes on *Scenedesmus sp.* and concluded that the process could achieve above 90 % of microalgal removal efficiency only if aluminium ions were present in the solution. Al-Yaqoobi & Al-Rikabey (2023) concluded that a maximum harvesting efficiency of 96 % was achieved in 12 min for aluminium and 14 min for graphite, with the addition of 2 g/L of NaCl.

2. Material and Methods

2.1. Lab-scale cultivation of microalgal biomass

The microalgae strains used in the current study, *Chlorella vulgaris* and *Scenedesmus sp.*, were supplied by the company Algen LLC. The algal culture was grown in Bold's Basal Medium with added nutrients (0.34 g/L NaNO₃) at a light intensity of 65 μ mol/m²s at room temperature. A photobioreactor (PBR) with a working volume of 27 L operated under semi-sterile conditions, with a stirring mechanism and controlled CO₂ supplementation. Electroconductivity (1200 μ S/cm) and pH (9.3) were measured with WTW multi 3620 IDS multi-parameter portable meter.







2.2. Pilot-plant cultivation of microalgal biomass

Environmental samples were obtained from the HRAP located at the Central Wastewater Treatment Plant Ajdovščina, Slovenia. HRAP with volume of 3 m³ was designed as a pilot plant to treat primary treated wastewater from the central wastewater treatment plant. It was operated in a batch mode receiving 300 L of every 3 days. HRAP was continuously steered with a paddle wheel and contained a mixture of bacteria and microalgal species, predominantly belonging to *Chlorella* sp., *Scenedesmus* sp., and *Pseudopediastrum* sp. that provided secondary (removal of organic substances) and tertiary (nutrient removal) treatment of wastewater.

The first sampling took place in mid-October 2024. The algae in the pond were visibly in poor condition and the solution was diluted due to the rainy weather. Therefore, 2 L of algae-bacteria sludge from the ultrasonic sedimentation tank was taken and mixed with 25 L water from the pond. The 2 L sample from the tank was sourced from the same HRAP system a few weeks earlier, during the period when algal biomass was in good condition. The second sampling took place at the beginning of November 2024. Samples were again taken from the ultrasonic sedimentation tank (algae-bacteria sludge, 2L), and HRAP (25 L). Due to the diluted media in the pond, the solution had a low electrical conductivity (200 $\mu \text{S/cm}$), which disabled the EC process. In the previous experiment, the electrical conductivity was approx. 1200 $\mu \text{S/cm}$. To achieve this conductivity in the second experiment and enable the EC process to function effectively, 0.5 L of a 2.5 % NaCl solution was added.

2.3. Electrochemical experiments

The experimental work was carried out using a 2 L glass beaker. Two types of electrode materials were used to compare the EC process: the sacrificial electrode (aluminium; Al) and the non-sacrificial electrode (graphite; Gr). When aluminium is used, the process involves electrocoagulation-flotation, whereas with graphite, the process is limited to electroflotation (Al-Yaqoobi & Al-Rikabey, 2023). Electrodes had dimensions of 10 cm × 5 cm × 0.3 cm. The distance between electrodes was 0.5 cm. There were three configurations of electrodes: 1) set of four Al electrodes; 2) set of four Gr electrodes and 3) set of two Al and two Gr electrodes arranged in the sequence: Al-Gr-Al-Gr. The applied constant voltage from DC power supply Basetech BT-3020 was set to 5, 8, and 10 V. A magnetic stirrer was used for constant mixing at a speed of 100 rpm. The experiments were performed with approx. 900 mL of initial sample at room temperature. The full setup of the laboratory experiment is shown in Figure 1a, and electrode sets on Figure 1b and 1c. During the process, the flocs of microalgae floated to the surface of microalgal suspension due to gases formed at the electrodes (H₂ and O₂). The samples were taken with a tube at the depth of approx. 5 cm and were collected at 4 and 8 minutes throughout the EC process.





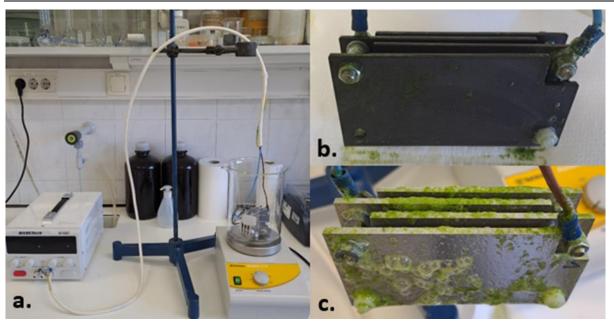


Figure 1. a) Laboratory setup. b) Graphite electrodes after EC process. c) Aluminium electrodes after EC process.

2.4. Determination of harvesting efficiency

The harvesting efficiency of the microalgae was calculated based on the change in optical density. Absorbance of the culture suspension was measured using UV-Vis spectrometer Nanocolor VIS (Macherey-Nagel, Germany), at 680 nm (Al-Yaqoobi & Al-Rikabey, 2023). The harvesting efficiency was subsequently calculated as Eq. (1):

harvesting efficiency
$$[\%] = \frac{(ODi - ODt)}{ODi} \times 100$$
 (1)

where *ODi* is optical density of the initial suspension, and *ODt* is the optical density of the suspension at a time t (Al-Yaqoobi &Al-Rikabey, 2023).

3. Results

3.1. Results for lab-scale cultivated microalgae

After calculating the efficiency based on absorbance at 680 nm, the maximum harvesting efficiency for lab-scale organisms is as follows:

Table 1. Summarized maximum efficiency results for harvesting lab-scale organisms based on the absorbance at 680 nm.

Electrode material	Voltage [V]	Time [min]	Efficiency [%]	Experiment
Al	10	4	58.0	October
Al-Gr	5	4	0.6	October
Gr	10	4	2.9	October
Al	10	8	31.0	November
Al-Gr	10	8	14.2	November
Gr	10	8	9.8	November
	•			

In the initial experiment in October, several aspects were not fully optimized. The time limit was initially set to 4 minutes, as this duration was sufficient to harvest the environmental sample during preliminary testing. However, after harvesting the model organism for the first time, it became apparent that extending the time would improve the process.





As a result, for the next three experiments, the time limit was set to 4 and 8 minutes. Additionally, during the first experiment, the mixing of the initial sample was not sufficient, resulting in a denser sample by the end of the test when using the combined Al-Gr electrode. This inadequate mixing led to reduced efficiency with the combined electrode. In subsequent experiments, this issue was addressed by optimizing the sample mixing process.

3.2. Results for pilot-plant cultivated microalgae (environmental samples)

Based on the absorbance at 680 nm, the highest harvesting efficiency results for environmental samples from the HRAP system are as follows:

Table 2. Summarized maximum efficiency results for harvesting environmental samples from HRAP system based on the absorbance at 680 nm.

•				
Electrode	Voltage [V]	Time [min]	Efficiency	Experiment
material			[%]	
Al	10	8	89.0	October
Al-Gr	10	8	92.1	October
Gr	10	8	17.4	October
Al	10	8	98.2	November
Al-Gr	8	4	96.8	November
Gr	10	8	66.5	November

The most efficient electrodes were Al and the combined Al-Gr, both achieving approx. 90 % efficiency. The optimal voltages were 10 V and 8 V, when the process duration was set to 8 minutes. Gr electrodes were less effective with 17.4 and 66.5 % of removal efficiency. The harvesting efficiency of environmental samples from the HRAP system was better compared to lab-scale biomass (max. 98.2 % for environmental; max. 58.0 % for lab-scale).

4. Discussion

Harvesting efficiency increased with higher voltage, as the stronger electric field enhanced the production of H₂ and O₂ when using all three types of electrodes. These gases enabled microalgae flotation by producing a continuous stream of fine bubbles in the solution. In addition, higher voltage resulted in the release of more aluminium ions when using Al and Al-Gr electrodes, which likely caused microalgae coagulation (Liu et al., 2018).

Increasing the harvesting time also improved harvesting efficiency by increasing the dissolution of metal ions from the anode and providing additional time for flotation with gas bubbles. However, as harvesting time can impact the final quality of the algal biomass, the process duration needs to be controlled (Visigalli et al., 2021).

The selection of electrode materials has a significant impact on the EC process. Despite identical operating conditions during experiments, differences were observed between Gr and Al electrodes. Liu et al. (2018) emphasized that neutralizing the electric charge on the microalgae cell surface is important to achieve high harvesting efficiency. This process can be achieved using positively charged metal ions such as Al³+ releasing from Al electrodes together with an electric field. Gr electrodes are not producing positively charged metal ions which explains their reduced harvesting efficiency. However, in our experiment on environmental samples, the Al-Gr electrode demonstrated relatively high efficiency, while reducing Al usage by 50 %.

Major differences in harvesting efficiency were also evident when comparing lab-scale cultivation with environmental (HRAP) samples. Efficiency of environmental samples was higher, likely due to the chemical and microbial composition of the algal medium.

Electroconductivity proved to be an important parameter for successful EC reactions, as low electroconductivity inhibited the process. Nageshwari et al. (2022) highlighted that







higher salinity of the electrolyte increases conductivity, thereby promoting ion release required for microalgal removal. However, the addition of large amounts of NaCl can increase costs and introduce several drawbacks related to utilization of both water and biomass. Additionally, Al-Yaqoobi & Al-Rikabey (2023) reported that increasing the NaCl concentration from 2 g/L to 3-5 g/L significantly reduced harvesting efficiency.

5. Conclusions

The laboratory-scale study showed that EC processes (flotation and coagulation) can achieve high harvesting efficiency also with combination of non-sacrificial electrodes. The highest harvesting efficiency for environmental samples using combined Al-Gr electrode was 96.8 %, achieved at 8 V and 4 minutes. This efficiency is comparable to the maximum achieved with Al electrodes alone, which reached 98.2 % at 10 V, 8 minutes. The use of graphite alone is less sufficient, achieving only 66.5 % efficiency at 10 V in 8 minutes. Significant differences are observed in the harvesting efficiency between lab-scale microalgae and environmental samples from the HRAP. Further research needs to evaluate the compounds present in the HRAP system that enhances harvesting efficiency compared to lab-scale cultivated microalgae.

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Research

Quantitative Determination of Total Phenolic Compounds in Plant Extracts Using Supercritical Carbon Dioxide and Water-ethanol mixture

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Abstract:

Natural products serve as an important source of bioactive compounds with significant applications in various fields of science. Among these, polyphenols are particularly noteworthy due to their antimicrobial and antioxidant properties and role in capturing free radicals. This study quantitatively determined the total phenolic content in plant extracts prepared using two distinct extraction methods: supercritical carbon dioxide (SC-CO₂) and water-ethanol maceration. The Folin-Ciocalteu method was employed for quantitative analysis, with results expressed as gallic acid equivalents per kilogram of plant material. The findings demonstrated that water-ethanol extracts consistently contained higher levels of phenolic compounds compared to SC-CO₂ extracts across all analyzed samples. For instance, rosemary extracts prepared with water-ethanol had the highest phenolic content, while thyme and marigold showed lower values. In contrast, SC-CO₂ extracts yielded significantly lower phenolic concentrations, likely due to the selective extraction of lipophilic compounds. Variations in phenolic content were attributed to factors such as sample polarity, plant type, and environmental conditions. These results highlight the importance of selecting appropriate extraction methods based on the desired application. Water-ethanol maceration proved superior for obtaining extracts rich in phenolic compounds, making it suitable for applications requiring high antioxidant activity. This study underscores the potential of plantderived polyphenols as natural, "green" bioactive ingredients for diverse scientific and therapeutic purposes.

Keywords: Plant; Supercritical carbon dioxide; Water-ethanol, Extracts; Polyphenols, Biologically active compounds; Medicine





1. Introduction

Phenols and their derivatives, including polyphenols, are a prominent group of secondary metabolites found in plants (**Figure 1**) (Cowan, 1999; Daglia, 2012). Polyphenols are characterized by the presence of one or more hydroxyl groups attached to a benzene ring, and their production is closely linked to plant growth and responses to biotic or abiotic stress. These compounds exhibit a wide range of biological activities, such as anti-inflammatory, anti-infective, anti-proliferative, anti-incrobial, and antioxidant effects (Luna-Guevara et al., 2018; Othman et al., 2019).

Polyphenols are particularly known for their antioxidant properties, as they scavenge and neutralize free radicals, thereby reducing or preventing oxidative cell damage, including oxidative DNA degradation (Jeran et al., 2021; Nisca et al., 2021). Due to their diverse biological effects, polyphenols have found applications as potential active pharmaceutical ingredients and are widely used in the textile, food, and cosmetics industries (Albuquerque et al., 2021).

In addition, plant and its derived polyphenols, have long been used to treat infections due to their antimicrobial properties. The antimicrobial efficacy of polyphenols is directly influenced by their specific composition and the extraction method used. For instance, methanol extracts of rosemary have demonstrated stronger antimicrobial activity against Gram-positive and Gram-negative bacteria, as well as yeasts, compared to water extracts (Moreno et al., 2006). Similarly, the antimicrobial effects of ginger extracts are attributed primarily to phenolic compounds such as eugenol, shogaols, zingerone, gingerdiols, and gingerols. Since these compounds are insoluble in water, organic ginger extracts tend to exhibit greater antimicrobial activity (Beristain-Bauza et al., 2019).

In thyme, the antibacterial activity of its extracts correlates with their total phenolic content, with key components such as thymol, carvacrol, and eugenol demonstrating antimicrobial efficacy in both planktonic and biofilm forms of pathogens (Mokhtari et al., 2023; Walsh et al., 2019). The choice of organic solvent also significantly impacts the extraction of antimicrobial compounds. For example, methanol extracts of *Calendula officinalis* L. showed larger zones of inhibition against several clinical bacterial pathogens compared to ethanol extracts. However, ethanol extracts demonstrated higher activity against specific Gram-positive bacteria such as *Staphylococcus aureus* and *Enterococcus faecalis* (Efstratiou et al., 2012).

Additionally, plant-derived polyphenols have shown synergistic effects when combined with conventional antimicrobial agents, leading to enhanced activity against clinical pathogens (Manso et al., 2021). For instance, phenolic compounds from *Mangifera indica* L. seeds significantly reduced the minimum inhibitory concentration (MIC) of penicillin G against Methicillin-Resistant *Staphylococcus aureus* (MRSA) (Jiamboonsri et al., 2011). Similarly, polyphenols from *Cuspidaria convoluta*, when combined with Ampicillin and Gentamicin, exhibited increased antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa* (Torres et al., 2019).

In the context of rising antimicrobial resistance, these findings highlight the potential of plant-based polyphenols as promising agents for combating multidrug-resistant bacterial infections (Ahmed et al., 2024). Moving forward, plant-derived extracts could play an essential role in developing innovative antimicrobial therapies.

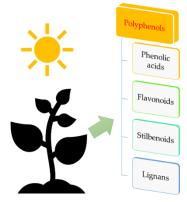


Figure 1. Types of polyphenols.





Due to the extraordinary properties of polyphenols and their versatile applications, especially in the fields of nutrition, pharmacy and medicine, their content in different samples of plant material was investigated in the present experimental work. The work was carried out with samples prepared using two different isolation techniques – waterethanol extraction and supercritical carbon dioxide extraction. The quantitative determination of the total phenolic content was used to investigate which of the extraction techniques used has a greater yield in leaching the total phenolic compounds. These findings will thus contribute to the development and progress of this highly interdisciplinary field of science. Throughout the work we will use the following model extracts: ginger, calendula, thyme and rosemary.

2. Selected plants

Calendula, rosemary, common thyme and ginger are traditionally used as home remedies both internally and externally.

Calendula (*Calendula officinalis*), also known as pot marigold has been cultivated since ancient times and has a long tradition of use as a medicinal plant (Kemper, 1999). The most important active ingredients of marigold are terpenoids such as triterpenoids (faradiol esters), polyphenols (phenolic acids and flavonoids), carotenoids and polysaccharides. The European Medicines Agency (EMA) recognizes calendula as a traditional herbal medicinal product for the treatment of mild inflammation of the mouth and throat and mild inflammation of the skin as well as to support the healing of minor wounds (European Medicines Agency, 2018).

Rosemary (*Rosmarinus officinalis*) is an aromatic herbal plant known for its high essential oil content. It is rich in terpenes, phenolic compounds, terpenoids and other phytochemicals. These are bioactive substances with specific antibacterial and antioxidant effects (Shankar et al., 2024). It is traditionally used to treat dyspepsia and cramp-like gastrointestinal disorders. Externally, it is used in remedies to treat mild muscle and joint pain as well as mild circulatory disorders (European Medicines Agency, 2011).

Common thyme (*Thymus vulgaris*) main active ingredient is the essential oil, of which the thymol it contains is responsible for its antiseptic and antimicrobial effect. It also contains flavonoids, phenolic acids, triterpenes and other compounds (Lee et al., 2005). Thyme is traditionally used to treat respiratory diseases such as bronchitis, coughs or inflammation of the upper respiratory tract. The thymol contained in the extract has antiseptic and antimicrobial effects (Sakkas & Papadopoulou, 2017).

Ginger (*Zingiber officinale*) has long been used in traditional Chinese medicine for its versatile therapeutic properties, especially its strong anti-inflammatory effect. The main source of the characteristic ginger aroma is the essential oil and non-volatile pungent chemicals. The rhizome contains the main active components, namely gingerols, shogaols, zingerone and gingerdiol (Gupta et al., 2025). Ginger extracts are traditionally used in herbal remedies to prevent nausea and vomiting, mild stomach cramps, flatulence, coughs and sore throats (European Medicines Agency, 2012). The gingerols in ginger have antioxidant and antimicrobial effects. Studies have shown an effect against cardiovascular diseases (Mao et al., 2019). Ginger extract can also be used externally in massage preparations to improve blood circulation and thus help to relieve tension (Polasa & Nirmala, 2003).

3. Methods

3.1. *Samples – Isolates from plant material*

The isolates (**Table 1**) with supercritical carbon dioxide were prepared by Flavex Naturextrakte GmbH (Germany), the water-ethanol extracts by Epo S.r.l. (Italy).





Table 1. Selected plant material for the experimental work.

Plant	Method of isolation	Part of the	Origin
		plant	(country)
Calendula	Supercritical carbon dioxid	Flowers	Egypt
	Water-ethanol extract	Flowers	Egypt
Rosemary	Supercritical carbon dioxide	Leaves	Spain
	Water-ethanol extract	Leaves	Italy
Thymus	Supercritical carbon dioxide	Leaves	Spain
	Water-ethanol extract	Leaves	Italy
Ginger	Supercritical carbon dioxide	Rhizome	Nigeria
	Water-ethanol extract	Rhizome	Italy

3.2. Determination of total phenolic content in plant samples

The content of total phenolic compounds in plant samples was determined according to the method of Jeran et al. (2023) and Zhang et al. (2010). The Folin-Ciocalteu reagent was used for the quantitative determination and the absorbance was measured by UV/Vis spectroscopy. Aliquots of the samples (2.5 μ L) were mixed with 10-fold diluted Folin-Ciocalteu reagent (2 M, Sigma Aldrich, St. Louis, MO, USA) (12.5 μ L) and 10 μ L of 7.5% aqueous sodium carbonate solution (ACS reagent \geq 99.5%, Sigma-Aldrich, Taufkirchen, Germany). The mixture was allowed to stand for 30 minutes in a dark place at room temperature. Then the absorbance at 760 nm was determined spectrophotometrically using Nanodrop One C (Thermo Scientific, Waltham, MA, USA). Ultra-pure water and 70% ethanol solution served as a blank. The calibration curve (**Figure 2**) was generated with gallic acid standard solutions at concentrations of 2 – 90 μ g/mL. The curve was treated as a linear function with the equation: y = 0.0129x + 0.0586, and the R^2 value of the line was 0.9959. The results were expressed as amount of gallic acid per plant weight. Each sample measurement was performed in triplicate and the average value was calculated, with the error bars presented in bar graphs.

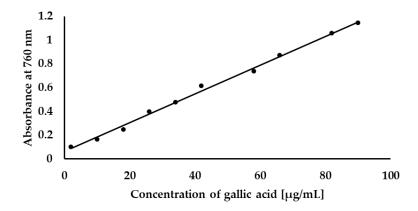


Figure 2. Calibration curve for determining the total phenolic content in plant material.

4. Results

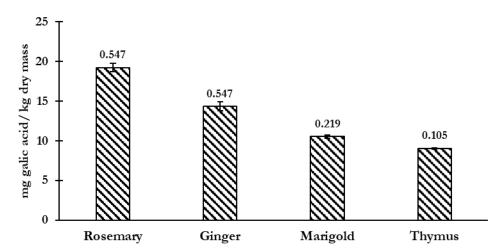
The total phenolic content was analysed using the Folin-Ciocalteu reagent and by measuring the absorbance at 760 nm. The total phenolic content of the analyzed plant samples, comparing the two evaluated extraction methods, is presented in **Figure 3**.





(a)

Water-ethanol extract



(**b**)

Supercritical carbon dioxide extract

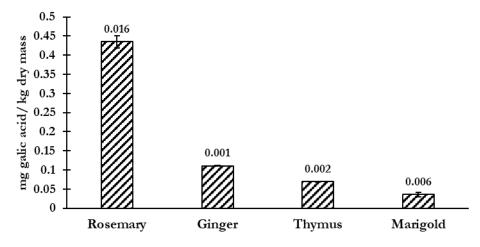


Figure 3. Total phenolic content of the (a): analyzed plant material in water – ethanol extract and (b): suprecritical carbon dioxide extract

The comparison of the two extraction methods—water-ethanol and supercritical carbon dioxide (SC-CO₂)—reveals important insights into the efficiency and selectivity of each method in extracting phenolic compounds from different plants.

For water-ethanol extracts, rosemary showed the highest total phenolic content, with a result of 19.25 ± 0.55 mg gallic acid/kg dry plant material. This high value indicates that rosemary contains a significant amount of phenolic compounds that are efficiently extracted using water-ethanol, which is known for its ability to solubilize polar compounds. Ginger followed with 14.34 ± 0.55 mg/kg, showing a slightly lower but still substantial phenolic content, with minimal variability, suggesting consistent extraction. Calendula and thyme, however, showed lower phenolic contents of 10.54 ± 0.22 mg/kg and 9.03 ± 0.11 mg/kg, respectively. While these values are lower, they still demonstrate that both plants contain polyphenols, though the concentration is inherently lower compared to rosemary and ginger. The relatively small errors in these values indicate that the method of analysis for these plants was precise, suggesting consistent results.





In contrast, the SC-CO2 extracts yielded much lower total phenolic content across all plant samples. Rosemary still had the highest total phenolic content in the SC-CO2 extracts, with a result of 0.44 ± 0.02 mg/kg, which is significantly lower than its water-ethanol extract. This dramatic reduction in phenolic extraction highlights that SC-CO2 is less effective for polar compounds as it primarily extracts non-polar substances such as terpenes and essential oils. Ginger exhibited 0.11 ± 0.001 mg/kg, which is much lower than the water-ethanol extract, suggesting that SC-CO2 cannot efficiently extract the polyphenolic compounds from ginger either. Similarly, thyme and marigold showed even lower values of 0.07 ± 0.001 mg/kg and 0.04 ± 0.006 mg/kg, respectively. These results suggest that both plants have significantly lower phenolic contents when extracted with SC-CO2, further reinforcing the idea that this method is ineffective for extracting polar phenolic compounds.

In addition, the lower errors in the SC-CO₂ isolates indicate that the analytical determination was more consistent, but the phenolic content simply could not be effectively detected.

8. Discussion and Conclusion

Recent research has increasingly focused on natural, "green" active ingredients for a wide range of scientific and therapeutic applications because plants possess the ability to synthesize numerous bioactive compounds. Among these, polyphenols stand out as compounds with significant antioxidant potential due to their chemical structure, characterized by one or more hydroxyl groups attached to a benzene ring. This structure not only enhances their free radical scavenging ability but also imparts a stronger acidic character compared to other alcohol groups, underlying their antioxidant properties (Jeran et al., 2021). Furthermore, Nisca et al. (2021) demonstrated a strong correlation between the total polyphenolic content and antioxidant activity, with higher polyphenolic concentrations leading to stronger antioxidant effects.

The difference between the two methods arises from the contrasting solubility properties of the solvents used. Water-ethanol, a polar solvent system, is highly effective at extracting polar compounds like polyphenols, which are abundant in plant tissues. In contrast, SC-CO₂, a non-polar solvent, excels at extracting lipophilic compounds such as terpenes and oils but is less effective for polar compounds. Consequently, water-ethanol extraction is better suited for obtaining phenolic-rich extracts, making it the preferred method for applications requiring high phenolic content, such as antioxidant and antimicrobial activity or nutraceutical production.

For applications requiring samples rich in antioxidants or phenolic compounds, the findings clearly indicate that water-ethanol maceration is the preferred extraction method. These results further emphasize the value of optimizing extraction techniques to harness the full potential of plant-derived polyphenols for use in research, healthcare, and other applications.

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Conflicts of Interest: The authors declare no conflict of interest.

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Review

Impact of Wind Turbines on Human Health and Safety

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Abstract:

The global challenges of climate change and energy security have highlighted the urgent need for renewable energy solutions, with wind energy making a crucial contribution. As nations strive to reduce greenhouse gas emissions and transition to sustainable energy systems, wind turbines provide a reliable and environmentally friendly alternative to fossil fuels. This article looks at the design, operation and benefits of wind turbines, but also addresses public concerns about noise emissions, infrasound, visual disturbance and electromagnetic fields. While some fears are due to misinformation and psychological factors, modern turbine designs have been shown to mitigate risks and adhere to strict safety standards. In addition, wind energy offers significant health benefits by reducing air pollution and related diseases while promoting economic growth and climate protection. By promoting education and transparent dialogue, wind energy can overcome societal barriers and cement its role in a sustainable and resilient energy future.

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Keywords: Wind energy; Renewable energy; Climate change; Health impacts; Noise emissions; Infrasound, Energy security; Sustainable development; Public perception; Wind turbines





1. Introduction

The global challenges of climate change and the ongoing energy crisis have highlighted the urgent need for a transition to sustainable energy systems. As fossil fuel resources dwindle and their environmental impact becomes increasingly severe, the push towards renewable energy has become a cornerstone of international climate and energy strategies. In Europe, initiatives such as the European Green Deal, REPowerEU and the National Energy and Climate Plans (NECPs) set out strategic objectives to tackle these challenges, including reducing greenhouse gas emissions, improving energy security and promoting sustainable economic growth (EU Monitor, 2023).

Among the strategic goals, wind energy has emerged as an important solution. The European Union aims to generate 50% of its energy from wind power by 2050 (**Figure 1**) (Wind Europe, 2024). To achieve this ambitious goal, significant challenges in the areas of energy supply, infrastructure and public acceptance must be overcome. The way to get there is through massive investment in wind energy technologies, grid integration and advances in monitoring and maintenance systems. However, a number of obstacles — including technical, economic, regulatory and social factors — threaten to hinder progress towards this goal (Wind Europe, 2021; Janipur, 2023).

Demand for electricity will more than double by 2050 with wind energy meeting 50%

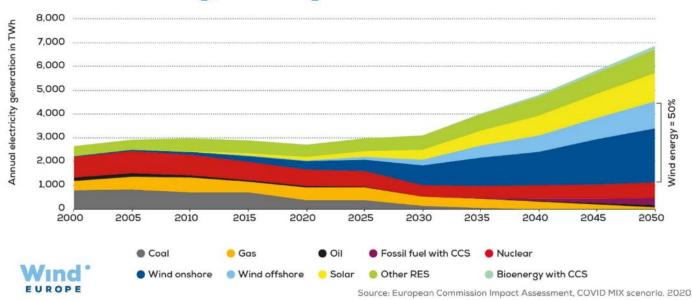


Figure 1. Wind energy meeting 50% by 2050 (Source: European Commission Impact Assessment, 2020; In: (Wind Europe, 2024)).

One major obstacle is public resistance, often fueled by misinformation and difficulties/problems of understanding about the health and well-being impacts of wind energy (Kegel and Jeran, 2025). Misinformation about visual impacts, noise, shadow flicker and psychological effects such as the nocebo phenomenon have fueled public fears and created barriers to the use of wind energy. These concerns highlight the importance of addressing the technical and social dimensions of wind energy development (Clark, 2022). This article aims to contribute to a broader understanding of wind energy by examining its role in life monitoring systems and addressing unwarranted public fears based on misinformation. By summarising the current state of knowledge and debunking common myths, the article aims to close knowledge gaps and promote a better-informed public dialogue that supports the EU's vision of a sustainable and resilient energy future (Thyssenkrupp, n. d.).





2. Wind turbines and their characteristics

Understanding the structure, materials and operation of wind turbines is important to understand their role in modern energy systems and address public concerns about their impact. By addressing the mechanics of converting wind to electricity, we can demystify the operation of wind turbines and emphasize their importance as a clean, renewable energy source (U. S. Department of Energy, n. d.). In this section, wind turbines are examined in detail as individual units in a wind farm, focusing on their design, operation and the technologies that ensure their efficiency and safety. A wind turbine converts wind energy into electricity through the aerodynamic forces acting on the rotor blades. Here is a simple explanation of how wind turbines work (U. S. Department of Energy, n. d.).

2.1. Aerodynamics and blade function

The wind flows over the wings and creates a pressure difference on each side. This difference creates lift (stronger force) and drag (weaker force), causing the rotor to turn. The spinning rotor drives a generator directly (direct drive turbines) or via a gearbox, which increases the speed of rotation (U. S. Department of Energy, n. d.). This is the common explanation but there is a more complex flow phenomenon behind it.

2.2. Electricity generation

The generator converts the mechanical energy of the rotor into electrical energy. The copper windings of the generator produce electricity by moving through an (electro)magnetic field (U. S. Department of Energy, n. d.).

2.3. Key components of a turbine

A modern Wind Turbine exists of the following key komponents in different size or small variations (U. S. Department of Energy, n. d.):

- Blades (usually made of fiberglass, they vary in size, with some modern blades being more than 100 meters long)
- hub and rotor (the hub connects the blades to the main shaft; together they form the rotor)
- gearbox or direct drive (increases speed for efficient power generation or connects the rotor directly to the generator in simpler systems)
- governor (starts the turbine at wind speeds of 11–17 km/h and shuts it down at speeds above 88–104 km/h to prevent damage)
- brake (stops the rotor for maintenance).

2.4. Tower and height advantage

Turbines are mounted on towers, often made of tubular steel, concrete and hybrid materials, to capture stronger and steadier winds at greater heights (U. S. Department of Energy, n. d.).

2.5. Pitch and yaw systems

To always achieve the optimum production outcome of the current wind situation, modern Wind Turbines use the following systems to adjust key component's angles (U. S. Department of Energy, n. d.):

- pitch system (adjusts the angle of the rotor blades to control power production and protects the turbine from too much wind by fogging the rotor blades)
- yaw system (rotates the nacelle the housing for the turbine's mechanical components to adjust to the wind direction).

2.6. Electricity transmission

Electricity flows from the generator to a transformer station via transformers that increase the voltage for efficient transmission over long distances. The voltage is then lowered for safe use in homes and businesses (U. S. Department of Energy, n. d.).

2.7. Wind farms

Several turbines are connected together at one location to form a wind farm. These farms generate electricity together and feed it into the grid to supply communities. This process





ensures that wind turbines provide clean, renewable energy with minimal environmental impact (U. S. Department of Energy, n. d.).

A detailed understanding of the design and operation of wind turbines reveals the precision and engineering behind these renewable energy systems. By examining their components, functions, and integration into wind farms, it becomes clear how wind turbines are designed to maximize energy production while minimizing environmental and social impact (U. S. Energy Information Administration, 2022).

3. General concerns about health effects

3.1. Noise Emissions from wind turbines

How is sound generated and how does it propagate? Sound is a fluctuation in air pressure that leads to vibrations of the eardrum; sound emissions from a certain source lead to fluctuations in the air particles and thus in the air pressure, and as soon as the fluctuations reach the eardrum, they are audible. Pressure fluctuations cannot only be perceived in the ear but also in other parts of the human body. The frequency of the sound is decisive for perception: the higher the frequency, the more vibrations reach the ear – the sound is perceived as a higher tone, and the unit is hertz (1 hertz is one vibration per second) (**Figure 2**) (IG Windkraft, 2022). Another important unit is sound pressure (intensity), which is measured in decibels (dB). dB corresponds to a logarithmic scale and is preferred to a linear rating scale as it reflects the behavior of the human ear — higher frequencies are perceived as louder than lower frequencies (IG Windkraft, 2022).

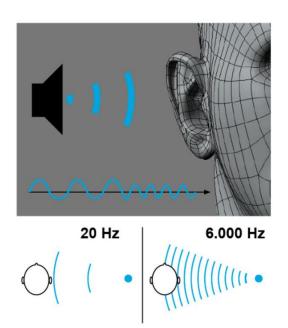


Figure 2. If a person is played a tone of 1,000 Hz, they perceive the volume of this tone as significantly louder than the volume of an actual tone of the same volume (*i.e.* with the same sound pressure) with a frequency of *e.g.* 50 Hz (IG Windkraft, 2022).

3.2. Sound sources in wind turbines and frequency ranges

The dominant source of noise in wind turbines is the rotor blades; the mechanical noise emissions from the gearbox, generator and other auxiliary units play a subordinate role in modern wind turbines and have even been reduced in recent decades (IG Windkraft, 2022). The noise is caused by the interaction of the incoming turbulent wind with the massive rotor blades, which results in a broadband noise with pronounced low-frequency sound pressure levels. The sound pressure level decreases towards higher frequencies (IG Windkraft, 2022).

Another source of noise is the interaction between the blades and the tower, which leads to low-frequency noise (infrasound source) (Oerlemans et al., 2007; Oerlemans, 2009).





Measurements with microphones along the rotor blades have shown that the main source of noise from modern wind turbines is primarily the outer part of the rotor blades, but not directly the tip (**Figure 3**) (Oerlemans et al., 2007).



Figure 3. Location and quantification of noise sources on a wind turbine (Oerlemans et al., 2007).

The measurement also revealed that the noise sources are distributed asymmetrically in the rotor plane, although the blades emit a constant sound pressure level during a complete rotation. The descending blade is perceived louder by an observer on the ground than the ascending blade. This is referred to as a hissing noise (Oerlemans et al., 2007). During one rotation of a three-bladed wind turbine, three different hissing noises can be perceived. For an observer standing directly in front of the wind turbine, the strongest hissing noise occurs at around the 3 o'clock position of the blade (Oerlemans et al., 2007).

3.3. Available methods and technical standards

In the case of wind energy, in addition to emissions directly at the turbine, noise immissions are also important. During the planning phase, the most exposed buildings/neighbourhoods are identified and an immission forecast is carried out (Schlömicher, 2013). The immission forecast is based on the loudest operating state of the turbine. In addition, the previous pollution from other technical systems or operations is taken into account. The immission forecast also takes into account other aspects such as buildings, vegetation or orography, which can influence sound propagation (BWE, 2018). Once the forecast has been completed and the sound levels have been determined, they are compared with specific limit values which may result from federal laws, state laws, technical standards, etc. These laws or standards may also contain instructions for determining sound immissions. For example, the WHO recommends that noise emissions from wind turbines near general residential areas should not exceed a value of 45 dB Lden: The day-evening-night level is a key value for measuring long-term noise exposure. It represents the average sound level over a 24-hour period, with different weighting factors for day, evening and night to take account of people's different sensitivity to noise at different times of the day (European Environment Agency, 2001). During the day (24-hour average of noise emissions) in order to avoid adverse health effects. They do not give recommendations for the night time, but thresholds can be found in several other sources such as the "TA Lärm", which is the technical set of rules and regulations commonly used in Germany (BWE, 2018).

3.4. Influence of noise emissions from wind turbines on human health

According to van Kamp & van den Berg (2021), a literature search was conducted comprising 12 reviews and 57 original articles (the literature was selected following a





screening of three scientific databases and the selection of publications according to a specific selection procedure which ensured a sufficiently high quality), which was also published in the report "Health effects related to wind turbine sound: an update", National Institute for Public Health and the Environment, 2020 (van Kamp & van den Berg, 2020). According to this research noise from wind turbines is low compared to other sources such as traffic (road, rail and air) or industry and is generally below 45 dB (van Kamp & van den Berg, 2021). Nevertheless, the noise from wind turbines is perceived as more disturbing than that from many other sources with the same sound level. Living near a wind turbine or listening to wind turbines can lead to chronic annoyance for residents. For other health effects such as sleep disturbances, insomnia or effects on mental health, the evidence is contradictory or insufficient. There is no evidence that the low-frequency component affects residents in any other way than the typical sound or that infrasound, which is far below the hearing threshold, can have an impact. The level and amplitude modulation of all wind turbine noise are the main causes of increased annoyance, not low-frequency sound or infrasound (van Kamp & van den Berg, 2021).

Although low-frequency noise emissions cannot be heard, they may still lead to annoyance, but the link between wind turbines and low-frequency noise has not been established. The causality and directionality of these effects remain unproven (McKenna at al., 2025).

3.5. What is infrasound?

Infrasound is an everyday phenomenon. It is in general sound with a frequency below 20 Hz (Ratzel et al., 2016). Almost every sound emission also contains infrasound components – the decisive factor is the intensity. In wind turbines the infrasound is generated during rotation: The rotor blades generate turbulent air flow and when the rotor blade passes the tower this turbulent flow is interrupted resulting in infrasound. For many sound sources such as traffic, wind itself, compressors (of refrigerators), motors, etc., the infrasound is more intense than for wind turbines. A comparison between a wind turbine and a car (inside with back windows open and with all windows open) is shown in **Figure 4**, where the intensity of infrasound emissions of the car is significantly higher than the intensity of the wind turbine. Also, the infrasound emitted by wind turbines is lower than the perception treshold (Fachagentur wind und solar, n. d).

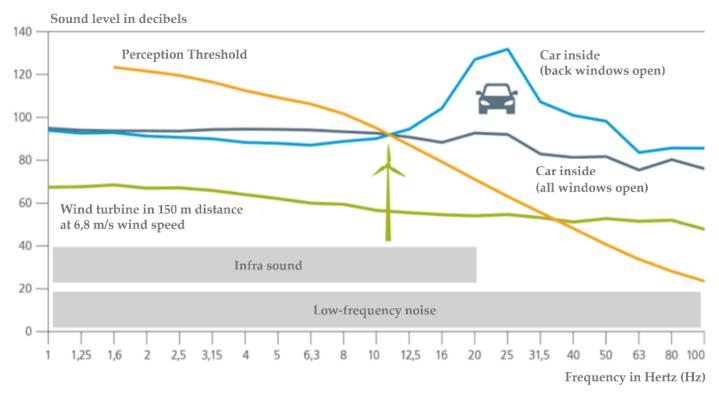


Figure 4. The other sound sources (car etc., are significantly louder than the wind turbine as a sound source (Fachagentur wind und solar, n. d)).





3.6. Electromagnetic fields (EMFs): How are they generated?

EMFs are made up of magnetic and electric fields. They are created when electrical voltages and currents change, causing an electromagnetic wave to propagate spatially. EMFs can be generated in nature or in technical devices and systems, *e.g.* in televisions, hair dryers or wind turbines. They can be characterized by their frequency, measured in Hertz (Hz), and their magnetic flux density, measured in milliGauss or Tesla (mG or T). Various sensors, such as Hall sensors, are available for measuring the EMF (McCallum et al., 2014).

Influence on human health and effect of EMF in wind turbines on people living near the wind farm. Several publications show that EMFs are only detectable in the immediate vicinity of wind turbines. In the study "Measuring electromagnetic fields (EMF) around wind turbines in Canada: is there a human health concern?« by McCallum et al. (2014), measurements of electromagnetic fields (EMF) were carried out and the authors found that the EMF could no longer be distinguished from the background within 2 m of the base. EMF levels measured within 2 to 3 m of the wind turbines were comparable to or lower than magnetic field measurements near typical household electrical appliances (Figure 5) (Knopper et al., 2014).

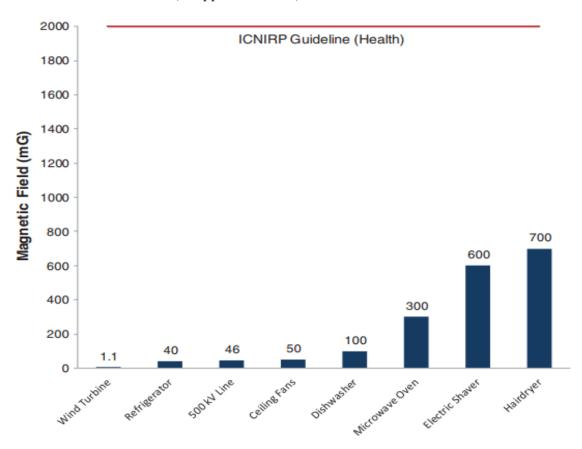


Figure 5. The picture shows that the EMFs are located significantly below the threshold given by the International Commission on Non-Ionizing Radiation Protection (ICNIRP) (Knopper et al., 2014).

4. Visual & psychological effects of wind farms

Wind turbines' visual and psychological impacts have been the subject of various scientific studies.

4.1. Visual impacts

Research, including seventeen primary studies on health effects due to visual characteristics of wind turbines, found no evidence of serious adverse health effects





reported for visual exposures. Most studies primarily investigated the frequency of annoyance to residents living up to 1200 m from the nearest wind turbine (Meyer, 2022). Depending on the type of visual exposure (*e.g.* shadow flicker, reflection) the prevalence of annoyance varied between 7 and 31% (Meyer, 2022). The pooled prevalence of high annoyance was the same for shadow flicker and altered visibility from wind turbines (6%) (Freiberg, 2019). If there is a possibility that impacts from shadow flicker may occur, mitigation measures can minimize or eliminate these impacts. Shadow flicker impacts can be reduced either by optimizing the placement of WTGs, by screening the line of sight between receptors and problematic WTGs (*e.g.* through landscaping), or by restricting the operation of WTGs during the critical periods when shadow flicker impacts are most likely to occur (Meyer, 2022).

4.2. Psychological effects & nocebo effects

The nocebo effect is also a psychological phenomenon in which negative expectations of a particular stimulus or situation lead to the perception of negative effects, even if there is no direct physical cause. In the context of wind turbines, this effect has been studied in detail to understand why some people report health complaints in the vicinity of wind farms, even though there is no conclusive evidence that wind turbines cause direct damage to health (Crichton et al., 2014).

"Wind turbine syndrome" refers to symptoms reported by people living near wind farms. These symptoms include headaches, dizziness, nausea, sleep disturbances, anxiety and other non-specific complaints. Studies such as the one published in *Noise & Health* in 2014 (Rubin et al., 2014), suggest that these symptoms are not directly caused by physical aspects of wind turbines (*e.g.* noise or infrasound), but may be related to the nocebo effect, where the belief that wind turbines are harmful produces actual physiological or psychological discomfort (Rubin et al., 2014).

4.3. Physical safety and health risks

Although wind turbines are generally safe and meet stringent safety standards, potential risks such as ice throw, mechanical failure and fire hazards are occasionally identified. Ice throw occurs when ice accumulates on the turbine blades in cold conditions and detaches during rotation, which can pose a localized safety hazard. However, modern turbine designs have advanced monitoring systems that detect and mitigate such conditions, significantly reducing the likelihood of accidents. Rare mechanical failures, including blade fractures, are also cited as a risk (Rubin et al., 2014). However, due to strict safety protocols, regular maintenance schedules and advanced engineering designs, these events are extremely rare. Wind turbines must comply with international safety standards while minimizing risks to the surrounding environment. In addition, potential fire risks are mitigated by fire-resistant materials and monitoring systems. Modern wind farms have advanced fire protection systems, including heat sensors, automatic fire extinguishing systems and fire-retardant coatings (Rubin et al., 2014). Detailed risk assessments are carried out prior to construction, including analyzing the potential for ignition and developing appropriate countermeasures. Wind farms in cold climates have de-icing technologies such as heating elements on the rotor blades or coating systems with substances that prevent ice formation. The use of these technologies significantly reduces the risk of ice drift. In areas with low temperatures, safety markings are used and protection zones are set up to protect people and property. Studies show that the risk of injury is extremely low if these measures are observed. Wind farms are designed with local weather conditions in mind and built to withstand extremely strong winds and other weather extremes. For example, the construction may be made of robust materials that are resistant to vibrations and weather influences. Studies show that the vibrations emitted by wind farms have no significant impact on surrounding properties or buildings. Modern turbines use vibration damping technologies to ensure stability and minimal impact on the surrounding area (Rubin et al., 2014).





5. Positive effects of wind energy on human health and beyond

Wind energy is one of the healthiest energy sources available today. Its use offers numerous health and environmental benefits, making it a key component of the transition to sustainable energy systems. Wind energy offers a sustainable and renewable energy solution and significant health benefits by reducing pollution, lowering healthcare costs, promoting community development and contributing to global ecological well-being (Local Government Association, n. d.).

5.1. The elimination of greenhouse gas emissions

Burning fossil fuels is one of the main causes of global warming, while renewable energy generates electricity without emitting carbon dioxide or other pollutants. According to the International Renewable Energy Agency (IRENA, 2022), switching to renewable energy could reduce carbon dioxide emissions by up to 70% by 2050, underlining its crucial role in combating climate change (IRENA, 2022). Unlike fossil fuels, wind turbines generate electricity without emitting greenhouse gasses. This freedom from emissions is crucial, as burning fossil fuels releases toxic and carcinogenic substances into the atmosphere that contribute to serious health problems. In the United States, for example, coal pollution is linked to around 50,000 premature deaths each year (IRENA, 2022).

5.2. Reducing health costs

The pollutants produced by coal-fired power plants have significant health impacts and lead to considerable economic burdens. In Australia, the cost of the health impacts of this pollution is estimated at AUD 2.6 billion per year (Biegler, 2009). Switching to wind energy can reduce these health risks and associated costs by providing a cleaner alternative (Climate Council, 2015).

5.3. Promoting the wellbeing of rural communities

Wind energy projects often provide economic opportunities for rural areas. Wind farm development can boost local economies through job creation and infrastructure investment. In addition, renewable energy initiatives empower rural communities to become self-sufficient in energy and reduce dependence on fossil fuels (Ionescu, 2024).

5.4. Ccontribution to global environmental health

The use of wind energy reduces carbon emissions, which plays an important role in combating climate change. Stabilizing the climate is crucial to prevent the escalation of extreme weather events and protect ecosystems. The use of renewable energy sources such as wind energy is a proactive step towards securing the long-term health of our planet (Local Government Association, n. d.).

5.5. Confirmation of safety by health authorities

Extensive reviews by health organizations have found no consistent evidence of a link between wind farms and adverse effects on human health. For example, the Australian government's National Health and Medical Research Council concluded that wind turbines pose no health risks, underscoring the safety of wind energy as an energy source. (Climate Council, 2015).

5.6. Energy security: using renewable energy sources for a more stable and cleaner energy supply Energy security has become a cornerstone of sustainable development, with renewable energy sources playing a central role in creating a reliable, clean and resilient energy system. Traditional energy systems that rely heavily on fossil fuels are vulnerable to price volatility, geopolitical conflict and resource depletion. Renewable energy sources mitigate these risks by enabling localized and diversified energy production. For example, solar and wind power plants can be deployed in different regions, reducing dependence on centralized, disruption-prone energy grids (IRENA, 2022). This decentralized approach increases the resilience of grids and ensures the availability of energy even in times of crisis. In addition, renewable energy technologies are not subject to the same supply chain constraints as fossil fuels. Unlike coal, oil or natural gas, which have to be extracted and transported over long distances, renewables use inexhaustible resources such as sunlight





and wind. This inherent stability enables countries to achieve greater energy independence and become less dependent on external market fluctuations. (IRENA, 2022).

5.7. Social acceptance of wind energy

The study "Social acceptance of wind energy in urban landscapes" by Westerlund (2020) examines the factors that influence the social acceptance of wind energy in urban environments. It highlights that positive perceptions are often associated with increased awareness and understanding of the benefits of wind energy, suggesting that educational initiatives can improve acceptance. The main findings are that there are different types of acceptance: Protagonists are highly supportive of urban wind energy projects; centrists are neutral or ambivalent towards such projects; and antagonists are opposed to the introduction of wind energy in urban areas (Westerlund, 2020). An important finding for project developers and political decision-makers is that active participation of the population in planning and decision-making can increase acceptance. Considering optimal distances from residential areas can also allay concerns about proximity. Tailored information and participation strategies for different demographic groups, e.g. by gender, can improve public awareness and support (Westerlund, 2020).

6. Conclusion

Wind turbines are one of the cornerstones of the global transition to sustainable energy systems and offer numerous environmental, economic and health benefits. While concerns have been raised about noise emissions, infrasound, ice throw and visual impacts, evidence shows that these issues are either negligible or effectively mitigated by modern technologies and stringent safety standards. Misinformation and psychological factors, such as the nocebo effect, often contribute to public concern and emphasise the need for education and transparent communication.

The health benefits of wind energy, such as the reduction of air pollution and associated diseases, far outweigh the potential risks. In addition, wind energy contributes to energy security, job creation and climate change mitigation, making it an indispensable element of a sustainable and resilient energy future. By removing technical, legal and social barriers and promoting an informed public dialogue, the potential of wind energy can be fully realised in line with global efforts to combat climate change and protect public health. This article aims to contribute to a broader understanding of wind energy, make sense of the often misunderstood theories of health impacts, and highlight the benefits of wind energy for policy makers and individuals, that can also contribute to stress reduction.

Conflicts of Interest: The authors declare no conflict of interest.

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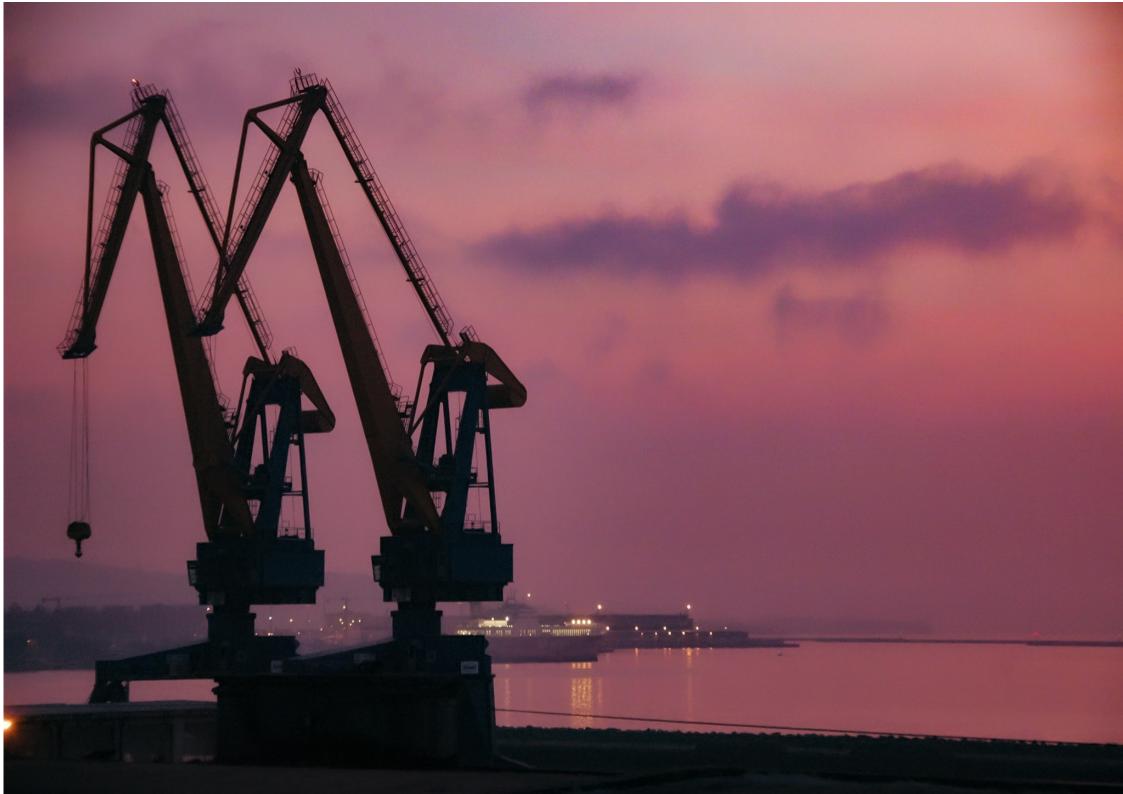
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Research

Polymer Materials in Orthotics and Prosthetics

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Abstract:

Polymer materials play a crucial role in the design and fabrication of orthotic and prosthetic devices, enhancing functionality, comfort, and aesthetics and thus significantly impacting patient mobility and quality of life. Their versatility, characterized by lightweight properties, biocompatibility, and customization potential, has transformed the field, particularly with the introduction of advanced manufacturing techniques such as 3D printing. Polymer materials are classified into thermoplastics, thermosetting plastics, specialty polymers, composites, and foams, each serving specific mechanical and structural functions. Thermoplastics enable reshaping during fittings, while thermosetting plastics provide necessary rigidity. High-performance materials like polyether ether ketone (PEEK) and carbon fiber composites offer superior strength and lightweight benefits, making them ideal for advanced applications. The integration of smart technologies further enhances prosthetic functionality, allowing for adaptive and responsive designs. However, challenges remain, including material durability, manufacturing waste, and economic barriers to adopting novel technologies. Research efforts focus on developing biodegradable and smart materials to improve sustainability and performance. The continued evolution of polymer materials in orthotics and prosthetics is poised to drive innovation, offering more effective and patient-centered solutions for individuals with mobility impairments.

Keywords: Polymer materials; orthotics, prosthetics; biocompatibility; smart materials; 3D printing





1. Polymer materials

1.1. Thermoplastics

Thermoplastics are a significant category of polymers used in orthotics and prosthetics due to their formability when heated. These materials become mouldable at specific temperatures and can be reheated multiple times, allowing for adjustments during fittings. Thermoplastics are categorized into low-temperature and high-temperature materials based on their melting points. Low-temperature thermoplastics, which can be moulded at temperatures less than 149°C, are often used for orthotic devices that provide temporary support and protection, such as Kydex, Orthoplast, and Polysar (Kogler et al., 2020). High-temperature thermoplastics require heating to higher temperatures and are typically moulded over a positive model of the patient's limb. Common examples include polyethylene, polypropylene, and acrylic (Kogler et al., 2020; Professional Plastic, 2024).

1.1.1 Low-Temperature Thermoplastics

Low-temperature thermoplastics can be molded directly onto a patient's limb without the need for casting, significantly reducing the time from measurement to the finished product. These materials are particularly suited for spinal and upper extremity orthoses, where flexibility and quick adjustments are necessary. Basic tools, such as an electric frying pan or a heat gun, are sufficient for their application, making them convenient for practitioners (Kogler et al., 2020).

1.1.2 High-Temperature Thermoplastics

High-temperature thermoplastics, including polycarbonate and acrylonitrile butadiene styrene, are utilized in scenarios requiring greater structural strength and durability. Unlike low-temperature variants, these materials cannot be reshaped once cooled, making them suitable for applications where permanent shapes are needed, such as in prosthetic sockets and components (Kogler et al., 2020).

1.2. Thermosetting Plastics

Thermosetting plastics are another essential category in orthotics and prosthetics. These materials are applied in a liquid state over a positive model and are chemically cured to maintain their shape. Common thermosets include acrylic, polyester, and epoxy, which are often laminated with various fabrics to enhance structural properties. Unlike thermoplastics, thermosetting plastics cannot be reheated for reshaping, which makes them less flexible but advantageous for producing rigid orthoses and prostheses (Kogler et al., 2020; Professional Plastic, 2024).

1.3. Specialty Polymers

Advanced polymers like PEEK (polyether ether ketone) and PEKK (polyether ketone ketone) are gaining traction in the field of orthotics and prosthetics due to their biocompatibility and mechanical strength. These materials are particularly useful for implantable devices and structural components requiring enhanced durability. Their ability to be processed through techniques like CAD/CAM milling and 3D printing opens new avenues for customization in prosthetic design (Ardelean et al., 2023; Huber et al., 2023; Thibodeau et al., 2022).

1.4. Composites

Composites combine two or more materials to produce a substance with enhanced performance characteristics. In orthotics and prosthetics, composites often involve reinforcing fibers such as fiberglass, carbon fiber, or Kevlar embedded in a matrix material like thermosetting plastics. These combinations provide superior strength, lightweight properties, and resilience against stresses compared to the individual components alone. Carbon fiber, in particular, is known for its high strength-to-weight ratio, making it a popular choice for high-performance orthotic and prosthetic devices (Kogler et al., 2020).





1.5. Polyurethane Foams

Polyurethane open-cell foams serve as alternative materials for top covers in foot orthoses. They are favored for their excellent shock absorption and heat dissipation properties, contributing to enhanced comfort for users. Some commercially available options include Poron, PPT, and Vylite (Kogler et al., 2020).

2. Properties of Polymer materials

Polymer materials play a critical role in the design and fabrication of orthotic and prosthetic devices, owing to their diverse properties that influence functionality, comfort, and durability.

2.1. Durability and Fatigue Resistance

Durability, particularly fatigue resistance, is essential for polymer materials used in orthotics and prosthetics. This property determines the material's ability to withstand repeated cycles of loading and unloading during typical activities, which can compromise strength and increase the risk of failure or fracture (Kogler et al., 2020). The interface between materials with differing characteristics can exacerbate fatigue resistance challenges, making it crucial to select compatible materials.

2.2. Density and Weight Considerations

The density of a polymer material significantly impacts the overall weight of an orthotic or prosthetic device. Lighter materials are generally preferred to minimize energy costs during functional activities; however, achieving optimal strength, durability, and fatigue resistance may necessitate using denser materials. For example, some advanced polymers, such as Kevlar, provide tensile strength properties that are significantly greater than those of steel, allowing for lighter designs without sacrificing strength (Kogler et al., 2020).

2.3. Corrosion Resistance and Moisture Imperviousness

Corrosion resistance is another vital characteristic of polymers in orthotic and prosthetic applications. Many materials used retain heat, which can exacerbate issues related to perspiration and moisture retention. Materials that are impervious to moisture are easier to clean and maintain compared to porous options, making them more suitable for devices used in potentially wet environments or by patients with incontinence concerns (Kogler et al., 2020).

2.4. Stiffness and Flexibility

The stiffness of polymer materials is crucial in determining their suitability for various applications within orthotics and prosthetics. Stiff materials provide stability and support, making them ideal for devices like fracture braces and rigid prosthetic frames. Conversely, flexible materials are necessary for components that must conform to body segments, such as in a posterior leaf-spring ankle-foot orthosis (AFO) or a flexible transfemoral prosthetic socket (Kogler et al., 2020).

2.5. Ease of Fabrication

The ease of fabrication is a practical consideration when selecting polymer materials. Thermoplastics, for example, can be reheated and reshaped multiple times, allowing for minor adjustments during fittings. Techniques like thermoforming, where sheets of thermoplastic are heated and shaped over molds, are commonly employed in the production of orthotic and prosthetic devices. In contrast, thermosetting materials require chemical curing, limiting their ability to be reshaped once set (Kogler et al., 2020).

2.6. Mechanical Properties and Applications

The mechanical properties of polymers, including strength, ductility, and yield strength, are critical in ensuring the reliability of orthotic and prosthetic devices. High strength-to-weight ratios are particularly important for components subjected to repetitive loading. Low-alloy and high-alloy steels are also utilized in conjunction with polymer materials for certain joint components, balancing the mechanical properties of both materials for optimal performance under load (Kogler et al., 2020).





3. Applications in Orthotics

Orthoplast is a material commonly used in orthotics, favored by occupational therapists, orthotists and prosthetists, orthopedic technicians, and physicians due to its direct application on patients, which eliminates the need for a negative impression. It is made from thermoplastic polymers, typically polyolefins like polyethylene (PE) or polypropylene (PP). These materials can be heated to become soft and flexible, allowing them to be molded around body parts (e.g., limbs) to create protective splints, braces, or orthotics. Once the material cools, it hardens and retains its shape, providing solid support and protection for injured or weakened areas of the body. Its thermoplastic properties make it easy to mold, while still maintaining strength and stability after cooling, making it ideal for orthopedic applications where flexibility and support are essential. This material is frequently employed in the fabrication of orthoses for the treatment of fractures, providing ease of molding and adaptability (Showers & Strunck, 1984; Drijkoningen et al., 2018).

3.1. Characteristics of Orthoplast

Orthoplast has the capability to be custom molded over two positive models, allowing for flexibility in design. While it is often utilized in various applications, it is not always the preferred choice due to its comparatively shorter life expectancy when juxtaposed with other more durable flexible sheet plastics. Despite this, orthoplast remains a popular choice for certain applications, such as the orthotic treatment of scoliosis using Milwaukee style orthoses or T.L.S.O. "body jackets" (Karimi & Kavyani, 2015). The material's ability to be easily adjusted with a heat gun post-fabrication enhances its usability, particularly for patients who require modifications after initial fitting.

3.2. Advancements in Orthotic Technology

The integration of thermo-plastics has marked significant advancements in orthotic practices. The process typically involves taking a negative impression of the body segment, followed by laboratory assembly to create a custom-molded orthotic device tailored to the patient's specific disability.

3.3 Other Materials and Techniques

In addition to orthoplast, other materials such as ortholene are also utilized in orthotics, particularly for posterior leaf spring ankle-foot orthoses (AFOs) designed for patients with weak dorsiflexors. However, the durability of ortholene has been questioned, leading to the exploration of subortholene, a newer material reported to offer improved longevity (Gatt et al., 2016).

4. Applications in Prosthetics

The application of polymer materials in prosthetics has revolutionized the design, functionality, and comfort of artificial limbs. Modern prosthetic devices leverage advanced polymers and composites to enhance user experience and performance.

4.1. Lightweight and Durable Design

One of the key benefits of polymer materials is their lightweight nature. Traditional prosthetics made from wood or metal often resulted in cumbersome and uncomfortable devices. In contrast, polymers like carbon fiber composites and medical-grade silicone provide significant weight reduction while maintaining structural integrity. This improvement allows users to wear their prosthetics for extended periods without discomfort, enabling a more active lifestyle (Howington, 2023).

4.2. Customization and Fit

Customization is essential in prosthetics, as each user's needs can vary widely based on factors such as the type of amputation and personal preferences. The use of 3D printing technology allows for rapid production of tailored prosthetic limbs that fit the unique contours of the user's body. This adaptability is crucial for enhancing the functionality and aesthetic appeal of prosthetic devices (Howington, 2023; Sakib-Uz-Zaman & Khondoker, 2023). Moreover, the ability to create complex designs with polymers can lead to improved





fit and performance, addressing the individual needs of each patient (Choo et al., 2020; Huber et al, 2023; Sakib-Uz-Zaman & Khondoker, 2023; Thibodeau et al., 2022).

4.3. Advanced Features

Recent advancements have seen the integration of smart technologies into prosthetic devices. Polymers facilitate the development of bionic and myoelectric prosthetics that utilize sensors to detect electrical signals from the user's residual muscles. This enables the prosthetic to respond to the user's intentions, allowing for more natural movements such as grasping and walking (Grace Prosthetic Fabrication, 2024; Howington, 2023, Shallal et al., 2019). Furthermore, incorporating antimicrobial properties into polymer materials helps reduce the risk of infection, especially for users in warm and humid climates (Grace Prosthetic Fabrication, 2024).

4.4. Cost-Effectiveness

In addition to performance benefits, polymer materials can also contribute to cost reductions in prosthetic manufacturing. The affordability of 3D printing and the low material costs associated with polymers make it possible to produce high-quality prosthetics at a fraction of the price of traditional methods. Reports indicate that 3D-printed prosthetic devices can be 56–95% less expensive than their laminated counterparts, making them accessible to a broader range of individuals (Sakib-Uz-Zaman & Khondoker, 2023; Huber et al, 2023; Thibodeau et al, 2022).

4.5. Future Directions

The ongoing research and development of polymer materials in prosthetics indicate a promising future for the field. Innovations such as topologically optimized designs and smart prosthetics are expected to further enhance the functionality and usability of these devices. As manufacturers continue to explore advanced polymers and composites, the potential for even greater improvements in comfort, safety, and performance is substantial (Grace Prosthetic Fabrication, 2024).

5. Advantages of Using Polymer Materials

Polymer materials have become increasingly prominent in the field of orthotics and prosthetics due to their unique properties and advantages.

5.1. Cost-Effectiveness

Recent developments in additive manufacturing (AM) have demonstrated that polymer-based prosthetic devices can be produced at significantly lower costs compared to traditional methods. Reports indicate that 3D-printed prosthetic devices can be 56-95% less expensive than those fabricated using conventional techniques (Sakib-Uz-Zaman and Khondoker, 2023). This reduction in cost, coupled with improved manufacturing efficiency, can increase access to these life-enhancing technologies for individuals who may otherwise be unable to afford those (Huber et al., 2023).

5.2. Durability and Fatigue Resistance

One of the primary benefits of polymers, such as polyethylene and polypropylene, is their excellent durability and fatigue resistance. These materials are capable of withstanding repeated cycles of loading and unloading, which is crucial for devices that endure significant mechanical stress during daily activities (Nagarajanet al., 2023). The long fatigue life of polyethylene, for example, makes it suitable for various applications, including prosthetic sockets and orthotic components, ensuring longevity and reliability.

5.3. Lightweight Design

The density of polymer materials allows for the creation of lightweight devices that enhance user comfort and mobility. Since lighter prosthetics reduce the physical burden on the user, polymers are often preferred over traditional metals, striking a balance between strength and weight. This is particularly important for patients who rely on these devices for mobility, as excessive weight can impede movement and increase fatigue.





5.4. Customizability and Design Flexibility

Polymers can be easily formed, die cut, and machined, allowing for a high degree of customizability in orthotic and prosthetic design (Professional plastics, 2024). Advanced manufacturing techniques such as 3D printing have further enhanced this ability, enabling the production of highly customized implants that ensure optimal fit and performance for individual patients. This customization is critical for addressing the unique needs of each user, potentially leading to better outcomes and increased satisfaction (Huber et al, 2023; Thibodeau et al, 2022).

5.5. Biocompatibility

Another significant advantage of polymer materials is their biocompatibility. Many polymers are well-tolerated by the human body, minimizing the risk of adverse reactions when used in orthopedic devices. This makes them suitable for both temporary and long-term applications, which is essential in the development of prosthetics and implants that integrate seamlessly with biological tissues.

6. Challenges and Limitations

The integration of polymer materials in orthotics and prosthetics presents several challenges and limitations that practitioners and manufacturers must navigate to optimize device efficacy and patient satisfaction. These issues can broadly be categorized into material properties, manufacturing processes, and economic factors.

6.1. Material Properties

The selection of appropriate polymer materials is crucial for the performance of orthotic and prosthetic devices. Durability, fatigue resistance, and corrosion resistance are key considerations. For instance, materials must withstand repeated cycles of loading without succumbing to failure, particularly in applications where high stress is anticipated, such as lower limb devices (Caselli, 2004). The challenge lies in balancing the need for lightweight materials against the requirements for strength and durability, as denser materials may provide necessary mechanical support but can compromise patient comfort and energy efficiency during use. Additionally, the viscoelastic properties of certain polymers can complicate the design process. For example, materials like Sorbothane and Viscolas, which exhibit stress relaxation and creep, must be carefully considered to ensure they provide adequate support while allowing for necessary movement and comfort (Caselli, 2004).

6.2. Manufacturing Processes

The traditional methods of fabricating orthotic and prosthetic devices often involve manual processes that can produce significant waste and pose health hazards due to dust from cutting and grinding materials like fiberglass and carbon fiber (Sakib-Uz-Zaman & Khondoker, 2023). These conventional approaches not only increase costs but also create environmental concerns. Recent advancements in additive manufacturing (AM) have shown promise in addressing these limitations by enabling more efficient use of materials and reducing waste. However, the transition to AM is not without its own set of challenges, including the need for substantial investment in new technologies and training for personnel to operate these advanced systems (Sakib-Uz-Zaman and Khondoker, 2023).

6.3. Economic Factors

Economic constraints represent a significant barrier to the widespread adoption of innovative materials and manufacturing technologies in the O&P field. Many small and medium-sized facilities may find it economically unfeasible to invest in expensive 3D printing equipment, which can hinder their ability to provide cutting-edge solutions. While some practices have begun outsourcing fabrication to larger manufacturers, this approach may not be sustainable or practical for all providers (Sakib-Uz-Zaman & Khondoker, 2023). The cost-benefit analysis of adopting advanced manufacturing techniques continues to be a pivotal factor that influences decision-making in the industry.





7. Recent Innovations and Trends

The field of orthotics and prosthetics has seen significant advancements in recent years, particularly through the application of innovative polymer materials. These developments not only enhance the functionality and comfort of prosthetic devices but also improve patient outcomes.

7.1. Advancements in Polymer Technologies

Recent innovations in polymers are transforming orthopedic medicine. Key breakthroughs include the introduction of bioabsorbable and shape-memory polymers, as well as antimicrobial materials. These advanced polymers are paving the way for more effective and sustainable orthopedic treatments, ensuring better integration with the body and reducing the need for additional surgeries.

7.2. 3D Printing and Customization

One of the most impactful technologies in this domain is 3D printing, which allows for the creation of highly customized implants and prosthetics. This technology enables the production of devices tailored to the unique anatomy of each patient, significantly improving fit and performance. Moreover, the ongoing research into new thermoplastics and composites aims to further enhance the strength, flexibility, and antimicrobial properties of 3D-printed orthotic devices (Choo et al., 2020; Huber et al., 2023; Thibodeau et al, 2022).

7.3. Smart and Biodegradable Polymers

The exploration of smart polymers—materials that can respond to external stimuli such as temperature and pH—holds great promise for future prosthetic development. These materials can adapt over time to meet the changing needs of patients, thus enhancing the usability of prosthetic devices (Shallal et al., 2019; Nagarajanet al., 2023). Additionally, research into biodegradable polymers is advancing, which aims to create prosthetic components that safely degrade within the body, thereby improving patient recovery and reducing the need for surgical removal of temporary implants.

8. Challenges and Future Directions

Despite the promising advancements, several challenges remain in the use of polymers for prosthetic devices. Material degradation due to prolonged exposure to physiological conditions poses risks to the durability and effectiveness of these devices. Additionally, ensuring a perfect fit through customization remains a critical aspect of prosthetic design. However, the continued evolution of polymer technology, coupled with innovative manufacturing techniques such as CAD/CAM milling and light-curing, presents exciting opportunities for the future (Ardelean et al., 2023).

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Review

Biocompatible Materials in Dentistry

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Abstract:

This article presents a mini-review of biocompatible materials used in dental medicine. The main characteristic of biomaterials is that they do not cause any immunological reaction in the host, their use on living tissue is suitable. In general, biomaterials represent a broad field that is constantly being upgraded. This article presents four main groups of biomaterials (metals and metal alloys, dental ceramics, polymers, and composites), the most frequently used materials in dentistry, their improved versions, and innovations of biomaterials. The integration of novel biomaterials, such as nanoparticles, is also dis-cussed. Also, the article describes glass-ionomer cement, an irreplaceable material in pedi-atric dentistry, impression materials, sealers, luting materials, and other materials repre-senting essential parts of clinical work

Keywords: Biocompatibility; Metals and metal alloys; Dental ceramics; Polymers; Resin composites; Nanomaterials





1. Introduction

The main property that allows the use of a biomaterial on living tissue is biocompatibility, which refers to the material's ability to perform its intended function without causing harmful local or systemic reactions in the recipient of the material (Perroti et. al., 2017). Biomaterials in modern medicine have a huge range of applications such as biosensors, medical implants, systems that promote the healing or regeneration of human tissues, drug-delivery systems, etc. The applicability of biomaterials is defined by a long list of their characteristics. The most important properties of biomaterials are biocompatibility (non-toxic, non-carcinogenic, non-allergenic, etc.), desirable physical, chemical, and mechanical properties (tensile and yield strength, elastic modulus, corrosion, wear, and fatigue resistance), stable durability for some time, processable with available techniques (CAD/CAM systems, 3D printing,), sterilizable without changing his structure, accessibility, etc. (Kiran & Ramakrishna, 2021).

2. Topical materials in dentistry

The largest number of different materials in dentistry is applied in the field of prosthetic and restorative dentistry. In general, we can divide materials in dentistry into four main groups: metals and metal alloys, ceramics, polymers, and resin composites. Well-known aesthetic materials resin composites, which have replaced long-used amalgams, today are used for direct, indirect, and provisional restorations, veneers, core buildups, cement, and sealants (Sakaguchi, 2019). The properties of composites have changed significantly in a positive way by infiltrating the latest technology of using nanoparticles in the production of composites. The contemporary group of materials also includes compomers, which combine the advantages of composites and glass-ionomer cement (Sakaguchi & Mitra, 2019). The composition of composites also includes polymers, which have a major role in most areas of prosthetic and restorative dentistry. They are also components of impression materials, resin, glass-ionomer cement, dentin bonding agents, which enable a strong bond between the composite and the tooth structure (Sofan et al., 2017), pit and fissure sealants, and root canal sealants. The most commonly used polymer material is polymethyl methacrylate (PMMA). PMMA is a strong, tough, lightweight material (Rokaya et al., 2018), which is used for the fabrication of artificial teeth, dentures and denture bases, obturators, orthodontic retainers, occlusal splints, temporary or provisional crowns, and for repairment of dental prostheses (Zafar, 2020). To reduce bacterial and fungal colonization, different nanoparticles are incorporated into PMMA, such as silver or platinum nanoparticles (Rokaya et al., 2018). Polyetheretherketone (PEEK) has excellent properties and close elastic modulus to the bone, which their use makes possible in many fields of dentistry such as orthodontics for orthodontic wires, maxillofacial surgery for obturators (Hamsho et al., 2022), prosthodontics for fixed and removable prostheses, temporary abutments, etc. (Parate et al., 2023). Polymeric films (PMFs) are also used in dentistry. For example, coating the surface of hydroxyapatite, which is part of the tooth structure, with a copolymer containing 50% MPC (2-methacryloyloxyethyl phosphorylcholine) inhibits bacterial adhesion to the tooth surface (Rokaya et al., 2018). Glass-ionomer cements are important materials in pediatric dentistry and are the materials of choice in patients with high caries risk due to their ability to release fluorides. GIC is used in atraumatic restorative treatment, for small lesions, a temporary filling, open and closed sandwich technique, for cementing crowns and bridges, and as a liner or base in all deep lesions. There are two main groups of GIs: Conventional GIs and Resin-Modified GIs, also known as hybrid materials. Some nanoparticles were added to RMGIs to obtain nano ionomers (Sakaguchi &.Mitra, 2019) Impression materials in dentistry, which are used to copy oral tissue, can be divided into two main groups: elastic (polysulfides, addition and condensation silicones, alginate, polyether, and agar) and inelastic/rigid material (impression waxes, impression compound, zinc oxide eugenol) (Gupta & Brizuela, 2023). In endodontic therapy, sealers play an essential role in ensuring that the root canal system is properly sealed after cleaning and shaping. The most used sealer is zinc-oxide eugenol sealer. There are also glass-ionomer-based, calcium hydroxide-based, resin-based, and bio-ceramic sealers (Gasner & Brizuela, 2023). Gutta-Percha is a natural polymer with excellent sealing properties. The choice of luting materials depends on factors such as the type of restorations and the clinical situation. They could be classified as water-based (zinc-oxide eu-







zinc-polycarboxylate,..) or resin-based luting cement (conventional and self-adhesive resin cement) (Leung et al., 2022). Metals and metal alloys are used in almost every aspect of dental practice, including the dental laboratory. The American Dental Association (ADA) describes three categories of dental casting alloys based on the content of noble metals: (1) high noble (Au-Ag-Pt), (2) noble (Ag-Pd), and (3) predominantly base metal (Ni-Cr, Co-Cr, and Ti-alloys) (Messer et al., 2002). In restorative dentistry, we use them as filling materials (amalgam), in prosthetics for the construction of metal cores of crowns and dental bridges, for the metallic framework of removable partial dentures, for dental implants, in orthodontics for orthodontic wires, brackets, etc. For example, stainless steel or titanium alloys are used for different dental instruments. Dental ceramics have excellent aesthetic properties, are chemically inert in the oral environment, and show high biocompatibility with oral tissues. They are used for various types of dental restorations, such as inlays, onlays, veneers, crowns, and bridges. Dental ceramics are mainly composed of crystalline minerals and glass matrix (Babu et al., 2015). The last classification system categorizes ceramic restorative materials into three families: (1) glass-matrix ceramics - which contain a glass phase (feldspathic ceramics, synthetic glass ceramics, glass-infiltrated ceramics), (2) polycrystalline ceramics – which contain only crystalline phase (alumina, zirconia), and (3) resin-matrix ceramics (hybrids) - which consist of an organic matrix highly filled with ceramic particles. The "Resin Nano Ceramic" contains nanoceramic particles bound in a highly cross-linked polymeric matrix. The hybrids are the most suitable materials for making crowns over implants where the periodontal ligament is lost (Bajraktarova-Valjakova et al., 2018). As preventive and intermediary materials, we utilize pit and fissure sealants (glass-ionomers and flowable composites), glass-ionomer cement for prevention of the progression of decay, calcium hydroxide cement, mineral trioxide aggregate (MTA), biodentine, fluoride-based materials (varnishes, mouth rinses, toothpaste, gels) (Marinho et al., 2003) and others. Calcium hydroxide is the material of choice in deep caries therapy. It is also used for direct and indirect pulp capping, as an intracanal medication, and in endodontic treatment of teeth with incomplete root growth (Sakaguchi & Ferracane, 2019). The most commonly used materials in oral and maxillofacial reconstructive surgery are metal and metal alloys (stainless steel, Co-Cr alloys, Titanium and Ti-alloys) (Pacifici et al., 2017) ceramics (zirconia and alumina for dental implants, calcium phosphate (CaP) for bone grafts), resorbable suture materials (gut, chromic gut, polyglycolic acid, and polydioxanone) and non-resorbable suture materials (silk and polyester) (Davis & Smith, 2023), hemostatic agents (hemostatic collagen, chitosan,...) (Scarano et al., 2023), tissue adhesives (e.g. cyanoacrylate) (Habib et al., 2013), resorbable and non-resorbable membranes and polymer materials. Titanium is the metal of choice in maxillofacial surgery due to its biocompatibility, which results in the formation of an oxide layer on its surface. Titanium is combined with other elements to create alloys suitable for the manufacturing of dental works (Ti-6Al-4V). Its ability to integrate Osseo has been used to make Ti-based implants, which represents the gold standard for endo-osseus dental implants. The materials used in periodontal regenerative therapy include bone grafts (e.g., hydroxyapatite, tricalcium-phosphate), membranes (resorbable and non-resorbable) which keep the soft tissue out of the periodontal or bony defects, growth factors and cell-based materials (cell sheets, stem cells) (Darby, 2011). There are four major metal alloys to produce elements in orthodontics (wires, bands, brackets): stainless steel, cobalt-chromium, NiTi, and beta-titanium alloys (Brantley, 2020). Nitinol, also called Nickel-Titanium-based shape memory alloy, is a wrought Ni-Ti alloy. Nitinol is widely used in orthodontics due to its unique properties, its shape memory, and super elasticity (Fernandes et al., 2011). Orthodontic brackets can also be ceramic, and adhesives are used to bond this element to the tooth. Polymers used in therapy are acrylic resins for mobile orthodontic devices, elastomeric materials for active elements (elastomeric ligatures, elastomeric chains and power chains, elastomeric O-rings), and others. Today, nanomaterials are part of almost every field in dentistry. They could be used in the diagnosis of malignant and precancerous lesions or periodontal diseases, for example, the use of gold nanoparticles (AuNPs) in the research of cancer diagnosis and therapy (Zhang et al., 2022).

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Review

Society & Science: High Pressures for Innovative Pro-Health Foods

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Abstract:

The report addresses quests related to innovative methods of food preservation in the context of the Industrial Revolutions epoch and the assisted population boost. Particular attention is paid to the High-Pressure Preservation/Processing (HPP) of foods that uniquely combine the pro-health expectations of 21st-century societies with the pro-environmental requirements of Sustainable Development. HPP technology also correlates with producers' and logistics' expectations due to the long period of fresh product quality preservation. Beneficial features of HPP technology and related physical foundations of high-pressure impact on foods are presented. HPP is related to "cold & high-pressure pasteurization" at near ambient temperature. This report also considers an innovative sterilization concept, linking colossal barocaloric effect (CBE) & HPP. It is related to compressing up to ~600MPa and high temperatures (90 °C – 120 °C) controlled by the heat associated with CBE phenomenon. Further, the supplementary concept of 'cold sterilization' under medium-high pressures, related to $P \sim 200$ MPa & $T \sim -20$ °C, supported by the reversed CBE, is presented. Finally, socioeconomic consequences of these unique technologies are discussed. They address 5th Industrial Revolution & Sustainable Development targets and expected new market niches.

Keywords: Industrial Revolutions; Food preservation; High-pressure processing (HPP); High-pressure sterilization; Physical origins; Socio-economic impacts





1. Population, Food & Malthusian Trap

In 1798, Thomas Malthus presented the model showing population growth (dynamics), exemplified by a scaling equation with the constant population growth rate coefficient r (Malthus, 1798; Galor, 2000; Weil and Wilde, 2010):

$$P(t) = P_0 \exp(rt) \implies \left[\frac{\mathrm{d}\ln P(t)}{\mathrm{d}t} = r \implies \frac{\mathrm{d}P(t)}{\mathrm{d}t} = rP\right] \implies \frac{\mathrm{d}P(t)/P(p)}{\mathrm{d}t} = G_P(P) = r,\tag{1}$$

where P(t) stands for population changes, P_0 is its value at the onset time t = 0, and r = const. The left part is sunject to basic Malthus' scaling, including the coupled differential equation. The right part shows the new form associated with the population growth factor $G_P(P)$ - a quantity whose significance has been stressed recently (Sojecka and Drozd-Rzoska, 2024, Lehman et al., 2021).

Malthus explicitly declared inspiration by Isaac Newton's legacy, demonstrating the Scientific Method's extraordinary "cognitive power" (Malthus, 1798, Anstey, 2004; Gauch, 2012). He indicated that seemingly separate phenomena can be described via common scaling equations, supported by differential counterparts revealing their formal origins and discussed the significance of food resources, assuming the linear pattern of changes (Malthus, 1798):

$$F(t) = a + bt (2)$$

where a, b = const. **Figure 1** presents the schematic interplay analysis of Eqs. (1) and (2). Malthus commented it as follows (Malthus, 1798): "The population increases in geometrical ratio and the subsistence rises only linearly, which finally leads to times of vice and misery", i.e., the Malthus Trap (Catastrophe) - indicated in the plot. Malthus advised population constraints or extra rise in subsistence (food) to escape the trap disaster. Unfortunately, the simplistic escape concept, namely robbery or conquest of other countries, has often been implemented (Reuveny, 2012).

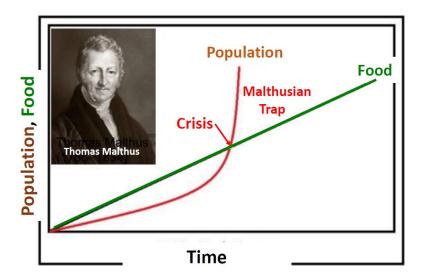


Figure 1. Malthusian human population and food "resources" changes, related to model scaling relation, Eqs. (1) and (2), and leading to the Malthusian Trap (Crisis, Catastrophe).





The experimental validation of conceptual model concepts is essential for the Scientific Method (Gauch, 2012). For Malthus, it could be observations of extraordinary changes in England, the World Leader in developing 1st Industrial Revolution, also named the Steam Age (Crump, 2007; Allen 2012). Numerous large industrial centers with huge demands for labor appeared. They were full of hopes for a new, better life, but also fierce economic relations and over-exploitation. Great wealth, hopes, and expectations accompanied dramatic poverty and decline. The general driving force of the Industrial Revolution was innovation: for the 1st Industrial Revolution, they explored coal, a new and effective energy source. Worth stressing are feedback interactions between technological innovations and the evolving political, economic, and social issues, yielding 'innovative & supporting' socio-economic surroundings.

Figure 2 shows global population changes during Industrial Revolutions times, based on the authors' data obtained by collecting data from different sources and their numerical filtering. It yielded the "analytic set" of global population data, for which the derivative analysis was possible (Sojecka and Drozd-Rzoska, 2024 and 2025). The nonlinear pattern of changes in **Figure 2**, which uses the semi-log scale, is in explicit disagreement with Malthus model predictions, for which the linear behavior is obligatory (Eq. (1).

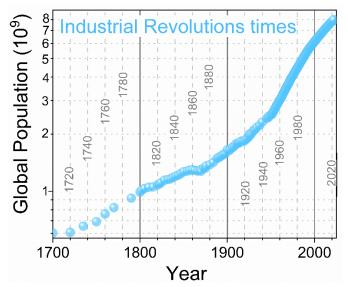


Figure 2. Global population growth during the Industrial Revolutions, based on the authors' data (Sojecka and Rzoska, 2024).

2. Super-Malthus Population Growth and Food Resources

For portraying non-Malthus changes in global population growth, the following extension of Eq. (1) was suggested (Sojecka and Drozd-Rzoska, 2024):

$$P(t) = P_0 \exp(rt) \Rightarrow P(t) = P_0 \exp(r(t)t) = P_0 \exp\left(\frac{t}{\tau(t)}\right)$$
 (3)

In this dependence, for which the name Super-Malthus (S-M) equation was proposed (Sojecka and Drozd-Rzoska, 2024), the time-dependent rate coefficient r(t) has been introduced. Equally important is the introduction of the relaxation time $\tau(T) = 1/r(t)$, the parameter commonly used in the physics of complex systems. It offers a reader-friendly interpretation, i.e., the estimation of the time required for the population to change by 50% from the value of P(t) at a given moment in time: $t_{50\%} = \tau(t) \ln 2$.

Unfortunately, the non-defined functional form of $\tau(t)$ evolution can suggest a seemingly impossible portrayal of P(t) data directly via Eq. (3). Nevertheless, one can focus on the relaxation time itself, using Eq. (3) for calculating $\tau(t)$ changes: $\tau(t) = t[\ln(P_0/P(t))]$. (Sojecka and Drozd-Rzoska, 2024). Such behavior, associated with data presented above, is shown in **Figure 3**. The plot reveals two characteristic time domains. In the huge time interval, ranging between years ~1100 and ~1700, the relaxation time changes are described





by the horizontal line, i.e., $\tau(t) = \tau = \text{const}$, which also means r = const. Consequently, in this period, the basic Malthus model (Eq. (1): left part) yields the dominant description for the global population evolution, with a disturbance correlating with the Black Death pandemic times. Starting from year ~1700, which can be associated with the Industrial Revolutions times onset, the dominant trend changes: $\tau(t) = a - bt$ (Sojecka and Drozd-Rzoska, 2024).

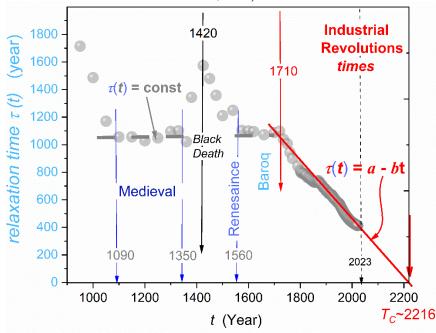


Figure 3. The evolution of the relaxation in the last millennium, related to the general Super-Malthus Eq. (3). Subsequent epochs and relevant events (Black Death) are indicated.

The substitution of $\tau(t) = a - bt$ into Eq.(3) yields:

$$P(t) = P_0 \exp\left(\frac{t}{a-bt}\right) = P_0 \exp\left(\frac{t/b}{a/b-t}\right) = P_0 \exp\left(\frac{ct}{T_C-t}\right),\tag{4}$$

where c = 1/b = const and $T_C = a/b \approx 2216$ years.

This new global population growth pattern, introduced in (Sojecka, Drozd-Rzoska, 2024) correlates with dynamics of constrained and frustrated systems considered in Complex Systems Physics. Regarding constraints, one can indicate global-scale spatial limitations and the environment's ecological carrying capacity. Frustrations include numerous political and economic disturbances, which can rapidly grow up to a global scale.

3. Innovative Food Preservation Methods in Industrial Revolutions times

During the Industrial Revolutions times, global and local populations have grown dramatically, following the Super-Arrhenius pattern discussed above. In 1700, there were ~610 million people on Earth, after millennia since the Anthropocene onset (10 000 BCE). By 1800 it was ~1 billion, and by 1900: 1.6 billion (Sojecka and Drozd-Rzoska, 2024). These rapid changes were accompanied by the emergence of populous and increasingly numerous industrial centers, for which providing a sufficient amount of health-safe food was a critical challenge. Generally, human populations have struggled with the problem of pro-health food safety since the beginning of its history, which has led to the creation of many "preservation techniques" and products that enrich our cuisine to this day. However, a new generation of preservation methods has become necessary for the growing global population and industrial centers. They should not change basic features noted by consumers, like the view and taste, and be suitable for highly processed "industrial food", massively "fabricated".

The scientific basis and target for these methods could be defined due to the breakthrough research of Louis Pasteur's (1863). He linked the fast loss of food properties to





spontaneously rising contamination by dangerous microorganisms – bacteria (Hunter, 2014). Pasteur found a general remedy: heating the product to a temperature of 84 - 86 0 C, for several minutes. This process is known as the thermal pasteurization. For decades, it has been one of the essential preservation methods: the basic examples can be milk or juice in stores. Currently, this process is defined by 5-decade reduction in the number of microorganisms to the level of 10^{-5} in comparison to the native state. Using the commonly used jargon: it is 5-log reduction (Rahman, 2007; Sun, 2005).

For many food types, essential is the very long microbiological safety guaranteeing negligible microbiological contamination, and simply applicable for solid and liquid products. For this purpose, many chemical preservatives or their complexes have been worked out. For decades, it has remained one of the leading food and beverage preservation methods (Rahman, 2007). Today, chemical preservatives are present on store shelves as a dominant part of products.

In the 20th century, there was also a mass implementation of preservation techniques based on products cooling to a temperature of $6-8\,^{\circ}\text{C}$ or freezing at temperatures reaching $-30\,^{\circ}\text{C}$. A significant novelty was the appearance of large refrigerated facilities for mass cooling/freezing, often associated with a unique oxygen-free atmosphere (Rahman, 2007). A significant breakthrough was the development of home refrigerators and coolers, mandatory equipment of every home nowadays. This was possible after implementing the physical concept of circulating, suitably selected liquid, in which adiabatic decompression yields a strong decrease in temperature. Significant was finding a liquid where it is associated with the exceptional colossal entropy change $\sim 400\,\text{JK}^{-1}\text{kg}^{-1}$, which is the process metric. They are hydrofluorocarbons, especially their most famous representative with the trade symbol HF134a (Sojecka et al., 2024).

Another modern and widely used preservation method is the impact of ionizing radiation. It is the basic method for very dry products, such as spices (Rahman, 2007).

In the 21st century, however, significant side effects of the above-mentioned food preservation technologies have appeared:

- Numerous food preservation chemical agents are the cause of the obesity epidemic, allergies, and even some types of cancer diseases. They are also linked to fatal, intestinal gut complications (Anand and Sati, 2013; Reardon, 2015)
- Thermal pasteurization, and much more sterilization can deprive food and beverages. From 60% to 80% of their nutritional and bioactive values are lost, and these numbers increase during storage (Sojecka et al., 2024).
- Freezing, due to the appearance of ice crystals, can partially destroy the product's texture (Bald, 2012).
- Freezing/cooling equipment like air conditioning and most of the heat pumps use the above-mentioned thermodynamic cycle with circulating and rapidly compressed/decompressed 'fluid agents'. However, hydrofluorocarbons (inluding their 'soft, eco-friendly' counterparts) are hundreds of times or more harmful to Global Warming than CO₂ which dominates discussions in mass media (Sojecka et al., 2024).

The innovative food preservation methods implemented since the 19th century have greatly impacted the current state of food abundance on store shelves and the elimination of many disease threats. They have also supported efficient logistics in numerous populated urban centers.

However, in the 21st century, the cumulated 'side effects' of these methods have appeared, creating new, global-scale threats. The fundamental way to solve such challenges in Industrial Revolutions times was/is the next Grand Innovation, avoiding the abovementioned problems and responding to consumers' expectations and producers' logistical requirements.

It turns out that a method with such unique features exists. It can be the High Pressure Processing / Preservation (HPP) (Sojecka et al., 2024).

4. High Pressure Processing (HPP) of foods

High Pressure Processing (HPP), also called cold or pressure pasteurization in canonical version, is related to compressing in the range of p = (300MPa - 600MPa) for





several minutes. Comparing cold 'pressure pasteurization' with the classic/standard thermal pasteurization, the Super-Arrhenius (SA) and Super-Barus (SB) relations describing temperature and pressure-related dynamics in complex systems, like foods, are worth mentioning (Drozd-Rzoska et al., 2008; Drozd-Rzoska et al., 2023):

$$k\langle r^2\rangle d \propto \exp\left(-\frac{E_a(T)}{RT}\right)$$
 (SA: $p = \text{const}$)

$$k\langle r^2\rangle d \propto exp(cP) = \exp\left(-\frac{PV_a(T)}{RT}\right)$$
 (SB: $T = \text{const}$)

where k stands for chemical reaction rate, also related to bondings, d is the diffusion rate, $\langle r^2 \rangle$ describes the average fluctuations of molecules around the equilibrium position, R denotes the gas constant, c = const; $E_a(T)$ is the apparent (temperature dependent) activation energy and $V_a(p)$ is the apparent (pressure-dependent) activation volume.

For $E_a(T) = E_a = {\rm const}$ one obtains the simple Arrhenius equation when substituting to Eq. (5). Heating increases $k\langle r^2\rangle d$ values, and the effect can be further increased for the SA $(E_a(T) \neq 0)$ dynamics. For proteins, with their multi-level complex structures, these factors lead to their permanent intrastructural changes when passing the denaturation border. **Figure 4** shows the schematic view of this process. Bonds breaking, mainly in secondary and tertiary intra-structural protein parts, is related to rising thermal fluctuations impacting $k\langle r^2\rangle$ matched with the activation energy changes.. This leads to irreversible changes associated with denaturation. For food products, it means eliminating the dominant part of the living microbiological contaminations and permanent changes in properties. An example would be the specific taste of milk purchased in stores.

The standard HPP technology is related to isothermal pressure changes, the consequences of which are illustrated by Eq. (6). Essential is the impact of compressing on the free volume. For foods, it is related to intermolecular volumes and between different macromolecular and multimolecular assemblies with varying degrees of complexity. The activation volume in Eq. (6) is the effective metric of these compressing impacts on all these 'free volume components'. The mentioned sub-elements are also characterized by strongly different local compressibilities, i.e., sensitivity to compression, expressed by the change in the local volumes they occupy or the distances between individual components. For food, the basic ingredient is water, the compression of which to $p \sim 600 \text{MPa}$ leads to an 18% decrease in density, which further means that the average inter-element distances decrease to ~93% from the reference state. It can be much larger locally for macromolecules and multi-molecular assemblies. Consequently, strong gradients of local shear forces induced by local compressibility gradients must appear inside a food product. They can be extreme for such very complex Soft Matter systems as living microorganisms.

Local shear forces associated with compressing must lead to breaking bonds between local sub-elements of food and even more for extremely complex microorganisms. As the pressure increases, the rising impacts microorganisms appear, starting from the largest multi-molecular structures, such as cellular walls or intracellular structures. Their irreversible damage is the significant cause of parasitic microorganisms' destruction in HPP technology. The above shows that an optimally designed HPP implementation can almost exclusively affect undesirable microorganisms, preserving fresh food product features when decompressing. Further and stronger compression must lead to denaturation, an irreversible process. The undesirable destructions of products in the pressure range used for HPP technology have little or no effect on viruses, which relatively less complex structure can explain in comparison to bacteria, yeast,...





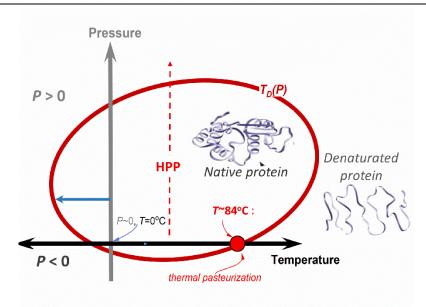


Figure 4 The scheme of the denaturation curve $T_D p$, in pressure – temperature (p-T) plane. The thermal pasteurization temperature (under atmospheric pressure), the standard HPP path (vertical, dashed red arrow), and the possibility of the 'mild' pressure-assisted pasteurization or sterilization (horizontal blue arrow) are indicated. Note positive and negative (isotropic stretching) pressure domains. The plot schematically shows native and denatured proteins related to domains separated by the elliptic denaturation $T_D(p)$ curve.

In conclusion, denaturation is the dominant mechanism responsible for standard thermal pasteurization under atmospheric pressure. For HPP, the pressure dependence of the denaturation curve, with the elliptic curve in the pressure-temperature plane, is essential. Such form is the consequence of the extended Clausius – Clapeyron (C-C) equation, with the pressure-dependent changes of the volume $(\Delta V(p))$) and enthalpy $(\Delta H(p))$, (Sojecka et al., 2024):

$$\frac{dT_D}{dP} = T_D(\text{ref.}) \frac{\Delta V(P)}{\Delta H(P)} \qquad , \tag{7}$$

where $T_D(\text{ref.})$ is the reference onset temperature; ΔV and ΔH are for the volume and enthalpy change when passing a discontinuous phase transition – or, in the given case, the denaturation curve. Originally C-C equation was related to the melting (liquid – crystal) discontinuous transition and for pressures in the immediate neighboring of the atmospheric pressure, where $\Delta V = \text{const}$ and $\Delta H = \text{const}$. However, it can be linked to two borders of two states with significantly different ordering. This is also the case with denaturation curves, which separate states with essentially different conformational arrangements of proteins.

Bert Holmes Hite carried out the first studies on high pressures' impact on foods at the end of the 19^{th} century, focusing on microbiological contaminations in milk subjected to pressures up to p=670 MPa for 10 minutes and showed a 5-6 log reduction in total counts (Hite, 1899). He also tested beef meat treated with pressure p=530MPa for an hour. After three weeks of storage, only insignificant microbial growth was noted. In 1912, Percy W. Bridgeman reported egg albumin coagulation after compressing at p=590 MPa for 1hour (Brifgemenn, 1912). Bridgeman was honored with the Nobel Prize for extensive high-pressure studies in 1946 (Bridgeman, 1946).

These early works indicated basic features of HPP technology on foods, namely the reduction of parasitic microorganisms and the denaturation of proteins. However, it was not until 1989 that the first HPP-treated food products appeared in Japan (Sojecka et al., 2024). For decades, the barrier was the construction of large-volume pressure processors with chamber volumes of $V\sim50L$, 100L, and more. For the last decade of the 20^{th} century, market demands for new-generation, health-promoting food preservation methods also became notable. The HPP-treated food products market is worth at least USD 8.2 billion and is expected to grow by ~120% in the next decade globally (Sojecka et al., 2024).





Unique benefits of HPP technology (Sojecka et al., 2024):

- fresh product quality: nutritional properties, taste, flavor, texture, ...
- shelf-life extension from 2–3 days to even up to 180 days!
- high microbiological safety
- taste, flavor, and appearance of a fresh product
- vitamin composition of the fresh product
- maintained bioactive properties
- limited or even no chemical preservatives
- activation/deactivation of selected enzymes
- salt-free products
- sugar-reduction possibilities
- application to fluid, soft, and solid food
- application to packed food: secondary contamination risk avoided
- environment-friendly technology, namely: (i) $\sim 4x$ less electric energy required than for the thermal pasteurization; (ii) practical lack of waste
- reduction of the of expired products number: disposal costs environmental "costs" reduction
- "clean label" and innovative technology
- creation of new products and a new market niche
- products, including ready-to-eat meals, for customers/patients with special health needs
- immediate exposure of the entire volume of the product to high-pressure

The standard thermal pasteurization is related to the general principle applicable to each product, i.e., exposure to temperatures of $\sim 84-86$ °C for at least several minutes. HPP technology requires an individual implementation protocol for each product. This applies to (Sojecka et al., 2024):

- (i) Pressure pulse duration and its value (usually 300 600 MPa)
- (ii) Time profile of the pulse edges
- (iii) Processing temperature (standard range: 10 50 °C)
- (iv) In advanced applications: the number of HP pulses and their arrangement

The Photo in **Fig. 5** shows the HPP processor, with V = 1L pressure chamber volume, supported by a programmed control system for preliminary preparation of the implementation protocol for a food product. The subsequent test requires the pilot-scale facility, such as the one shown in **Fig. 6**, with the pressure chamber volume V = 50L, compressing up to P = 600MPa, with remote control of the cycle. It is the central part of the pilot line, composed of vehicle ramps, large-scale coolers at the entrance and exit, and the HPP processor in between. Additionally, the pilot line is assisted with the product preparation room. The facilities shown in **Figures 5** and **6** are parts of the X-PressMatter laboratory located in the Innovation Park of the Institute of High Pressure Physics of the Polish Academy of Sciences in Celestynów near Warsaw (Poland). X-Press Matter Lab is included in 'RoadMap' of Significant Research Infrastructure (Poland).

5. HPP – innovative frontiers

Standard HPP implementation is related to the 'cold pasteurization' for foods, pharmaceuticals, or cosmetics. It shows beneficial 'mild method' features, linking high microbiological safety with fresh (native) product quality. In some cases, even strengthening pro-health features is possible (Sojecka et al., 2024). Implementation of standard HPP technology means a qualitative reduction in the number of parasitic microorganisms to the reference level of the thermal pasteurization but without numerous adverse side effects of the latter.

The obvious path for developing HPP technology seems to be pressure-driven sterilization. This technology is widely used for thermal sterilization under atmospheric pressure for many products on store shelves, such as milk in cartons and numerous juices in their "healthy" versions.





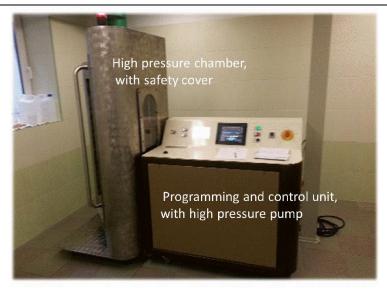


Figure 5. HPP lab scale processor with V = 1L pressure chamber, for basic research and preliminary establishing of implementation protocols for "HPP foods". X-PressMatter Lab, IHPP PAS (X-PressMatter, 2025).



Figure 6. HPP pilot processor with V = 50L pressure chamber for the industrial-scale test, finalization of implementation protocol, and market experiments. X-Press Matter Lab, IHPP PAS (X-PressMatter, 2024).

A typical thermal sterilization process is arranged in two versions (i) wet pasteurization at a temperature of 121 - 125 °C for ~15 minutes in the presence of saturated vapor, and (ii) short-tome (~1 second) treatment with a temperature above 150 °C (Teixeira, 2014). Notwithstanding, after such sterilization, the destruction of some nutritional properties and bioactivity of processed products must be even more deteriorated than after the thermal pasteurization carried out at $T \sim 86$ °C. It is suggested that increasing compression up to even ~1.4GPa in HPP technology can also destroy spores and viruses, thus leading to pressure sterilization. However, it also means the essential rise of HPP processor costs, limiting its durability and increasing the costs of service inspections. Extreme pressures can be destructive to essential food properties. In such context, the maximum pressure P = 600MPa of standard HPP industrial processors available today seems to be a favorable compromise (Koutchma, et al.; 2005).

When considering the impact of pressure, the simultaneous link of compressing at p = 600 MPa matched with heating in the range $90\,^{\circ}\text{C} - 120\,^{\circ}\text{C}$ is considered the High Pressure High Temperature (HPHT, HPT) sterilization technology. There are no essential problems in creating such conditions for the lab-scale, where simply the pressure chamber can be heated. For industrial-scale or even pilot-scale HPP processors, this is not possible. Probably the only commercial solution to this challenge is related to the thermo-isolated





container in which the product is pre-heated to ~90 °C. The adiabatic compressing up to p = 600MPa can further increase the temperature up to ~120 °C. In this way, HPT technological sterilization is achieved (Hyperbaric, 2024).

The mentioned solution is undoubtedly effective for high pressure & high temperature sterilization (HPT, HPHT), but it has obvious limitations. First, it requires a significant amount of energy comparable to the amount necessary for thermal sterilization. Second, the process requires a relatively large number of operations, and third, the volume available for the product inside the HPP chamber is reduced.

Very recently, the concept that significantly reduces the above problems was proposed (Sojecka et al., 2024). Its essence is the application of the barocaloric effect, originally considered an innovative physical concept for the new generation of refrigerators and airconditioners (Lloveras, et al.; 2019). During compressing, a strongly discontinuous phase transition from a disordered to an ordered phase is passed, releasing a significant amount of heat associated with the change in internal order (H_a in C-C Eq. (7)).

For the implementation proposed in (Sojecka et al., 2024), elements containing neopentyl glycol (NPG), the basic material exhibiting the so-called Colossal Barocaloric Effect (CBE) (Lloveras, et al.; 2019) is placed within the pressure chamber together with the processed product. A thermo-isolated layer, such as Teflon, covers the pressure chamber's walls. It converts the standard HPP processor to the facility to the adiabatic pressure chamber. The process design causes that heat from CBE elements is released only at pressures close to the planned stationary value, for instance, p = 600MPa. Additionally, the temperature is increased due to standard internal energy changes during compressing. Consequently, the "designed" temperature range 90 °C – 120 °C can be reached, and it appears only for the well-defined time of the maximal, stationary compressing. For the decompression stage, the temperature inside the pressure chamber immediately drops (Sojecka et al., 2024), and post-process product removed from the processor is again at ambient temperature, which facilitates handling.

Notable, that such an innovative CBE-supported concept of HPT technology requires an energy supply similar to the standard HPP applications, i.e., ~4x or lesser than for the "classic" thermal pasteurization under atmospheric pressure.

Notwithstanding, the general problem of HPT / HPHT technology remains - it is no longer a "mild & gentle" treatment of food processing, which is the essential prevalence of basic HPP "cold pasteurization". The crossover of the high temperature thermal pasteurization limit (84-86 °C) must always lead to a reduction of the essential values of food products, even when pressure-assisted. Nevertheless, HPT/HPHT process creates unique possibilities for the deactivation and activation of enzymes, which may be important for people with unique health problems and food limitations. For the CBE-supported HPP version, such processing can be controlled precisely, also offering new opportunities for pharmaceutical industry applications.

Figure 4 shows the denaturation curve for the *p-T* plane. The horizontal arrow (in blue) indicates the other possibility of CBE-supported & adiabatic HPP-based innovative sterilization/paseurization concept. For still limited cases of materials, the so-called inverse-CBE has been found (Zhang, et al; 2023). For such a system, compressing causes thermal energy absorption from the environment, i.e., a drop in surrounding temperature under adiabatic conditions. For decompressing, heat emission from the CBE element to the environment occurs.

The eutectic minimum for water (the main component of food), causes that it remains liquid under pressure $p \sim 200 \text{MPa}$ down to $T \sim -21\,^{\circ}\text{C}$. It means that, for instance, at process onset temperature $T \sim 10\,^{\circ}\text{C}$, one can carry out compression up to the "mild value" = 200 MPa withint "HP-adiabatic" process. Reaching this pressure can trigger the absorption of heat energy by the "inverse-barocaloric" elements. It can yield a rapid temperature decrease to $\sim -20\,^{\circ}\text{C}$. If necessary, one can now consider further compressing up to $p \sim 300\,\text{MPa}$ or more.

Notable, water inside the chamber and food products does not crystallize, thanks to the mentioned eutectic minimum. Therefore, there are no destructive effects of ice crystallites inside the product.

For the descibed process, $T_D(p)$ denaturation curve is passed, as indicated in **Fig.4** by the horizontal blue arrow. This t indicates the possibility of low-temperature "mild" pressure-





assisted HPP sterilizations – supported by the inverse colossal barocaloric effect (ICBE). Such innovative processing is related to a strictly defined exposure time and reduced temperature.

The described low-temperature "HPP-adiabatic" processing supported by the inverse CBE can offer a qualitatively new opportunity also for activating and deactivating some enzymes, which currently pose great challenges for the entire population, significantly related to chemical preservatives in food over-using.

6. Conclusions: Innovative H & Socio-Economic Impacts

There are disputes about the beginning of the Industrial Revolutions. It is considered between 1700 and 1750, with a significant number of researchers indicating the latter date. The analysis of global population changes (Fig. 3, Sojecka and Drozd-Rzoska, 2024) definitely points to the year ~1700, the time of the first industrial applications of coalfired steam engines/machines. This was also the beginning of the unique Enlightenment cultural period and the pan-European dissemination of the Scientific Method as a new cognitive method, happily supported by reformed political and social systems. In such unique circumstances, a growing avalanche of innovations appeared spontaneously: and were precursors to the Steam Age. Interestingly, their creators often were sons of blacksmiths, whose forges were scattered local "centers of advanced technology" in previous eras. Another important factor is exposed - the unprecedented social and economic advancement opportunities - driven by outstanding skills in science and technology only.

At the beginning of the 18th century, the innovations-driven civilization was referred to as the Industrial Revolutions epoch. In this New World, so different from previous epochs, the main factors creating progress and development were technological and scientific innovations, spontaneously and widely implemented, supported by feedback interactions with the socio-economic and political environment.

The current era of the 5th Industrial Revolution (IR) is defined as "harmonious human—machine collaborations, with a specific focus on the well-being of the multiple stakeholders" (Martin, 2023). This is a general and ambiguous definition compared to the earlier IR epochs, which referred to leading "emerging" technological aspects. The 5th Industrial Revolution is sometimes linked to artificial intelligence (AI). But isn't AI (nowadays) a continuation and consequence of the main challenge of the previous IR phase?

All the above is well visible in definitions of earlier Industrial Revolution (IR) stages (Allen, 2017):

1st IR: Steam Age

2nd IR: Electricity Age

3rd IR: Electronic Technologies & Computers Age

4th IR: Datafication and Internet Age

5th IR: New Generation of Energy Sources & Innovative Materials Age

For the the authors, this definition of the 5th IR can be better suited to support the Sustainable Society Development.

Food is the most important energy resource, directly and daily necessary for every human. Currently, losses in produced food at the processing and logistics stages can be estimated (at least) by ~30% (Marimuthu. et al., 2024). One can even expect to double this value at the final consumer use stage. An important source of losses is the quality of delivered products and their still limited durability, often extended at the cost of introducing agents unfavorable to health, as discussed above.

The HPP for food technologies described in this work can be considered the innovative material engineering, leading to better use of Earth's most important energy resource food. It creates products with qualitatively new features that benefit humans/consumers health, and the environment due to the qualitative reduction of losses and disposals.

Mass implementation of innovative methods for food processing and preservation, where HPP holds a special and already market-proven position, can qualitatively increase the amount of available food without all the problematic environmental issues related to its production.





In the Industrial Revolutions epochs, leading innovations yielded huge market niches associated with new jobs. The authors' would like to point out this aspect for this chapter, complementing the discussion in the recent report (Sojecka and Drozd-Rzoska, 2024).

Each country has many excellent local products, which in all their richness of taste - but also nutritional - are available only at the local level due to natural time constraints. In Poland, dozens, if not hundreds, of products and dishes can be enjoyed when visiting the country's central, coastal, eastern, or mountainous parts. Thanks to HPP technology and its advanced innovative developments, these excellent products and dishes may be available in every corner of Poland and even Europe.

Another possibility is for restaurants - to use high-pressure HPP technologies directly in their kitchens, which opens up new and previously unexplored creative options for chefs. Many of the essential kitchen products could be delivered in small packages, previously subjected to HPP technology, without preservatives and with the benefits of a fresh product. It could qualitatively change the style of work and reduce losses.

The above examples indicate possible emerging market niches and "innovative" jobs that can arise within the pro-health and environmentally sustainable economy supported by "HPP food engineerig" innovations.

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Research

Domain Patterns in Quenched Confined Nematic Liquid Crystals

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Abstract:

This study explores the Imry-Ma theorem and the Kibble-Zurek mechanism in the context of quenched confined nematic liquid crystals (NLCs). The Imry-Ma theorem explains domain formation under the influence of random fields, while the Kibble-Zurek mechanism describes defect dynamics during fast enough symmetry breaking phase transitions. We demonstrate that considering both mechanisms one could explain the equilibrium domain structure observed in NLCs confined within a plane-parallel cell.

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Keywords: Imry-Ma theorem; Kibble-Zurek mechanism; Nematic liquid crystals; Phase transitions; Topological defects

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1. Introduction

1.1. Context and importance

The interplay between theoretical frameworks and experimental observations is essential for understanding complex systems like nematic liquid crystals. Liquid crystals (LC) , with their unique combination of fluidity and ordered molecular alignment, are sensitive to impurities and external influences, making them an excellent platform for exploring the effects of disorder on structural properties and phase transitions (Kléman et al., 2003). For simplicity we limit to thermotropic rod-like LC molecules whose local orientational order is at the mesoscopic scale described by the nematic director field \vec{n} which points along the average orientation. Here the states $\pm \vec{n}$ are physically equivalent. In thermotropic LCs phase behavior is controlled by varying temperature. At relatively high temperatures LC display liquid-like order, forming the so-called isotropic (I) phase. Thermotyropic nematic liquid crystal (NLC) phase represents the simplest LC phase which can be reached starting from the isotropic phase by lowering temperature. The equilibrium nematic (N) phase exhibits long range orientational order in which \vec{n} is homogeneously aligned alog a single symmetry breaking direction.

Of our interest is recent experimental study (Pišljar et al., 2024) in which domain-type is observed in NLC confined with a plane-parallel cell of thickness h, where the confining substrates enforce the so-called isotropic tangential anchoring. In the latter case nearby LC molecules prefer to lie within the confining plate where all directions are equivalent. However, the experimental study shows that in equilibrium a domain-type nematic structure exists, where the characteristic domain size $\xi_d \propto h$.

Below we demonstrate that such behavior could be explained using universal Imry-Ma (Imry & Ma, 1975) and Kibble-Zurek (Kibble, 1976; Zurek, 1985) mechanism which are valid for phases which are reached via a continuous-symmetry breaking phase transition (Sethna, 1992).

1.2. Theoretical background

The Imry-Ma (IM) theorem, first introduced in the context of magnetism, provides a foundational framework for studying the impact of disorder on long-range order which is reached via a continuous symmetry breaking phase transition. It predicts that random fields can destabilize order in systems described by continuous symmetry breaking phase transitions, leading to the formation of domains whose sizes depend on the strength of the disorder (Imry & Ma, 1975). Initially formulated for ferromagnets, the theorem has since been adapted to other systems, including liquid crystals (Ranjkesh et al., 2014). Furthermore, if a continuous symmetry breaking phase transition is fast enough it could temporarily exhibit domain-like structure due to the finite speed of information propagation. The resulting size of initially formed domains is determined by the Kibble-Zurek (KZ) mechanism (Zurek, 1985) which was originally derived in cosmology (Kibble, 1976).

1.3. Application to liquid crystals

In confined nematic liquid crystals, domains could be temporarily formed in a fast enough (Zurek, 1985) I-N phase transition. Namely, in such cases in distant parts of LC symmetry is in general broken in different directions because of finite information velocity propagation (Kibble, 1976). In bulk NLC domains grow with time and terminate in a single domain which exhibits the lowest parallel cell of thickness h, whose non-treated confining plates do not favor any in-plane direction. We claim that a domain pattern, formed slightly after the nematic phase was reached, could be imprinted into confining plates due to the memory effects (Kléman et al., 2003; Kralj & Sluckin, 1994). Namely, it is well known that LC molecules nearby a confining plate could be »frozen-in« orientationally if LC-substrate interaction is stronger that LC-LC configuration. In the following we show that the combination of IM argument and KZ mechanism could qualitatively explain recent experimental observations (Pišljar et al., 2024).





2. Material and Methods

2.1. Theoretical framework for Imry-Ma theorem

The Imry-Ma theorem explains the emergence of domain structures in systems experiencing continuous symmetry breaking, particularly in the presence of random fields(Imry & Ma, 1975). The elastic energy term, which enforces homogeneity, plays a critical role in this process. The key term enforcing homogeneity in field-type theories is approximated by a single elastic modulus, given by (Kléman et al., 2003)

$$f_e \sim \frac{K}{2} |\nabla \vec{n}|^2 \quad , \tag{1}$$

where K is the nematic elastic constant and $\nabla \vec{n}$ represents the spatial gradient of the director field \vec{n} . This energy term drives the system toward minimizing distortions and maintaining uniform alignment. When geometrical constraints are enforced, elastic energy becomes sensitive to characteristic distances within the system, such as the domain size ξ_d . It roughly holds

$$f_e \sim \frac{K}{2} \left(\frac{1}{\xi_d}\right)^2 \quad . \tag{2}$$

Equation (2) highlights how smaller domains increase elastic energy, favoring larger domains in the absence of competing forces.

2.2. Influence of random fields

Random fields disrupt the uniform alignment enforced by the elastic term. In our illustration we present a random-type disorder by spatially randomly varying local random field direction \vec{u} . This local field enforces a preferred local alignment on the director field \vec{n} . In NLCs the energy contribution from the random field could express as (Bradač et al., 2011)

$$f_i \sim -WSP_2 \ (\vec{n}.\vec{u}) \ , \tag{3}$$

here, W is the random field strength, S represents the degree of molecular order, and the second Legendre polynomial $P_2(x) = (3x^2 - 1)/2$ is dependent on the alignment between \vec{n} and \vec{u} . The random field leads to the formation of domains, as it introduces localized disruptions to the system. In the continuation we assume that W is spatially constant and orientations of unit vectors \vec{n} are randomly distributed.

The balance between elastic energy (f_e) and random field energy (f_i) determines the characteristic domain size. The resulting domain size ξ_d is given by (Ranjkesh et al., 2014)

$$\xi_d \sim \frac{1}{W^{\frac{2}{4-d}}} \quad , \tag{4}$$

where d is the dimensionality of the system. This equation shows that stronger random fields lead to smaller domains, while larger elastic constants promote larger domain sizes what is illustrated in **Figure 1**.





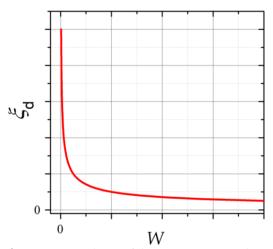


Figure 1. Dependence of domain size ξ_d on the strength of a random field W in d=3 systems. The graph illustrates how an increasing random field strength impacts the characteristic domain size.

2.3. Kibble-Zurek mechanism

The Kibble-Zurek mechanism provides a framework for understanding how defects form during continuous phase transitions. As the system is cooled through its critical temperature T_c , the relaxation time τ and coherence length ξ diverge. These quantities are close to T_c expressed as (Zurek, 1985)

$$\tau = \tau_0 |r|^{-\eta} \quad , \tag{5}$$

$$\xi = \xi_0 |r|^{-\nu} \quad , \tag{6}$$

where $r=(T-T_c)/T_c$, η and ν are critical exponents and τ_0 , ξ_0 are values of τ and ξ_0 deep below the phase transition. In NLCs it roughly holds $\eta=1$ and $\nu=1/2$ (Bradač et al., 2011) what we consider in the continuation. In the classical KZ approach one assumes that the quench is realized linearly in time, i.e. $r=-t/\tau_Q$, where τ_Q is the characteristic quench time. At the freeze – out time t_z , the system transitions from an adiabatic to a non–adiabatic regime as shown in **Figure 2a**. It is defined by (Zurek, 1985)

$$t_z \sim \tau_0 |r_z|^{-1} \tag{7}$$

and the corresponding coherence length at freeze-out is:

$$\xi_z \sim \xi_0 |r_z|^{-\frac{1}{2}}$$
 (8)

One assumes that the maximal cluster size formed by fluctuations is frozen-in at the time $|t_z|$ above T_c and remains frozen-in till the time $-|t_z|$ below T_c . The largest frozen-in domains, given by ξ_z , in this modelling then represent the so-called *protodomains*, which, below T_c , nucleate individual domains whose orientational distribution is randomly distributed. From description above it follows (Bradač et al., 2011; Zurek, 1985) that the characteristic size of protodomains $\xi_p \sim \xi_z$ exhibits the following scaling law in NLCs:

$$\xi_p \sim \xi_0 \left(\tau_Q / \tau_0\right)^{1/4}.\tag{9}$$

Faster cooling rates result in higher defect densities, while slower rates allow the system to form fewer defects. It roughly holds that the defect density when the protodomains are formed scales as $n \propto 1/\sqrt{\tau_Q}$ on varying τ_Q , see **Figure 2b**.





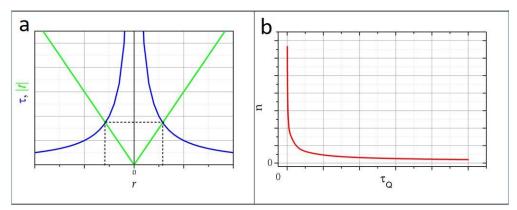


Figure 2. a: Dynamics of the system as a function of temperature. The blue color shows the change in the relaxation time (τ) as a function of the change in temperature. The green color shows how the system temperature changes over time (t). The dashed line shows the part where the system goes through the freeze-out process, **b**: Defect density n at the time when protodomains are formed as a function of the characteristic quench time τ_0 .

3. Results

In the following we apply the KZ and IM approaches to explain experimental observation reported in (Pišljar et al., 2024) with respect to typical domain length ξ_d as a function of the confining cell thickness h.

We describe the essential free energy contributions of the confined NLC as

$$F = \int \frac{K}{2} |\nabla \vec{n}|^2 dV - \int WSP_2 (\vec{n}.\vec{u}) dA.$$
 (10)

In the first term the integration is performed over the LC body. In the 2^{nd} term we consider conditions at the confining boundaries. We assume that the conditions at both substrates are identical and that they do not enforce any preferred direction within the confining plates in the isotropic phase. Therefore, this term begins to contribute in the nematic phase where it enforces a locally preferred direction due to the reasons explained below.

We assume that the system is quenched into the nematic phase with the characteristic quench rate τ_Q . Consequently, a domain-type structure is formed where the initial size of domains is estimated by ξ_p given by Eq.(9). Next, we assume that the pattern of LC molecules at the confining plates becomes frozen-in due to stronger LC-substrate interactions with respect to LC-LC coupling. Note that this phenomenon is not possible in the isotropic phase because the thermal fluctuations are two strong. After the pattern is imprinted at the substrates it remains relative stable which is supported by several observations (Kralj & Sluckin, 1994). Therefore, soon after the quench a domain-type pattern, which depends on τ_Q , is frozen-in at the confining substrates which effectively acts as a local random-field. In Eq.(10) we approximate this randomness by randomly distributed values of surface imposed unit vectors \vec{u} . Note that the average separation between two neighboring sites enforcing different orientations is given by ξ_p .

Our goal is to obtain an estimate on the characteristic size ξ_d of stable domains where they reflect the compromise between the elastic and random-field penalties. For this purpose we use similar approaches used in standard IM approach and apply it to our system.





With this in mind the average elastic penalty F_e reads

$$F_e \sim K/(2\xi_d^2)Ah \tag{11}$$

where A stands for the confining substrate area. On the other hand the average surface interaction penalty F_i of the systems reads

$$F_i \sim -WS < P_2 > A,\tag{12}$$

where <....> determines the average. Using the standard IM approach we assume that $< P_2 >$ depends on the domain size and that it is averaged out in very large domains. In finite domains it holds $< P_2 > \sim 1/\sqrt{N}$, wher N stands for the number of random sites within each domain. It roughly holds $N \sim (\xi_d/\xi_p)^2$ since the confining plates are two dimensional. Taking this into account one obtains from the »compromise« condition $F_i \sim F_e$ an estimate for a stable domain size:

$$\xi_d \sim h \frac{d_e}{\xi_p},\tag{13}$$

where $d_e = K/W$ is commonly referred to as the surface coherence length.

4. Discussion

Our simple derivation suggests that stable domain pattern could be observed in confined LCs where the imposed disorder is enabled by memory effects. Because disorder is in such a case enforced only by the confining plan-parallel surfaces the resulting domain size should linearly depend on the cell thickness. Note that this observation could be experimentally tested by changing the quench rate of the I-N phase transition. Our derivation suggests that $\xi_d \propto \frac{1}{\xi_p} \propto 1/\sqrt{\tau_Q}$.

5. Conclusions

Our study illustrates how knowledge in different branches of science could be transferred between disciplines. This transfer is possible even in strongly physically different systems if mathematical description is similar. In our study this transfer was possible because the unifying element was continuous symmetry breaking phase transition. In it we crossed from a phase exhibiting isotropic symmetry to a phase where the symmetry was broken along a single symmetry direction.

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Research

Importance of the Critical Point in Thermotropic Nematic Liquid Crystals in Terms of Their Sensitivity

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Abstract:

The critical point (CP) of liquid crystals has proven to be of immense significance in terms of their sensitivity and their ability to respond to even the slightest external stimuli, such as surface interactions and applied electric or magnetic fields. In this article we focus on the mathematical approach to achieving the the minimal free energy potential of a ther-motropic liquid crystal - which we prove to happen at the CP - via an applied external electric field and surface interactions caused by biomolecules at the interface. We graph-ically present the free energy behavior, which can be simplified to a 4^{th} order polynomial. We also explain how this phenomenon could be used to achieve extremely high sensitivity in liquid crystalline sensors, based on nematic liquid crystal droplets (temperature sensors, biodetectors, light-sensitive sensors). We also give a few plausible causes and mechanisms responsible for heightened sensitivity directly related to the CP in general.

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Keywords: Critical point; Liquid crystal sensors; Mesoscopic modeling





1. Introduction

Liquid crystals (LCs) have been present in complex biological organisms for millennia; however, the soft matter has been relatively thoroughly studied and explained in the last two centuries. Liquid crystalline formations (liquid crystal matter) are essential for the survival of most living creatures, including ourselves, homo sapiens sapiens, the modern human. The first liquid crystal to be discovered was myelin, which is a substance found in nerve cells and is responsible for the isolation and transmission of electrical impulses across nerves.

LCs are also building blocks of most cell membranes in form of phospholipid molecules. These, when exposed to water, turn their hydrophobic tails in a way as to make a construction commonly referred to as the lipid bilayer membrane, which exhibits LC behaviour (Seddon & Templer, 1991). This type of construction is energetically most stable. Primitive cell membranes can easily be formed with a combination of oil and water. This is useful for research purposes, and the classification of the properties exhibited by liquid crystals.

In this article, we shall take a deeper look at the fundamental physical properties of thermotropic LCs and how they react to external stimuli, such as an applied electric or magnetic field, and surface interactions caused by biomolecules such as phospholipids at the interface. We will mathematically explain the behaviour of the free energy potential and why the critical point is important for the sensitivity of liquid crystal sensors that use liquid crystal droplets as their means of sensing.

2. Liquid Crystal Sensors

The usefulness of LCs in terms of sensing is shown in an example of a thermotropic LC in an aqueous environment, where the long range orientational order is described by the order parameter S. In the isotropic phase, the order parameter is equal to S=0 and, in the nematic phase, is equal to S = 1, meaning we have complete order, and the long axes of LC molecules are oriented in a single direction. We call this a uniaxial state. A phase transition from an isotropic to nematic state is a symmetry-breaking phenomenon, and LCs in the low-temperature nematic phase become immensely sensitive to external perturbations. If we add organic molecules to an NLC in water, we notice this impacts the local change in the nematic director field \hat{n} , especially when LC droplets are present (Kaučič et al., 2004). The change in the nematic field orientation is directly correlated to the type of organic molecules the LC (or LC droplet) interacts with. We choose the type of LC based on the type of interaction between the LC and the organic molecules, which either classifies whether they are compatible or incompatible for a desired type of sensor (Lowe & Abbott, 2011). The conjuction of the properies of LCs (softness and directional order, susceptibility to external fields) and the properties of organic molecules (affinity to adhere to surfaces through polar interactions and their chemical similarity to other LCs) makes an effective sensory system. A system such as that described above is well known, however if we take into account the critical point, which is well defined by the phase behavior of water when pressure and temperature are not fixed variables, it is possible to enhance the sensitivity of such sensory systems to an unprecedented accuracy. The critical point is the point above which phase transitions are no longer discontinuous: they become phase transitions of the 2nd order. The enthalpy of a continuous phase transition above the critical point is negligible (Zid et al., 2024). At the critical point the free energy is highly dependent on the order parameter, which is proven by the extremely shallow energy potential in the vicinity of a steady state. When the system is at the critical point, in our case, the first three derivatives of the free energy function in terms of order parameter are equal to zero.





3. Mathematical Model

To mathematically describe the behavior of LCs at the critical point and their free energy potential, we begin by defining a traceless and symetric tensor \mathbf{Q} as follows:

$$Q = S\left(\hat{\boldsymbol{n}} \otimes \hat{\boldsymbol{n}} - \frac{I}{3}\right),\tag{1}$$

where \underline{I} is the identity tensor, S the order parameter and $\widehat{\boldsymbol{n}}$ the unit length of the nematic director $\overline{\boldsymbol{n}}$. If we add organic molecules such as phospholipids to the LC, the volume concentration ϕ is:

$$\phi = \frac{N_{om}v_{om}}{V},\tag{2}$$

where N_{om} is the number of organic molecules in a volume V and v_{om} the average volume of an organic molecule. Considering the symmetrically relevant energy contributions obtained by the Taylor series, we write the free energy (F) of the system as:

$$F = \int_{V} f_{v} d^{3}\vec{\boldsymbol{r}} + \sum_{i} \oint f_{i}^{(j)} d^{2}\vec{\boldsymbol{r}}, \tag{3}$$

where the voulmetric free energy density ($f_v = f_c + f_e + f_f$) contains the energy contributions of condensation, elasticity and field, respectively. $f_i^{(j)}$ represents the free energy density at the organic molecule-LC interface and measures the local interaction strength of the j^{th} organic molecule and the closest LC molecule. We assume a spatially homogenious nematic field along which we apply an external electric field, and we assume the structural equivalence of all organic molecules, meaning each of them has the same surface free energy contribution. In this case, we write the energy contributions as:

$$f_c = \frac{3}{2}a_0(T - T^*)TrQ^2 - \frac{9}{2}b(TrQ^3) + \frac{9}{4}c(TrQ^2)^2,$$
(4a)

$$f_e \sim 0,$$
 (4b)

$$f_p = -\frac{3}{2}\varepsilon_0 \Delta \varepsilon \vec{E} \cdot Q \vec{E}, \tag{4c}$$

and

$$f_i^{(j)} = -\frac{3}{2}w^{(j)} \ \hat{\boldsymbol{e}}_i \cdot Q\hat{\boldsymbol{e}}_i. \tag{4d}$$

In subEqs. (4) the parameters a_0 , b and c represent the Taylor series coefficients, whereas T^* is the lowest supercooling metastable isotropic phase temperature (Zid et al., 2024), which is similar to the lowest water vapor temperature in the temperature region, where liquid phase is stable. $w^{(j)}$ is the anchoring energy density of an organic molecule along the unit vector $\hat{\boldsymbol{e}}_i$. The local nematic field orientation is closely dictated by the interaction between the LC and an external electric field $\vec{\boldsymbol{E}}$, which is described by subEq. (4c). In the subEq. (4c) we use a Legendre polynomial of the $2^{\rm nd}$ order $P_2(\cos\theta) = (3\cos^2\theta - 1)/2$, where θ is the angle between $\vec{\boldsymbol{E}}$ and $\hat{\boldsymbol{n}}$. To clarify, let us expand the final scalar product in subEq. (4c):





$$\vec{E} \cdot Q\vec{E} = (\hat{E} \cdot \hat{n})^2 - \frac{E^2}{3} = \frac{2E^2}{3} P_2(\cos\theta), \tag{5a}$$

which is the same as:

$$\left(\widehat{\boldsymbol{E}}\cdot\widehat{\boldsymbol{n}}\right)^2 - \frac{E^2}{3} = \frac{2E^2}{3}P_2(\cos\theta). \tag{5b}$$

We notice that the value of the expression in subEq. (5a) is highest when $\theta=0^{\circ}$. Subsequently the term $\Delta \varepsilon$ in subEq. (4c) enforces a nematic field orientation along the electric field's orientation. A few important average values of the 2^{nd} order Legendre polynomial are 0, 1 and -1/2. Values 0 and 1 are extreme values and mean that we may have complete alignement of LC molecules with the direction of an applied electric field or no alignment at all. In subEq. (4d) they also measure the angle between organic molecules and the surface of an LC droplet. We express the average free interaction energy of organic molecules (F_i) as:

$$F_i = N_{om} a_{om} \bar{f}_i^{(j)}, \tag{6}$$

where $\bar{f_i}^{(j)} = w^{(j)} S \overline{P_2}$ is the average interaction free energy of one organic molecule and a_{om} the area of the organic molecule. If we take into consideration Eqs. (2) and (4d), we can rewrite Eq. (6) as follows:

$$F_i = -V\phi \frac{a_{om}}{v_{om}} w^{(j)} S \overline{P_2}. \tag{7}$$

Considering the relation $F_i/V = \overline{f_i}$, we can expand the average interaction free energy densitiy as:

$$\overline{f_i} = -\phi \frac{a_{om}}{v_{om}} w^{(j)} S \overline{P_2}. \tag{8}$$

Now, the system's effective free energy (\tilde{f}_v) is approximately expressed as $\tilde{f}_v \sim F/V$, which is the same as:

$$\widetilde{f}_{v} \sim \overline{f}_{c} + \overline{f}_{f} - \phi \frac{a_{om}}{v_{om}} w^{(j)} S \overline{P_{2}(\overline{e_{i}} \cdot \overrightarrow{n})}.$$
 (9)

We assume that all organic molecules take up the same area and $\overline{P_2(\vec{e_i} \cdot \vec{n})}$ is the average orientation of an organic molecule at the interface. $\overline{e_i}$ is the anchoring direction, locally enforced by the organic molecule. For general purposes we now switch to a dimensionless notation by introducing the dimensionless order parameter s as follows:

$$s = \frac{S}{S_0},\tag{10}$$

where $S_0 = b/2c$. We then rewrite the subEq. (4a) like so:

$$f_c = a_0 (T - T^*) S^2 - b S^3 + c S^4$$
 (11)

and, while taking into account Eq. (10) also as:

$$f_c = cS_0^4 \left[\frac{a_0 (T - T^*)}{cS_0^2} s^2 - \frac{b}{cS_0} s^3 + s^4 \right].$$
 (12)





Considering the expression $S_0 = b/2c$ and Eq. (12), we can define the reduced temperature r^{ef} as:

$$r^{ef} = \frac{a_0(T - T^*)4c}{b^2},\tag{13}$$

which also satisfies the phase transition condition at the value $r^{ef}=1$. Eq. (13) can otherwise be expressed as:

$$r^{ef} = \frac{T - T^*}{T_{IN} - T^*}. (14)$$

We now obtain a relation $T_{IN} = T^* + b^2/(4a_0c)$, which is the phase transition temperature from an isotropic to nematic phase in a thermotropic liquid crystal and it's droplets, formed at the interface. The sum of $\overline{f_t}$ and $\overline{f_t}$ can be writen as:

$$\overline{f_f} + \overline{f_t} = \left[-\varepsilon_0 \Delta \varepsilon E^2 - \phi w \frac{a_{om}}{v_{om}} \right] \overline{P_2} S, \tag{15}$$

Which is equal to:

$$\overline{f_f} + \overline{f_L} = a_0 (T_{IN} - T^*) S_0^2 [-\sigma_E - \sigma_w] \overline{P_2} s, \tag{16}$$

where the following relations hold:

$$\sigma_E = \frac{\varepsilon_0 \Delta \varepsilon E^2 S_0}{a_0 (T_{IN} - T^*) S_0^2} \tag{17a}$$

and

$$\sigma_{w} = \frac{\phi w a_{om} S_{0}}{v_{om} a_{0} (T_{IN} - T^{*}) S_{0}^{2}}.$$
(17b)

From Eq. (17b) it's clear, that the value of the parameter σ_w is heavily dependent on the geometry (a_{om} and v_{om}) of the organic molecule (can also be a nanoparticle or other impurity), material properrties (a_0 , T_{IN} , T^* and S_0) and the concentration of oorganic molecules (ϕ). The effective control parameter is now $\sigma^{ef} = \sigma_E + \sigma_w$. Considering Eqs. (12) and (13), we can rewrite Eq. (9) as:

$$\tilde{f}_v = f_0(r^{ef}s^2 - 2s^3 + s^4 - \sigma^{ef}s). \tag{18}$$

We now introduce the dimensionless effective free energy density $\tilde{f} = \tilde{f}_v/f_0$, where $f_0 = a_0(T_{IN} - T^*)S_0^2$, as:

$$\tilde{f} = r^{ef} s^2 - 2s^3 + s^4 - \sigma^{ef} s. \tag{19}$$

Eq. (19) can be expanded as:

$$\tilde{f} = A(s - s_1)^2 (s - s_2)^2 + B, \tag{20}$$

where A and B are constants, and s_1 and s_2 the effective free energy function zero values. If we equate the the coefficients in Eqs. (19) and (20), we obtain the next relations:

$$\sigma^{ef} = 2A(s_1^2 s_2 + s_1 s_2^2), \tag{21a}$$





$$r^{ef} = A(s_1^2 + 4s_1s_2 + s_2^2), (21b)$$

$$s_1 = 1 - s_2 \tag{21c}$$

And

$$A = 1. (21d)$$

We now minimize the system's free energy by demanding that the first derivative of function given by Eq. (19), being $\tilde{f}(s)$, is equal to zero. We now obtain three possibilities: a minimum, a maximum or curve. The minimum is determined by the second derivative condition $\partial^2 \tilde{f}/\partial s^2 > 0$. The third derivative gives the rate of change in curvature and at the CP it hold, that it is also equal to zero, meaning it's a flat line. The conditions which are unequivocally true at the CP are as follows:

$$\frac{\partial^2 \tilde{f}}{\partial s^2} = 0 \tag{22a}$$

and

$$\frac{\partial^3 \tilde{f}}{\partial s^3} = 0. {(22b)}$$

The 1st order pahse transition conditions are given by:

$$r^{ef} = 1 + \sigma^{ef} \tag{23a}$$

And

$$\sigma^{ef} < \sigma_{cp}^{ef}. \tag{23b}$$

At the *CP* it then holds: $r_{cp}^{ef} = 1 + \sigma_{cp}^{ef}$ and $\sigma^{ef} = \sigma_{cp}^{ef}$. When conditions given by subEqs. (23) are realised, the following is also true:

$$s_c^{(-)} = \frac{1 - \sqrt{1 - 2\sigma^{ef}}}{2} \tag{24a}$$

and

$$s_c^{(+)} = \frac{1 + \sqrt{1 - 2\sigma^{ef}}}{2},\tag{24b}$$

where $s_c^{(-)}$ and $s_c^{(+)}$ are the minima of the $\tilde{f} = \tilde{f}(s)$ function. The relation $s_c(\sigma^{ef})$ is shown in **Figure 1**.





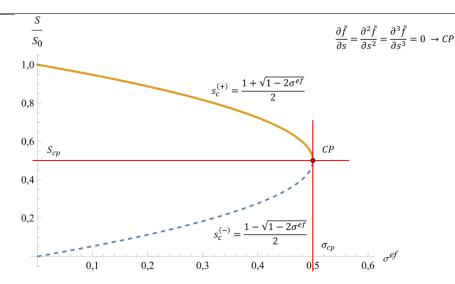


Figure 1. The relation between the normalized order parameter s and the effective control parameter σ^{ef} . The orange curve represents values of $s_c^{(+)}$ and the blue one the values of $s_c^{(-)}$. The black dot marks the critical point CP which has the coordinates (1/2, 1/2). S_{cp} is the critical value of the order parameter S and σ_{cp} the critical value of the effective control parameter σ^{ef} . Red lines help in marking the CP's coordinates.

The minimization of the effective free energy, as shown in this example, can only be used when the system is completely homogenious; the parameters in Eqs. (15-17b) are spatially unrelated. At the *CP* the condition $s_c^{(+)} = s_c^{(-)}$ is true. Higher values of σ^{ef} than σ^{ef}_{cp} cause a supercritical phase behavior (Immanuel et al., 2019). Our system is at the *CP* when the following is true:

$$\sigma_{cp}^{ef} = \frac{1}{2}, r_{cp}^{ef} = \frac{3}{2}, s_{cp} = \frac{1}{2}.$$
 (25)

Normalized values of the free energy density function $\tilde{f}(s)$ are shown in Figure 2.

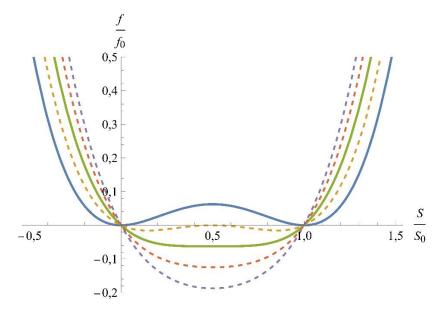


Figure 2. Free energy density during phase transition. Dark blue curve: $\sigma^{ef} = 0$, orange slitted curve: $\sigma^{ef} = 0.75$, green curve: $\sigma^{ef} = 0.75$, red slitted curve: $\sigma^{ef} = 0.75$, purple slitted curve: $\sigma^{ef} = 1$.

We observe that the potential becomes incredibly flat at the *CP*. Subsequently, this enables strong fluctuations in the LC molecule orientation, which in turn gives a very sensitive





system, the phase of which can be changed by the slightest external perturbation, which can easily be observed qualitatively. Similar techniques and phenomena could be useful in future sensory systems, where precision and sensitivity are of utmost importance (in medicine for dosing antibiotics, in food industry for dosing aditives or in biology for measuring the concentrations of biomolecules present in a sample).

4. Experimental Relevance

Experimentally, the sensitivity of LC droplets is usually measured in an aqueous environment where liquid crystal droplets have formed. We may introduce low-wavelenght light or impurities such as nanoparticles or biological molecules into such an environtment. We then observe at what intensity of light or concentration of impurities the LC droplets reconfigure form usually bipolar to radial configuration and obtain an effective measuring system that can further be used in real-scenario sensors that are sensitive to light or different molecular structures. If, in such experiments, the LC was at its critical point, the sensitivity would cerainly be higher, because the free energy pottential for a phase transition is already at it's minimum, which causes the energy difference between competing structural configurations to also be minimized. In the following paragraphs, we shall discuss additional causes for enhanced sensitivity at the *CP*.

At the *CP*, a few very important mechanisms are responsible for enhancing the sensitivity of LC droplets and LCs in general. The first one is the divergence of correlation length. This is the distance over which fluctuations in molecular alignment are correlated. Because these distances become large, the result is the collective mollecular behavior over macroscopic distances, making discussed sensory systems highly responsive to small perturbations in temperature, external fields and stress (Nicastro, 1984).

The relaxation time for fluctuations increases dramatically at the *CP*, meaning the system takes longer to return to equilibrum state after being perturbed. This phenomenon amplifies the effects of external perturbations, acting as a magnifying glass in terms of sensitivity. We call this critical slowing down (Zid et al., 2024).

Thermal fluctuations in order parameter (S), such as the degree of molecular alignment in LCs, are amplified and make the system more sensitive to external forces, which can significantly alter the phase state that the system is in.

Finally, at the *CP*, the response of an LC to external fields oftern becomes nonlinear, due to the system's inherent instability and the competition between different phases.

5. Conclusion

Liquid crystal formations have been around for millenia, and while incredibly important for living beings, they are also very useful in sensory systems. Their susceptibility to external magnetic and electric fields or impurities makes them excellent candidates for electrical appliances such as LC displays. It also makes them great test subjects in environments, where our goal is to measure a certain concentraion of nanoparticles or biomolecules in an aqueous environment and the sensitivity of certain electric fields and magnetic fields.

The mathematical model is very important in determining what mainly causes an LC to change its phase and how to finely tune its sensitivity through the mentioned perturbations. It also shows that a complex free energy system can easily be generalized by a 4th order polynomial to show the change in its potential below, at and above the critical point. Through this we can easily determine where the system is the most sensitive.

Conflicts of Interest: The author declares no conflict of interest.







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Research

Volume and Surface Phase Transitions in Confined Nematic Liquid Crystals

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Abstract:

We study bulk and surface phase transitions in confined nematic liquid crystals exhibiting 1st order isotropic-nematic phase transition in bulk systems. We demonstrate analytically that confinement could in general quantitatively and even qualitatively modify phase behavior. Furthermore, we show that confining substrates could enable surface phase transitions. Studied phase behaviors could be applied also to other condensed matter systems displaying 1st order continuous symmetry order-disorder phase transition if the relevant order parameter amplitude is linearly coupled to a local surface ordering field.

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1. Introduction

Liquid crystals (LCs) exhibit both liquid and crystal-type of order (Kleman & Lavrentovich, 2004). They consist of weakly interacting anisotropic molecules in which relatively fast noncollective fluctuations average out microscopic details. The resulting system exhibits orientational or even translational order that could be well theoretically described by mesoscopic order parameter fields. Due to microscopic averaging the resulting phase behaviour on varying a relevant control parameter (temperature, pressure, concentration of LC molecules...) often exhibits several universal features, which depend mostly on the symmetry properties of order parameter.

In this contribution we focus on thermotropic nematic LCs consisting of rod-like molecules which in bulk systems (where confinement effects could be neglected) exhibit on varying temperature 1st order isotropic (I) – nematic (N) phase transition. The isotropic phase possesses liquid-like isotropic behavior, where all directions in space are equivalent. This phase is stable at relatively high temperatures. On decreasing temperature, the nematic phase is formed which in bulk equilibrium exhibits spatially homogeneous uniaxial order along a single symmetry breaking direction. On the other hand, the center of mass of molecules could flow like in a liquid. Note that due to the continuous symmetry breaking any spatial direction could be chosen. For this reason, the system could exhibit the so-called Goldstone modes (Kleman & Lavrentovich, 2004), due to which LC order could be relatively easily excited by various perturbations. Because of this susceptibility to various stimuli, LCs are also referred to as soft materials. Furthermore, uniaxial orientational order of transparent LC molecules yield optic anisotropy. Therefore, nematic LCs are particularly useful for sever applications which rely on fast enough external electric or some other field driven optical changes.

In this contribution we illustrate analytically how a confinement of nematic LC within a plan-parallel cell could affect its phase behavior which could be quantitatively and even qualitatively different from bulk behavior.

2. Material and Methods

We use a minimal model to describe the impact of confinement on the I-N phase behaviour. We use a Landau-de Gennes type modelling where one determines the nematic orientational order by the scalar order parameter S and the nematic director field \hat{n} . This unit vector defines a local uniaxial direction of nematic order, where the states $\pm \hat{n}$ are physically equivalent. The amplitude S determines the degree of nematic order and S=1 corresponds to the maximal nematic order, where LC molecules would be firmly aligned along \hat{n}

In our approximate treatment we allow spatial variations along a single coordinate, say x-axis of the Cartesian coordinate system (x,y,z). We use a one-dimensional system which is enough to identify key mechanisms that can impact I-N phase behavior. For simplicity we parametrize the director as $\hat{n} = (\sin\theta, \cos\theta, 0)$. In terms of S(x) and $\hat{n}(x)$ we express the free energy of the system, where LC is confined between parallel plates placed at x = 0 and x = h, where h stands for the cell thickness. The free energy F per surface area A is given by (Ranjkesh et al., 2014)

$$\frac{F}{A} = \int_{0}^{h} \left[f_c + f_e + f_f \right] dx + f_i(x = 0) + f_i(x = h) \tag{1}$$

in terms of the condensation (f_c) , elastic (f_e) , external field (f_f) , and interfacial (f_i) free energy densities. Here we assume that an external electric field $\vec{E} = E\hat{e}$ is also present, enforcing orientation parallel to \hat{e} . The condensation term determines S in bulk equilibrium, the elastic terms penalize spatial variations in orientational order, the field term tends to align LC molecules along the external field direction for LCs with positive dielectric anisotropy, and the interfacial term determines conditions at LC confining substrates. We express the free energy densities in the simplest form, where we include only the key terms affecting the LC phase behavior. It follows (Ranjkesh et al., 2014)

$$f_c = a_0 (T - T^*) S^2 - b S^3 + c S^4, (2a)$$





$$f_e = \frac{L}{2} \left(\frac{\partial S}{\partial x} \right)^2 + \frac{LS^2}{2} \left(\frac{\partial \theta}{\partial x} \right)^2, \tag{2b}$$

$$f_f = -\frac{\Delta \varepsilon \varepsilon_0 E^2}{2} SP_2(\hat{n}.\,\hat{e}),\tag{2c}$$

$$f_i = -\frac{wS}{2}. (2d)$$

Here a_0 , T^* , b, c, L are positive material parameters, $P_2(x) = (3x^2 - 1)/2$, ε_0 is the vacuum permittivity and $\Delta \varepsilon$ is the LC dielectric anisotropy. We set that the interfacial term locally supports nematic, order. Consequently, the interaction (wetting) strength constant w is positive. For simplicity we set that the conditions at both confining substrates are the same.

3. Results

3.1. Spatially homogeneous system

Firstly, we consider systems in which w is relatively weak. To simplify the problem, we neglect spatial variations in S and set $P_2 = 1$ (i.e., \hat{n} is aligned along \vec{E}). Furthermore, we rewrite the free energy into a dimensionless form. For this purpose, we introduce scaled quantities

$$s = \frac{S}{S_0}, u = \frac{x}{h}, r = \frac{T - T^*}{T_{IN} - T^*}.$$
 (3)

where $S_0 = b/(2c)$ is the equilibrium order parameter at the bulk I-N phase transition temperature $T_{IN} = T^* + \frac{b^2}{4a_0c}$. Here s, u and r stand for the scaled nematic order parameter, dimensionless Cartesian coordinate and temperature, respectively.

Furthermore, we introduce the material dependent lengths (Kleman & Lavrentovich, 2004) that characterise the nematic response to different perturbations:

$$\xi_e = \sqrt{\frac{LS_0^2}{\Delta \varepsilon \varepsilon_0 E^2 S_0}}, \xi_{IN} = \sqrt{\frac{L}{a_0 (T_{IN} - T^*)}}, d_e = \frac{LS_0}{w}.$$
 (4)

Here ξ_e , ξ_{IN} and d_e stand for the external electric field extrapolation length, nematic order correlation length and interface extrapolation length. All these distances characterise bulk LC responses and could be measured. They are temperature dependent and for this reason we express them at T_{IN} .

Taking this into account we obtain the following expression for the scaled free energy

$$\tilde{f} = \frac{F}{A \ a_0 (T_{IN} - T^*) S_0^2 h}$$

$$\tilde{f} = r_{eff}s^2 + 2s^3 + s^4 - \sigma_{eff}s. {5}$$

In this expression we introduced the scaled effective temperature r_{eff} and the scaled effective interaction σ_{eff} :

$$r_{eff} = r + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e, \tag{6}$$

$$\sigma_{eff} = \frac{\xi_{IN}^2}{\xi_e^2} + \frac{\xi_{IN}^2}{h \ d_e},\tag{7}$$

where

$$\tilde{g}_e = \int_0^1 \left(\frac{\partial \theta}{\partial u}\right)^2 du \tag{8}$$





One can obtain phase transition conditions analytically by assuming that at a phase transition the free energies of the competing phases are equal. Therefore, \tilde{f} could be expressed as

$$\tilde{f} = A(s - s^{(+)})^2 (s - s^{(-)})^2 + B \tag{9}$$

where $s^{(+)}$ and $s^{(-)}$ determine equilibrium values of order parameter just above and below the phase transition. By equating Eq.(5) and Eq.(9) we got a system of equations by requiring that coefficients in front of s, s^2 , s^3 and s^4 are equal. The resulting phase transition conditions read

$$r_{eff}^{(c)} = r_c + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e = 1 + \sigma_{eff}, \tag{10a}$$

$$s^{(-)} = \frac{1 + \sqrt{1 - 2\sigma_{eff}}}{2},\tag{10b}$$

$$s^{(+)} = \frac{1 - \sqrt{1 - 2\sigma_{eff}}}{2} \tag{10c}$$

providing the effective interaction σ_{eff} is weak enough:

$$\sigma_{eff} < \sigma_c = 0.5. \tag{11}$$

Here $r_{eff}^{(c)}$ and r_c determine values of r_{eff} and r at the transition. In the supercritical regime, where $\sigma_{eff} > \sigma_c$, the phase transition is replaced by gradual variation of orientational order on varying the temperature.

3.2. Nonhomogeneous system

Next, we consider regime where w is relatively large. Consequently, we assume that above each confining substrate a thin surface layer (we refer to it as an *interface*) exists where the degree of order is larger than in the remaining LC volume (i.e., *volume* in continuation). We set that $s \sim s_i$ and $s \sim s_v$ in the *interface* and *volume* part of the system. Furthermore, we assume that the *interface* width is roughly given by the nematic correlation length ξ .

The average dimensionless free energy in the *volume* part of the system is roughly given by

$$\tilde{f}_{v} \sim r_{eff}^{(v)} s_{v}^{2} - 2s_{v}^{2} + s_{v}^{2} - \sigma_{eff}^{(v)} s_{v}, \tag{12}$$

where

$$r_{eff}^{(v)} = r + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(v)}, \sigma_{eff}^{(v)} = \frac{\xi_{IN}^2}{\xi_e^2}.$$
 (13)

On the other hand, the phase behaviour of interfaces is roughly described by

$$\tilde{f}_i \sim \Delta u \left[r s_i^2 - 2 s_i^3 + s_i^4 + s_i^2 \tilde{g}_e^{(i)} \frac{\xi_{IN}^2}{h^2} - s_i \frac{\xi_{IN}^2}{\xi_e^2} \right] - s_i \frac{\xi_{IN}^2}{d_e h},\tag{14}$$

where $\Delta u \sim \xi/h$. It follows

$$\frac{\tilde{f}_i}{\Delta u} \sim r_{eff}^{(i)} s_i^2 - 2s_i^2 + s_i^2 - \sigma_{eff}^{(i)} s_i.$$
 (15)

$$r_{eff}^{(i)} = \frac{T - T^*}{T_{IN} - T^*} + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(i)}, \sigma_{eff}^{(i)} \sim \frac{\xi_{IN}}{d_e}.$$
 (16)

In expressing $\sigma_{eff}^{(i)}$ we assumed $\xi_{IN} \sim \xi$.

Phase behaviours in the *volume* and *interface* part can be solved analytically as described in the previous subsection. In the *volume* part subcritical behaviours are obtained for $\sigma_{eff}^{(v)} < \sigma_c \equiv 0.5$. In this regime a *volume* 1st phase transition occurs when the condition





$$r_{eff}^{(v,c)} = \frac{T_c - T^*}{T_{IN} - T^*} + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(v)} = 1 + \sigma_{eff}^{(v)}$$
(17)

is fulfilled. If $\sigma_{eff}^{(v)} > \sigma_c$ the *volume* LC exhibits noncritical behaviour, i.e., s_v varies gradually on varying the temperature.

Similarly, the *interfacial* part exhibits a 1st order phase transition if $\sigma_{eff}^{(i)} < \sigma_c$ and the phase transition is determined by

$$r_{eff}^{(i,c)} = \frac{T_c - T^*}{T_{IN} - T^*} + \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(i)} = 1 + \sigma_{eff}^{(i)}.$$
(18)

We analyzed analytically phase behavior is system where the surface interaction is relatively weak or strong. In the first case one sensibly assume a homogeneous order in the whole system and the effective free energy is given by Eq.(5). Minimization of free energy reveals that the system exhibits 1st order phase transition only in the regime σ_{eff} < 0.5. In this case the phase transition temperature T_c is given by Eq.(10a), yielding

$$\Delta T_c = (T_{IN} - T^*) \left(\frac{\xi_{IN}^2}{\xi_e^2} + \frac{\xi_{IN}^2}{h \ d_e} - \frac{\xi_{IN}^2}{h^2} \tilde{g}_e \right), \tag{19}$$

where $\Delta T_c = T_c - T_{IN}$ measures the temperature shift with respect to the bulk system. One sees that the external electric field and the surface wetting interaction tend to increase the phase transition temperature. Note that the surface contribution scales with 1/h. Hence, its impact increases on decreasing h. Furthermore, elastic distortions decrease T_c . Furthermore, in the subcritical regime (i.e., $\sigma_{eff} < \sigma_c$) the system exhibits finite ordering also above T_c providing $\sigma_{eff} > 0$, to which one commonly refers as the *paranematic* order. Values of nematic and paranematic order just below and above T_c are given by Eq.(10b) and Eq.(10c), respectively. Note that for $\sigma_{eff} = 0$ it holds $s^{(-)} = 1$ and $s^{(+)} = 0$, where the paranematic phase is replaced by isotropic order.

In the supercritical regime, where $\sigma_{eff} > \sigma_c$, the phase transition is replaced by a gradual variation of orientational order on varying the temperature. The condition separating critical and subcritical regime is determined by

$$\frac{\xi_{IN}^2}{\xi_a^2} + \frac{\xi_{IN}^2}{h \ d_a} = 0.5. \tag{20}$$

This condition can be reached either by a strong enough electric field or by a large enough surface interaction strength, where in the latter case the critical interaction strength decreases with decreasing *h*.

On the other hand, if w is relatively strong then one should distinguish between volume and interface LC contribution. In general, they could exhibit different phase behaviour what has been also experimentally observed (Boamfa et al., 2003). In such cases the volume part exhibits a 1st order phase transition if $\frac{\xi_{IN}}{\xi_{\rho}^2}$ < 0.5 (see Eq.(13)) where

$$\Delta T_c^{(v)} = (T_{IN} - T^*) \left(\frac{\xi_{IN}^2}{\xi_e^2} - \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(v)} \right). \tag{21}$$

If $\frac{\xi_{IN}^2}{\xi_e^2} > 0.5$ systems exhibit gradual evolution of order on varying the temperature. On the contrary, interfaces exhibit 1st order phase transitions if $\frac{\xi_{IN}}{d_e} < 0.5$ (see Eq.(17)) where it holds

$$\Delta T_c^{(i)} = (T_{IN} - T^*) \left(\frac{\xi_{IN}}{d_e} - \frac{\xi_{IN}^2}{h^2} \tilde{g}_e^{(i)} \right). \tag{22}$$

For $\frac{\xi_{IN}}{d_a} > 0.5$ interfaces exhibit gradual phase behaviour. Note that in such cases $T_c^{(v)}$ and $T_c^{(i)}$ (i.e., volume and interface phase transitions) are in general different. Furthermore, it might happen, for example, that the volume part exhibits the noncritical behaviour and the interface subcritical behaviour.







5. Conclusions

In this contribution we addressed phase behavior in confined nematic liquid crystals using a minimal model. We focused on key features that quantitatively or even qualitatively alter the bulk discontinuous I-N phase transition. Note that the approach shown could be applied also to other systems in which order parameter could exhibit different symmetries. For example, nematic LC possesses an axial symmetry and similar approach could be used to model ferromagnetic or ferroelectric systems, where one can use a vector field for the mesoscopic order parameter field. The following conditions need to be fulfilled to apply the illustrated approach to a different system: (i) the system should in bulk exhibit 1st order phase transition, (ii) continuous symmetry breaking should be realized in the phase transition (i.e., the symmetry broken phase should exhibit infinity different but equivalent ground states), (iii) the order parameter amplitude, measuring the strength of order (i.e. *S* in our illustration) should be linearly coupled to the relevant ordering field.

Conflicts of Interest: The authors declare no conflict of interest.

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Research

Symmetry Breaking and Patterns

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Abstract:

In the present study we analyse different patterns commonly encountered in nature. Firstly, we review patterns that display the hexagonal symmetry and triply periodic minimal surfaces. We analyse hexagonal honeycomb configurations and analyse advantages of such an architecture. Next, we use a simple Landau-type order parameter field model to illustrate how diverse patterns could be generated via the symmetry breaking mechanism.

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We study bulk and surface phase transitions in confined nematic liquid crystals exhibiting 1st order isotropic-nematic phase transition in bulk systems. We demonstrate analytically that confinement could in general quantitatively and even qualitatively modify phase behavior. Furthermore, we show that confining substrates could enable surface phase transitions. Studied phase behaviors could be applied also to other condensed matter systems displaying 1st order continuous symmetry order-disorder phase transition if the relevant order parameter amplitude is linearly coupled to a local surface ordering field.

Keywords: Liquid crystals; Phase transition; Confined nematic liquid crystal; Surface phase transition





1. Introduction

Our surrounding world displays an enormous complexity and an infinity of diverse patterns. One would expect that it is difficult to describe key mechanisms enabling such diversity. However, it seems that the basic principles of matter originate in symmetry and symmetry breaking (Kibble, 1976; Sethna, 1992; Zurek, 1985). For example, the standard model of particles is based on symmetry breaking of local quantum fields and it seems that conserved quantities in nature could be explained by taking into account relevant systems' symmetry properties (Kibble, 1976; Sethna, 1992; Zurek, 1985).

Different materials forming solid and soft materials (Kleman et al., 2003) in our world can be viewed as being built using only three different "building blocks", i.e., electrons, protons and neutrons. These are combined in atoms following quantum mechanical rules and furthermore, atoms could assemble in further higher-hierarchical configurations. Furthermore, it seems that key natural behaviors could be explained using geometrical concepts (Imry and Ma, 1975; Kibble et al., 1976; Sethna, 1992; Zurek, 1985).

In the following we present some i) systems exhibiting hexagonal symmetry or ii) systems, that are making use of the minimal surface principle to illustrate how structures are related to different emerging functionalities.

1.1. Hexagonal lattices

We first focus on hexagonal lattices which are ubiquitously realised in nature. The typical lattice consists of hexagons as shown in **Figure 1**. One see that blue and yellow coloured elements, which form the lattice, are not connected the same way. Blue atoms have a neighbour horizontally to the right and two more diagonally to the left. The situation is inversed for the yellow ones. This lattice can be described in terms of three base vectors **a**_i, via which each lattice point could be located. Hexagons are also one of three shapes that can cover a 2D plane just by themselves, the other two being triangles and squares. But hexagonal tiling is the closest packing way to arrange circles in two dimensions (the best way to divide a surface into regions of equal area with the least total perimeter) which was proven by Thomas C. Hales (Hales, 2001).

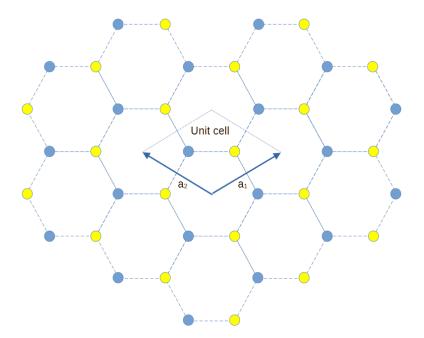


Figure 1: Hexagonal lattice in 2D in which we show the unit cell and the corresponding base vectors.





Hexagonal lattices are widely used in various applications. One example are hexagonally shaped fence from galvanized steel, also known as the chicken mesh. Furthermore, Mebrahtom et al. (2024) compared the chicken mesh with welded square mesh and expanded metal mesh in tests, where they added mentioned meshes to concrete beams. They tested their energy absorption (free fall), compressive strength (capacity of a material to withstand loads that try to reduce its size) and flexural strength (the ability of a material to withstand bending under the application of external force). The chicken mesh outperformed the other two in all the tests. A single layer increased the flexural strength of a beam by 8 times (compared to 6 and two times for other two), reduced the compressive strength by less than 1% (better than over 1% for welded mesh and 8% reduction for expanded mesh), while it could absorb 255 J of energy from free fall (compared to 198 J and 188 J for other two). That in combination with its cheap price makes it a great contender for reinforcing concrete beams.

Furthermore Carbon, which is the base of life on Earth, is often found forming hexagonal lattices in forms like graphite, graphene or nanotubes. Latter two forms have high ultimate tensile strength, up to 130,000 MPa for graphene (Lee et al., 2008) and (theoretically) up to 300,000 MPa for nanotubes (Yu et al., 2000). For comparison, different forms of steel can be found to have around 2,000 MPa. Graphite on the other hand is thought to be one of the first crystals to ever condense, possibly only behind diamond (Hazen et al., 2013). Graphite could be transformed into diamond only under pressure of a few GPa, depending on the temperature (1 GPa = 10,000 bar). At 5000 K and 12 GPa a triple point of carbon was found (where carbon is present in liquid, diamond and graphite form)(Bundy et al., 1994). One layer of graphite yields graphene, the miracle material. This is partly due to the fact that for decades, it was believed that 2D materials were non-existent in nature (Ding et al. 2019). Part of the problem is that the bending stiffness (the resistance of a member against bending deflection/deformation) based on continuum mechanics theory is proportional to the cubic power of the thickness. Therefore it should be negligible for 2D materials. But the discovery of graphene in 2004 changed the narrative. Ding et al. (2019) found that when it comes to 2D lattices, hexagonal is superior to triangular and square. This is because, in contrast to the other two, the hexagonal lattice experiences variation of the bond angles during bending, providing finite bending stiffness and thus great stability. With their model they also found that repulsive moment of atoms arranged in a hexagonal lattice increases the structural stability of the lattice. The same cannot be said for triangular and square lattices, as their attractive inner moment renders them unstable, which was shown by using the same model.

Back to graphene, it is said to conduct electricity better than any other material known to science, and is known for its material strength. Its heat conductivity was also believed to be beyond any other material, however Han and Ruan found that this is not the case (Han and Ruan, 2023). Many papers put the thermal conductivity of graphene between 3,000 and 5,000 W/mK, but Ruan's team measured it to be around 1,300 W/mK, which falls short of diamond (2,000 W/mK). This figures are all huge, as thermal conductivity of iron for example is around 80 W/mK at room temperature. But graphene is still the front runner for superconductivity of electricity. Superconductivity is the phenomenon when the electrical resistance of the material drops to zero and magnetic fields are expelled from it. This only happens once the temperature of the material drops under the critical temperature. One way to make graphene into a superconductor is to take two sheets of the magical material and twist them for the "magic angle". At the temperature of under 1,7 K under an angle of roughly 1.05° the material becomes superconducting. The term magic angle was suggested in 2011 (Bistritzer and MacDonald, 2011), although it wasn't known back then it would lead to superconductivity, rather it was known that band structures appear in which electrons are in a way isolated from each other. More options for superconductivity are made with more layers of graphene. With three layers, twisted by 1.5° relative to each other, the material becomes superconductive at roughly 2.5 K. The mechanism behind this exact observation remains unknown (Garisto, 2023).





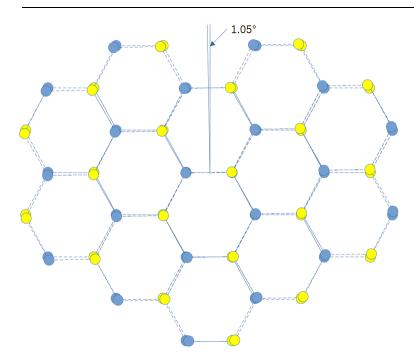


Figure 2: Two graphene lattices, twisted by 1.05°.

Finally, that same lattice that makes concrete stronger and graphene into a miracle material, is also used by animals such as bees. They build cylindrical cells that later transform into hexagonal prisms, achieving a maximal surface/perimeter ratio, through a process that it is still debated, even though the honeycomb has been a subject of thought since the antiquity, when it caught the eye of the Roman philosopher Marcus Terentius Varro. It could be explained partially by action of physical forces, as well as behaviour of bees, as they use their legs to measure distances (Nazzi, 2016). They also use their bodies to keep the temperature of the wax at around 35 °C, which is a bit below the temperature where wax exists in a liquid-solid equilibrium at 40 °C, which still allows bees to modify wax in hexagonal patterns, suggesting that behaviour of bees could be the answer (Bauer and Bienefeld, 2012). Further articles (Nazzi, 2016) talk about a similar idea of bees themselves creating hexagonal pattern. The bees were shown to mostly start constructing a new cell from the groove formed by the two previous cells. However, bees also build honeycombs starting from many different points, and this is where constructing a perfect hexagonal lattice becomes impossible (Smith et al., 2021). But they have evolved to change their behaviour in order to sew parts of the honeycomb together with pentagons and heptagons. They can also tilt individual hexagons in order to fill out the pattern. Keep in mind that the bees have limited global awareness and make decisions based on local information. And they still get it right, so we can conclude that bees are some of the most skilled architects out there.

1.2. Triply periodic minimal surfaces

Triply periodic minimal surfaces or TPMS are structures with two main defining features. The first is their minimal surface trait. In topology, for a surface to be deemed minimal, it has to satisfy a simple requirement: the mean curvature at each point on the surface is zero. In other words, they are equally convex and concave at all points or saddle-like or hyperbolic in shape. The name comes from the fact that, given a fixed boundary, their surface is the smallest compared to other surfaces constructed under the same boundary conditions. The second feature is the triply periodicity. This means that the structure has three base vectors and thus repeats periodically in 3 directions or is in other words a three-dimensional structure. Most TPMS are non-intersecting and divide space into separate volumes. These volumes can be tailored to be different in size in relation to each other. In our cases there will only be two of these volumes.





There are many different types of TPMS with new ones still being investigated or just theorized. The main types of TPMS structures are the primitive (P), diamond (D) and gyroid (G) types. They can be depicted in multiple ways. The simplest is in the form of a skeletal structure. The skeletal structures represent the volumes that are separated by the surfaces. These lattices are the same in our cases, with them being rotated and intertwined. The primitive TPMS has a simple cubic skeletal structure that represents one of the volumes in red and another simple cubic structure that represents the other volume in blue as seen in **Figure 3**.

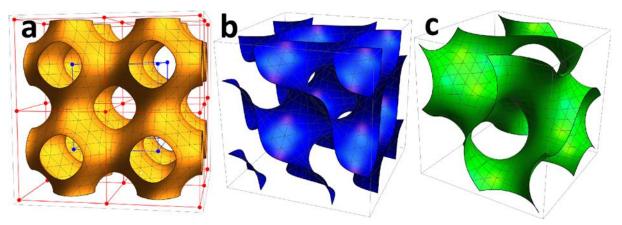


Figure 3: Examples of TPMS: a) primitive TPMS with lattices as depiction of volumes; b) diamond TPMS; c) gyroid TPMS.

In the following we list some of peculiar features enables by TPMS structures. There are two ways of creating a coloured surface. The first is with the use of dyes. These work by simply absorbing or reflecting certain parts of the spectrum of visible light, due to their chemical make-up. The second are structural colours. These are produced due to the way material is shaped. These structures use physical phenomena such as interference or diffraction to selectively reflect different wavelengths of light at different intensities. In the case of TPMS the phenomenon that we see happen is mainly Bragg scattering. Bragg scattering is a phenomenon that was first observed in atomic lattices. The interaction between the atoms and the waves, when they enter a crystal, can be modelled by creating parallel planes at a distance d from one another off which light would bounce. If the wavelength of light and angle of the incident waves is just right the constructive interference occurs. Otherwise, we get destructive interference or there is no change in intensity. This means that a peak of intensity of certain wavelengths of light is created. The peak is what we primarily then interpret as the colour of the object. The colour of parts of the wing or even the whole wing can change as we view it from different directions, because of the angle dependence of the incoming and thus reflected light. We also see a metal-like shine due to the method of colouring, that is not common in dyes.

The coloured part of butterfly wings are really scales, that are attached to the main body of the wing. Butterflies use many different techniques to attain the colours of these wings. In some cases, they use simple dyes and in others they use different types of structural colouring methods. What we do see is that in the latter cases, traditional dyes like melanin are also in some cases used as a base. These dyes cause the structural mechanisms to work better, as they also help to absorb light.

Firstly, to determine what method of colouring is used. Saranthan et al. used a method of imaging called small angle X-ray scattering or SAXS to take measurements of the nanostructure of certain species of butterfly wings, which were previously thought to have TPMS structures, to determine which type of structure they use. They found that the five species they imaged all used the gyroid structure. The structure is comprised of chitin and air. They also then, using this information, predicted what the reflective spectra of the butterfly wing scales would be. They found that the real and simulated spectra both match up well (Saranathan, 2010).





Beetles also use TPMS structures, that are created in the form of dots on their carapace, to display colours. Galusha (2008) investigated the scales of the weevil Lamprocyphus augustus using high-resolution imaging techniques and simulations to determine that these weevils use a diamond TPMS structure as their colouring method. They also found that these structures have a sort of grainy composition to them. The structure is not perfect like we would expect to see in a crystal but shows domains of periodicity. They also found that it is thanks to these non-uniform assemblies that the beetles have a near angle-independent coloration, which means that you can see these colours from any angle (Galusha, 2008). TPMS is encountered also in the mitochondria of amoeba (Chaos carolinensis). The cell is commonly known as a sort of ball of living material floating around in an environment. The only thing dividing these two environments is the membrane on the outside of the cell. And just as we have smaller masses called organs inside of us that have their own specific function and are separated from the rest of the body by different layers, so does the cell too have its own organelle. These have their own outer and inner layers. These layers have recently in some cases been found to be cubic membrane structures which are analogous to TPMS. Examples from the work of Landh included the smooth endoplasmic reticulum, mitochondrial inner membrane and chloroplast thylakoid membranes (Landh, 1995). Deng et al. have used transmission electron microscopy and simulations of TPMS structures to find the structure used in the cells of the giant amoeba Chaos carolinensis (Deng et al., 2017). They found that the structure best aligns with a double diamond structure, which is a more complex variant of the normal diamond structure. The exact reasons for these structures are still not fully known although a few theories and advantages have been found. One theory, as proposed by Deng et al. is that because these amoebae do not have a way of going into a dormancy state, the mitochondria constantly must produce energy (Deng et al., 2017). The most common source of this energy during starvation are the membranes of the mitochondria. Because the mitochondria still need to uphold a certain surface to volume ratio while their membrane is being consumed, they take the form of a very efficient structure (Deng et al., 2017).

Deng et al. have also found another possible use of these structures in mitochondria. They tested the oxidative damage that was dealt to RNA in the presence of two different mitochondria structures. They used the mitochondria of a 7-day starved amoeba and those from a mouse liver, which do not have TPMS structures. As another control, they tried comparing the effects from the mitochondria of starved and non-starved amoeba. The RNA with the cubic structure mitochondria had accumulated less damage than the RNA in the other two cases. Thus we can assume there to be another reason for mitochondria to take the cubic membrane structure (Deng et al., 2017).

In this contribution we present two different illustrations of geometrically driven phenomena. Firstly, we consider hexagonal patterns engineered by bees and analyze resulting beneficiaries. Next, we present a simple minimal model which explains how symmetry breaking could generate attractive or repulsive interactions among "objects". We demonstrate that infinity of different configurations could emerge although equilibrium configurations of such unperturbed systems are expected to be featureless.

3. Methods

We use a simple Landau-type approach in which phase and structural behaviors are described in terms of order parameter field (Kleman et al., 2003; Sethna, 1992). We consider a 3D system exhibiting a continuous symmetry breaking phase transition on varying a relevant driving parameter (temperature in our case). We assume that a long-range axial orientational order is formed in the symmetry broken phase, which is spatially homogeneous in bulk (i.e., large enough unperturbed system where boundary conditions could be neglected) equilibrium. Hence, we focus on a system which exhibits isotropic symmetry (i.e., it does not possess any kind of order) in the higher temperature phase and below the critical temperature T_c displays a long-range orientational order. The latter is in bulk equilibrium spatially homogeneous along an arbitrary symmetry breaking direction.

For example, such a phase transition could be realized in thermotropic nematic liquid crystals (Kleman et al., 2003) (NLCs) which we use as an illustrating "toy model". Namely, the theory of NLCs is relatively well developed (Kleman et al., 2003) and phenomena





analyzed in this contribution could be experimentally observed. For simplicity we consider LCs formed by anisotropic rod-like molecules, whose local order could be presented by an axial field, whose orientation is commonly referred to as the nematic director field \hat{n} . The states $\pm \hat{n}$ are physically equivalent to mimic the axial symmetry. In thermotropic NLCs the orientational describing local uniaxial order is given by the symmetric and traceless tensor order parameter which can be expressed in terms of \hat{n} as

$$\underline{Q} = S\left(\hat{n} \otimes \hat{n} - \frac{l}{3}\right) \tag{1}$$

The additional quantity in Eq.(1) is the scalar nematic order parameter S which describes the degree of orientational order. In the isotropic phase, which exhibits isotropic symmetry and is stable above the critical temperature T_c , the nematic order is absent and S=0. In the uniaxial nematic phase, which is stable below T_c , S>0 and in bulk equilibrium the tensor order parameter is spatially homogeneous.

For general conditions the order parameter field spatial configuration minimizes the free energy *F* which we express as

$$F = \iiint (f_c + f_e)dv + \sum_i \iint f_i^{(j)} da.$$
 (2)

The first integral in Eq.(2) is carried out over the LC body, where dv stands for the volume element, f_c is the condensation and f_e elastic free energy density. The second integral sums contributions at interfaces of a confining surface (or surfaces) and other interfaces, for example, LC-immersed particle interfaces. The quantity $f_i^{(f)}$ describes the local interaction free energy contribution at j-th interface and da is the interface surface element. In our analysis we will assume that either super-micrometre (colloids) or nano-sized objects (nanoparticles) are immersed in LC liquid. Such additional symmetry breaking sources could enormously increase the pallet of different stable NLC configurations. We use the simplest minimal model which is needed for our analysis in which we express the free energy densities entering Eq.(2) as

$$f_c = \frac{3}{2}a_0(T - T^*)\operatorname{Tr}\underline{Q}^2 - \frac{9}{2}b \operatorname{Tr}\underline{Q}^3 + \frac{9}{4}c\left(\operatorname{Tr}\underline{Q}^2\right)^2 = a_0(T - T^*)S^2 - b S^3 + cS^4, \tag{3a}$$

$$f_e = \frac{L}{2} \left| \nabla \underline{Q} \right|^2 \sim \frac{L}{2} |\nabla S|^2 + \frac{\kappa}{2} |\nabla \widehat{\mathbf{n}}|^2, \tag{3b}$$

$$f_i^{(j)} = -\frac{3}{2} w \hat{\mathbf{e}}. Q \hat{\mathbf{e}},$$
 (3c)

The condensation term determines the equilibrium degree of nematic order $S = S_{eq}$ in bulk equilibrium, where a_0 , T^* , b and c are positive material dependent constants (Kleman et al., 2003). The minimization of Eq. (3a) yields

$$S_{eq}/S_0 = \frac{3 + \sqrt{9 - 8\frac{T - T_c}{T_c - T^*}}}{4} \tag{4}$$

for
$$T \le T_c$$
 where $S_0 = \frac{b}{2c} = S_{eq}(T_c)$ and $T_c = T^* + \frac{b^2}{4a_0c}$.

The elastic term penalizes spatially non-homogeneities in ∇Q , where L>0 and $K\sim LS^2$ stand for the representative elastic constant resisting spatial variations in S and \hat{n} , respectively.

The interfacial term enforces the alignment along the so-called local easy direction \hat{e} if the anchoring constant w is positive.

4. Results

Below we present our analysis. We first show key factors why bees engineer hexagonal patterns. Next we show using our simple modelling how diverse patters could be obtained in nature via symmetry breaking mechanisms.





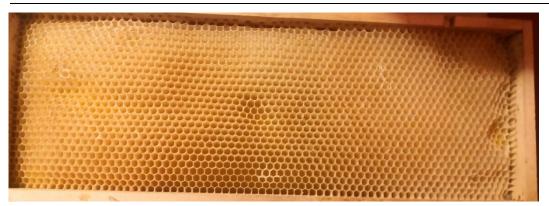


Figure 4: An example of honeycomb. There are no shapes other than hexagons, as there is a matrice below for bees to use.

4.1. Hexagonal honeycombs

We analysed a honeycomb shown in **Fig.4** of area 37.6 cm times 12.9 cm. We measured that the base of one hexagonal cell is 3.25 ± 0.06 mm. We found out that the combined length of all the hexagons in the honeycomb is around 17.2 m, which is way more than one would anticipate. The area of the cell is $A = \frac{3\sqrt{3}a^2}{2}$, where a is the base. This gives $A=27\pm1$ mm². From that we can conclude that the side length of a square with the same area is around 5.2 mm. If bees used squares instead of hexagons, then the total length of the perimeter would be 18.2 m. So we can see that the Honeycomb conjecture holds true. If we assume the height of the cells to be 2.8 cm and the width of the walls to be 0.3 mm, than we can calculate that the bees save around 8 cm³ of wax per honeycomb.

4.2. Symmetry breaking generated patterns

In the following we illustrate how the simple Landau-type approach presented reveals that simple mechanisms could generate several patterns and interactions that we observe in nature.

As already mentioned, the free energy given by Eq.(2) favors spatially homogeneous patterns aligned along a single symmetry breaking direction in the absence of interfacial interactions that would break the symmetry. We emphasize that there exists an infinite number of equivalent equilibrium states each pointing along a different symmetry direction. The competition between these states is crucial for the diversity of different complex patterns that could emerge.

We first consider simple possible excitations in a bulk equilibrium nematic structure where we set $f_i^{(j)} = 0$. For simplicity we restrict to 2D and parametrize the director field by an angle θ as

$$\hat{\mathbf{n}} = (\mathsf{Cos}(\theta), \mathsf{Sin}(\theta)) \tag{5}$$

and assume a constant value of S equal to S_{eq} , given by Eq.(4) which minimizes f_c . Minimization of the free energy yields the Euler-Lagrange equation

$$\nabla^2 \theta = 0 \tag{6}$$

and the possible solutions read [17]

$$\theta = m \operatorname{ArcTan}\left(\frac{y}{x}\right) + \theta_0 = m \operatorname{ArcTan}(\varphi) + \theta_0.$$
 (7)

Here $\varphi = \operatorname{ArcTan}\left(\frac{y}{x}\right)$ is the azimuthal angle in polar coordinates, m is the so-called winding number and θ_0 is a constant. Note that the condition $\hat{\mathbf{n}}(\varphi = 0) = \hat{\mathbf{n}}(\varphi = 2\pi)$ must be obeyed, which restricts values of m to half integers (due to the axial symmetry the states $\pm \hat{n}$ are equivalent):

$$m \in \{0, \pm \frac{1}{2}, \pm 1, \pm \frac{3}{2}, \dots\}$$
 (8)





Solutions corresponding to m=0 represent the competing equilibrium states $\hat{\mathbf{n}}_{eq}=(\text{Cos}(\theta_0), \text{Sin}(\theta_0))$ aligned along a symmetry breaking direction given by $\theta=\theta_0$. Furthermore, the solutions with $m\neq 0$ correspond to topological defects (TDs) whose centers are localized at (x,y)=(0,0). Some examples are shown in **Figure 5**. In 2D the winding number m is equivalent to the so-called topological charge, which is a conserved quantity, similarly as the electric charge in electrostatics. Namely, if the boundary conditions along any closed path are fixed then the total value of m within the enclosed region is conserved. Furthermore, defects bearing m>0 and -m are referred to as defects and antidefects. Such a pair experiences mutual attraction and tends to annihilate into a defectless state. **Figure 6** illustrates cases including more TDs. Because Eq.(6) is linear, linear combinations of solutions are also solutions. A pattern emerging from N defects, where an i-th defect possessing a charge x_i is centered at (x_i, y_i) , could be described by

$$\theta = \sum_{i=1}^{N} m_i \operatorname{ArcTan}\left(\frac{y - y_i}{x - x_i}\right) + \theta_0. \tag{9}$$

For example, the top two patterns in **Figure 5** present defects bearing m=1/2 (left) and m=-1/2 (right). These two defects act as a pair {defect, antidefect} and tend to mutually annihilate into a defectless state in which the director pattern is spatially homogeneous. The first left panel of **Figure 6** illustrates such a nearby pair. Note that the total charge of the system equals to zero and such a state is topologically equivalent to a spatially homogeneous pattern (i.e., ground state) which has the lowest free energy. Therefore, if one starts with a pattern of N defects, where the total topological charge of the system equals to zero, then such a pattern would gradually transform into a defectless ground state via annihilation of defects and antidefects. This behavior is reminiscent to the system of particles and antiparticles bearing positive (e>0) and negative (e<0) electric charges, where m mimics the role of e. Similar as in electrostatics, TDs bearing opposite (same) sign mutually attract (repeal) and the total charge is in both cases conserved.

How a complex nonuniform pattern could be stabilized? Let us assume that colloids or nanoparticles are present in the system, to which we henceforth refer as particles. For simplicity we assume that particles are spherical and their interaction with the enclosing LC is described by Eq.(3c). If the coupling constant is relatively weak, such particles do not strongly affect the surrounding LC as it is schematically depicted in Figure 7a. Let us assume that easy axes of particles are radially oriented, so that particles would act similarly as m=1 defects (see the TD pattern in the left middle panel of Figure 5) on larger length scales in the strong interaction limit w>>0. In such case the particle will create an antidefect bearing *m*=-1 in order to reduce the total deformation in the system. This is illustrated in Figure 7b, where the far nematic director field (with respect to the particle position) tends to be spatially homogenous along the vertical direction. Note that in three dimensions the accompanying antidefect (described by *m*=-1 in 2D) could form a topologically equivalent defect line which is shown in Figure 7c. However, despite topologically equivalent structures shown in Figure 7b and Figure 7c, the symmetry and resulting interactions strongly differ. Namely, in these figures the effective nematic pattern acts as a topological dipole (Figure 7b) and topological quadrupole (Figure 7c). Therefore, in the presence of several particles different deformed nematic patterns could emerge depending on particles' geometries (size, shape) and particle-LC interaction character.





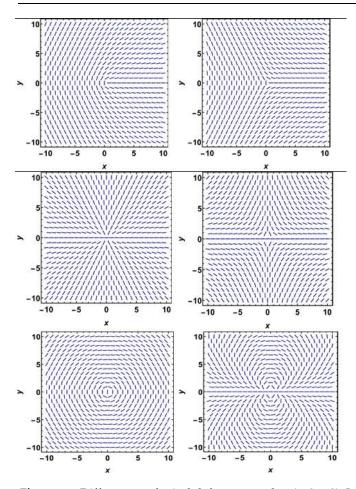


Figure 5: Different topological defects centred at (x=0,y=0). In most case θ_0 = 0 if not stated otherwise. Top panel: (left) m=1/2; (right) m=-1/2. Middle panel: (left) m=1; (right) m=-1. Bottom panel: (left) m=1, θ_0 = π /2; (right) m=2.

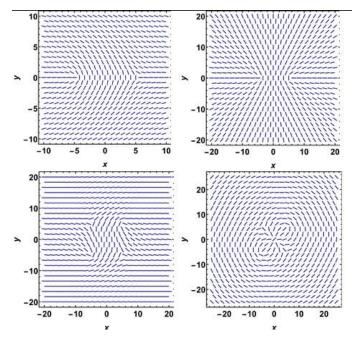


Figure 6: Assemblies of TDs. Their total charge is given by m_{tot} . Top panel: (left) a pair $\{m=1/2, m=-1/2\}$, $m_{tot}=0$; (right) two m=1/2 TDs, $m_{tot}=1$. Bottom panel: (left) two anti-parallel pairs $\{m=-1/2, m=1/2\}$ of TDs, $m_{tot}=0$; (right) four |m|=1/2 and one m=1 TDs, $m_{tot}=1$.





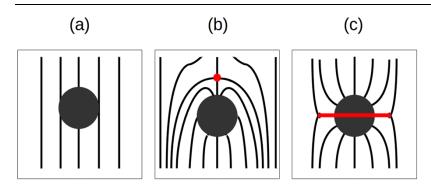


Figure 7: A spherical particle immersed in a LC body. (a); *w*=0, spatially homogeneous nematic structure. In (b) and (c) we have strong interfacial coupling and consequently an antidefect needs to be introduced in LC patterns. Their cores, where nematic order is essentially melted, are red coloured; (b): point-like antidefect and dipolar LC symmetry; (c): line-like antidefect and quadrupolar LC symmetry.

Let us estimate the condition in 3D for which a configuration in **Figure 7c** is more favorable than an undistorted pattern like the one shown in **Figure 7a**. For this purpose, we roughly compare LC free energy penalties of the competing configurations. In the undistorted pattern there are no elastic penalties. However, the surface penalty is relatively high, because LC molecules are not aligned along directions favored by $f_i^{(j)}$. The total free energy penalty of the system is then roughly given by

$$F^{(non)} \sim W A$$
 , (10)

where $A=4\pi R^2$ is the surface of the particle of radius R and the superscript (non) refers to the undistorted LC pattern. On the other hand, in **Figure 7c** the interfacial conditions are perfectly obeyed and consequently a line antidefect of radius r must be formed in order to immerse the distorted LC region in an uniform nematic alignment. We assume that the key "volume" LC penalty comes from the melting of LC order at the defect line. Namely, at the center of defects the orientational order is not uniquely defined and must be locally melted. The radius of the melted region is roughly given by the so-called nematic order correlation length $\xi \sim \sqrt{L/(a_0(T_{IN}-T))}$ (Kleman et al., 2003). Therefore, the dominant free energy contribution of the distorted configuration reads

$$F^{(dis)} \sim a_0 (T_{IN} - T) S_0^2 (2\pi r) (\pi \xi^2) \sim L S_0^2 4\pi r. \tag{11}$$

In this estimate we took into account Eq.(4) and expressed the resulting free energy close to the phase transition temperature and (dis) labels the distorted pattern. Assuming $r \sim R$ we obtain from the "compromise" criteria $F^{(dis)} \sim F^{(non)}$ the condition

$$\mu = \frac{Rw}{K} \sim 1 \tag{12}$$

where $K = LS_0^2$. Therefore, in the regime $\mu < 1$ and $\mu > 1$ the undistorted and distorted patterns are realized, respectively.

5. Discussion

Our study illustrates that symmetry breaking mechanisms could very efficiently generate various complex patterns particularly in system where a continuous symmetry is broken. In such cases systems are very susceptible to various perturbations, because they have in general a rich palette of different ways to respond. Furthermore, continuous symmetry breaking enables formation of topological defects (Kibble, 1976; Sethna, 1992; Zurek, 1985). Such excitations in many cases behave like flexible bodies and one can assign to them topological charges which are conserved quantities. In interactions and recombination of such entities could be predicted based on group theory (Kibble, 1976; Sethna, 1992; Zurek, 1985) and systems exhibiting similar symmetries share several similarities in mathematical behavior although they could be physically completely different. Axial symmetry enables in





addition to point defects also topologically stable line defects (Bradač et al., 2011; Kleman et al., 2003). For example, in magnetic systems, where a vector order parameter could serve as the order parameter, line defects could not be topologically stable. Therefore, systems like nematic liquid crystals could for this purpose exhibit even larger complexity than for example magnetic systems, in which several pioneering solid state studies have been made (Imry and Ma, 1975; Sethna, 1992).

We demonstrated that different patterns in nematic liquid crystals can be efficiently formed by dispersing various particles within them. We illustrated that both geometry, in particular topology, and interfacial interactions play an important role. However, the history of samples could be another rich generator of additional patterns which we did not address. For example, if the isotropic-nematic phase transition is fast enough then a dense tangle of topological defects is formed just after the transition. Namely, if the phase transformation is too fast for different regions to communicate, in distant parts the symmetry is in general broken in different directions. In such cases the concentration of TDs strongly depend on the quench rate (Kibble, 1976; Zurek, 1985; Bradač et al., 2011) (i.e., the time in which the transition is realized). If particles are present in LC medium, then additional different and complex configurations could emerge whose average behavior could be tuned by diverse tunning parameters (Bradač et al., 2011; Pišljar et al., 2024; Ranjkesh et al., 2014) (e.g., quench rate, topology of particles, geometrical properties of particles, LC confining geometry...). This will be the focus of our future planned research.

6. Conclusions

We analysed different patterns that are commonly realised in nature. We reviewed configurations displaying hexagonal symmetry and exploiting triply periodic minimal surfaces in different physical systems. Furthermore, key advantages of such patterns are reported. We studied in more detail honeycomb hexagonal structure and show the key advantages of such configurations. In addition, we presented a simple Landau-type model to show how a relatively simple symmetry breaking mechanism could generate a rich diversity of different patterns. In our illustration we used nematic liquid crystal pastern as an experimentally approachable system where such phenomena could be directly observed using relatively simple optic methods (e.g., polarising microscopy).

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Conflicts of Interest: The authors declare no conflict of interest.

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Research

A simple Statistical Mechanical Description of the Atmosphere Composed of Small Particles around Massive Spherical Body

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Abstract:

The atmosphere of small particles around a spherical massive body is theoretically described starting from statistical mechanical principles combined with the field approach. It is assumed that the system is in thermodynamic equilibrium and that the small particles are explicitly independent and therefore subjected to the Boltzmann statistics. The model system is the atmosphere of small particles that are attracted to the spherical massive body while their complete approach to the surface of this body is hindered by thermal motion. The gravitational attraction between the molecules and the massive object is described by introducing a potential of the attractive field and by considering that the massive object is the source of the field. The above assumptions lead to the formulation of the variational problem based on the 2.nd law of thermodynamics in the form of the consistently-related system of differential equations for the gravitational potential and the distribution of the molecules within the atmosphere.

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Keywords: Gravitation; Atmosphere; Gravitational potential



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1 Introduction

The objective of modeling atmosphere in deeper space is the study of extraterrestrial bioenvironment including molecules, micro and nano-organisms, and extracellular particles and vesicles. Formally similar models were constructed to model ionic atmosphere around charged bodies (see for example Hill, 1986). In this work we derive the expressions for the gravitational potential, density of the number of small particles and gravitational field in a system composed of a central spherical massive body surrounded by very many small particles which are subjected to thermal motion.

2 Theory

2.1 Field of the mass m

We take that the source of the field G is mass m,

$$\nabla \cdot \mathbf{G} = \varrho, \tag{1}$$

where

$$\rho = \frac{\mathrm{d}m}{\mathrm{d}V},\tag{2}$$

and V is the volume. For convenience of scaling, we introduce a constant γ so that

$$\mathbf{G} = \Gamma \gamma \tag{3}$$

and

$$\nabla \cdot \mathbf{\Gamma} = \frac{\varrho}{\gamma},\tag{4}$$

We assume that The field Γ has no vortexes,

$$\nabla \times \mathbf{\Gamma} = 0, \tag{5}$$

and introduce gravitational potential φ_{Γ} ,

$$\Gamma = \nabla \varphi_{\Gamma}. \tag{6}$$

Gradient in spherical coordinates reads

$$\nabla = \left(\frac{\partial}{\partial r}, \frac{1}{r} \frac{\partial}{\partial \varphi}, \frac{1}{r \sin \varphi} \frac{\partial}{\partial \varphi}\right). \tag{7}$$

Combining Eqs.(4) and (6) yields the Poisson equation

$$\nabla^2 \varphi_{\Gamma} = \frac{\rho}{\gamma}.\tag{8}$$

2.2 Poisson-Boltzmann equation

Assuming the thermodynamic equilibrium in a system with constant temperature and volume, and the validity of the Boltzmann statistics, the equality of gravitochemical potential at two different points (one at a chosen distance r from the massive body's center and the other at the surface of the massive body) reads

$$-m_1\varphi_{\Gamma} + kT \ln(\frac{\rho}{\rho_0}) = -m_1\varphi_{\Gamma}(r_0), \tag{9}$$





where ρ_0 is the density of small particles at the surface of the massive body, k is the Boltzmann constant and T is the temperature. After some rearranging of Eq.(9) we get the Boltzmann distribution

 $\rho = \rho_0 \exp\left(\frac{m_1}{kT}(\varphi_{\Gamma} - \varphi_{\Gamma}(r_0))\right). \tag{10}$

The expression for ρ (Eq.(10)) is inserted into Eq.(8) to obtain the Poisson-Boltzmann differential equation for φ_{Γ}

 $\nabla^2 \varphi_{\Gamma} = \frac{\rho_0}{\gamma} \exp\left(\frac{m_1}{kT} (\varphi_{\Gamma} - \varphi_{\Gamma}(r_0))\right). \tag{11}$

As for the spherical geometry, we use the spherical coordinate system so that Eq.(11) transforms into

$$\frac{1}{r}\frac{\mathrm{d}^2(r\varphi_{\Gamma})}{\mathrm{d}r^2} = \frac{\rho_0}{\gamma} \exp\left(\frac{m_1}{kT}(\varphi_{\Gamma} - \varphi_{\Gamma}(r_0))\right)$$
(12)

or

$$\frac{1}{r} \frac{\mathrm{d}^2(r(\varphi_{\Gamma} - \varphi_{\Gamma}(r_0)))}{\mathrm{d}r^2} = \frac{\rho_0}{\gamma} \exp\left(\frac{m_1}{kT}(\varphi_{\Gamma} - \varphi_{\Gamma}(r_0))\right)$$
(13)

2.3 Consistently related solution of the Poisson-Boltzmann equation

Solving the Poisson-Boltzmann equation (Eq.(13)) yields the gravitational potential φ_{Γ} in dependence on r, which is then used to calculate the distribution of small particles ρ and the gravitational field Γ . For convenience, we introduce dimensionless quantities

$$y = \frac{m_1(\varphi_\Gamma - \varphi_\Gamma(r_0))}{kT},\tag{14}$$

$$x = \frac{r}{r_0},\tag{15}$$

and

$$\rho = \rho(r_0) \exp(y). \tag{16}$$

The Poisson-Boltzmann equation (11) transforms into

$$\frac{1}{x}\frac{\mathrm{d}^2(xy)}{\mathrm{d}x^2} = \kappa^2 \exp(y),\tag{17}$$

where

$$\kappa = \sqrt{\frac{\rho_0 r_0^2 m_1}{k T \gamma}}. (18)$$

This is a nonlinear differential equation of the second order and to our best knowledge, it does not have an analytic solution. We explore the limiting case where y is small and the exponential function can be expanded,

$$\exp y = 1 + y,\tag{19}$$

so that

$$\frac{1}{x}\frac{d^2(xy)}{dx^2} = \kappa^2(1+y). \tag{20}$$

Eq.(20) can be solved analytically,

$$y = C_1 \frac{\exp(-\kappa x)}{x} + C_2 \frac{\exp(\kappa x)}{x} - 1, \tag{21}$$





where C_1 and C_2 are constants. As the density of small particles in not expected to rise to infinity far away from the massive sphere, C_2 is set to 0,

$$C_2 = 0, (22)$$

so that

$$y = C_1 \frac{\exp(-\kappa x)}{x} - 1. \tag{23}$$

At the surface of the massive body, x = 1 and we chose the value of the potential to be 0. Therefore,

$$C_1 = \exp(\kappa), \tag{24}$$

so that

$$y = \frac{\exp(-\kappa(x-1))}{x} - 1. \tag{25}$$

It follows from Eq.(25) and Eq.(16) that

$$\rho = \rho_0 \exp(\frac{\exp(-\kappa(x-1))}{x} - 1). \tag{26}$$

The dimensionless gravitational field is obtained by

$$\frac{\mathrm{d}y}{\mathrm{d}x} = \frac{(\kappa x - 1)}{x^2} \exp(-\kappa (x - 1)). \tag{27}$$

.

2.4 Estimation of parameters

In the integral form, Eq.(1) reads

$$\oint \mathbf{G} \cdot d\mathbf{S} = m, \tag{28}$$

where **S** is the area. In the case of spherical massive body with mass m, the surface around the mass is taken to be a spherical shell. In this case, only the radial component of the vector $\Gamma = (\Gamma_r, \Gamma_\theta, \Gamma_\phi)$ will give a nonzero contribution

$$\Gamma_r \gamma 4\pi r^2 = m,\tag{29}$$

so that

$$\Gamma_r = \frac{m}{4\pi\gamma r^2},\tag{30}$$

where

$$\frac{1}{4\pi\gamma} = G \tag{31}$$

is the gravitation constant $G = 6.67 \times 10^{-11} \text{ m}^3/(\text{kg s}^2)$. Then,

$$\gamma = \frac{1}{4\pi G} = 1.2 \times 10^9 \frac{\text{kg}^2}{\text{Nm}^2}.$$
 (32)





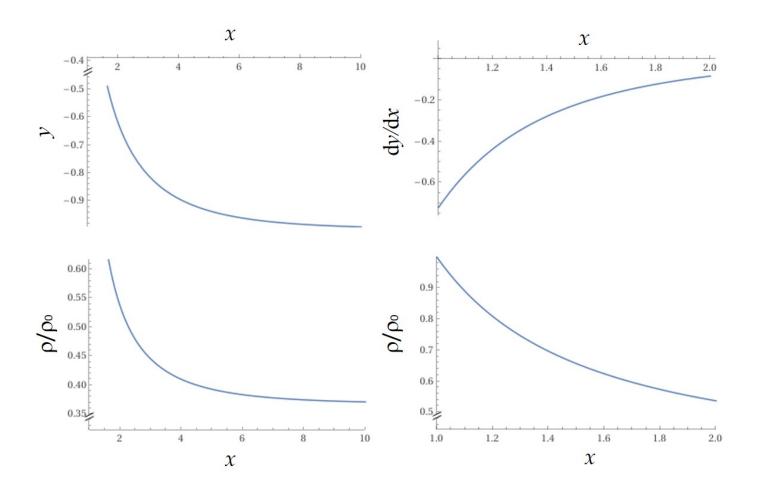


Figure 1: Calculated dimensionless gravitational potential y, dimensionless density of small particles ρ/ρ_0 and dimensionless gravitational field in the radial direction $\mathrm{d}y/\mathrm{d}x$ in dependence on the distance from the center of the spherical massive body x for $\kappa=0.28$. The gravitational potential is the solution of the variational problem describing thermodynamic equilibrium of the atmosphere of small particles around the massive spherical body. The atmosphere develops from the surface of the massive body (x=1) and expands to infinity.

3 Results

As an example we consider that the massive body has a mass of 6×10^{24} kg and radius is 6×10^{6} m. We take that the density of particles at the surface of the massive body is $\rho_0 = 1 \text{kg/m}^3$ and that the mass of the small particles m_1 is 10^{-26} kg. We take that the temperature is 280 K. Considering the above,

$$\kappa = \sqrt{\frac{\rho_0 r_0^2 m_1}{k T \gamma}} = 0.28. \tag{33}$$

Figure 1 shows calculated dimensionless gravitational potential y, dimensionless density of small particles ρ/ρ_0 and dimensionless gravitational field in the radial direction dy/dx in dependence on the distance from the center of the spherical massive body x for $\kappa = 0.28$.







4 Conclusions

The field diminishes to 0 at large distances while the potential approaches a constant value as the value at the surface of the massive body was set according to the density of small particles at the surface. Consequently, in this model the density of the small particles diminishes, but does not vanish at large distances.

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6 Conflicts of Interest

The author discloses no conflict of interest.

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Review

How to Hide: Camouflage from Ultraviolet to Infrared

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Abstract:

Effective concealment in outdoor environments requires an understanding of basic camouflage principles and the specifics of light spectrum in which detection may occur. Light is mainly reflected and absorbed (from ultraviolet (UV) to near-infrared (NIR)) or emitted (from mid-infrared (MIR) onwards) for a subject with a typical surface temperature of 37°C. In the visible spectrum, concealment is achieved by matching the background reflectance (using appropriate colours) or using disruptive patterns to break up recognizable shapes. These patterns can be applied manually or generated using specialized algorithms that allow for rapid adaptation to different environments. Camouflage in the UV range requires highly absorbing coatings that remain transparent in the visible spectrum, while in the NIR, similar effects can be achieved using absorbing metallic flakes in a polymer binder or carbon black nanoparticles. Beyond the NIR, emittance becomes the dominant factor, and concealment requires controlling an object's thermal signature. This can be done by lowering its temperature, improving thermal insulation, or utilizing selective heat emission through radiative cooling in the atmospheric window (5-8 µm). By carefully considering these principles, effective multispectral camouflage can be achieved.

Keywords: Camouflage; Light spectrum; Pattern generation; Diffuse reflectance; Emittance; Thermal signature





1. Introduction

Camouflage, as defined by the Merriam-Webster dictionary, is concealment by means of disguise. It signifies one's goal to remain hidden from unwanted eyes. Motivations for such behaviour are diverse. Prey species increase their chances of survival by minimizing their visibility within their environment or by hindering attempts to distinguish individual members within a group. Similarly, predators gain an advantage by getting closer to their prey without being recognized as a threat. Surveillance for scientific purposes requires the observed subjects not changing their behaviour upon the realization that they are being monitored. All these instances can benefit considerably by using effective camouflage that follows these basic principles:

- Background matching: blending with the environment is achieved by matching the surrounding colours (**Figure 1a**). This technique is effective only for specific environments and seasons. Some animals circumvent this problem by having a whiter winter coat that replaces the grey and brown colouring of the warmer seasons.
- Disruption: elements with strong contrast break up the outlines of an object and hide its actual shape, therefore hindering recognition (**Figure 1b**). An example is a zebra hiding among other members of its herd or a snake hiding among ground fallen leaves. Additional enhancement is possible by making the edges of darker areas even darker and the same for lighter areas, thus achieving a 3D effect (Sharman et al., 2018).
- Multi-scale patterns: fractal features on micro-, midi- and macro-scale allow effective concealment at various distances while preventing isoluminance. This undesirable effect occurs when patches of similar colour blend together as they are viewed from a distance, making the object appear uniform in colour and therefore stand out considerably from the environment.

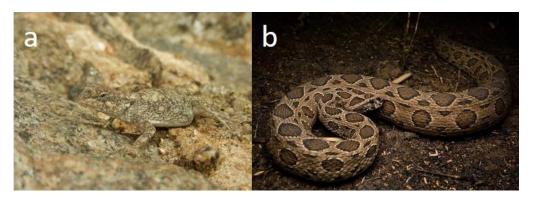


Figure 1. a) Lizard camouflaged on a stone (Photograph by Praveen Illa / CC BY-SA 4.0). b) Disruptive colouring of a Siamese Russell's viper (Photograph by Tontan Travel (www.tontantravel.com) / CC BY-SA 2.0).

2. Light detection

To develop an effective concealment, one must understand some of the basic properties of light. It can be described as an electromagnetic wave with specific energy and therefore a specific wavelength (it can be described as a particle as well, however, the wave-particle duality will not be important for this work). Wavelengths from 380 to 700 nm correspond roughly to what we call the visible spectrum (VIS). Lower wavelengths correspond to ultraviolet (UV) and higher to infrared (IR) light. Here, the focus will mostly be on the UV-A part from 315 to 380 nm, in the near- (NIR, 700–1500 nm), mid- (MIR, 1500–5600 nm) and far-infrared (FIR, >5600 nm). Light can be absorbed or scattered in the material as well as reflected (Figure 2a). Every object also emits light (Figure 2b) and its spectrum depends on the surface temperature (e.g., a body with surface temperature of 37°C emits mostly in the IR, with a peak at around 10 μ m).





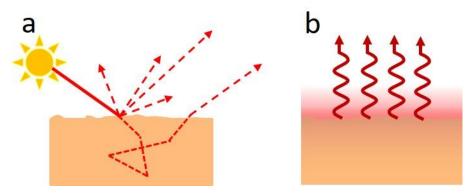


Figure 2. Types of light interaction with a material. a) Specular and diffuse reflection of light (UV to near-IR). Specularly reflected light is reflected off the surface while diffusely reflected light enters the material, where it is absorbed and scattered before it exits again. b) Emission of light due to black body radiation (mid- to far-IR).

For the light to be detected, it has to reach a suitable sensor (e.g., eye, camera) and interact with it. Humans are equipped with photoreceptors capable of detecting visible light (cones and rods on the eye's retina), and the range can be extended further. Individuals with the eye lens removed report being able to see further into the UV (Griswold & Stark, 1992), since the lens absorbs a significant amount of UV light to protect the retina. Extending the range into the NIR requires the use of photoreceptor-binding upconverting nanoparticles (Dhankhar et al., 2020; Ma et al., 2019) or the exchange of vitamin A1 for A2 in the subject's diet (Science For The Masses, 2013), with a side effect of diminished sensitivity to blue light. It is easier to achieve a wider range of detection with man-made sensors. CCD (charged-coupled device) and CMOS (complementary metal–oxide–semiconductor) sensors are used for light wavelengths spanning from 300 to 1000 nm, InGaAs sensors are used from 1000 to 2500 μ m and thermal cameras are used for higher wavelengths.

The range of detectable light in the animal kingdom also outperforms that of a human. Animals such as reindeer (Hogg et al., 2011), blue tits (Rajchard, 2009) or butterflies (Finkbeiner & Briscoe, 2021) can detect UV light. The mantis shrimp has a staggering 12 different types of cones, being able to detect light from the UV to the far red (Thoen et al., 2014). Detection of heat signatures in the MIR and FIR is possible with specialized organs (e.g., pit vipers (Goris, 2011), vampire bats (Kürten & Schmidt, 1982) or mosquitos (Zermoglio et al., 2017)).

3. Camouflage

The range of detection discussed in the previous section shows that it is not sufficient to hide in the visible, but it is also necessary to consider the UV and IR spectral ranges. One way to successfully blend with the surroundings in the visible spectrum, is to use a combination of pigments that match the desired reflectance (Asofiei et al., 2021; Goudarzi et al., 2012). A spectral VIS–NIR analysis of the colours of two camouflage patterns is shown in Figure 3, with a comparison to the spectrum of the environment with fresh and dry green leaves. Most of the darker measured camouflage colours correspond well to the environment in the VIS spectrum. A considerable difference in spectral behaviour can be ascertained in the NIR region, where the MultiCam camouflage shows little contrast between patches of different colour. This better corresponds to the spectra of green leaves than the Multi-Terrain Pattern and could lead to better concealment in such environments.

Patterns used for camouflage can consist of flat coloured geometric shapes of varying sizes, pixelated patches, or gradients. For greater ease in pattern adaptation for other environments, a generating algorithm can be programmed. There are several existing open source algorithms; CamoEvo is an artificial camouflage evolution experiment using genetic algorithms to determine patterns that increase concealment in a specified environment with each additional generation (Hancock & Troscianko, 2022), CamoGen is a digital camouflage generator (Lederrey, 2023), while CONCEAL creates a digital/fractal camouflage

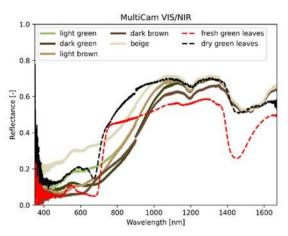




based upon provided images of the environment and supplied required colours (Peters, 2023).







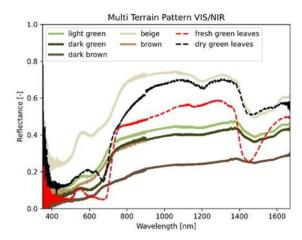


Figure 3. Diffuse reflectance of two camouflage patterns, a) MultiCam and b) Multi-Terrain Pattern. Circles represent sections of different colour used for the measurements. The dashed lines represent measurements on fresh and dry green leaves. The spectra were measured for the project MULKAM – Development of multispectral camouflage for vehicles, equipment and soldiers for the Ministry of Defence of the Republic of Slovenia.

Achieving acceptable camouflage in the UV region depends on the environment reflectance. Snow exhibits large reflectances (up to 80 %), which can be simulated with a coating of white pigment dispersions (e.g., calcium carbonate, barium sulphate (Wilhelm, 1967)). Other environments, e.g., forest or desert, require a decrease in reflectance, which can be achieved with the use of a UV absorbing agent. Available products are U-V-Killer spray (Atsko, USA), UVRCTM (UVR Defense Tech Ltd., USA) or chemical sunscreen based on organic molecules that absorb UV light, to hide exposed skin.

To change absorption in the NIR region, pigments such as perylene black, phthalocyanine blue, isoindoline derivatives, carbon black (soot), IRT black (Standard Colors, NC, USA) are used on the fabric. For coatings with low emissivity, metal pigments with an organic binder are often used, usually in the form of flakes (e.g. aluminum pigments in flakes with a water-based polymer binder (Hallberg et al., 2005); with titanium dioxide (TiO₂ in the form of rutile and anatase) (Wong et al., 2015); from polyurethane, aluminum and nanopigments (Liang et al., 2018); polyvinyl butyral with nanoparticles of tungsten disulphide (Samolov et al., 2021).

Living creatures, with a temperature around 37°C, begin emitting light in the MIR and FIR regions, making them stand out in the often colder surroundings. To hide this thermal signature, different approaches can be undertaken. One option is to manipulate the surface emissivity, either with active cooling or thermal insulation (e.g., silica aerogel) with multilayer films acting as wavelength-selective emitters (high emittance in the non-atmospheric window, where absorption in the atmosphere is significant (5–8 μ m) and low in the atmospheric window (3–5 μ m and 8–14 μ m) (Jiang et al., 2023; Xu et al., 2020; Zhu et al., 2020)) or tri-layer structures with overall low emittance (Woo et al., 2022). One group (Hong et





al., 2020) developed a flexible thermoelectric device capable of responding to the background temperature change via thermoelectric cooling and heating. A plethora of approaches is covered in several review articles (Degenstein et al., 2021; Su et al., 2023). An additional tactic, suitable for the whole range of wavelengths, is to self-decorate by covering oneself with materials from the environment, which is utilized by masked hunter bugs or a ghillie suit. A Chinese group (Xie et al., 2023) also developed chlorophyll microcapsules that could be embedded in clothing and can mimic leaf reflectance better than traditional pigment combinations.

4. Conclusion

Concealment poses a difficult challenge, particularly when one considers the spectral range at which detection is possible. Achieving effective camouflage in the range across UV to IR necessitates adherence to basic principles of camouflage (matching the environment reflectance at multiple distances with the incorporation of disruptive patterns), as well as careful consideration of the specifics of background reflectance at these wavelength ranges. Various strategies exist for attaining more or less effective camouflage, with continuous developments each year indicating this specific field remains open to further research and improvements.

Conflicts of Interest: The author declares no conflict of interest.

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Review

Society & Science: Doomsday Criticality for the Global Society

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Abstract:

The paper discusses global population changes from the beginning of the Anthropocene onset till 2024, based on the 'tools' and model concepts of Soft Matter dynamics. The 'bottom-up' approach, focused on the population data themselves and the distortion-sensitive validation of portrayal via a given scaling relation, is applied. Two exponential Super-Malthusian dependences are considered, namely (i) the empowered exponential one, and (ii) containing the time-dependent relaxation time. The latter follows the dominant linear pattern in the Industrial Revolution times, leading to 'constrained and frustrated' – type critical scaling for the global population growth. Finally, the evolution of the relative growth rate versus the population itself is considered. The link to the empowered exponential behavior is indicated.

This contribution presents a new cognitive path for studying global population growth, exploring the Soft Matter dynamics approach. It offers a reliable fundamental base of derived scaling equations, including the meaning of relevant parameters. Studies revealed the non-monotonic nature of global population growth, where temporal local events can significantly influence leading trends. For Industrial Revolutions, the essential meaning of technological innovation in feedback alliance with socio-economic innovatively re-shaping surrounding is noted.

Keywords: Global Population; Scaling Equations, Modelling, Soft Matter, Dynamics, Universalistic Features.





1. Soft Matter and Global Population as the Socio-Economic Counterpart

In 1991, Pierre G. de Gennes constituted the Soft Matter category in the Nobel Prize lecture (1991) entitled 'Soft Matter' (de Gennes, 1991; de Gennes and Badoz, 1996). There is no general definition of Soft Matter, but it is related to microscopically distinct systems showing common, universalistic features, namely: (i) dominance of mesoscale assemblies, (ii) extreme sensitivity to perturbations, (iii) spontaneous mesoscale self-assembling and self-organization, (iv) unique phase transitions, (v) common scaling functional patterns despite essential microscopic differences.

The canonic Soft Matter includes: liquid crystals, colloids, micellar systems, critical liquids, polymers, supercooled liquids, nanocolloids, vesicles based fluids, bio-systems, micellar systems ... and also some semi-solids such as plastic crystals, ... (de Gennes and Badoz, 1996; Brochard-Wyart, 2019; Roland et al., 2008; Drozd-Rzoska, et al., 2008; Rzoska, et al., 2001; Drozd-Rzoska, et al., 2013).

De Gennes' pointed out a large set of systems, earlier considered mostly in frames of material engineering, that can linked in this category due to universalistic features for 'isomorphic' physical properties. In subsequent decades, the concept of Soft Matter expanded. Currently, foods are considered the Complex Soft Matter (Mezzenga, et al., 2005). Living Soft Matter is focused on biosystems from DNA to bacteria and viruses, ..., including their assemblies (Sinha, 2024). Quantum Soft Matter has become a special category (Thedford, et al., 2022). Focused liquid crystals and their nanocolloids studies revealed fascinating parallels to the elementary particles world: bosons, fermions, or Higgs fields (Jelen, et al., 2024). The concept of 'Soft cosmology' has emerged to interpret some exceptional properties in Universe sectors (Saridakis, 2021).

Soft Matter concept opened up extraordinary possibilities for experimental modeling of the mentioned systems on the Laboratory Table, with the support of materials engineering and monitoring by various physical methods. The Soft Matter cognitive advances yielded analytical tools and universal modeling concepts that could be implemented in various specific systems.

Global Population is composed of humans with an inherent tendency to interact, self-assemble, and spontaneously create ordered local structures - from families, tribes, and cities to countries/states & empires. It is related to the rising range of 'interactions' related to developing management. Notable is the extreme sensitivity to endogenic and exogenic perturbations, often leading to qualitative transformations of societies (,phase' transitions?). Hence, the question arises (Sojecka and Drozd-Rzoska, 2024, 2025): Isn't the Global Population a unique Socio-Economic Soft Matter system?

2. Global population scaling: selected reference models

An adequate and fundamentally justified description of global population changes, using model-validated scaling relations, is essential for the insight into past and future-focused reliable predictions. It can deliver cardinal data supporting effective socio-economic planning and global governance.

The 'classic' reference for Population Growth is the Malthus equation (Malthus, 1798; Weil and Wilde, 2010):

$$P(t) = P_0 exp(rt) \Rightarrow lnP(t) = lnP_0 + rt \Rightarrow \frac{dP(t)}{dt} = rP(t)$$
 (1)

where time t refers to the onset time t_0 , coupled with the prefactor N_0 , and r = const is the Malthus rate coefficient.





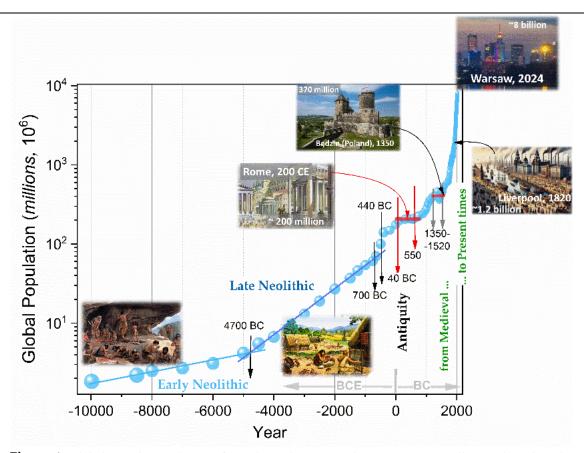


Figure 1. Global population changes from the Holocene (Anthropocene) onset till nowadays, based on data introduced in (Sojecka and Rzoska, 2024). Linear dependence is related to the basic Malthus behavior and red-horizontal lines to ~constant population in the given time domain. Subsequent historical epochs are indicated by characterizing them with pictures, which include messages regarding global population values.

The right part of Eq. (1), shows the basic assumption, namely that the population rise in the subsequent time domain is proportional to the current population, following the constant rate coefficient r. It directly leads to the exponential equation, shown in the left part of Eq. (1). For the Malthus model, one should expect linear population changes when using the semi-log plot, as shown in the middle part of Eq. (1).

Figure 1 presents global population changes from the Anthropocene onset (10 000 BCE) till nowadays, using the new data set introduced in (Sojecka and Drozd-Rzoska, 2024 and 2025). It is based on collecting global population data from a few sources and their subsequent numerical filtering to obtain an analytic set for which data-related derivative analysis is possible. The global population followed the linear behavior, in the semi-log scale, described by Malthus' Eq. (1) for ~9 millennia, - related to Neolithic times, splitting into domains: Early & Late'. The crossover can be linked to climate changes. From the end of the Bronze Era, via Antiquity, Medieval, till nowadays - the increasingly nonlinear behavior appears in **Figure 1**. For such pattern, beyond the basic Malthus model (Eq. (1)), the name Super-Malthus (S-M) behavior was proposed (Sojecka and Drozd-Rzoska, 2024 and 2025). The significant feature of the discussed S-M equations was the relaxation time τ introduced instead of the Malthus rate coefficient, namely $\tau = 1/r$. It allows a simple estimation of 50% change from the given P(t) value: $t_{50\%} = \tau \ln 2$.

In the first half of the 19th century, Verhulst proposed the model extending the Malthus approach by including the factor directly addressing required resources, such as food. It is explicitly visible for the model-related differential equation (Verhulst, 1847; Vandamme and Rocha, 2022):

$$\frac{\mathrm{d}P(t)}{\mathrm{d}t} = rP - sP^2 = P(r - sP) = P(t)\left(r - \frac{P(t)}{K}\right) \Rightarrow K \gg P(t) \Rightarrow \frac{\mathrm{d}P(t)}{\mathrm{d}t} = rP \tag{2}$$





where s is the Verhulst parameter describing available resources in the given systems. The carrying capacity coefficient K = 1/s was introduced by Pearl (Pearl, 1927; Volterra, 1928).

Eq. (2) shows that the carrying capacity coefficient K can be interpreted as the maximal population that can be hypothetically maintained in the given system with a given amount of resources. The right-hand part of Eq. (2) shows that for $K \gg P(t)$ it reduces to the basic Maltus Eq. (1).

For $sP \rightarrow r$, increasing distortion from the unlimited Malthus-type rise appears. Finally at sP = r, related to dP(t)/dt = 0, the population rise terminal is reached.

For the Verhulst equation, one can consider the following scenarios occurying after the first Malthusian growth stage (Sojecka and Drozd-Rzoska, 2025):

- (i) For infinite resources, only permanent Malthiusian-type population growth occurs.
- (ii) A constant amount of renewable resources (s(t) = s = const) is available. It remains constant despite the population rise. The condition dP(t)/dt = 0 means the transition to a plateau where P(t) = const.
- (iii) A constant amount of non-renewable resources is available. It irreversibly decreases with the population rise. The rise terminal is related to $dP(t=t_{max})/dt=0$, and for $t>t_{max}$ the population declines.
- (iv) The population is 'specifically sustainable', i.e., it limits demands for 'resources' by restricting needs. Consequently, the amount of resources 'relatively increases', postponing the terminal condition $dP(t=t_{max})/dt=0$.

A model manifestation of the option (iv) can be the evolutionary size reduction of animals on isolated islands. For the human population, it can be associated with lesser demands for food, raw materials, and limited harmful environmental impacts, which correlates with general requiremen0st of the Sustainable Society.

The integration of the differential Eq. (2) leads to the basic Verhulst scaling equation (Verhults, 1847; Sojecka and Drozd-Rzoska, 2025):

$$P(t) = \frac{K}{1 + CK \exp(-rt)} = \frac{1}{1/K + C \exp(-rt)} \implies (K \to \infty) \implies P(t) = P_0 \exp(rt)$$
 (3)

where $C = P_0 - (1/K)$. The right-hand part indicates the simplification to the Malthus Eq. (1). Malthus and Verhulst scaling equations remain a significant reference for population studies. Nevertheless, the 'empirical' pattern of global population growth does not correlate with the abovementioned characterization. However, they are an essential and proven tool in microbiology, general biology, epidemiology and medicine (Vandamme, 2021).

There are currently many model concepts for describing global population changes (Umpleby, 1990). For the authors, worth noting is the 'hyperbolic. Doomsday' equation (von Foerster, et al., 1960):

$$'P(t) = \frac{1.79 \times 10^{11}}{(2026.87 - t)^{\gamma = 0.99}} \propto \frac{1}{D - t}$$
 (4)

where D = 2026.87 was linked to Friday, 13 November 2026, Doomsday.

It was validated via the log-log scale plot analysis for 28 global population data ranging from ~500 CE till 1958. The title of von Foerster et al., (1960) report recalling 'Doomsday', and the suggestion of the 'human extinction' before 2026, led to decades of commentary and remarks in the general mass media, but also criticism from population change researchers, who mainly criticized the lack of model justification (Umpleby, 1990; Yakovenko, 2025). However, an excellent descriptive agreement, with the surprising simplicity of Eq. (4), remained a surprising fact.

From the 1970s, more historical estimates of global population data became available, and depending on the data set tested and the time period selected, the power exponent $\sim 0.7 < \gamma < 1$ was reported for Eq. (4) (Taagapera, 1979).

In the last decade, Taagapera and Nemčok developed a scaling function that avoids the 'Doomsday' singularity and can be reduced to the von Foerster scaling functional form. They suggest 'stationary, terminal phases' as the generic feature of global population development: 1st at Roman Empire times, & 2nd in the last decades. They were scaled as follows (Taagapera, 2014; Taagapera and Nemčok, 2024):





$$P(t) = \frac{A}{[\ln(B+E)]^M}$$
 , $E = \exp[(D-t)/\tau]$ (5)

associated with D = 100 CE for the 1st (Prehistoric-Roman Empire) period and D = 1980 for the second (early Medieval – Nowadays).

Equation (5) offers an excellent reproduction of the global population changes. However, it contains 5 parameters for each mentioned period and requires nonlinear fitting. Notable, that the experience gained in Soft Matter systems for similar patterns of data change led to the general indication that optimal scaling equations should rely on no more than 3-4 parameters.

A significant shortcoming of the analysis trend initiated by the work of (von Foerster et al., 1960) seems to be the lack of fundamental justification for such a unique characterization. Nevertheless, the unique descriptive efficiency seems to remain the fact (Drozd-Rzoska et al., 2023).

3. Localized & Soft Matter view on Global Population Growth

The standard analytic method for testing population changes relies on fitting data via a given scaling equation in an *ad hoc* selected time domain, often using nonlinear routines. This report presents a novel bottom-up approach to Global Population dynamics. It does not focus on fitting via a given scaling equation in an arbitrary time domain but on population data itself. It is related to the following basic issues (Sojecka and Drozd-Rzoska, 2024, 2025):

- 1. For the optimal description of P(t) changes, and reliable forecasts, it is necessary to analyze trends over a sufficiently long period.
- 2. The crucial problem for global population growth data constitutes multiple estimates of population data, particularly when shifting to the past. The authors reduced this problem by implementing numerical filtering to a large data set collected from different sources.
- 3. The latter led to the unique set of 198 global population data from 10 000 BC 2024, with an analytic pattern. i.e., the derivative analysis is possible.
- 4. The linearized & derivative-based analysis, recalling 3-parameters model equations, has been carried out for the new generation data set.
- 5. The latter yielded distortions-sensitive insights into local distortions, revealing time domains where a given scaling equation can be used. It also derived optimal values of relevant parameters consequently the nonlinear fitting routine was avoided.
- 6. The Soft Matter base of model scaling relations offers a fundamental reference meaning of relevant parameters.

In refs. (Sojecka, Drozd-Rzoska; 2024, 2025) two Super-Malthus (S-M) equations have been developed and implemented to discuss Global Population Growth. The first one is the 'empowered' S-M1 equation:

$$P(t) = P_0 \exp\left(\frac{t}{\tau}\right)^{\beta} \quad \Rightarrow \ln P(t) = \ln P_0 + \frac{t^{\beta}}{\tau^{\beta}} \tag{6}$$

where P_0 is the prefactor related t = 0, in the given report related to the mentioned Anthropocene onset; parameters τ , $\beta = const$.

The right-hand part shows that the simple semi-log plot analysis, successful for the basic Malthus Eq. (1), does not yield relevant parameters due to the essential non-linearity, related to the exponent β . Nevertheless, one can consider the derivative of the right-hand part of Eq. (6) and, subsequently implement the log-log scale analysis:

$$\log_{10} \left[\frac{\mathrm{dln}P(t)}{\mathrm{d}t} \right] = \log_{10} G_P = \log_{10} \left(\frac{\beta}{t^{\beta}} \right) + (\beta - 1)\log_{10} t = A + B \times x$$
 (7)

where $x = \log_{10} t$.

The plot of transformed P(t) data via Eq. (7), namely $y = \log_{10} G_P = \log_{10} [dln P(t)/dt]$ vs. $x = \log_{10} t$, validates domains of S-M1 description via the linear behavior, for which the linear regression yields optimal values of relevant parameters, namely τ and β .





Eq. (6) simplifies to the basic Malthus Eq. (1) for the power exponent $\beta=1$. Recalling consideration related to Soft Matter, it can be related to systems with a single, dominant relaxation process. A continuous distribution of multiple relaxation times is related to $\beta \neq 1$. It can be associated with the feedback process amplification for $\beta > 1$ and the system 'internal energy' dissipation for $\beta < 1$ (Sojecka and Drozd-Rzoska, 2024). The second Super-Malthus relation (S-M2) is associated with the following dependence (Sojecka and Drozd-Rzoska, 2024):

$$P(t) = P_0 \exp\left(\frac{t}{\tau(t)}\right) \quad \Rightarrow \quad \tau(t) = t \times \ln[P_0/P(t)] \tag{8}$$

In the above relation, the concept of the local and time-dependent relaxation time and then growth rate $r(t) = 1/\tau(t)$, has been introduced. It simplifies to the basic Malthus Eq. (1) for $\tau(t) = \tau = 1/r = \text{const}$ in the given time domain.

The direct analysis of P(t) data via S-M2 relation (left-hand part of Eq.(8)) might seem to be impossible because of the *apriori* unknown $\tau(t)$ functional form. Nevertheless, one can determine $\tau(t)$ temporal changes, as shown in the right-hand part of Eq. (8). The result of such analysis is shown in **Figure 2**.

The plot reveals that the relaxation time $\tau(t)$ and growth rate r(t) are constant for the extreme period reaching ~700 years, from the mid-Medieval Age till the Enlightenment epoch onset, as shown by horizontal dashed lines. It means that in this extreme time domain, the global population growth dominantly followed the simple Arrhenius pattern (Eq. (1)), with a huge distortion matched with the Black Death pandemic times. From the beginning of the 18th century till nowadays, a new general pattern is visble:

$$\tau(t) = -a + bt \qquad \Rightarrow \qquad r(t) = 1/(-a + bt) \tag{9}$$

where constant parameters a, b > 0.

Substitutrig above to Eq. (8) one obtains the exponential relation with internal critical-like singular change (Sojecka and Drozd-Rzoska; 2024):

$$P(t) = P_0 \exp\left(\frac{t}{\tau(t)}\right) = P_0 \exp\left(\frac{t}{-a+bt}\right) = P_0 \exp\left(\frac{ct}{T_C - t}\right) = 610 \times \exp\left(\frac{1.62t}{2216 - t}\right)$$
(10)

where P_0 is related to the Industrial Revolutions times onset at t = 1710, detected in **Figure 2**.

Notable is the link to the reference hyperbolic/Doomsday Eq.(4) (Von Foerster et al., 1960). Namely, when applying the Taylor series expansion for Eq. (10) one obtains (Sojecka and Drozd-Rzoska, 2024) the following relation:

$$P(t) = P_0 \exp\left(\frac{ct}{T_C - t}\right) = P_0 \left(1 + \frac{ct}{T_C - t} + \cdots\right) \propto \frac{1}{D - t}$$

$$\tag{11}$$

Equations (9), (10), and **Figure 2** indicate the singularity at the year $T_C \approx 2216$, whereas for von Foerster et al. (1960) Eq. (4) the 'Doomsday year' appeared $D \approx 2026$. Eq. (11) shows that this difference can result from neglecting higher-order terms in Eq. (4).

The model scaling Eq. (10) parallels relations characterizing dynamics in constrained and frustrated critical systems, with inherent spontaneously appearing multi-element critical fluctuations. Hence, for Eq. (10) and von Foerster (1960) Eq. (4), the name 'critical' instead of 'hyperbolic' seems to be more appropriate.





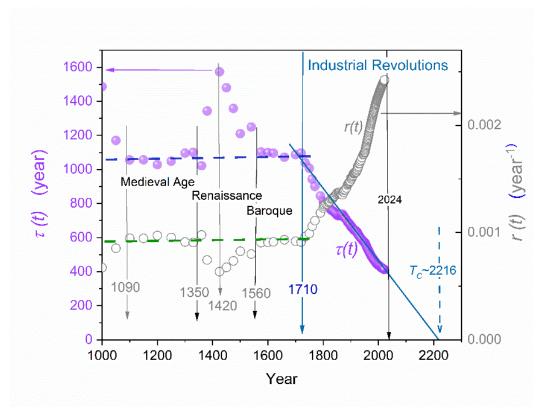


Figure 2. Temperature changes of the population growth rate r(t) coefficient and the relaxation time $\tau(t) = 1/r(t)$. Related historical epochs and dates are noted. Horizontal dashed lines are related to the basic Malthus behavior (Eq. (1)), with r = const and $\tau = const$.

The above reasoning considered the time-related growth of the global population. However, one can consider the relative population growth (RGR) vs. the population rise itself. In ref. (Lehman et al., 2021) considered the application of Verhulst Eq. (3) via the application of Pearl (1920) and Volterra (1927) concept of using a set of (r,s) parameters in subsequent time domains, and then a set of Verhulst equations for which next steps transformations $(r_i, s_i) \rightarrow (r_{i+1}, s_{i+1})$ occurred, before reaching the 'saturation phase' for the previous one. Lehman et al. (2021) explained these steps by overcoming subsequent bio- and eco-barriers during the global population development since the Anthropocene onset. In ref. (Lehman, et al., 2021), particular attention was drawn to the population relative growth rate (RGR) $G_P(\Delta t_i, \Delta P_i)$. Lehman, et al. (2021) considered changes in this parameter via the discrete analysis of population changes ΔP_i in subsequent time domains Δt_i . The resulted plot $G_P(\Delta t_i, \Delta P_i)$ vs. P_i revealed two linear dependences with crossover ~1965, namely:

$$G_P(t,P) = a + bP(t) \tag{12}$$

where a, b = const and the slope parameter b > 0 for periods 10 000 BCE < t < 1965 and 1965 < t < 2010. The crossover is associated with population $P \approx 3.2$ billion. In ref. (Lehman et al., 2021) such behavior was discussed as the argument supporting the portrayal of the global population changes via Verhulst model (Eqs. (2) and (3)), with

multi-parameter crossover introduced by Pearl (1927) and Volterra (1928). Very recently Sojecka and Drozd-Rzoska (2025) introduced the analytic $G_P(P,t)$ factor, instead of the discrete form earlier used. It is based on the new analytic set of global population data mentioned above (Sojecka and Drozd-Rzoska, 2024), namely:

$$G_P(t_i, P_i) = \frac{1}{P_i(t_i)} \frac{\Delta P_i(t_i)}{\Delta t_i} \implies G_P(t, P) = \frac{1}{P(t)} \frac{\mathrm{d}P(t)}{\mathrm{d}t} = \frac{\mathrm{d}P(t)/P(t)}{\mathrm{d}t} = \frac{\mathrm{d}\ln P(t)}{\mathrm{d}t} \qquad . \tag{13}$$





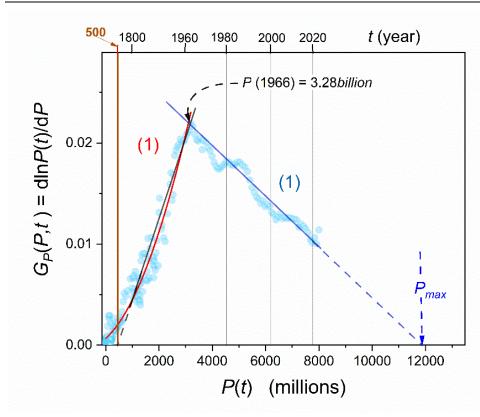


Figure 3. The global population-related changes of relative growth rate (RGR) coefficient since the Anthropocene onset till nowadays expressed vs. the global population values itself. The upper scale presents the coupled time scale. The brown vertical line indicates the Roman Empire terminal, which is also considered the early Medieval Age onset. Two domains (1) and (2), and the crossover between them are indicated. For domain (1), extended down to 10 000 the complete portrayal is nonlinear: in the plot via the second-order polynomial. Prepared using data shown in **Figure 1** and ref. (Sojecka and Drozd-Rzoska, 2025). The linear description of the $G_P(P)$ seems to be limited to ~2 centuries in domain (1).

Figure 3 shows the results of such analysis. It reasonably agrees with a similar plot presented in ref. (Lehman, et al., 2021) where the discrete definition $G_P(P_i, t_i)$ was used. The agreement includes two linear domains appearing for the eye-view test in **Figure 3**, as in ref. (Lehman et al.,2021). However, following Eq.(2), parameters describing the linear behavior via Eq. (12) should explicitly determine key parameters for the Verhults Eq. (3), namely: r = a and s = b. It means that following the above reasoning, related to **Figure 3** and Eq. (12), only two sets of parameters $(r_1 = a_1, s_1 = b_1)$ and $(r_s = a_2, s_2 = b_2)$ for domains (1) and (2) in **Figure 3** are allowed. It does not agree with the multi-parameter concept by Pearl (1927) and Volterra (1928). Also notable that the substitution of (r_1, s_1)) and (r_2, s_2) parameters to the Verhulst model Eq. (3) does not describe global population data P(t) in the subsequent two domains.

The above can question the ability of the Verhulst model for scaling global population growth.

The behavior presented in **Figure 3** based on the mentioned new generation data offers a higher resolution than earlier tests. It can indicate that the linear domain of $G_P(P)$ changes are relatively limited (black line in **Figure 3**). For the period reaching 10 000 BCE, the nonlinear evolution (2nd order polynomial) seems to offer better portrayal (red curve in **Figure 3**). Worth stressing problem of data presented in Figure 3 is the 'compression;' and superimposition for a colossal time domain covering 10 millennia, till ca. 500CE.

Figure 4. shows RGR evolution, presenting data from **Figure 3** in the log-log scale, two overcome the 'compression problem' mentioned above.





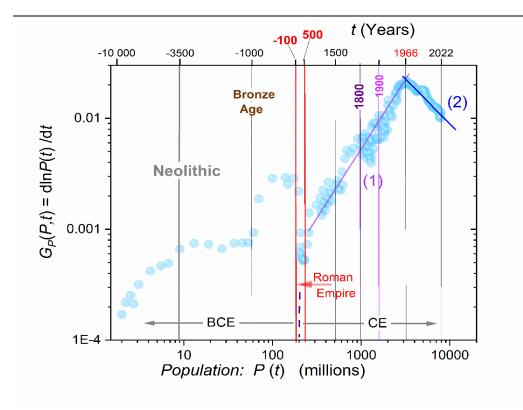


Figure 4. Relative Growth Rate (RGR) parameter changes vs. global population, for basic data from **Figure 3** presented in the log-log scale. The upper scale presents the coupled time scale. Some characteristic epochs are indicated.

The emergence of the 'complex structure' for Antiquity and Pre-Antiquity times and the grand RGR collapse for the Roman Empire times is notable. The linear portrayal of domains (1) and (2) is notably better for the log-log plot in **Figure 4** than for the reference 'direct' presentation in **Figure 3**. The extension of the fair linear behavior from the Roman Empire collapse to the crossover at ~1966 is notable.

Taking into account the definition of the 'analytic' RGR parameter (Eq. (13)) and log-log scale in **Figure 4**, one obtains the direct link to Eq. (7) associated with the empowered super Malthus S-M1 Eq. (6). Following the latter, the crossover year $t_{cross} \approx 1966$, related to the global population $P \approx 3.26$ billion, can be associated with the power exponent crossover in S-M1 Eq. (6): $\beta > 1 \Rightarrow \beta < 1$.

Following general features of the empowered Super-Malthus S-M1 equation, it can suggest the transition from a world where feedback interactions between different globally relevant factors amplified the 'Global Human Energy (GHE)' to a world where this energy is dissipated. It can mean that starting from the year ~1966, the global population began to spontaneously perceive the impact of grand constraints related to reaching real planetary borders – spatial and ecological.

4. Conclusions

This report presents a resume and new conclusions related to the recent works of the authors (Sojecka and Drozd-Rzoska, 2024 and 2025) and the report by Lehman et al., (2021). Complex Soft Matter Science methodology and tools not previously used in global population research have allowed a new discussion related to a new model scaling equation associated with fundamentally defined relevant parameters. The distortions-sensitive analysis showed significant local-temporal variability of global population changes. This casts doubt on attempts to study global population change based on the assumption of the possibility of monotonic description over long time intervals. The temporal variability and locality of global population growth are related to specific exogenous and endogenous disturbances, indicating that attempts to estimate future trends for the more distant future may be unreliable.







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Research

Haematology-Derived Inflammatory Markers in French Bulldogs with Brachycephalic Obstructive Airway Syndrome

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Abstract:

Brachycephalic obstructive airway syndrome (BOAS) is a chronic obstructive respiratory disorder that raises significant welfare concerns for affected breeds. Canine BOAS can be compared to obstructive sleep apnoea (OSAS) in humans, as both disorders cause intermittent hypoxia which can alter the metabolic, immune and inflammatory systems. Haematological markers and their ratios can be used to access systemic inflammatory status and have been proved to be changed in patients with OSAS. In this retrospective study we compared haematological parameters and haematology-derived inflammatory markers between French Bulldogs diagnosed with BOAS and nonbrachycephalic dogs. The study involved a total of 30 client-owned dogs; the BOAS group consistent of 15 French Bulldogs that were evaluated as BOAS grade 2 and 3 and the control group consisted of 15 healthy non-brachycephalic dogs. Significant differences (p < 0.05) were observed in several investigated markers. BOAS patients had a significantly higher platelet count, platelet-to-lymphocyte ratio and systemic immuneinflammation index, as well as significantly lower lymphocyte-to-monocyte ratio values compared to non-brachycephalic dogs, which suggest the presence of systemic inflammation in BOAS-affected dogs. Our study provides new insights into the inflammatory state in BOAS affected French Bulldogs and highlights the need for further studies in larger groups of BOAS patients of different brachycephalic breeds including evaluation of the effects of surgical treatment on haematology-derived inflammatory markers.

Keywords: Brachycephaly; Inflammatory markers; BOAS; Veterinary; Haematology; Systemic immune-inflammation index





1. Introduction

Brachycephalic obstructive airway syndrome (BOAS) is a chronic obstructive respiratory disorder that is predominantly observed in the brachycephalic dog breeds, one of the most popular being the French Bulldog (Mitze et al., 2022). BOAS arises from the skeletal changes that have evolved during selection of brachycephalic dog breeds that cause narrowing of the air passages. BOAS is related to respiratory and thermoregulatory problems (Žgank et al., 2023) as well as gastrointestinal (Freiche, 2021), dermatological (Hobi et al., 2023), ophthalmic (Palmer et al., 2021) and reproductive disorders (Ekenstedt et al., 2020) and therefore there is a significant welfare concern for brachycephalic breeds.

BOAS itself can be diagnosed with identification of specific anatomic changes and by concurrent observance of respiratory clinical signs (Packer et al., 2015). An accurate diagnosis is usually made with endoscopic evaluation of the upper respiratory tract (Fasanella et al., 2012). Respiratory or physical performance can also be used to determine the severity of BOAS, as well as an assessment of blood oxygenation (Bernaerts et al., 2010; Arulpagasam et al., 2018; Riggs et al., 2019). All these tests can be time consuming, expensive and require specific equipment, knowledge and trained personnel, as well as animals willing to cooperate.

Canine BOAS shares features with obstructive sleep apnoea syndrome (OSAS) in humans (Niinikoski et al., 2023). OSAS is a complex syndrome where muscle relaxation results in partial or complete blockage of pharynx during sleep, which results in apnoea or hypopnea. This leads to reduced blood oxygen saturation and chronic intermittent hypoxia that can cause altered metabolic, immune and inflammatory system responses (Topuz et al., 2022). Most of the systemic consequences of BOAS are similar to those of OSAS (Mellema and Hoareau, 2014).

Haematology-derived inflammatory markers, such as the neutrophil-to-lymphocyte ratio (NLR, calculated by dividing neutrophil count (NEUT) to lymphocyte count (LYMPH)), platelet-to-lymphocyte ratio (PLR, calculated by dividing platelet count (PLT) to LYMPH), monocyte-to-lymphocyte ratio (MLR, calculated by dividing monocyte count (MONO) to LYMPH), lymphocyte-to-monocyte ratio (LMR, calculated by dividing LYMPH to MONO), systemic inflammatory response index (SIRI, calculated using the formula: NEUT × MONO/ LYMPH) and systemic immune-inflammation index (SII, calculated using the formula: NEUT × PLT/LYMPH), are popular, inexpensive and easy accessible markers of systemic inflammation derived from routine haematological analysis. Selected haematology-derived inflammatory markers, such as the NLR, PLR, MLR, white blood cell count (WBC)-to-mean platelet volume (MPV) ratio (WMR), SIRI and SII, have confirmed the presence of systemic inflammation in OSAS patients and were proven to predict or even evaluate OSAS severity (Nadeem et al., 2013; Wu et al., 2018; Topuz et al., 2022; Kim et al., 2023; Elfeky et al., 2024).

The clinical significance and the potential to use the above-mentioned markers as diagnostic markers has already been indicated in canine inflammatory bowel disease (Marchesi et al., 2024), canine myxomatous mitral valve disease (Tuna, 2024), canine pyometra (Yazlık et al., 2022), gastric cancer patients (Karra et al., 2023) and as prognostic factors in canine sepsis (Pierini et al., 2020) and canine monocytic ehrlichiosis (Ozalp et al., 2024).

To date, there are no published studies reporting haematology-derived inflammatory markers in brachycephalic dogs with advanced stage of BOAS. Therefore, this study aimed to evaluate selected haematology-derived inflammatory markers—specifically, NLR, PLR, MLR, LMR, SII and SIRI in French Bulldogs with advanced stage of BOAS (grade 2 and 3) in order to identify systemic inflammation in this breed.

2. Materials and methods

A total of 30 dogs were included in this retrospective study, in which we evaluated haematological markers in 15 client-owned French Bulldogs diagnosed with BOAS that were admitted for surgical treatment of BOAS at the Small Animal Clinic of the Veterinary Faculty in Ljubljana, between the year 2021 and 2022. Data for the BOAS group was retrieved from the electronic internal medical database, including details on age, sex, breed, weight, BOAS grade, surgical records, and results of haematological analyses. The included dogs





were free from any systemic diseases, had not undergone medical treatment or vaccination in the preceding month, were diagnosed with BOAS, and had no prior history of upper airway surgery.

Upon initial examination, a thorough history was gathered for each BOAS patient. A preoperative questionnaire, completed by the owners, assessed various clinical signs associated with BOAS (respiratory problems, exercise intolerance, sleep disturbances and gastrointestinal symptoms). The diagnosis of BOAS was determined based on clinical symptoms of upper airway obstruction and the presence of anatomical abnormalities. BOAS severity was categorized according to the grading system outlined in previous studies (Dupré and Heidenreich, 2016). BOAS patients were classified into three grades based on the degree of airway narrowing at the nasopharynx, oropharynx, laryngopharynx and larynx. Grade 1 patients showed minimal to no airway narrowing, grade 2 patients exhibited around a 50% reduction in airway radius and grade 3 patients had near-complete obstruction at one or more of the before mentioned airway levels (Erjavec et al., 2021; Erjavec and Nemec Svete, 2023). Dogs were included in the study if they were evaluated as BOAS grade 2 and 3. Written informed consent was obtained from all dog owners.

The control group consisted of 15 non-brachycephalic dogs that were admitted for elective neutering. All dogs in the control group were clinically healthy and showed no signs of respiratory or chronic diseases. To be considered healthy, control dogs had to exhibit normal findings on physical examination, as well as the results of haematological and biochemical analyses (data not shown) within established reference ranges.

Venous blood samples were drawn from the cephalic vein prior to the surgical procedure and before administering any medications to the animals. For haematological analyses, the blood was collected in tubes containing EDTA anticoagulant (BD Microcontainer, Becton Dickinson, Franklin Lakes, NJ, USA). The samples were then analysed within one hour of collection using an automated laser-based haematology analyser ADVIA 120 (Siemens, Munich, Germany) to ensure accurate and timely results. Inflammatory markers derived from the haematological analyses were calculated using white blood cell differentials and PLT.

Statistical analysis was performed using commercial software (IBM SPSS 28, Chicago, Illinois, USA). The Shapiro-Wilk test was performed to determine the distribution of data. Based on the findings, parametric test or non-parametric test was used to compare data between control group of non-brachycephalic dogs and BOAS patients. Accordingly, independent t-test (for normally distributed data) or Mann Whitney test (for non-normally distributed data) were used to compare variables (age, weight, haematology parameters, haematology-derived inflammatory markers) between the two groups of dogs. Normally distributed data are reported as means \pm standard deviation (SD), whereas non-normally distributed as a median and interquartile range (IQR – 25th to 75th percentile), respectively. A value of $P \le 0.05$ was considered significant.

3. Results

3.1. Demographic data

The BOAS group involved 8 dogs that were diagnosed with BOAS stage 2 and 7 that were diagnosed with BOAS stage 3.

BOAS and control groups were equally represented by both sexes. The average age in months of the control group was 38.00 ± 21.01 and in BOAS group 41.27 ± 22.89 . There was no significant (p = 0.687) difference in age between the groups of dogs; however, the dogs in the control group were significantly (p = 0.028) heavier compared to the BOAS group, 17.7 ± 9.7 kg and 11.5 ± 2.5 kg, respectively.

3.2. Hematological parameters and haematology-derived inflammatory markers

The haematological parameters and haematology-derived inflammatory markers of dogs included in the study are presented in **Table 1**. BOAS patients had significantly higher PLT, PLR and SII and significantly lower LMR than control group. Other parameters, such as WBC, NEUT, LYMPH, MONO, MLR and SIRI demonstrated no significant differences between the groups. The NLR, MLR and SIRI were higher in the BOAS group compared





to the control group; however, these differences did not reach statistical significance, with p values approaching 0.05.

Table 1. Haematological parameters and haematology-derived inflammatory markers of dogs included in the study

Group	BOAS	Control	P value
WBC (× 10 ⁹ /L) ^a	10.87 ± 3.98	8.78 ± 2.43	0.094
PLT (× 10 ⁹ /L) ^a	431.4 ± 65.9	251.8 ± 55.5	< 0.001*
NEUT (× 10 ⁹ /L) ^a	6.96 ± 2.98	5.83 ± 2.40	0.264
LYMPH (× 10 ⁹ /L) ^a	2.42 ± 0.68	2.39 ± 0.82	0.922
MONO (× 109/L)b	0.45; 0.32–0.59	0.34; 0.22–0.48	0.125
NLRa	2.84 ± 0.69	2.38 ± 1.07	0.174
LMRa	5.40 ± 1.88	8.14 ± 3.67	0.016*
PLRa	189.3 ± 52.6	103.1 ± 29.4	< 0.001*
MLR ^b	0.20; 0.14-0.24	0.13; 0.09–0.22	0.051
SII (× 10 ⁹ /L) ^b	1264.2; 983.6–1404.7	508.3; 324.1–1004.8	< 0.001*
SIRI (× 10 ⁹ /L) ^b	1.24; 0.77–1.74	0.74; 0.32–1.54	0.071

Legend: *Significant difference (p < 0.05) between BOAS patients and control dogs; a - data presented as mean and standard deviation; b - data presented as median and interquartile range (25th to 75th percentile). WBC - white blood cell count, PLT - platelet count, NEUT - neutrophil count, LYMPH - lymphocyte count, MONO - monocyte count, NLR - neutrophil-to-lymphocyte ratio, LMR - lymphocyte-to-monocyte ratio, PLR - platelet-to-lymphocyte ratio, MLR - monocyte-to-lymphocyte ratio, SII - systemic immune-inflammation index, SIRI - systemic inflammatory response index. Statistically significant differences are marked bold.

3. Discussion

To our knowledge, this is the first study to investigate hematology-derived inflammatory markers in French Bulldogs with BOAS, revealing significant differences that highlight the systemic inflammatory impact of this condition. Our findings provide new insights into the systemic inflammatory state in BOAS affected dogs with notable changes in PLT and blood cell ratios, such as PLR, LMR and SII.

As previously mentioned, canine BOAS shares characteristics with OSAS in humans (Ni-inikoski et al., 2023). Wu and colleagues (2018) reported that MLR and PLR are elevated in OSAS, suggesting monocyte-driven inflammation, which contributes to the pathogenesis of OSAS-related complications. In our study, MLR was higher in the BOAS group, although the difference was not statistically significant. However, PLR values were significantly increased in BOAS dogs compared to non-brachycephalic dogs. Similarly, Gölen and colleagues (2024) observed significantly elevated PLR, as well as NLR, and SII values in OSAS patients compared to people without OSAS-

In our study, BOAS group showed significantly lower LMR values compared to control group. This might be associated with a heightened inflammatory response, as an increased proportion of monocytes relative to lymphocytes indicates systemic inflammation.

BOAS dogs exhibited a significantly increased SII compared to controls. SII, which incorporates neutrophil, platelet and lymphocyte counts, provides a comprehensive measure of systemic inflammation. Similar to our results, increased SII values were also reported in OSAS patients (Gölen et al., 2024; Güneş and Günaydın, 2024). Kim and colleagues (2023) showed that the SII can be a robust marker of immune and inflammatory activity in patients with OSAS. The higher SII values in our study are most likely the result of significantly higher PLT and the absence of lymphopenia in BOAS patients.

A significantly elevated PLT was noted in the BOAS group compared to controls. This finding might suggest a pro-inflammatory state in dogs with advanced BOAS (Margraf and Zarbock, 2019). Platelet activation has been associated with chronic inflammation and hypoxia, which are common in BOAS due to obstructed airflow and oxygen deprivation





(Delaney et al., 2021). Similar results have already been published in BOAS dogs, where PLT, MPV and plateletcrit were found significantly higher compared to non-brachyce-phalic dogs (Erjavec and Nemec Svete, 2023). As mentioned before, BOAS group exhibited a markedly higher PLR values, which is most probably a consequence of high PLT.

Interestingly, not all inflammatory markers showed significant differences between the groups. For example, WBC were slightly higher in BOAS patients compared to control dogs. Similarly, NLR and monocyte counts did not differ significantly between groups. This contrasts with findings in OSAS patients, where studies reported significantly higher NLR values in OSAS patients compared to healthy individuals (Gölen et al., 2024; Güneş and Günaydın, 2024).

Although we expected significantly higher SIRI values in the BOAS group compared to control group, the difference did not reach statistical significance. Interestingly, Pau and colleagues (2023) demonstrated a strong correlation between lower oxygen saturation and higher SIRI in OSAS patients. The absence of a significant difference in SIRI in our study may be attributed to low statistical power due to the small sample size.

Despite the compelling findings, the study is limited by its small sample size (15 dogs per group), which may affect the generalizability of the results. Therefore, larger studies in different brachycephalic breeds with BOAS are needed to offer better insight into the complex interplay between systemic inflammation and respiratory dysfunction in BOAS-affected dogs. Additionally, a deeper evaluation of the immune and inflammatory responses in these animals is essential to fully understand the underlying mechanisms contributing to the condition. Given the significant differences between the groups in our study, there is a solid base for further research in these areas.

In conclusion, the results of our study suggest the presence of systemic inflammation in BOAS-affected French Bulldogs. Further studies in larger groups of BOAS patients including evaluation of the effects of surgical treatment on haematology-derived inflammatory markers are warranted.

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Institutional Review Board Statement: The work described in this paper involved the use of non-experimental (client-owned) animals. Established internationally recognised high standards ('best practice') of veterinary clinical care for the individual patient were always followed. Due to retrospective nature of the study, ethical approval was not specifically required.

Conflicts of Interest: The authors declare no conflict of interest.

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Review

Can we Strengthen the Anterior Cruciate Ligament with Passive Knee Joint Loading?

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Abstract:

Passive knee joint loading has been proposed as a safe method to enhance knee stability during early postoperative rehabilitation following anterior cruciate ligament (ACL) reconstruction. However, its effects on anterior and mediolateral rotational laxity remain unexplored. Evaluating whether passive knee joint loading in the direction of anterior tibial translation improves knee stability post-surgery is essential. Current research primarily focuses on active loading, such as quadriceps strengthening exercises, but concerns about excessive anterior tibial translation and graft compromise make early postoperative loading controversial. Our review builds on previous studies on passive knee joint loading in healthy individuals and post-ACL reconstruction cases. Unlike active loading, passive anterior tibial translation via the DYNEELAX® arthrometer enables controlled mechanical stimulation within a safe force range, potentially aiding early graft adaptation. Preliminary findings suggest passive loading could be a valuable addition to ACL rehabilitation, though further research is needed to determine optimal parameters such as force magnitude, frequency, and duration. Methodological challenges complicate the study of passive loading in clinical trials. Future research should focus on integrating controlled passive loading into rehabilitation protocols while distinguishing its contribution to recovery. Understanding the biomechanical effects of passive loading will help refine ACL rehabilitation and broader musculoskeletal treatment strategies, ultimately improving long-term knee stability and reducing graft failure risk.

Keywords: Knee; ACL reconstruction; Passive loading; Stability; Postoperative rehabilitation; Joint mechanics





Introduction

In Slovenia, we initiated research to determine whether passive loading of the knee joint affects its stability in healthy women (Vauhnik et al., 2015). In this study, physiotherapists applied anterior translation forces of 100 N and 170 N to the knee joint, and the results showed no statistically or clinically significant effect on knee joint stability. This remains the only published study investigating the effects of passive knee joint loading in healthy individuals. Regarding anterior cruciate ligament (ACL) injuries, two case studies have been published (Rugelj et al., 2021a; Rugelj et al., 2021b), whereas no studies have yet explored the effects of passive knee joint loading on its stability in participants following ACL reconstruction surgery.

In contrast, research on active knee joint loading provides insight into its effects on knee stability. Barcellona et al. (2015) found that active knee loading through quadriceps strengthening exercises in an open kinetic chain improved knee joint stability in participants with acute ACL injuries. The group performing exercises involving anterior tibial translation demonstrated statistically and clinically significant reductions in knee joint laxity compared to the control group. Similarly, Perry et al. (2005) investigated the effects of quadriceps strengthening exercises in an open kinetic chain on knee joint loading in participants following surgical ACL reconstruction. Loading initiated 8 weeks postoperatively, applying a force of 270 N, did not increase knee laxity.

Mechanical loading is essential for maintaining and adapting the structural, histological, and functional properties of connective tissue (Kjaer, 2004; Woo et al., 2006). Exposing the newly reconstructed ACL to controlled mechanical loading before 8 weeks postoperatively is critical for its functional adaptation. However, excessive loading may overstress the graft, highlighting the need for a carefully regulated approach. One of the primary forms of mechanical loading is anterior tibial translation, which can be applied either passively (e.g., manually via the Lachman test or instrumentally using knee arthrometers such as KT1000, KT2000 and DYNEELAX®) or actively (through quadriceps strengthening exercises in an open kinetic chain). Morrissey et al. (2009) reported that loading forces between 160 N and 850 N improve knee stability, while forces exceeding 850 N increase laxity and may lead to instability.

Knee joint stability, a key indicator of successful ACL reconstruction, should be assessed through a combination of patient-reported stability and objective measurements using knee arthrometers such as the DYNEELAX® (Figure 1). This device has demonstrated high intra-rater reliability (Mihalinec et al., 2024; Nascimento et al., 2024) and excellent interrater reliability (Mihalinec et al., 2024), reinforcing its value in clinical practice.

The DYNEELAX® arthrometer allows for the precise determination of anterior knee laxity, measured in mm/N. The results are presented as a force-displacement curve (Figure 2A), where the slope represents the tensile strength of the ligament. Additionally, mediolateral rotational laxity is quantified in °/Nm and displayed as a torque-angle curve (Figure 2B), with the slope similarly reflecting the ligament's tensile strength (Genourob, n.d.).



Figure 1. The DYNEELAX® arthrometer set up to measure the laxity of the right knee joint (Genourob, n.d.).





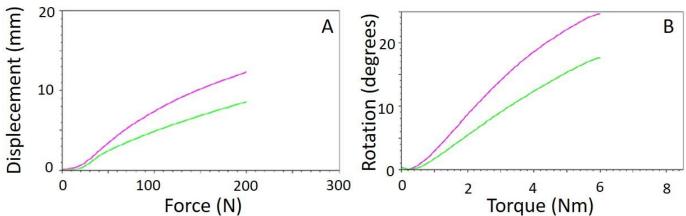


Figure 2. A: Force-displacement curve for anterior knee joint laxity obtained from the DYNEELAX® arthrometer. **B**: Torque-angle curve for medial rotational knee joint laxity obtained from the DYNEELAX® arthrometer. The pink line represents a pathological knee joint, while the green line indicates a healthy knee joint (Genourob, n.d.).

A critical component of ACL rehabilitation is the identification of safe and effective loading strategies that facilitate graft adaptation. The early postoperative phase is particularly sensitive, as excessive anterior tibial translation can compromise graft integrity. As a result, active loading exercises—such as quadriceps strengthening in an open kinetic chain—are typically avoided during the first eight weeks postoperatively. This limitation highlights the need for alternative approaches, with passive loading emerging as a promising strategy for controlled mechanical stimulation.

Passive loading may provide sufficient mechanical stimulation to promote the functional adaptation of the ACL graft while minimizing the risk of excessive strain or graft failure, thereby contributing to a reduction in knee joint laxity. Unlike active loading, which imposes high forces on the graft, passive anterior tibial translation using the DYNEELAX® arthrometer enables controlled application of mechanical stress, ensuring gradual exposure within a safe range. If validated by future research, these findings could support the incorporation of passive knee joint loading into early rehabilitation protocols, helping to refine post-surgical treatment strategies, optimize outcomes, and reduce the risk of graft failure and other postoperative complications.

Taken together, these findings highlight the importance of further research into passive loading as a potential strategy for enhancing ACL rehabilitation. Determining the optimal parameters—such as force magnitude, duration, and frequency—will be crucial for safely integrating passive loading into clinical practice. Given the ethical constraints of clinical studies, it would not be ethically appropriate to implement passive loading alone within rehabilitation protocols. Future research should focus on rehabilitation programs that incorporate controlled passive loading, ensuring that its effects are distinguished from other therapeutic interventions. This approach would allow for a comprehensive understanding of passive loading's potential role while maintaining ethical standards and patient safety in clinical settings.

Investigating the role of passive knee joint loading not only holds promise for improving post-ACL reconstruction outcomes but also contributes to a deeper understanding of knee joint biomechanics and the optimization of rehabilitation strategies for other musculoskeletal conditions. Addressing these gaps through well-designed studies will help establish evidence-based guidelines that enhance clinical decision-making. Ultimately, each new insight in this field represents a step toward more effective and safer rehabilitation, improving long-term knee stability and reducing the risk of re-injury.

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Conflicts of Interest: The authors declare no conflict of interest.







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Review

Vladimir Shtiftar (1880-1942) and his Views on the Formation of the Architectural and Cultural Landscape of the Eupatoria Resort

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Abstract:

The paper considers activities and theoretical heritage of Vladimir Shtiftar, a pedagogue and social activist of Slovenian ancestry, living in the early 20th century in a Black Sea town of Eupatoria. Shtiftar's works devoted to the local landscape aesthetics, can be considered as one of the first attempts to ponder over an interaction between natural factors, urban planning, history of arts and healing practices. His interdisciplinary studies give us an insight into a vibrant context of a changing socio-economic structures in the pre-war Black Sea region. What inspired this passionate classical scholar to formulate a holistic outlook for a health resort aimed at preserving its rich cultural heritage, which was endangered by rapid modernization? How effective were V.Shtiftar's efforts? Why do his views and works remain relevant today?

Keywords: Vladimir Shtiftar; Eupatoria; Kerkynitis; archaeology; landscape





Much has been said about the importance of landscapes and landmarks in the development of cultural activities aimed at health improvement, psychotherapy, and wellness (Souter-Brown, 2015; Rose, 2012). Rapid economic development worldwide has increased the demand for effective policies to preserve landscapes from uncontrolled destruction (Fancourt et al., 2019). Today, the international community unanimously agrees that poorly regulated urban development has damaged and impoverished the cultural and aesthetic heritage of entire regions. In response, various preventive measures have been proposed, including the supervision of all types of public and private construction. These structures should be designed to meet specific aesthetic standards while avoiding superficial imitation of traditional forms. Additionally, they should harmonize with the overall atmosphere of the site that is intended to be preserved. Civic control mechanisms and architectural watchdogs gain importance when discussing the development of historical settlements with recreational potential, as these areas are particularly vulnerable to economic pressures at the local level. Their activities promote the best principles agreed upon at the international level. One of the most notable examples of this approach is the 1962 UNESCO Recommendation concerning the Safeguarding of the Beauty and Character of Landscapes and Sites.

However, protecting heritage requires continuous improvement and upgrading of policy tools, making further research into past best practices essential. This task becomes even more significant when considering that Slovenians were among those who formulated and defended these advanced principles. This paper seeks to revive the legacy of Vladimir Shtiftar (Figure 1), a remarkable teacher and public figure of Slovenian origin. Born in 1880 in the Russian city of Kaluga, he was the son of France Štiftar (Figures 2, 3, and 4), widely recognized as the father of mountain tourism in Logar Valley.



Figure 1. Vladimir Shtiftar in gymnasium teacher uniform, decorated with the Order of Saint Stanislaus, 3rd class, early 1910s.





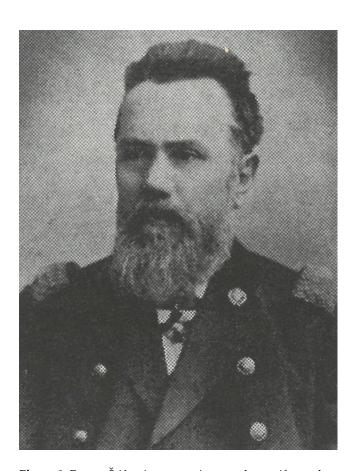


Figure 2. France Štiftar in gymnasium teacher uniform, decorated with the Order of Saint Anna, 2rd class, early 1910s.

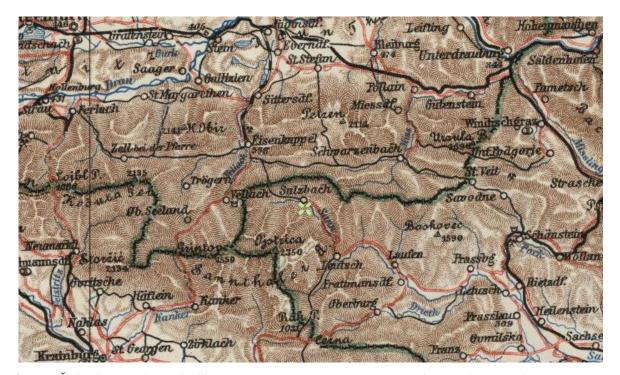


Figure 3. Štiftar family village of Solčava appearing under its German name of Sulzbach on the old Austro-Hungarian map, early 1900s (marked with a green cross).







Figure 4. Solčava on today's map of the Slovene-Austrian borderland.

As a graduate of the Historical and Philological Lyceum in the city of Nizhyn, Vladimir Shtiftar secured a position as a teacher of ancient languages at the gymnasium in Eupatoria. Situated on the Black Sea coast, Eupatoria captivated the young teacher, sparking his deep passion for architecture and archaeology. He devoted himself wholeheartedly to preserving the city's unique urban landscape, which harmoniously blended Greek, Ottoman, Oriental, and European elements. Engaging his students in hands-on historical inquiry, Shtiftar led the excavation of an ancient burial mound near the city. Determined to preserve his findings for future scholars, he meticulously documented his observations, performing schematic measurements of the stone masonry and describing the collected artifacts. Through this work, he contributed significantly to solving a key scholarly question of the time—the localization of the ancient Greek colony of Kerkynitis.

Shtiftar's ambitions, however, extended far beyond personal curiosity. Building on substantial theoretical knowledge and excavation findings, he fundamentally reimagined the architectural identity of Eupatoria. He envisioned it not merely as a developing resort city but as a "park city" and a "garden city," united by a cohesive architectural style.

The quintessence of his vision is articulated in his 1916 book, Suburban Architectural Landscape of Eupatoria. He proposed replacing the city's chaotic, unstructured development with centralized urban planning inspired by the symmetry, proportion, simplicity, and elegance of 5th-century BCE Greek temples. This influence is hardly surprising, given Shtiftar's background as a scholar of philology. However, his retrospective aesthetic choices can also be seen as a reflection of the "Zeitgeist", the spirit of the Historicism movement that dominated European and, particularly, Russian architecture at the time.

The late 19th and early 20th centuries saw a resurgence of Greek Revival architecture, particularly in Moscow, where designers and patrons incorporated refined elements of the Athenian Erechtheion (421–406 BCE) into the city's architectural landscape. Interestingly, this trend largely bypassed St. Petersburg, the imperial capital. It is possible that Shtiftar's embrace of Greek Revivalism was influenced by his native Kaluga, situated in close proximity to Moscow, where the movement thrived.

Tragically, Shtiftar did not live to see his vision realized. He and his family perished in a mass execution carried out by the Nazis in 1942. However, his contributions were not in vain. His invaluable descriptions of archaeological excavations laid the foundation for future research on Kerkynitis. Furthermore, the Neo-Classical architectural principles he championed were eventually embraced by post-war Soviet architects tasked with recon-







structing war-ravaged Black Sea cities such as Sevastopol and Odessa. More broadly, Classical architectural principles were integrated into Socialist Realism and, later, into Postmodern Contemporary Classical architecture, ensuring the enduring influence of Shtiftar's ideas. Slovenia has every right to be proud of this remarkable son—one of the pioneering researchers of ancient civilizations in the Northern Black Sea region. It is time to celebrate his multifaceted legacy and reinstate him as a visionary advocate for healthy urban land-scapes, a thinker who was clearly ahead of his time.

Shtiftar's creative and intellectual contributions remain highly relevant in today's era of rapid advancements in landscape design, urban planning, and territorial branding. His central idea - that a well-planned, welcoming resort landscape must be integrated into urban development strategies from the very beginning - has never lost its significance.

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Research

The Performer as a Multi-Expressive Artist in Current Flute Experimentation in Argentina.

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Abstract:

The proposed theme of the discussion is the actual musical experimentations in Argentinian flute scene. Specifically, the approach to music pieces where the performer plays the flute, recites in different languages, uses the voice while playing the instrument and realizes various movements on the stage.

Keywords: Flute, Experimentation, Expressiveness, Performer, New virtuosity, Creativity.

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1. Introduction

Artistic practices have changed under the influence of new technologies and the expressive use of the body of the musical performer and the instruments in works that can include stage movement, use of one or several musical instruments and the human voice with recitations of fragments of poetic texts, and guttural, onomatopoeic, phoneme, etc., sound emissions. In this way, in the last centuries in the Western academic music scene, artistic productions have begun to ask the performer to appropriate a role of creative centrality, most of the time, sharing with the composer the "formative intention" (Eco, 1970). In this position, great flexibility is required, greater participation in the creation of the work and a complex use of the body and its expressiveness, to create a kind of new instrumental virtuosity typical of these epochal experimentations characterized by the emergence of the performer as a multi expressive actor.

We investigated two pieces by Argentine composers that ask the performer for an energetic reading of a highly complex sound material in which sound aspects of the instrument are interwoven: recitation or vocalization and the use of new technologies. The pieces selected for analysis are two pieces for flute dedicated to the Argentine flutist Ana Ligia Mastruzzo: "ubicua" by Juan Carlos Tolosa and "Coyolyantototl" by Federico Núñez.

2. "ubicua" by Juan Carlos Tolosa

The piece "ubicua" by Juan Carlos Tolosa (**Figure 1**) is presented as a paradigm of the use of new technologies in art. It is a piece for four flutes (performed by one performer) and bluetooth devices. Tolosa thought of the idea of ubiquity, of being in different places at the same time, as a projection of what happens with the live flute and the ghostly flutes (prerecorded flute sounds) that emerge from Bluetooth. Regarding the idea of ubiquitous, he also thought about what this meant: at the right place, at the right moment. Everywhere: For this, he devised a stage arrangement in which the flutist was moving through four stations. In four spaces: Each of these spaces was going to be attributed to a particular flute: Station I (flute in G): located approximately in the center and slightly to the left of the stage (always from the audience's perspective); then it moved to the right and to the back, Station II (piccolo): the movement occurred forward and a little more to the left, Station III (flute in C): finally it moved to the extreme left, quite far forward but less than the C flute, Station IV (bass flute): where the work concludes.

The performers of Bluetooth devices are in the audience, as far away from each other as possible and, if there are seats, they should avoid sitting in the same row. As for lighting, if there are lighting devices in the room, only the station in which the flutist is playing should be illuminated. The rest of the stage must be in darkness. In Figure 2 we can see Ana Ligia Mastruzzo playing on the stage The flute player's movements from one lectern/Station to another will occur in total darkness. Phosphorescent adhesive tapes are planned for the stage floor. To turn the different lights on or off it is necessary to follow the score. In the event that the illuminator does not know how to read music, an assistant will be required with the score to indicate which station to illuminate. At the end of Station IV, where indicated in the score, the general stage lights and those in the stalls must be turned on abruptly and simultaneously. If there are no lighting devices in the room, lectern lights will be used at each lectern/station, with the entire performance of the work remaining on.

The performers of the Bluetooth devices must have a cell phone or a Tablet or iPad – devices that have Bluetooth – with the 10 tracks that the work needs. They must also be provided with a small Bluetooth speaker to amplify the device.







Figure 1: Score of "ubicua" by Juan Carlos Tolosa. Detail of piccolo score that belongs to the Station II. Movements of flute player on the stage, correspondence between flute and the devices, musical and time instructions are indicated in the score.





2.1 Reflexion about "ubicua" piece

The cell phone, tablet or iPad, any device chosen to use in the performance of the piece, serves to project what happens with the live flute and the ghostly flutes that emerge from the Bluetooth, operating as a "ritual object" (Ulm, 2021) that transmits from different points of the auditorium sounds that were pre-recorded, created and manipulated by the composer based on the sounds of the flute instrument and those created electronically. Thus, Tolosa captures, retains and makes available for any performer and for the user the fugitive in Edison's way, retains the sound created in the device and with this material experiments and breaks perceptual thresholds. The work is a sound experience (Ulm, 2021) in which the viewer does not listen to a finished piece, but rather it is an open musical experience as conceptualized by the semiologist (Eco, 1984) in which there is no a certain place for perfect listening, all spaces in the auditorium are possible places for effective listening. In the case of "ubiquitous", the composer does not create a finished art object to be observed or enjoyed as a whole, but rather uses technology and everyday devices such as the cell phone to make them act in a way illicit, by users who adjust them to new functions. It is a highly human operation as the Italian composer Luciano Berio conceives (Berio, 2023): "However, music remains a human activity: it does not exist in nature. Therefore, it is not possible to conceive a musical discourse without referring to those means that man has invented or adapted for musical purposes." ("Sin embargo, la música sigue siendo una actividad humana: no existe en la naturaleza. Por lo tanto, no es posible concebir un discurso musical sin hacer referencia a aquellos medios que el hombre ha inventado o adaptado con fines musicales". (The tradition of the author).

Each Bluetooth device performer produces a performance that will be distributed in the sound space. The cell phone or technical device used allows the sensitive to be transmitted through small speakers from different points of the auditorium in which the musical experience takes place, making the miniaturization of the devices and the digitalization of sound essential for the realization of the experience. The random arrangement of the technique means that there is no ideal place to perceive, in this way, the listener (Saavedra, 2013) will be able to create his or her particular and unique sound space. In the score, in the text, the performance is written, the way of acting, the map of more or less arranged, regulated actions, and the map of the energy flow. What is written is an energetic text that the artist translates into ritual, the interpreter performs and puts into action a form that sounds. (In **Figure 2** we can see Ana Ligia Mastruzzo performing in a dark stage at the Brazialian premiere of the piece at the UNESP, Universidade Estadual Paulista). The cell phone is outside its context of use and becomes a device of sensitivity. The technical object is outside the norm, it emerges as a new artifact, creating unprecedented spaces of legitimacy, a new space of ritual.

2. Coyolyantototl by Federico Núñez

2.1. About the aesthetics and performance

The piece Coyolyantototl (**Figure 3**) (in the Nahuatl language which means place of action of the rattle bird) is written for pre-Columbian ceramic double flute, flute and recitation in Nahuatl (by an interpreter).

This is a piece composed of three parts: 1 – "Teotl (God of movement); 2 – "Oncuicatinemi" (Go sing); 3 – "Totol" (Bird) and invites listeners to immerse themselves in an imaginary Mesoamerican past through the combination of the timbres of the original language, the replica of a Mesoamerican clay flute and a modern Western instrument such as the transverse flute. In this way, the aesthetic idea consists fundamentally in exploring timbral combinations in the scenic play that develops during the piece.

We can notice an alternation of onomatopoeia and Nahuatl language. These onomatopoeias serve as a link to acquire the continuity of the piece and allow the performer to move from one instrument to another. It is recommended to use amplification that allows you to balance the sound planes of the voice and the instruments. The texts were provided by the Mexican poet Cuauhtémoc Vite.





It is highly recommended that performers have experience and knowledge in recitation and theater to better address this work and any other that requires the use of the voice by the performer.

The work contains a series of issues to take into account when studying and interpreting it. As for the recitation, this is always clarified in the score with a rhythm that serves as a guide for the cadence of the words and syllables, without having to be strict or too rhythmic. It should flow naturally. In the first piece, the voice is recited in a whisper, respecting the indicated rhythm and using onomatopoeia "nu" that are interspersed between the sounds of the flute and the double flute. Here the performer must achieve a sound environment of lightness and fluidity, thinking about the God of movement that gives its name to the piece. There is a non-linear timbral transfer between the flutes where finally the double flute together with the onomatopoeia closes the number. In this way, a kind of timbral modulation from the transverse flute to the double flute is generated, carried out gradually, where the Nahuatl language functions as a link or as a distracting element. In the second part the recitation is spoken and functions as an introduction that begins the section playing exclusively with the double ceramic flute. The spirit here is that of joy, it is an ode to beauty, to birds, to song, to love. It is very important to feel confident with the fingerings and explore the sounds of this new instrument to convey the spirit of the piece. It is recommended to memorize the double flute section to become comfortable with this flute. The last part has an interpretation indication: «mechanical», here the performer must be very rhythmic and forget about the lyrical issues of the previous part. It is a fast piece that requires a lot of skill to move quickly from one flute to another and from the flute to the whispered sections with onomatopoeia or the mouth closed. The text, like the previous piece, is inserted during the course of the work.



Figure 2: Ana Ligia Mastruzzo playing







Figure 3. Coyolyantototl manuscript.





2.2. Reflection on Coyolyantototl

Given the characteristics of the piece, we find ourselves facing a *new virtuosity* (Lavista 2010) that is not the traditional one of romantic pieces of European academic origin, but rather is typical of the world of contemporary music that shows new musical expressions that seek to break with tradition without denying its existence. Local features are mixed with the vestiges of European tradition and compositional techniques.

Here the prominence is given by the replica of the ceramic instrument that provides the piece with its organological characteristics and with it its own interpretative complexity. The double flute is in no way intended to resemble and imitate the transverse flute, but, on the contrary, the piece is structured around it in such a way that the transverse flute and the voice modulate and transform to create a sound unity, amalgamated.

The musical piece Coyolyantototl represents a paradigmatic creation of the rapprochement between Mesoamerican and European cultures that challenges the performer with endless questions to address, from interpretative-performative to cultural aspects. It is also true in regarding the treatment of stylistic and instrumental resources, since it marks an unprecedented way of connecting different musical cultures through its own new musical grammar that is manifested throughout musical discourse. Language, in this way, is freed from all rigidity and acquires formal relationships that are legitimized in creation itself. The interpreter opens up a huge and rich panorama of topics to know.

3. Conclusion

As we mentioned at the beginning, it has been more than a century since Artists (authors, performers) have worked in search of the extreme limit of the instruments' potential, building their own aesthetics on those frontiers. New timbral searches have repudiated all positivist principles of homogeneity and equality, the parameter of timbre became increasingly predominant and the instruments became true laboratories for these explorations. With the digital era, new technologies began to occupy more space in artistic productions, causing the need to amplify the flute, transforming it into an "electric flute." Effect pedals were used to alter the source sound, devices such as Bluetooth were used and electronic and mixed music was composed, in works in which various disciplines also dialogue and the limits between them were blurred and fluidized. More and more musical creators have needed to use materials that were not exclusively sound, at the same time that they required expanding the possibilities of traditional instruments towards unknown frontiers. It remains for future research to investigate the deep causes of these current sound searches and aesthetic experimentations, the musical reasons why we find this predominance of works destined for multi-expressive flutists.

Conflicts of Interest: The author declares no conflict of interest.

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Research

Nannies and Governesses in Russia and Russian Literature of the 19th Century

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Abstract:

This article describes the role of nannies and governesses in Russian noble families in the 18th and 19th centuries. It presents briefly the guidelines for the upbringing and education of children in the aristocratic families and the influence from European countries, mainly France and to some extent England. The Russian nobility hired foreign teachers, governesses and governors to raise their children in the "Western way". In contrast to them, the nannies who took care of the children in their earliest childhood, were Russian women, mostly of peasant origin. The article provides examples of the portrayal of governesses and nannies in the literary works of some famous Russian writers. It mentions some real nannies of future writers who played an important role in their lives and influenced them with their moral examples, as well as by telling fairy tales and stories. The article also touches on the topic of depictions of nannies in painting and sculpture.

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Keywords:

Governess; nanny; European influence; national culture; upbringing and education; Nobility; Russia of the 18th and 19th centuries





1. Introduction

1.1. The upbringing and education of noble children in Russia in the 18th and 19th centuries

As Marina Surenskaja writes, upbringing and education in noble families depended mainly on the financial situation of the family and on the gender of the child, but the choice of schooling could also be influenced by the long distance from educational institutions. Boys were taught by home teachers or attended military schools, boys from wealthier families could also attend private fee-paying schools. Very important families could send their children to special elite schools, such as the famous Tsarskoye Selo Lyceum, which Pushkin, among others, attended. Girls were educated at home or in so-called institutions of virtuous virgins.

Governesses first appeared in Russia in the families of the tsars, as Olga Solodjankina writes, but in the period between the 18th and 19th centuries they were a common phenomenon in all noble families. The main educators of children in the 18th century were governors and governesses, who were mostly foreigners, writes Ljudmila Konšina (Konšina 2016). As Konšina writes, in the rich noble families of the 18th century, even two governors were hired, one of whom was usually French. One educator took care of the child's upbringing, while the other took care that the child studied and read books with him - this was usually a Frenchman who, of course, also taught French. For other subjects, they hired other teachers. The governess was responsible that the child was diligently learning with the teacher, whether it was a teacher of dance or other subjects.

The nobles who could afford such pedagogues were very well educated. They read a lot, even in foreign languages, and could recite a lot by heart, writes Ljudmila Konšina (Konšina 2016).

As Marina Surenskaja writes, in the first half of the 19th century it was very common for children to be educated at home. They could be taught by parents or older brothers or sisters; in this case, they remained poorly educated, even almost illiterate, writes Marina Surenskaja. In the first half of the 19th century, education got more value. The families often hired home teachers. There were very few Russian teachers, so the children were mainly taught by foreigners. Finding a good teacher was quite difficult. As we learn on the culture.ru portal, already in the time of Peter I, governesses and governors were often not properly educated to teach children. They were hired mainly because the children could learn a foreign language with a native speaker. Those teachers taught the children other subjects as well, even though they were not pedagogues. According to Ljudmila Konšina, it often happened that French immigrants looked for work in other professions, for example as cooks or hairdressers, and ended up working as governors, since the salary of governors was quite high even by the European standards of that time, and they got also a free accommodation and meals. As the aforementioned author writes, there were also excellent teachers among foreigners, but there were very few of them. The emphasis in domestic education was on learning a foreign language, French. They learned some geography and early history, but learning Russian grammar with foreign teachers was no longer possible.

As stated on the website culture.ru, the domestic education was regulated by a decree from 1755 by Empress Elizabeth I, when foreign teachers had to pass an exam at the Academy of Sciences in St. Petersburg or at Moscow University. The person who hired a governess without an exam was fined for not following this rule. As Ljudmila Konšina writes, at the end of the 18th century, home teachers or governors could already be high-quality educators. Such were also teachers in educational institutions for nobles.

According to Marina Surenskaja, girls' education was often deficient. In poor families, girls were mostly taught by their mothers or older sisters, while rich families hired governesses or even teachers to educate their daughters. A good education for noble girls meant good upbringing, i.e. correct behavior in society, and included playing of some instrument, singing, drawing and dancing. Of course, there were exceptions: existed well-educated girls from simpler families and perfectly educated aristocrats. Young noblewomen could only go out into the street accompanied by a person older than them or a governess. The rules of etiquette included, for instance, that they were not allowed to laugh loudly and







speak loudly or talk to strangers. The main goal of education was for noblewomen to behave well at dances and in society, while poorer girls were raised to be good housewives, the mentioned author emphasizes. As Ekaterina Kolosova writes, the first public educational institutions for girls appeared only in the 19th century; in 1858, public high schools appeared. (Kolosova, p. 68).

As Ekaterina Kolosova states, educators and teachers at the beginning of the 19th century were mainly church dignitaries, university professors (who were often foreigners), students or educated servants. Depending on social status and education, teachers or educators were divided into so-called home teachers, nannies and governesses. The title of teacher was higher than that of governor. The teacher had to be a graduate of a high school or faculty, or had to pass special exams to obtain the title of home teacher (Kolosova, p. 69). In 1834, the Native Teachers Ordinance was issued, requiring teachers to be Christians and resident in Russia. The emphasis in education was on foreign languages; subjects as history, geography, mathematics, physics were studied according to how important these subjects were to the child's parents and to the children themselves; religious education was also important (Kolosova, p. 70). Teaching was thus distinctly individual, adapted to each student individually. As Kolosova writes (p. 71), boys could be educated at home until the age of twenty, while girls could be educated at home until they got married. Teachers had to be supported financially throughout their lives. According to culture.ru, the governess stayed with the girl until she found a husband. If she did not marry, the governess could remain with her as a companion. From 1853, old governesses received small public pensions. The profession of governess was widespread in Russia until the revolution of 1917 (culture.ru).

As we learn from various sources, the parents handed over all their obligations to the children to the governesses. The nobles did not even see much their children, as they lived their cosmopolitan lives, and they did not even have the time or desire to raise with their children. Immediately after birth, the child was placed in the care of a nanny or a nurse and they assumed all maternal obligations towards the child.

The period of Tsar Peter I (reigned between 1672 and 1725) was a time of reforms. In 1703, Tsar Peter I., named also Peter the Great, had built from a swamp by the Neva River a city of St. Petersburg. The construction of the city, which was supposed to be a "window to the West", was based on European models. Saint Petersburg later became the Russian capital instead of Moscow (it remained a capital until 1918). (Wikipedia, Peter the Great). In that time the Russian nobility began to follow the European way of life and thinking. Many aristocrats also wanted to raise their children in a "Western" way and hired foreign governesses and governesses.

2. Governesses in Russian literature

2.1. European influence in the education in the work of Ivan Sergeyevich Turgenev

The writer Ivan Sergeyevich Turgenev, who spent a lot of time abroad, especially in Germany and France, was widely read and admired by his contemporaries, but at the same time he was also criticized. He was labeled as too much "European" writer, who even did not love much his homeland. As Blaž Podlesnik writes, Europe was for progressive-oriented intellectuals of the 19th century "a progressive model that Russia should follow"; on the contrary, conservative, "authentically Russian" oriented thinkers claimed that this very Europeanness "is the source of all problems " for Russia (Podlesnik 2024, p. 115). Dostoevsky wrote in his visionary essay about the World Exhibition in Vienna in 1873, which he visited, that for Russians Europe is a "land of holy wonders": a place where all Russians want to go. At the same time, Europe will never recognize Russia as a part of itself, because Russia, with all its Asian parts, is too different from Europe.

Turgenev in his novel A House of Gentlefolk described a noble family that raises its children with the help of foreign, European educators: »When the time came to teach him languages and music, Glafira Petrovna engaged, for next to nothing, an old maid, a Swede, with





eyes like a hare's, who spoke French and German with mistakes in every alternate word, played after a fashion on the piano, and above all, salted cucumbers to perfection. In the society of this governess, his aunt, and the old servant maid, Vassilyevna, Fedya spent four whole years.« (Turgenev 1906, pp. 68-69).

»She spoke modestly of Paris, of her travels, of Baden [...] taking off her gloves, with her smooth hands, redolent of soap à la guimauve, she showed how and where flounces were worn and ruches and lace and rosettes.« (Turgenev 1906, p. 245). The child's father then decided to undertake the upbringing and declared to his wife: »I want above all to make a man, un homme, of him' he said to Glafira Petrovna, 'and not only a man, but a Spartan.' Ivan Petrovitch began carrying out his intentions by putting his son in a Scotch kilt; the twelve-year-old boy had to go about with bare knees and a plume stuck in his Scotch cap. The Swedish lady was replaced by a young Swiss tutor, who was versed in gymnastics to perfection. Music, as a pursuit unworthy of a man, was discarded. The natural sciences, international law, mathematics, carpentry, after Jean- Jacques Rousseau's precept, and heraldry, to encourage chivalrous feelings, were what the future 'man' was to be occupied with. He was waked at four o'clock in the morning, splashed at once with cold water [...] Ivan Petrovitch on his side wrote him instructions in French in which he called him mon fils and addressed him as vous. In Russian Fedya called his father thou but did not dare to sit down in his presence. The 'system' dazed the boy, confused and cramped his intellect [...]« (Turgenev 1906, pp. 70-71).

2.2. Influence of Germany

Knowledge of foreign languages was considered to be part of education, so the nobles welcomed into their homes teachers who were native speakers of foreign languages. During the period of the reign of Peter I, German-speaking teachers were most valued. German governesses were said to be meticulous and practical in raising future mothers and wives who will be able to manage a household perfectly.

In his Writer's Diary for the year 1873, Dostoevsky named Turgenev's story Rudin to be the most German of all Turgenev's works. The main character Rudin loved German romantic poetry and philosophy and spent two years in Germany.

2.3. Influence of France

In the middle of the 18th century, during the reign of Catherine the Great (between 1762 and 1796), a fashion for everything French appeared. Catherine II. or Catherine the Great, a German on the Russian throne, was correspondance with Diderot, Voltaire and D'Alembert. She quickly learned Russian, read a lot and was fascinated by the European Enlightenment. (Wikipedia, Katarina Velika).

Many terms for luxury items used by aristocrats, such as jewelry, alcohol, elite clothing, in Russian are from French origin. As Olga Solodjankina writes, Paris was an aesthetic example not only for Russia, but also for Europe. As we learn on the culture.ru website, France became the leading model that the Russian nobility followed in their way of life, fashion, and literary taste. Every well-educated Russian nobleman had to master the French language. As Ljudmila Konšina points out, even at the beginning of the 19th century, children from noble families first learned to speak French, and only later Russian. It happened sometimes that nobles knew French better than Russian, as they thought in a foreign language (Konšina 2016).

In his story or novel Rudin, Turgenev distorted many quotations of the aristocrats' conversations in French and thereby wanted to show the poor knowledge of French in the Russian upper circles.

A typical Russian noblewoman writes Turgenev in his novel *Fathers and Sons*, Bazarov's mother Arina Vlasyeva» [...] in her youth she had been comely, and a player of the clavichord, and able to speak a little French« (Turgenev 1929, p. 165).





A typical Russian noblewoman, fascinated by France, was Varvara Pavlovna from Turgenev's *A House of Gentlefolk*: she spoke French, and from her trip to France she brought a maid from Paris with her to Russia. She wrote a long letter in French to her husband Lavrecki. She read only French books: "George Sand drove her to exasperation, Balzac she respected, but he wearied her; in Sue and Scribe she saw great knowledge of human nature, Dumas and Féval she adored. In her heart she preferred Paul de Kock to all of them, but of course she did not even mention his name. To tell the truth, literature had no great interest for her."

(Turgenev 1906, p. 258). "[...] till a late hour of the night, the lofty apartments of the house and even the garden re-echoed with the sound of music, singing, and lively French talk." (Turgenev 1906, p. 291).

Governesses and governors from France came to Russia. As Olga Solodjankina writes, governesses were mainly women. As we learn on the website culture.ru, French women were distinguished by good manners and good aesthetic taste, but they were quite beautiful, so Russian ladies were worried that they would not be a temptation for their husbands. For that reason noblewomen preferred to hire older governesses. Thus, in Tolstoy's novel *Anna Karenina*, the husband of the Oblon family had a relationship with a French woman who worked in their house them as an educator. As Olga Solodjankina points out, French governesses were "a typical figure of the Russian noble family before the reforms in Russia."

In the novel *A House of Gentlefolk*, Turgenev portrayed the character of Liza, who was raised by a teacher from Paris, Mlle Maureaux, until she was ten years old. This French woman was **a tiny wrinkled creature with little bird-like ways and a bird's intellect In her youth she had led a very dissipated life, but in old age she had only two passions left — gluttony and cards. When she had eaten her fill, and was neither playing cards nor chattering [...] Either as a result of her frivolous youth or of the air of Paris, which she had breathed from childhood, a kind of cheap universal scepticism had found its way into her, usually expressed by the words: tout qa dest des betises. She spoke ungrmmatically, but in a pure Parisian jargon, did not talk scandal and had no caprices — what more can one desire in a governess? Over Lisa she had little influence; all the stronger was the influence on her of her nurse, Agafya Vlasyevna.* (Turgenev 1906, pp. 213-214). In this novel, we cannot read the most beautiful words about governesses. They were cold towards the pupils, so they did not stay in the best memories.

A governess in Turgenev's novel Rudin, » [...] Mile Boncourt, in spite of her forty years' residence in Russia understood Russian with difficulty« (Turgenev p. 90).

Governesses also appear in 20th-century prose, such as, for example, in Pasternak's novel Doctor Zhivago, the governess of Countess Zhabrinska's daughter, Mlle Fleury. Pasternak describes her as a »Gray-haired, pink-cheeked, and dishevelled [...] She told long stories in her broken Russian, swallowing the ends of her words in the French manner [...]. (Pasternak 1958, p. 212).

2.4. Influence of England

The upper classes of the Russian nobility in the 18th and 19th centuries became enthousiastic also about England, writes Olga Solodjankina. Russian nobles thus began to admire the English way of life, English parks, artists and writers. Most young Russians did not know real life in England, but they were fascinated by everything English: things, fashion, food, literature; they were also attracted by the "free English spirit" or dandies, writes the aforementioned author. An excerpt from Turgenev's *A House of Gentlefolk* depicts such a Russian nobleman who admires England - it was Ivan Petrovič, Fedja Lavrecki's father: "Ivan Petrovitch returned to Russia aft Anglomaniac. His short-cropped hair, his starched shirt-front, his long-skirted pea-green overcoat with its multitude of capes, the sour expression of his face, something abrupt and at the same time indifferent in his behaviour, his way of speaking through his teeth, his sudden wooden laugh, the absence of smiles, exclusively





political or politico-economical conversation, his passion for roast beef and port wine — everything about him breathed, so to speak, of Great Britain.« (Turgenev 1906, p. 64).

A hero from Turgenev's Fathers and Sons, the nihilist Bazarov, mocks the obsession with England: »In my room there is an English washstand, yet the door will not shut! But such things (English washstands I mean) need to be encouraged; they represent 'progress.' « (Turgenev 1929, p. 21).

As it is written on the culture.ru portal, at the beginning of the 19th century, English governesses start to work as governesses in the aristocratic families. They were trying to raise Russian girls according to the example of English "Misses" and were never spoiling them. They were supposed to be typical "English ladies", such as the heroines of novels by Jane Austen, the Brontë sisters or Samuel Richardson, which English women liked to read. Often, English governesses were born in Russia and had no contact with England, and did not speak Russian well. Such a typical English governess, Miss Jackson, was portrayed by Pushkin in the story *The Squire's Daughter*.

An experience with the English governess was described in the memoirs of Sofia Vasilyevna Kovalevskaya, an excellent mathematician (born Korvin-Krukovskaya). As Lyudmila Saraskina writes, fifteen-year-old Sofia was in love with Dostoevsky, who read and praised her children's poems. Dostoevsky corresponded with her older sister Ana for almost a year, and he even desired to marry her, but she rejected him (Saraskina 2021, pp. 352-353). Kovalevskaya writes (quoting from: Solodjankina) that the English governess's daily schedule was quite strict. Sofia's brother had a Polish governor who was not as strict as the English lady. The girl got up early in the morning and after morning tea, music and other subjects were on the schedule. After the midday breakfast, there was an hour and a half long walk with the teacher, if it was warmer than ten degrees. After lunch, the pupil prepared for the next day's lessons and was all the time with the governess, who did not let her join the rest of the family in the afternoon meetings. According to Kovalevskaya's memoirs, the governess was strict and cold towards her. She focused only on raising the girl, because she was quite lonely in Russia, where she never really felt at home.

2.5. Russian teachers

According to culture.ru, in the 19th century, Russian pedagogues who had graduated from teacher training schools also began to be hired. Girls were prepared for this profession in special institutes. Mostly, those girls came from poor families and could earn good money as home teachers. They lived with families where they were employed, and their position was higher than that of servants, as they were educated and free women. Their job was to accompany their pupils all the time, take care of their development, bring them up and teach good behavior. They were also travelling with them and were even choosing books, which should be suitable for them. The governesses themselves were a model of behavior for their pupils and were constantly reminding them of the right behaviour.

In the Turgenev's novel *On The Eve*, there is a governess of the main character Elena. She » [...] was a Russian, the daughter of a ruined bribe-taker, graduate of a Government Institute [...] This governess had been very fond of literature, and was herself in the habit of scribbling bad verses; she imbued Elena with a taste for reading [...]« (Turgenev pp.46-47).

A famous example of a Russian governess in the literature is Dunya, a sister of Rodion Raskolnikov from Dostoevsky's *Crime and Punishment*.

Many governesses and nannies appear in Chekhov's novels and plays. For example, in the novella *The Duel:* »Marya Konstantinovna, who had been a governess in aristocratic families and who was an authority on social matters, said: "Oh yes! Would you believe me, my dear, at the





Garatynskys' I was expected to dress for lunch as well as for dinner, so that, like an actress, I received a special allowance for my wardrobe in addition to my salary." (Chekhov 1922, p. 37).

In Pasternak's famous novel *Doctor Zhivago*, Lara becomes an educator in the family of a great businessman Kologrivov. She has been their daughter Lipa's teacher for more than three years, and her parents and sister love her (Pasternak 1986, p. 115).

In the story *Nevsky Prospect*, Nikolai V. Gogol described how educators walked with their pupils on this famous street of St. Petersburg: »*Governesses*, pale English misses and rosycheeked Slav maidens, walk in stately fashion behind their lightsome, fidgety girls, commanding them to hold their heads high and backs straight; in other words, at this time Nevsky Prospect is a pedagogical thoroughfare.« (Gogol 1995, p. 5).

3. Russian nannies

3.1. Roles of nannies

In contrast to the teachers, governors and governesses, mostly foreigners, who were often strict and cold towards the children, the nannies had a much greater influence on them. There were also men who raised young children. They were called ДЯДЬКИ (uncles) in Russian. Nannies were benevolent, warm, caring, loving, often wise and religious Russian women who sincerely loved children and were faithful to their families.

The nannies also introduced national culture to children. With the stories and fairy tales they were telling, they also influenced the literary work of many famous Russian writers and poets.

Usually, the nannies were poor peasant women who were very dedicated to their work, with which they could live without financial worries for their entire lives, since the nannies stayed with the family even after the children grew up. They could become nannies for the children of the next generation. Nannies were not servants, but were treated as part of the family.

There is a well-known true story about Fyodor Mikhailovich Dostoevsky's nanny, Alyona Frolovna, who wanted to donate all her savings when the Dostoevsky family's possessions burned down. Dostoevsky also remembered her as a teller of wonderful stories (Saraskina 2020, p. 59).

According to Marina Melija, nannies were a kind of intermediaries between children and parents - the parents brought up the children only with their presence; about everything else, education and health of the children, took care the nannies, who watched over them every moment of the day. That is why nannies were respected and appreciated by everyone. Children were thus more attached to their nannies than to their own mothers, as the nannies showed them sincere love, attention and care. Being a nanny was a vocation in the true sense of the word, or rather, for poor peasant women, a "gift of fate", as Melija writes. If a nanny had children of her own, they were like brothers and sisters of the other children, of whom she was taking care. This was the case, according to the aforementioned author, for example, for the children of the nanny of Tsar Nicholas I.

As Marina Melija writes, not only representatives of the elite had nannies, but also children in other families who could afford it.

In larger cities, there were even specialized four-year schools for nannies. In the 20th century, during the revolution of 1917, most nannies emigrated together with the families in which they were working, writes Marina Melija. Philosopher Nikolai Berdyaev wrote that nannies in Russia even represented a special social class (quoting from: Melija). The





tradition of nannies in Russia continues into the 21st century; in many well-to-do families nannies help to take care of the children.

3.2. Nannies in literary depictions of some Russian classics

3.2.1. Nanny of Alexander Sergeyevich Pushkin

The most famous nanny in Russian literature is certainly Alexander Sergeyevich Pushkin's nanny, Arina Rodionovna. The poet mentioned his beloved nanny several times in his work; the poem 'Nanny' is a literary memorial to a dear person from the poet's childhood. He mentions his nanny with nostalgia in the poem 'Winter Evening', where he calls her "Sweetheart of my youthful springtime,"

Thou true-souled companion dear« (Pushkin, poets.org).

In Pushkin's novel in verse *Eugene Onegin*, the old nanny Filipyevna is Tatyana's confidant and advisor. Tatyana wants advice from her nanny's life experience; the nanny helps and supports her in difficult moments. In his work, Pushkin shows the spiritual connection between a person from a modest family (nanny) and an aristocrat.

3.2.2 Nannies in the work of Anton Pavlovich Chekhov

From the literary work of Anton Pavlovich Chekhov, we can cite an example of a nanny from his story (according to some definitions, the only novel) *The Shooting Party*. Chekhov's dramas often feature nannies or governesses: in *The Cherry Orchard* there is a governess Charlotte Ivanovna, in *Three Sisters* a nurse, an 80-year-old woman called Anfisa, in *Uncle Vanya* there is an old nurse Marina. In *Three Sisters*, Natasha wants to banish the old nanny, saying she is useless, and treats her harshly.

3.2.3. Nannies in Turgenev's A House of Gentlefolk

Turgenev in the novel A House of Gentlefolk describes life of a nanny Agafya Vlasyevna. "»This woman's story was remarkable. « (Turgenev 1906, p. 214). Agafya was Lisa's nanny, who took care of her from the age of five to eight and had a significant influence on the girl. »Lisa was at first frightened by the austere and serious face of her new nurse; but she soon grew used to her and began to love her.« [...] She was Afraid of her father; her feeling towards her mother was undefinable, she was not afraid of her, nor was she demonstrative to her; but she was not demonstrative even towards Agafya, though she was the only person she loved. Agafya never left her. It was curious to see them together. Agafya, all in black, with a dark handkerchief on her head, her face thin and transparent as wax, but still beautiful and expressive, would be sitting upright, knitting a stocking; Lisa would sit at her feet in a little arm-chair, also busied over some kind of work, and seriously raising her clear eyes, listening to what Agafya was relating to her. And Agafya did not tell her stories; but in even measured accents she would narrate the life of the Holy Virgin, the lives of hermits, saints, and holy men. She would tell Lisa how the holy men lived in deserts, how they were saved, how they suffered hunger and want, and did not fear kings, but confessed Christ [...] Lisa listened to her, and the image of the all-seeing, all-knowing God penetrated with a kind of sweet power into her very soul, filling it with pure and reverent awe; but Christ became for her something near, well-known, almost familiar. Agafya taught her to pray [...]« (Turgenev 1906, pp. 217-218). When three years later, the nanny Agafya was replaced by a French governess, » [...] the frivolous Frenchwoman, with her cold ways and exclamation, tout ça c'est des bêtises could never dislodge her dear nurse from Lisa's heart; the seeds that had been dropped into it had become too deeply rooted. Besides, though Agafya no longer waited on Lisa, she was still in the house and often saw her charge, who believed in her as before.« (Turgenev 1906, p. 219). Later, the old nanny went on a pilgrimage and, according to some rumors, stayed in a monastery. At the end of the novel, Lisa also goes to a monastery and becomes a nun.





The nannies influenced the children with the fairy tales and folk tales they were telling them. An example of such a nanny was Ivan Aleksandrovich Goncharov's nanny, Ana Mihailovna. He described his memories of her in a famous novel *Oblomov*. A characteristic of the hero Oblomov is that he spent his whole life behind the stove and did not work. "On a long winter's evening he was pressing close to his nurse, and she was whispering of some unknown country where neither cold nor darkness were known, and where miracles took place, and where rivers ran honey and milk, and where no one did anything the year round [...] Goncharov 1915, p. 109). So artfully did the nurse or tradition eliminate from the story all resemblance to everyday life that the boy's keen intellect and imagination, fired by the device, remained enthralled until, in later years, he had come even to man's estate. As a matter of fact, the tale which the nurse thus lovingly related was the legend of the fool Emel — that clever, biting satire upon our forefathers and, it may be, also upon ourselves. [...] (Goncharov 1915, p. 110). Russian folk tale about the fool Emel, who is extremely lazy, but managed to catch a pike, which gave him magical power: everything he commands is fulfilled. (Wikipedia, At the Pike's Behest).

To the same story had his father and his grandfather listened as, shaped according to the stereotyped version current throughout antiquity, it had issued from the mouths of male and female nurses through the long course of ages and of generations. (Goncharov 1915, p. 111). With a simplicity, yet a sincerity, worthy of Homer, with a lifelike similitude of detail and a power of clear-cut relief that might have vied with the great Greek poet's, she fired the boy's intellect and imagination to a love for that Iliad which our heroes founded during the dim ages when man had not yet become adapted to the sundry perils and mysteries of nature and of life — when still he trembled before werewolves and wood demons, and sought refuge with protectors like Alesha Popovitch (one of the three brave knights from Russian folklore (Wikipedia, Alyosha Popovich, At the Pike's Behest.) from the calamities which surrounded him — when air and water and forest and field alike were under the continued sway of the supernatural. (Goncharov 1915, pp. 11-112). as little Oblomov listened to his nurse's legends concerning the Golden Fleece, the great Cassowary Bird, and the cells and secret dungeons of the Enchanted Castle, he became more and more fired to the idea that he too was destined to become the hero of doughty deeds. Tale succeeded to tale, and the nurse -pursued her narrative with such ardour and vividness and attractiveness of description that at times her breath choked in her throat. For she too half-believed the legends which she related; so that, during the telling of them, her eyes would shoot fire, her head shake with excitement, and her voice attain an unwonted pitch [...]« (Goncharov 1915, pp.114-115).

3.2.5. Similarities and differences of nannies in Russian literature

Literary depictions of nannies in the quoted cases have a different function. For the most part, the characters of nannies are personifications of goodness, love and loyalty, as is evident in Pushkin, who even dedicated a poem to his nanny. Nanny Filipyevna in *Eugene Onegin* is a confidant and moral support for the heroine in difficult moments.

In Chekhov's drama *Three Sisters*, the nanny is something left over from the past and is no longer necessary in the present, as the heroine wants to kick the old woman out of the house.

In Turgenev's *A House of Gentlefolk*, the nanny is a moral example that even seals the life path of the heroine.

In Goncharov's novel *Oblomov*, nanny connects different generations with the help of folk tradition and influences the fate of the hero, to whom she appears in his dreams as a precious memory from his childhood.





3.2.6. Examples of depictions of nannies in sculpture and painting

In addition to literature, nannies were also depicted in other arts. Famous are monuments to Pushkin's nanny Arina Rodionovna, there are several in various Russian cities, and new ones appear from time to time. Thus, as we read in the online newspaper "Родина" (Cygankova, 2024), in October 2024, a new monument to Arina Rodionovna was erected in the Pushkin Mountains in the Pskov region (these are three estates, Mikhaylovskoye, Trigorskoye and Petrovskoye, which are connected with the life of the great poet, and the Svyatogorsky Monastery, where he is buried.

Nowadays, there is a museum - reserve of Aleksandr Sergeyevich Pushkin (source: Guzeva, 2020). Arina Rodionovna in this statue is sitting on a bench, with a cat next to her.

In painting, there is a well-known portrait of the Russian impresario in Paris, Sergei Diaghilev, with his nanny. This portrait was painted in 1906 by Léon Bakst, who created scenes and costumes for Diaghilev's Ballets Russes. Léon Bakst (1866-1924), Russian Jewish painter who studied in Paris between 1893 and 1896. After returning to Russia, he was a member of the "World of Art" group, which published a newspaper of the same name with the aim of introducing new trends in art. In 1909, Bakst returned to Paris, where he began creating exotic, colorful sets and costumes for Diaghilev's Ballets Russes (cited from: Kuiper). In the foreground of the picture is a portrait of Sergei Diaghilev, and behind him in the background on the left is his old nanny (see Virtual Russian Museum).

Among the paintings of governesses in Russian art there is a well-known painting by Vasily Perov 'Arrival of a New Governess in a Merchant House' from the Moscow's Tretyakov Gallery.

4. Conclusion

Nannies and governesses had an important role in the upbringing and education of the children of nobility in Russia in the 18th and 19th centuries. While the governors and governesses were generally foreigners, mostly strict and unyielding, the nannies were Russian women who were distinguished by kindness, indulgence, love for children, wisdom and a sense of belonging to their families. The task of governesses was primarily to teach a foreign language, their mother tongue, and to remind the children of the rules of etiquette. The nannies, on the other hand, had an important influence on children, because they were like their second mothers, who took upon themselves parental responsibilities and raised kids with love, indulgence and benevolence. They were able to tell wonderful stories.

Governesses and nannies were often mentioned by writers and poets in their works. The characters of the nannies, with their love, loyalty and self-sacrifice, reflect the character traits of real nannies, who played an important role in shaping the characters of future writers and poets.

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Reflection

On the Ethics in Monteverdi's Coronation of Poppaea

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Abstract:

We reflect on the scope and design of the opera Coronation of Poppaea composed by Claudio Monteverdi according to the libretto of Francesco Busonello that was first performed in 1654 during the carnival period in Venice. Matching timeless music, the opera is distinguished also by poetic lyrics on Fortune, Virtue and, in particular, Love. Surprisingly, the story is related to historic persons – Roman empress Sabina Poppaea the Younger (Poppaea) and emperor Nero Claudius Caesar Augustus Germanicus (Nero). As reports on Nero indicate multiple murders, including murders of those that were closest to him, the libretto rises questions on the necessity of Virtu in achieving happiness. Our considerations indicate that the main characters in the opera are ethically sound, however, the artistic changes have made them essentially different from the underlying real persons.

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Keywords: Baroque opera; Roman history; Venice carnival; Nero Claudius Caesar Augustus Germanicus; Sabina Poppaea the Younger; Commedia del Arte





1. Historical considerations

In composing the evidences on Poppaea and Nero, we rely on writings of historians Tacitus (56–cca 120) (Tacitus, 115–120), Suetonius (cca 69–122), (Suetonius, 121) and Lucius Cassius Dio (165–235):

1.1. On Nero

For most of this period, Roman emperors were chosen according to hereditary rule: they were born in the right family. The empire started through violence and depended on force. Civil war had brought Caesar to power. Once emperor and without an heir, he had adopted Augustus and thereby established hereditary succession. However, the society was harsh and emperors could only survive if their people believed they could outforce anyone and everyone.

Nero Claudius Caesar Augustus Germanicus (Nero) was born in 37 to Julia Agrippina (Agrippina the Younger) and Gnaeus Domitius Ahenobarbus. He was three when his father died. Agrippina the Younger became the third wife of emperor Tiberius Claudius Nero Germanicus (Claudius) in 49 and Claudius adopted Nero who became Claudius' eldest son and the heir to the empire. Annaeus Seneca, a member of the senate, was appointed to educate him. After the death of Claudius in 54, Nero became emperor. At the beginning, he ruled under a strong influence of his mother but with time, he became an absolutist and a tyrant. He was overthrown by Servius Sulpicius Galba and Marcus Salvius Otho and committed suicide at the age of 30 in 68.

Agrippina the Younger was the daughter, the sister, the wife and the mother of emperors. She was displaying her position as an equal partner in the power gained by her ancestors. Tacitus, Annals, Book 12: "Everything was subject to the control of a woman. Rome was now enslaved by a controlling and almost masculine dominance. In public, Agrippina showed a serious, often arrogant face; in private, there was no sign of immorality, unless it helped her in her search for power; she had an enormous desire for money which was excused with the reason that money was a means to power."

1.2. On Nero's marriages

A marriage was arranged between Nero (then 16) and Claudius' daughter Claudia Octavia (then 13) in 53. Since the two were legally brother and sister, Octavia had to be adopted into another family before Nero could marry her. It was implied that this marriage was never consummated and there were no children. By arranging the marriage between Claudia Octavia and Nero, Agrippina and Claudius gave an advantage to Claudius' daughter over his son.

In about 58, Nero began an affair with the aristocratic woman Poppaea Sabina (her parents were Poppaea Sabina the Elder and Titus Ollius) who was 7 years older than him and had been married earlier to Rufrius Crispius and then to M. Salvus Otho. Nero's affair with Poppaea raised the question of divorce from Octavia. Agrippina's determination to preserve the dynastic marriage that she and Claudius had arranged induced struggles between her and Nero. Nero attempted to assassinate his mother several times and when that failed, he had her executed for treason.

When Poppaea Sabina got pregnant, Nero divorced Octavia with the reason that she was unable to give him an heir. Octavia was sent to Campania, and twelve days later Nero married Poppaea (Saetonius). However, Octavia remained a threat as the previous emperor's daughter. Admiral Anicetus, who had collaborated also in Agrippina the Younger's murder, confessed to an affair with Octavia and she was suffocated and beheaded in 62.

Poppaea and Nero's daughter Claudia Augusta was born in 63, but she died at the age of only four months and Nero deified her. In 65, Poppaea became pregnant again, but miscarried and died thereafter at the age of 35. "Poppaea died from casual outburst of rage in her husband, who felled her with a kick when she was pregnant. Her body was not consumed by fire according to Roman usage, but after the custom of foreign princes was filled with fragrant spices and embalmed, and then consigned to the sepulchre of the Julii. She had, however, a public funeral, and Nero himself from the rostra eulogized her beauty,





her lot in having been the mother of a deified child, and fortune's other gifts, as though they were virtues". (Tacitus). Poppaea Sabina also perished at this time through an act of Nero's; either accidentally or intentionally he had leaped upon her with his feet while she was pregnant. (Cassius Dio, Epitome of Book LXII.27.4) After the close of the festival, Poppaea met her end through a chance outburst of anger on the part of her husband, who felled her with a kick during pregnancy. (Tacitus, Annals XVI.6.1).

Since Nero still had no heir, he promptly set about finding another wife after Poppaea's death. Suetonius reports that he considered marrying Claudia Antonia, the daughter of Claudius and his second wife Aelia Paetina and half-sister of Octavia Claudia that he had divorced and executed three years earlier. When Antonia refused this proposal, Nero had her executed. Nero, meanwhile, contracted his third marriage in 65, to Statilia Messalina. Messalina had been married to the consul Vestinus, whom Nero executed for involvement in Piso's coup. This marriage also produced no children.

1.4. Reports on Nero

Compared to today's rich records, the written evidences on Nero and his contemporaries are scarce. Historians Tacitus (56–cca 120), who was for some years the only Nero's contemporary, and Suetonius (cca 69–122) could still experience the impact of Nero while Lucius Cassius Dio (165–235) lived a century after the death of Nero.

At first, Suetonius exposes positive facts on Nero: As a boy, Nero was interested in almost all the literary and artistic subjects; but his mother kept him from philosophy, warning him that it was unsuitable for a future emperor. Seneca, his teacher, stopped him from reading works by the earliest orators, in order to make his admiration for Seneca himself last longer. In the game of Troy at the circus games, he gave a very good performance which was well-received. He happily turned his attention to poetry and wrote poems quite easily, and he did not, as some think, publish others' works as his own. There have come into my hands notebooks and papers with some well-known poems of his, written in his own handwriting so that it is clear that they were not copied or noted down when someone was speaking, but rather noted down by him while he himself was thinking and composing. There are many examples of words and phrases rubbed out or written over and added to or written above. He also had a considerable interest in painting and sculpture.

He began his reign with an appearance of family loyalty and duty. At a splendid and expensive funeral, he spoke in praise of Claudius, and then deified him. He gave the greatest honours to the memory of his father Domitius. He let his mother manage everything, public and private. On the first day of his reign, he even gave to the tribune on guard-duty the password "The Best of Mothers", and afterwards he often rode with her through the streets in her litter.

Suetonius then moves onto a devastating attack on Nero's character: There is a story that Seneca dreamt that he was teaching Gaius Caesar [Caligula], and Nero soon gave proof that the dream was real, when he showed his natural cruelty early on in his life. Because his brother Britannicus had, after his adoption, greeted him as usual as Ahenobarbus, he tried to prove to Claudius that Britannicus was not his son at all. In addition, when his aunt Lepida was on trial, he was a witness against her, to please his mother, who was making every effort to have her found guilty.

He corrupted free-born boys and seduced married women; he even forced himself on the Vestal Virgin Rubria. He almost married the freedwoman Acte, after bribing some ex-consuls to swear on oath that she was of royal birth. He castrated the boy Sporus and actually tried to turn him into a woman; then Nero held a marriage ceremony in the usual way, with a dowry and a bridal veil, took him home, accompanied by a crowd of followers, and treated him as his wife. He took this Sporus, all dressed up like an empress and carried along in a litter, to the assemblies and markets of Greece; soon afterwards, he took him around the image-market in Rome, repeatedly kissing him. No one doubted that he wanted sexual relations with his own mother, and was prevented by her enemies, afraid that this ruthless and powerful woman would become too strong with this sort of special favour. What added to this opinion was that he included among his mistresses a certain prostitute who they said looked very much like Agrippina. They also said that, whenever he rode in a litter with his mother, the stains on his clothes afterwards proved that he had





indulged in incest with her. Claudius was the first member of Nero's family to be murdered; Nero may not have been the one to arrange it, but he certainly knew about it. He did not hide the fact because later he used to praise mushrooms (the poison was administered to Claudius in a dish of mushrooms), as "the food of the gods", in the words of the Greek proverb. In fact, after Claudius's death, he made fun of him with every insult he could accusing him of stupidity and cruelty; he used to joke that Claudius had stopped "being the idiot" among men, by lengthening the first syllable of the word, and he treated many of his decrees and acts as worthless as if they had been done by a crazy fool; as a final humiliation of Claudius, he neglected his tomb providing it with nothing except a low-walled enclosure. Nero tried to kill his half-brother Britannicus with poison; he saw him as a rival in singing, since his voice was pleasanter than his own, but an equal motive for the murder was the fear that he might at some point gain more influence and support because men remembered his father. He got the poison from a certain well-known poisoner, Locusta. When the poison worked more slowly than expected, since it only gave Britannicus a serious stomach-ache, he had the woman summoned and he beat her himself. He accused her of giving him a remedy instead of the poison. Her excuse was that she had given Britannicus less in order to keep the crime secret and save Nero from being suspected. Then in his own room, he forced her to cook up the fastest possible potion to take effect instantly. He tested it on a kid-goat, which took 5 hours to die; he had her cook it again so that it was stronger still, and he gave it to a pig. The pig dropped dead at once. He ordered the poison to be taken to the dining-room and to be given to Britannicus who dined with him that day. The boy fell dead the moment he tasted it, but Nero lied to his guests saying that he had been attacked by an illness, which he had suffered from for some time. The very next day Nero had Britannicus quickly buried in a simple funeral in the middle of a thunder storm. He gave Locusta a pardon and large estates as a reward for her good work. He also provided her with some pupils. To the murder of his mother, he added the killing of his aunt, Domitia. He visited her when she was ill with severe constipation. She was stroking his beard - he was already a grown-up - and by chance said kindly: "As soon as I receive this, I want to die." He turned to those next to him and said, apparently as a joke, that he would shave it off immediately. He ordered the doctors to give the ill woman a dose of medicine which was too big in order to empty her stomach. He took over her property before she was actually dead and cancelled her will, so that he lost nothing. There was no sort of family relations which he did not damage by one crime or another. He killed Antonia, daughter of Claudius, when she refused his offer of marriage after Poppaea's death, on the charge that she was organising a plot against him. Likewise, he dealt with all others in any way related to him by family or by marriage. He insisted that Rufrius Crispinus, still only a boy, his stepson and the child of Poppaea, was drowned in the sea by his own slaves while he was fishing because they said he pretended to be a general and played being an emperor. He banished his nurse's son Tuscus, because, during his governorship of Egypt, he had bathed in the bathhouse built for Nero's visit to the province. He forced his tutor Seneca to commit suicide. He sent poison to Burrus, the Praetorian Prefect, having promised to send medicine for his throat. He used poison, either in their food or their drinks, to get rid of the old, rich freedmen who supported his adoption and his accession, and given their guidance when he was emperor (Suetonius). Moreover, publicly blaming Christians and forcing them to be fed to animals, ignited while alive, or nailed to a crucifix was another misstep. Mockery of every sort was added to their deaths. Covered with the skins of beasts, they were torn apart by dogs and perished, or were nailed to crosses, or were doomed to the flames and burnt, to serve as a nightly illumination, when daylight had expired. Nero offered his gardens for the spectacle, and was exhibiting a show in the circus, while he mingled with the people in the dress of a charioteer or stood aloft on a car (Tacitus, Annales, book 15, chapter 44). Christians were not a popular group in Rome at that time. Yet, the treatment and torture of the religious group was so horrific that even the Romans who believed them to be responsible thought it extreme.





1.5. Evidences on Poppaea

Tacitus: Poppaea would reproach the emperor with incessant vituperation and sometimes call him in jest a mere ward who was under the rule of others, and was so far from having an empire that he had not even his liberty. "Why," she asked, "was her marriage put off? Was it, forsooth, her beauty and her ancestors, with their triumphal honours, that failed to please, or her being a mother, and her sincere heart? No; the fear was that as a wife at least she would divulge the wrongs of the Senate, and the wrath of the people at the arrogance and rapacity of his mother. If the only daughter-in-law Agrippina could bear was one who wished evil to her son, let her be restored to her union with Otho. She would go anywhere in the world, where she might hear of the insults heaped on the emperor, rather than witness them, and be also involved in his perils." Such and similar complaints, rendered impressive by tears and by the cunning of an adulteress, no one checked, as all longed to see the mother's power broken, while not a person believed that the son's hatred would steel his heart to her murder (Tacitus, The Annals, 14.1).

Next to records that are indicating Nero's guilt to murder, in particular those that were his close relatives, there are records of Nero's sweetness: "Wearing Augustus's sun-hat amongst our courtyard plants I sit and eat and drink with you, Poppaea. Together we grow old and fat." (Lehmann, 1981).

2. The opera Coronation of Poppaea

Poppaea and Nero were chosen to be protagonists of the Monteverdi/Bussonello's opera with the focus on love. Claudio Monteverdi (1567-1643) was an Italian composer, choirmaster and string player. He composed sacred and secular music, introduced novelties in the opera and is considered a crucial transitional figure between the Renaissance and Baroque periods of music history. Francesco Bussonello (1598-1659) from Venice was a lawyer by profession, and a renowned writer of librettos. The Coronation of Poppaea was first performed during the 1643 carnival season at the Teatro Santi Giovanni e Paolo in Venice, where Monteverdi was the choirmaster at the Basilica di San Marco. The Coronation of Poppaea was one of the first operas to use historic events and people. Eight years later (in 1651), the opera was put on stage in Naples, Italy, and was then revived after more than 200 years. The original manuscript of the score was not found; two surviving copies from the 1650s show significant differences between them. However, this can be expected as the scores had to be adjusted to the cast. How much of the music is actually Monteverdi's, and how much of others, is a matter of dispute. The score features 28 singing characters. The original Venetian production may have used role-doubling, allowing the opera to be staged with no more than 11 singers: two female sopranos, three male sopranos (castratos), two contraltos (castratos), two tenors and two basses.

2.1. The story and the characters in the opera Coronation of Poppaea

While the libretto is based on actual historic persons, there are artistic differences introduced by the writer: The prologue sets the philosophical question of the utmost importance: is it Fortune, Virtue or Love? From experience, one could guess that the answer to this question is that all three are a prerequisite, but not necessarily sufficient for success. Yet, the writer inclines in the story that Love is the most important one. The scope of the story is Poppaea's ambition to become empress. Both Nero and Otho love Poppaea. Poppaea responds to Nero as he can make her empress which Otho cannot. Poppaea and Nero become lovers at the outrage of Otho and Nero's wife Octavia who feels humiliated and betrayed - for herself and for what she represents as the daughter of the emperor and a part of the aristocracy. Seneca, the moral pilar, and those close to Octavia also feel offended. Octavia urges Seneca to stand against Nero's adultery. Seneca challenges Nero and their discourse ends by Nero forcing Seneca to suicide. Then Octavia bids Otho to murder Poppaea. Otho approaches Poppaea asleep in the garden, but Love prevents his deed. Having learned about the complot, Nero banishes Otho and Octavia from Rome and crowns Poppaea empress. Overwhelmed with success and happiness, Poppaea and Nero celebrate the victory of Love.





2.2. Differences between the story and the evidences

There is a difference in the scope with respect to reality; to our best knowledge, there are no historical facts of Poppaea's ambition to the crown. The above arguments given by Tacitus indicate that Poppaea was complaining about Nero being manipulated by Agrippina the Younger and that after she was forced out of marriage with Otho she did not wish to be Nero's mistress but expected that he would marry her. This is essentially different from having an ambition to the crown, as it points to the interest of Nero to be with Poppaea as the main driving element. Tacitus reports that Poppaea would rather be reunited with Otho than be Nero's mistress, pointing to Virtu instead of Fortune or Love. In the story, Poppaea and Nero love each other; the libretto presents beautiful poetic expressions of their devotion. It seems that in reality it would be probable for Nero to love Poppaea, but not so convincing that she loved him. The above historical evidences indicate that Nero was taking advantage of his power to get what he wished regardless of others' feelings. The evidence on the deeds and opinions of Poppaea is scarce. There are records from both Tacitus and Suetonius that it was likely that Nero killed Poppaea and Poppaea's unborn child and also Poppaea's child from the previous marriage. Therefore, it seems unlikely that she could love someone with such attitude which was evidently not instantaneous. Although some historians think that there were prejudices about Nero, there are not many positive facts that would prove his gentleness and kindness. In the story, Claudia Octavia complots a murder of Poppaea. Moreover, she forces Otho to perform the deed. In reality, there is evidence on such intentions or actions from Claudia Octavia. In the story, the ethical bias is on instrumental marriages which were made for political motifs on the account of human misery. Instead, it seems right that Poppaea and Nero, who were fortunate to find love, come together in marriage. Also, in the story, Octavia, in spite of her complot plan, stays alive due to the generosity of Nero and we are left thinking that the marriage between Poppaea and Nero is a happy one.

The idea to worship Love linked to someone for whom there is a reasonable doubt that he killed among others also his mother, brother, sister, mentor, and ultimately his love and unborn child seems so odd and bold that one may pose a question whether it was meant to be a parody. Deriving from the records on the carnival festivities in Venice, where Coronation of Poppaea was first performed:

"Audience members sometimes struggled to follow the plot or identify with the characters, which led to the introduction of comic scenes. These originated in the Italian semi-improvised form of the commedia dell'arte (comedy of professional artists) and provided some light relief. Comic characters who usually held a low social position were a means to appeal to a wider public who could better identify with them. Comic arias or duets appeared, an early example being Monteverdi's 1642 opera, L'in-coronazione di Poppea (The Coronation of Poppea), in which the main character, Damigella, shares a comic love duet with her page, Valletto" (Maynes, 2024).

3. Conclusion

By the changes introduced in the libretto of the Coronation of Poppaea, the story on love between Poppaea and Nero seems ethically rather sound. Besides focusing on Love, it is supported also by Fortune and Virtu. However, it essentially does not have much in common with what happened to the real persons who most likely lacked all three elements facing loveless relationships and premature death for political reasons and glory.

There are many lucid timeless observations found in the libretto such as:

"One might add that our prince robs everyone to line the pockets of a few. The innocent suffer while criminals are doing very nicely."

"To have dealings with princes is perilous. Love and hate counts nothing with them: their emotions are governed by pure self-interest."

"Oh, unhappy female sex. Born free by nature and the will of heaven marriage chains us up in slavery."







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Research

Artmaking as Bravery: A Virtue-Based Perspective

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Abstract:

In this article we will reflect on the relation of art to ethics. The matter has been investigated by numerous thinkers since Plato, and a few distinct philosophical positions have been codified, ranging, as is known, from the refusal of any moral evaluation of art (autonomism) to the position that moral considerations are in fact part and parcel of any aesthetic evaluation of art (moralism). We will, however, focus on some ethical aspects of artmaking itself, not on the moral assessment of artworks and of their impact. Our considerations will be informed by the approach to ethics known as virtue ethics, which means avoiding a narrow conception of ethics, especially a legalistic one that interprets morality as compliance with rules or commandments, and returning to an Aristotelian conception of the virtues as excellent character traits expressed in action. Furthermore, we will incorporate some contributions from the emergent field of virtue aesthetics. We will discuss the role of the virtue of courage in artmaking and conclude that virtue ethics and virtue aesthetics offer a promising new perspective on art and ethics.

Keywords: Virtue ethics; Virtue aesthetics; Artmaking; Aristotle; Moral evaluation of art; Art and fear

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1. Introduction

1.1. Art and morality

The question of the relation of art to morality has been debated for a very long time, at least since Plato's famous stern judgment over the arts in the Republic. Two opposite "strong" positions can be identified in this regard: moralism, that is the view that moral evaluations prevail over or subsume aesthetic considerations; and autonomism, which rejects any evaluation of art that is not purely aesthetic. In recent times, however, the main question seems to be not whether moral evaluations of art are appropriate, but whether they contribute to the assessment of the aesthetic value of the artwork (Peek, 2025).

1.2. Radical moralism vs. radical autonomism

Pure or radical moralism will reduce the aesthetic assessment of art to its moral assessment; it will exclusively concern itself with an artwork's moral quality and may therefore become unable to distinguish art from other cultural products (Peek, 2025). Radical autonomism (aestheticism) claims that art may be assessed only on the basis of merely aesthetic and formal standards; but to defend this approach, to defend the view that aesthetic assessment is superior to any other kind of assessment, and indeed self-sufficient, the proponents of autonomism will need to resort to moral arguments; they will, for example, exalt tolerance, curiosity, openness, individualism; eventually, Ella Peek notes: "Aestheticism, in its most extreme form, could almost be seen as a version of radical moralism"! (Peek, 2025).

1.3. Moderate positions

A few "weak" positions have then emerged, like moderate autonomism, moderate moralism, and others, which seem in practice more helpful than the intransigent "strong" positions. Moderate autonomism and moderate moralism move away from the assumption that, in the evaluation of artworks, ethical and aesthetic judgments may never be completely disentangled; only, the interpreter will tend to privilege, respectively, aesthetic or moral considerations. Moderate positions may be preferable to the radical or "strong" positions; still, they are concerned with the evaluation of artworks and of their likely moral influence on the viewer or on the society; they have little to say about the production of artworks. Furthermore, the moral considerations that we may make about art in a similar perspective are not different in nature from the moral considerations that we may make about any other cultural product that carries a morally relevant message, but is not art. We might benefit from a different approach to the ethical evaluation of art that recognizes the specific character of art as opposed to other vehicles of morally relevant messages, and that escapes a stern, fault-finding approach to art.

2. Virtue ethics

2.1. What is virtue ethics?

Virtue ethics is an approach to moral philosophy that emerged in the second half of the 20th century out of dissatisfaction with the then dominant approaches of deontology and consequentialism. The seminal text is generally considered to be the Oxford philosopher Elizabeth Anscombe's paper "Modern Moral Philosophy" (Anscombe, 1958). Anscombe's main reason for discontent with the prevalent schools of thought was their legalistic character, their emphasis on laws, rules, and obligations, which not only determined a rigid, stifling approach to morality, but was also unsuitable to a secularized society that had lost sight of a supreme legislator. Among other key philosophers in the development of the new approach to ethics are Bernard Williams (Williams, 1985) and Alasdair MacIntyre (MacIntyre, 1985). Interestingly, Bernard Williams differentiated between morality and ethics in the sense that morality is represented by Kant's duty- and obligation-based approach to moral philosophy, while ethics is a broader concern akin to the Ancient Greeks' quest for the good life (Williams, 1985). It is in a similar, broader understanding of "ethics" that I submit my reflections (about virtue ethics see Slote, 2013; Russell, 2013).





2.2. Virtue

What does "virtue" mean, in this context? For Aristotle, virtues are desirable character traits that move a person closer to their ultimate purpose, their *telos*. Virtues are excellent character traits, which manifest themselves in admirable behavior, whereby a person demonstrates control over a particular emotion in a way that is appropriate to the concrete circumstances and is therefore able to act in accordance with reason. Virtues, for Aristotle are situated in a middle area between two corresponding excesses; for example, in relation to the emotion of fear, the virtue of courage stands somewhere between an excess of fear (the vice of cowardice) and an excess of confidence (rashness or temerity). Virtue can also be the appropriate behavior in relation to situations, for example, in relation to giving money, the virtue of generosity is somewhere in the middle between deficiency (stinginess) and excess (profligacy). It should be noted that Aristotle's view that virtues are found somewhere in the middle between two excesses must not be interpreted as an encouragement to mediocrity, because in different circumstances, very variable levels of effort might be required to raise to the virtuous Golden Mean (about Aristotle's ethics see Gottlieb, 2013).

2.3. From virtue ethics to virtue aesthetics

So, does a virtue-based approach have anything to say about art? Judging by the literature on Aristotle, virtue ethics and virtue aesthetics, yes. In a seminal paper, David Woodruff proposed to add to the catalog of traditional moral and intellectual virtues a set of properly aesthetic virtues (Woodruff, 2001). Aesthetic virtues enable both the making of art and its appreciation. The most important of such virtues are insight, sensitivity, vision, creativity, persistence, and courage (Woodruff, 2001). What makes such virtues "aesthetic" is their motivation and their orientation to the objective of creating something for aesthetic appreciation. Analogous virtues, indeed, may be involved in activities, which are not meant to create something for aesthetic appreciation; but in that case they are moral or intellectual virtues, not aesthetic.

2.4 Peter Goldie's contribution

An important contribution to the virtue-based study of art was made by Peter Goldie (Goldie, 2007). According to Goldie, "artistic activity, as expression of the virtues of artmaking and art appreciation, will, along with ethical activity and what Aristotle called contemplative activity, be a constituent part of what goes to make up human well-being [...] art, like ethics, is not a luxury good: without art, as without ethics, one cannot do well: one's life would be profoundly impoverished" (Goldie, 2007). Goldie saw in virtue ethics an opportunity to reconnect artmaking to an understanding of ethics "as concerned with the deeper question of what makes a good life, and not with some parochial, more superficial, notion of morals or morality" (Goldie, 2007). He, however, drew an analogy between art and such a conception of ethics, whereas it is unclear whether it is necessary to speak of a mere analogy. Artmaking is, indeed, a way to pursue a good life (as Goldie himself argues); it is a way to pursue the humans' telos; it is, in sum, ethical action. We appreciate, however, Peter Goldie's choice to focus on the making of art and not on the work of art: "we need to work with a notion that will help us to see why art, like ethics, matters to us as human beings, and, for this purpose, to give definitional priority to the concept of artwork, however broadly conceived, runs the risk of our being concerned only incidentally and instrumentally with the various activities, intentions, dispositions, feelings, and so on, that are involved in the whole practice of the production and appreciation (including valuing) of artworks" (Goldie, 2007). According to Peter Goldie, then, the making of art and the appreciation of art are necessary to a good life for each human being, on variable levels and for different specific art forms (Goldie, 2007). But why is it so? What makes art beneficial and indeed necessary? The answer is probably that "art, when successful, can bind





us together as fellow humans—can appeal to, and reveal, our shared experiences and our shared emotional responses to those experiences" (Goldie, 2007). I share this understanding and appreciation of art, but it should also be noted that it was not expressed by Aristotle himself.

3. Art and courage

3.1 Fear

Of all the virtues that may be relevant or necessary to artmaking, I would like to reflect in particular on the virtue of courage. It has often been observed that making art requires courage, in many ways. First, art is often self-expression, which exposes the artist's vulnerability. The artist must overcome the fear of criticism (even internalized criticism) and the fear of failure. They must face their own technical and aesthetic limitations and overcome self-doubt. The unbridgeable chasm between ideal and execution will always trouble the artist. There is no shortage of testimonies about fear in artmaking. In their book "Art & Fear", David Bayles and Ted Orland put it clearly: "Making art can feel dangerous and revealing. Making art is dangerous and revealing. Making art precipitates self-doubt, stirring deep waters that lay between what you know you should be, and what you fear you might be" (Bayles and Orland, 2023).

3.2 Risk

Furthermore, artmaking always involves risk. The risk may be a creative risk, when undertaking a creative activity, the outcome of which is by definition uncertain; when experimenting with a new medium or new techniques. The risk may be also a personal risk of stigma or dereliction, when the artist challenges artistic conventions, social norms, or even entire political regimes. The risk may be symbolic, psychological, or social; may be also a concrete risk of persecution. Many artists struggle also to support themselves and their families economically. David Bayles and Ted Orland identified the source of the modern artist's uncertainty and anxiety in their isolation, their lack of a social and cultural anchoring point, in contrast to the experience of artists in past ages: "Other people, in other times and places, had some robust institutions to shore them up: witness the Church, the clan, ritual, tradition. It's easy to imagine that artists doubted their calling less when working in the service of God than when working in the service of self. Not so today. Today almost no one feels shored up. Today artwork does not emerge from a secure common ground [...]. Making art now means working in the face of uncertainty; it means living with doubt and contradiction, doing something no one much cares whether you do, and for which there may be neither audience nor reward. [...] This is not the Age of Faith, Truth and Certainty" (Bayles and Orland, 2023).

3.3 Anxiety

Even in optimal circumstances, the artist often must confront the fear of rejection of one's work. Often the artist must face the fear of one's own emotions and sometimes even trauma; in other words, face one's demons. The artist may have to face troubling existential questions, like human suffering or the problem of evil; may have to face difficult philosophical or religious problems. That, too, requires courage. Furthermore, feverish competition is often present in artmaking, possibly with the attending fear. Also, as famously argued by Harold Bloom, the artist (the poet) must overcome the fear of never expressing one's originality and remaining forever captive to an earlier, more influential artist's influence, which entails a kind of symbolic death (Bloom, 1973). For an artist, a period of inactivity or of creative silence might represent a sort of symbolic annihilation; the fear of such experience is often the relentless spurn behind an artist's extreme productivity (Bayles and Orland, 2023). Finally, the artist's fear of one's own irrelevance or futility is compounded by the ultimate fear of the possible futility of art itself, or of its definitive sunset. So, in





many ways making art involves fear and overcoming fear; in other words, it requires courage. Aristotle made interesting observations about courage, and although he did not discuss courage in relation to artmaking, we think it would be worthwhile now turning to the Aristotelian account of courage.

4. Aristotle

4.1 Courage in the Nicomachean Ethics

Aristotle discusses courage in the Nicomachean Ethics, in the context of war (Nicomachean Ethics, Book II). In accordance with his famous theory of the mean, Aristotle describes courage as being equally distant from fear (cowardice) and from excessive confidence (rashness, temerity). Although Aristotle discusses courage on the battlefield, his notion of courage applies also to other contexts (Vigani, 2017). According to Aristotle, however, courage is invoked not in all fear-inducing circumstances, but only in relation to risks that are voluntarily chosen; courage is not relevant to fear-inducing situations that one has not caused and that one can only passively endure, like illness or poverty, or a storm at sea. Furthermore, those who are not afraid because they are unaware of the risks facing them are not courageous either. In his Ethics, again taking examples from war and military operations, Aristotle differentiates between the courage of those who are not aware of the actual dangers before them and therefore look brave but fly as soon as reality appears, and the courage of those who are fully aware of the dangers before them (Nicomachean Ethics, Book III, Chapter XI).

4.2 Courage in artmaking

We believe this description of courage may apply to the artist, in the sense that the knowledge of the risks involved in artmaking often increases with experience and with artistic maturity; and therefore, artmaking becomes an ever more fearful enterprise, requiring more courage and resilience to the experienced artist than to the beginner. We think that not only Aristotle's account of courage is relevant to artmaking, but that his famous theory of the Golden Mean is relevant, too. An excess of fear would prevent the artist from sharing or even creating any artwork; rashness, or an excess of confidence, might induce the artist to technical mistakes or to an excess of originality or of radicalism that would make the artwork unintelligible or aesthetically unacceptable. There are sure many other ways in which the making of good art depends on avoiding excesses and getting something "just right".

4.3 Artmaking as an ethical activity

There is another aspect of Aristotle's account of courage that is relevant to artmaking. According to Aristotle, courage enables us to perform noble actions, and is therefore necessary for human flourishing, or *eudaimonia*. Courage is essential also because it is a prerequisite or a point of anchorage for the manifestation of other virtues of artistic creation like authenticity, integrity, and truthfulness (Wilson, 2020). We propose, that artmaking, besides of course expressing aesthetic values, is essentially a manifestation of (moral) courage and of other virtues enabled by courage and should therefore be considered as an ethical activity. Artmaking is not only a constitutive element of a good life and is not merely analogous to ethical action (as claimed by Peter Goldie (Goldie, 2007)); instead, we suggest that artmaking is itself a virtuous activity.

4.4 Courage in the appreciation of art

Is courage (moral, aesthetic) necessary for the appreciation of art, too? Definitively yes. In his seminal article on virtue ethics, David Woodruff briefly explains why it is so: "Aesthetic courage, like creativity, is not merely involved in the production of works, though is most evident there. One must also have aesthetic courage to evaluate works and look for new





insights in works that have been previously considered. One feature common to many great works is that they tell us about ourselves and about reality. Courage is needed to face what the work is saying. This overlaps with moral courage, but it is an aesthetic virtue when motivation is to face what the work tells us about ourselves in order to appreciate the work. Aesthetic courage fits between the vices of timorousness and recklessness" (Woodruff, 2001).

5. Conclusions

Both aesthetic courage and moral courage are necessary to make art (and to appreciate it). Therefore we argue that art should be regarded not only as an aesthetic activity, where the artist exercises aesthetic virtues; but also as an ethical activity because of the inevitable need to exercise moral virtues, too (like courage). Art is an activity that moves us closer to our *telos*.

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Reflection

Truth, Beauty, and Ethics in Art, Science, and Health: Interdisciplinary Reflections and Philosophical Perspectives

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Abstract:

Since the beginning of history, humans have felt compelled to try to understand the reality we live in. Scholars, scientists, and humanists have tried to systematize knowledge and spread it for the benefit of society at large. Today, universities are the institutions where knowledge from various disciplines is gathered so that the interaction among them allows a better understanding of reality. No single discipline contains a specific truth in itself. Rather, different disciplines observe and study the same reality, but they approach it form different and specific points of view. The truth expressed by art, the truth studied by science or the truth explored by health disciplines are not different truths. They are the understanding of the same reality studied from different approaches. Greater communication and integration among the different disciplines that coexist in a university enhance the ability to deepen and broaden our understanding of reality. Investing in cooperation in research and teaching work among the different faculties and departments of the university fosters interdisciplinary knowledge that enriches the entire university. This talk offers some reflections in the field of art, science, and health with some ethical and philosophical considerations.

Keywords: Aesthetics; Goodness; Ethics; Reality; Values; Interdisciplinarity





1. Introduction

1.1. University and universal knowledge

When the first universities appeared in Europe in the Middle Ages, they took the name "university", meaning that the knowledge taught there was not isolated knowledge, but related to each other. That is to say, humanistic, scientific, artistic, and mathematical knowledge, among others, is brought together in the same institution and allows students to acquire knowledge from different integrative perspectives. This greatly enriches student's knowledge.

Although in today's society there is increasingly greater specialization in work, and this allows for further study in different fields, the role of the university still includes the function of uniting different specialties and building bridges between the different branches of knowledge, in order to achieve a more comprehensive knowledge of reality.

1.2. The different objects of knowledge

In this short article we will try to make a philosophical reflection on the relationship that exists, in the university environment, among three important types of knowledge: science, arts and medicine. To achieve this, it is necessary to analyze the objects of these types of knowledge and see the relationship that exists among these objects of knowledge. Sometimes we talk about certain types of knowledge without giving a concrete definition of the knowledge to which we refer, and this brings with it the consequent problem of moving away from the topic of discussion and not being able to set limits on our debates. For this reason, the first step will be to properly define some terms.

2. The objects of study of science, art and medicine

2.1. Science

Aristotle calls "science" (episteme) all the set of knowledge that aims to understand reality. Thus he recognizes the existence of a science that studies what is as it is, and is not identified with any of the sciences we call particular (Aristotle, 1933).

Scientific knowledge is obtained in a methodological manner through observation and experimentation. It follows a specific methodology, requiring a hypothesis, experimentation, and analysis of data obtained to reach conclusions.

2.2. Art

We could explain what art is by referring to simple definitions to try to understand knowledge: "art is that human activity that produces beauty," or "that activity that represents or reproduces reality", "that creates forms", "that expresses", "that produces aesthetic experience".

Heidegger states in one of his essays: "The essence of art would be, then, this: putting the truth of the entity into operation" (Heidegger, 1988). In this definition of the German existentialist philosopher, we can already observe the relationship between truth and essence of art, which we will delve into later.

2.2.1. Medicine

From birth and throughout our lives, we have visited doctors and hospitals, even without studying medicine. It is therefore not difficult to understand that the aim of medicine and health sciences is to find the causes of diseases in order to resolve them, curing patients and providing them with well-being. The health of the patient is the good that medicine seeks for man.

3. Truth, goodness and beauty.

In 13th century scholastic philosophy, Thomas Aquinas defined transcendentals as certain necessarily derived aspects of being (Aquinas,). In a simple way we could say that the





transcendentals of being are fundamental properties that everything that exists shares, because they derive directly from its existence. Traditionally, four properties are considered: unity, truth, goodness and beauty. Let us analyze the last three.

3.1. The truth and the good

The Greek philosopher Plato, in his epistemological theory, considers that the Good is the supreme idea, which is above all others. It is the ultimate source of all knowledge, truth and reality. The good is the goal of knowledge (Plato,). If we ask ourselves what makes a doctor's diagnosis or a judge's sentence good, we will conclude that only one thing makes them good: the truth, that is, that the doctor's diagnosis tells us what really happens to us, and that the judge's verdict conforms to what really happened. In this way we can say that acting well is acting in accordance with the truth.

A question that Pilate already asked Jesus, and that every man asks himself is: What is truth? And in the words of Thomas Aquinas, truth is "the correspondence between understanding and reality" (Aquinas,).

Thus, we will not say that a person is tall or nice because everyone thinks so, but because that person is tall or nice, all those who affirm it conform to the truth. It is reality that constitutes the foundation of truth, and knowledge is true when it manifests and declares the being of things. For this reason, error is not knowledge, since knowing something falsely is equivalent to not knowing it.

Opinions are not a source of truth; we must strive to understand the reality of things well and not just be informed of opinions.

Returning to Platonic theory, we can conclude that good, the highest knowledge, is achieved when the truth is known and respected.

3.2. The good and the beauty

For Aristotle, educating a man was teaching him to have good taste in acting: to love the beautiful and hate the ugly (Lorda, 1993).

We call an entity good because of its relationship to appetite in that it possesses the characteristics of the perfect and perfectible of others. Things in which we perceive perfection do attract us. The truth and goodness of things cause pleasure to those who contemplate them. This property of causing pleasure in this way is what we refer to when we say that something is beautiful.

The level of pleasure that we experience when we see a piece of handmade ceramic is not the same as when we see Rembrandt's famous painting "The Anatomy Lesson of Dr. Tulp". In the ceramic piece we observe a sensible beauty, because the harmony of the forms and colors attract us. But in Rembrandt's painting, in addition to sensible beauty, we experience an intelligible beauty contained in what the famous painter wants to convey to us.

Intelligible beauty is linked to truth and moral goodness; hence ugliness is proper to error, ignorance or vice. All good things produce joy when they are attained, but things that are beautiful engender a special pleasure in the mere fact of knowing them.

Although the contemplation of beauty always brings with it a delight, beauty is not pleasure or pleasantness, but those properties that make its contemplation pleasant. Like goodness, beauty is not an attribute that has its origin in the will of the one who wants it, but a perfection of the desired object: things would continue to be good or beautiful even if there were no men to appreciate or desire them (St. Agustine,).

3.2.1. Objective beauty and the perception of beauty

Although we say that things are good in themselves, we have experience that some good things are not good for everyone. Consider, for example, a chocolate cake. The cake is good for one person, but harmful for another who is allergic to lactose or gluten. But this does not mean that good is subjective: the need to drink water is not a whim, it is a truth independent of our subjective opinion. Similarly, objective values such as peace, justice, friendship will always remain valuable for all humans. In this sense we can say that good is what suits a thing, what perfects it.





Something similar happens with beauty: all beings possess greater or lesser beauty depending on their perfection; however, for a person to appreciate the beauty of things there must be a proportion between that person's cognitive abilities and the beauty this person perceives. For this reason, just as not all humans have the same capacity to understand concepts and reality, not all humans have the same capacity to be amazed by a work of art. A certain aesthetic education is necessary to perceive the beauty of some artistic manifestations.

Although sometimes, in the artistic field subjectivity seems to predominate, beauty is an attribute of beings. The beauty of an object does not depend on what each person likes. If it were so, it would make no sense to talk about beauty and ugliness.

3.3. Truth and beauty

Is art a luxury for human beings to embellish their lives or is it a necessity? As a starting point, we could say that a man has been moved, by his own interiority, to represent or express something of the external reality or of his interior since the beginning of human existence.

It happens that sometimes we watch a film in the cinema and we come out excited by the experience we have had during the screening. Let us suppose that what we have seen is an adventure and fantasy film. We can safely say that if we saw the same film again, but this time changing its original soundtrack for the soundtrack of a melancholic film, the impression would be absolutely different. The same story with music that does not suit it well, distorts the film. Relaxed music does not express the agility of the adventures of a fantasy film.

If, while admiring Michelangelo's statue of David, I simply thought it was beautiful, but if it gave me the impression that I was looking at Emperor Napoleon Bonaparte instead of King David, the statue would not fulfill the purpose for which the sculptor sculpted it. With his sculpture, Michelangelo wanted to convey beauty, but also a real historical fact.

We have said above that beauty is not an attribute that has its origin in the will of the one who likes it, but in the observed object. This helps us to understand that to the extent that we are able to know the observed object, that is, reality, we will be able to understand the beauty contained in that object.

4. Conclusions

There is no one truth in medicine or health sciences, another truth in painting or literature, and another truth in physics or chemistry. The reality studied by different disciplines is the same, but each discipline approaches it from its own unique perspective. When studying the human body, a doctor is more interested in health, an artist in proportions, and a biologist in the composition of tissues. The connection between all knowledge, of the human body in this example, from different perspectives is what makes this study comprehensive and more complete.

The study of a question made by several disciplines: what they have in common with the reality being studied, will be a good and beautiful study to the extent that the knowledge we acquire through it corresponds to the reality studied.

The more disciplines there are that study the same reality, the more complete our knowledge will be, making it richer, more beautiful, and better.

When medicine, art, and science approach a better understanding of the object of their studies, they become more perfect. Therefore, the connected study of these disciplines also helps to improve them as branches of the study of knowledge.

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Research

Maternal Dynamics: A New Ethical Paradigm

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Abstract:

This work develops an original conceptualisation of maternal ethics in relation to philosophical discussions of the temporality of the ethical act. Marked by the tension between continuity and disruption, maternal experience provides a model for ethical creativity and action. This unique temporal tension reflects a moral responsiveness that advocates a novel understanding of maternal ethics centered on empathy and courage. The work combines analyses of Husserl, Levinas, Jankelevitch and Cavell with readings of testimonies of Second World War survivors which I collected in Slovenia.

Keywords: Maternal; Ethics; Temporality; Phenomenology; Second World War; Testimony





1. Introduction

This work argues that the complex temporality of the maternal experience constitutes an affective tension within the maternal voice which accounts for the ethical singularity of the maternal figure. In support of this thesis, we explore the 'double' register of competing temporalities which resonate within the maternal voice; we argue that the unpredictable experience of disruptive temporality tangentially coupled with an impossible promise of temporal continuity creates an affective tension within the maternal experience which elicits a creative, improvisational, and singularly moral responsiveness. We show that the enigmatic and paradoxical relation between a maternal compassionate empathy and an austere maternal ethics of obligation produces a temporally lived tension which manifests paradigmatically as a temporally tensed attentive inclination of the maternal. We further show that maternal singularity not only exceeds essentialist stereotypes, but that the maternal should be more properly understood in terms of an improvisational responsiveness, temporally tensed between an empathetic ethics of care and an austere ethics of obligation. With the help of Vladimir Jankelevitch's discussion of the oscillation of time between "the interval and the instant," we attempt to think the tangential yet transformative relation between the continuity of inherited generational temporality, which we articulate in terms of Edmund Husserl's internal time consciousness (1) and the temporal disruption of subjectivity which Emmanuel Levinas² presents as the necessary condition for ethical consciousness, the paradigm of which is the maternal figure.

For Jankelevitch, both the consciousness of temporal continuity and the instantaneous moral act of conscience are connected by an imperceptible thread of the "je ne sais quoi ou le presque rien" of "the instant." Jankelevitch's unique development of his understanding of "the instant" allows him to pivot from a metaphysical discussion of the consciousness of the irreversibility of time to an elaboration of one's conscience which appears and disappears throughout one's life. According to Jankelevitch's theory of virtue, ethics cannot be a contemplative affair based on pre-given categories; rather virtue is created through the act, the doing, the instant, and intuition; hence, morality cannot be prescribed. Instead, the ethical demands to be continually created and re-created through an improvisational spontaneity and capture of the "presque rien" which is lived affectively as moral virtue, that is, as moral resistance, courage, fidelity and empathy. Jankelevitch writes of "how the pure Act is beyond the fission of being and non-being," of "the primordial primacy of the Doing that is pure of any being,"4 and of the "making-be of that is more and better than being."5 Accordingly, we suggest that Jankelevitch's account of virtues, as performatively and passionately lived acts, provides us with a new framework or "blueprint" within which we might explore the compelling nature of maternal empathy and courage. By amplifying the resonance, resiliency, and audacity of maternal voicing, this work hopes to articulate a new way to think the "tensed time" of the maternal experience and to pave a new way to listen to the exigency of the maternal voice.

Building on Husserl's description of the continuity of time and Levinas's account of the diachronous "rupture of time" as theoretic alternatives of the maternal temporal experience, we rely on Jankelevitch's theory of the "instant and the interval" as well as his theory of virtues in order to formulate a new account of maternal ethics. We propose that the oscillating relationship between empathy and courage captures the most significant insights from both Husserl's and Levinas's discussion of temporality, while supporting a new theory of maternal ethics which resists either reducing the maternal to a figure of

¹ Husserl, Edmund. On the Phenomenology of the Consciousness of Internal Time (1893–1917). Springer Science & Business Media, 2012.

² Levinas, Emmanuel. The Theory of Intuition in Husserl's Phenomenology. Northwestern University Press, 1995.

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³ Jankelevitch, Vladimir. *Philosophie première*, Presses Universitaires de France, Paris: 1953, p. 184.

⁴ Ibid., 187.

⁵ Ibid, 259.





martyrdom or to one of consolation, but which rather promotes the maternal as a paradigm of empathetic courageous action and attunement.

Yet why should temporality have a privileged methodological status for describing the maternal experience? While spacial descriptions of the maternal provide obvious metaphors for the experience of pregnancy and giving birth, temporal descriptions of endurance and disruption have the advantage that they evoke the ethical tension of becoming a mother to one's child or to another, without relying on the visual trope of the withdrawal, replacement, or disappearance of the mother. Not only does the temporal description reflect the ongoing, enduring, continuous relation of the mother to a child, it likewise marks disruptions and everyday events of separation belonging to the maternal experience. We argue that because competing forms of temporality belong to the temporal experience of the mother, the tension between these senses of temporality creates unforeseeable and ethically demanding situations. Accordingly, by explicating the temporal complexity of the maternal voice, we hope to articulate the ethical weight that the maternal voice bears. By challenging the ancient philosophical privilege afforded to the stable, the certain, and the eternal, and by redirecting us or "converting" us to think the "newness" of deformalized time, this work shows that the singularity of the maternal lived experience of time provides us with a pathway to think "newness" and "innovation" within time itself.

Following Henri Bergson (2019), we argue that the traditional category of the possible represents a paradigm of a poorly posed philosophic problem with significant consequences: in the history of philosophy, the category of the possible is assumed to contain less ontic value than that of the actual or, in other words, we assume that possibility is necessarily lesser than its real counterpart. However, for Henri Bergson, the exact opposite is true; it is the possible that contains more ontic value than the actual. Accordingly, the possibility of knowledge only becomes real through an act of thought that projects it backwards: as such, the possible contains the real and the retrospective act of thinking that constitutes it (Bergson,2019). Behind the illusion of the possibility of knowledge lies an error that threatens the very task of philosophy. Henri Bergson (2019)therefore advocates a new direction for philosophy, one where philosophy undertakes a conversion. The task of conversion which Bergson lays out requires philosophy to learn to turn its gaze away from the eternal; and in lieu of disregarding or disdaining the existential experience of time, philosophers would benefit by turning their attention toward movement, toward newness in the making, and finally should attempt to think time as becoming.

Philosophy's historical challenge of giving conceptual determination to existence is, according to Henri Bergson (2019), a direct consequence of its desire to grasp what is eternal in time and to conceive of time as finalistic. According to this view, what comes to exist, that is, what is born in time, must have its law, reason or cause of existence elsewhere, that is, outside of time. Because the movement of time is traditionally understood as the realization of a pre-existing possibility outside of time, temporal existence is seen as an inadequate incarnation of a pre-existing possibility, where the movement of time is understood as the realization of a pre-existing possibility. In contrast, Henri Bergson(2019) contends that the category of the possible be conceived as the becoming of temporal existence which contains more reality than the actual itself.

This is where our thinking of the maternal experience of time becomes critical. Rather than thinking the maternal category of possibility as "less than" or as "in service" to the patriarchal idea of the eternal, we propose that the maternal experience of time provides us with a paradigm for the power and agency of a new way to think the becoming of time. Time can no longer be thought of in terms of an external framework within which events unfold, but must instead be conceived as truly active and as a creative force. Consequently, we argue that becoming must be understood as a non-finalistic, non-teleologic conception of time, open to a non-determined, non-causal future, which moves between the alternative experiences of continuity and disruption. The conversion that Henri Bergson(1998) so hopes for is a conversion of philosophy from a teleological desire for eternity to an existential time. In alignment with Henri Bergson's (1998) proposal, we submit that the maternal experience of time provides us with a paradigmatic model for temporal becoming insofar as it captures the power, agency, and creativity of time – which is "invention or it is nothing at all." (Bergson,1998).





Because time is active in and of itself, the being of time is nothing apart from its specifically active, creative power, that is, its own agency. Were we to follow conventional phallogocentric thought, which adheres to the idea of eternity and from which it follows that the possibility of things precedes its existence, our thinking would entail a denial of the reality of newness and, as such, a denial of time's creative force. However, by exploring the thinking which maintains that the category of possibility does not precede their existence, we avoid the trap set by the idea of the eternal; instead, we gain epistemic and experiential access to the newness of becoming and the invention of maternal creativity. In order to make a case for the process or "birth" of such a conversion, our research develops the notion of a maternal, cathartic, and transformative temporal tension which belongs to the maternal existential experience which I formulate as the impossible coupling of time as alternating between continuity and as disruption. Insofar as the maternal temporal experience provides us with a paradigm for a transformative experience, we argue for the powerful agency of a new maternal ethics, lived as alternatively empathetically creative and improvisationally courageous.

2. Original Contribution of Thesis

The maternal experience is likewise lived as a continual resistance to a series of lifelong separations that motherhood imposes on the female psyche and body and which is expressed as maternal mourning and lament, culminating in the inseparable linkage between laments and motherhood. Laments are above all about separation and the severing of ties between mothers and their children. The extreme contrast between the intuitive, wishedfor inseparability of the mother-child relationship and the separation caused by death constitutes the epicenter and the gravitas of maternal lamentation. In the face of death, it is this subverted longing for life which accounts for the subversive tone associated with the female voice of lamenting and constitutes an ultimate expression of the continued resistance to the series of separations which defines motherhood. As Walter Benjamin suggests such expression is necessarily and "essentially intertwined with loss." "Deeply saturated with melancholy and loss, the lament thus functions both as an expression of loss and as a site for its recuperation." (Ferber, 2013) The lamenting language of mourning, as expressed by mothers in mourning, reflects a singular tension between the continual experience of separation which mothers experience and that unforeseen encounter with the most radical form of separation, namely the death of one's child. As such, the maternal lament works against the experience of mourning as a voice of resistance to death. It functions paradoxically to disrupt the experience of mourning through the very act of mourning itself. As such, the maternal lament is a paradigm of a radically subversive force which like empathetic courage expresses itself as pneumatically creative, at the border between language and action, between life and death.

3. As Gershom Scholem discerningly describes lament:

"[T]here is one language whose infinity is deeper and different from all others (besides the language of God). For whereas every language is always a positive expression of a being, and its infinity resides in the two bordering lands of the revealed and the silenced, such that it actually stretches out over both realms, this language [of lament] is different from any other language in that it remains throughout on the border, exactly on the border between these two realms. This language reveals nothing, because the being that reveals itself in it has no content (and for that reason one can also say that it reveals everything) and conceals nothing, because its entire existence is based on a revolution of silence. It is not symbolic, but only points toward the symbol; it is not concrete, but annihilates the object. This language is lament." (Scholem, 2014)

The lamenting invocations of mothers signal beyond themselves, beyond any verbal reference, exceeding the limits of ordinary language and are rather located at "the border…between… the revealed and the silenced." Lament, as a "language of the border," is situated between two spheres and exists in the tension between the "domesticating and the foreignizing" processes of language. (Ferber,2013) "Origin and border, as in birth in the sphere of life, converge in the sphere of language within lament. Birth and death, the fundamental symbolic poles of life, reappear in the internal structure of lamentation." (Scholem,2014) Retrospectively, the sense of the finality of death pushes the language





of laments backwards, to an unrealizable, inseparable phase that barely existed and was lost during life. The act of lamenting thus repeats and intensifies the tangential trajectory of the "presque rien" of moral resistance, courage and empathy. Yet in doing so, the object of lament, namely, mourning, is paradoxically destroyed through its performative expression. The language of lament is less a matter of melancholic withdrawal, than an attempt to productively confront the event of loss. As such the lamentation of maternal mourning provides us with an alternative window through which the power and agency of a new maternal ethics might be heard and voiced.

4. Original Contribution of Thesis

Philosophy has historically appropriated maternal tropes and metaphors in order to explain its generative and creative possibilities; this has repeatedly resulted in the exclusion or denigration of the embodied, material maternal experience from the philosophic register. Alternatively, recent feminist theory has striven to vindicate the experience and work of maternity while pillorying philosophical "co-opting" of the maternal metaphor. Rather than defending the maternal by engaging with philosophy through metaphorical or metonymical strategies, we argue that the singular temporal experience of becoming a mother provides us with the paradigm for a "new" philosophy which ideally oscillates between the "Je ne sais quoi" of intuition and the duration of discursive reasoning. As with the "organ-obstacle" relation which figures centrally in Jankelevitch's thought, neither the instant of intuition nor the interval of discursive reasoning can be abandoned; both are needed to avoid the dilettantism of relativity or the inevitability of religious absolutism. Accordingly, we contend that the maternal experience sketches out the deformalized movement of time and provides us with a road map to think the newness and creativity within time as becoming.

Although much recent work has been done in the intersecting fields of feminist and psychoanalytic theory regarding the maternal, we approach the "question of the mother" through a phenomenological lens, focusing on the dimorphic temporality of the lived maternal experience which carries with it ethical as well as affective, lived repercussions. Instead of challenging normative gendered concepts of subjectivity⁶, we direct our attention to the qualitative transformation enacted in "becoming another's mother." And finally, rather than investigating subjectivity through a predominantly static lens, we broach the subject of the maternal voice through a temporal analysis of creative and critical vitality which we integrate with performative speech act theory and which I then use to amplify the voices of those who during World War Two recounted the experience of "becoming another's mother." In doing so, this work theorizes the maternal voice while avoiding essentialist tropes which rely on or imply normative categories as well as tropes of ambiguity attached to the maternal which effectively dissemble and disable the force of the maternal voice.

Yet, why specify "maternal time" rather than "parental time"? Cannot both parents experience temporality in the same "tensed" way? Why privilege the maternal over the parental? The distinguishing feature of the maternal experience of time is due to the inherited relationality that mothers bear between generations: the mother is both born from a mother and bears the future generation - whether biologically, through adoption, or as a "second mother." And yet, the maternal is likewise the site of a radical non-self coincidence, in which the maternal subject fins itself temporally out-of-sync with itself and temporally disrupted. On account of such temporally lived incongruity, the maternal provides us with a singular lens through which we might envision the power of an intergenerational sense of possibility which is coupled with the inevitable disruptions and ruptures of temporal experience. As "doubled yet divided" the maternal experience reflects the paradoxicality of moral dilemma, choice, and action.

It does not follow that fathers or women who choose not to have children are excluded from the "maternal." Quite to the contrary: we are all born from mothers, and, therefore, we each have the potential to carry over or translate the inherited temporal experience

⁶ I have in mind the disruptive interventions of space in Julia Kristeva's pre-Oedipal Khora, the doubling found in Luce Irigaray's work on sexuation, and Bracha Ettinger's concept of the Matrix.





which we share with our mothers to the next generation. The point is not that the maternal is an exclusive category determined by a privileged access to a particular experience; rather, the point is that the maternal provides us with a model of relationality that deserves our attention. My research thus argues for a broad and inclusive understanding of maternal experience which is not biologically determined, yet which, nevertheless, resonates within the lived experience of the mother.

The originality of my contribution to this challenging field thus advances a theory of maternal experience which grows out of a phenomenological analysis of the inherent temporal tension and moral paradoxicality of the maternally tensed experience and which defends the maternal as a signature "event" of ethical action. This work likewise integrates the recorded testimonies of four Slovenian witnesses who survived through the war years - either as prisoners in concentration camps, as fighters, or as hidden children in Slovenia. Since the survivors whom I interviewed were children or adolescents during the war years, our research was able to bears witness to the unique and different ways that each experienced and enacted becoming a mother or becoming another's mother during this horrific period. In doing so, we highlight the resistance, courage, and empathy of each of these survivors and those who enabled their survival.

We believe the direction of our research is important because it initiates a discussion of the singularity of mothers - or those who acted like mothers - without reducing mothers to agents of a patriarchal system. Rather than seeing mothers primarily as care givers for the family unit, tribe, or State, we are interested in defending a view of the maternal which vindicates the singularity of the maternal by highlighting the lived tension between an empathetic inclination and courageous resistance of those who act like mothers. We show that this transformative tension, which characterizes the maternal phenomenological experience, is reflected in the heroic agency and improvisational talent of mothers and those who acted like mothers during the war. We focus on the heroic actions of those who acted like mothers - nannies, 'stranger' mothers, and children who became mothers to their own mothers, in an effort to defend an account of maternal virtue, where the mother is figured as moving between empathy and courage, constantly recreating herself, and therein "becoming a mother to another." This philosophical interrogation therefore supports an alternative account of the maternal, that is, a new account of maternal ethics which allows us to speak about mothers, without relying on stereotypical maternal tropes or minimizing the heroic paradigm of mothers, and to which we attribute the singularity of the maternal temporal experience.

5. Tracing temporality between Husserl, Levinas, Cavell, and Jankelevitch

Our research argues that the experience of maternal temporality accounts not only for the improvisational and disruptive audacity as well as the empathetic affect of the maternal voice, but in doing so, challenges philosophic references to the maternal which either figure the maternal in absolutist terms or dismiss the maternal due to essentialist categories. Against phallocentric voices which portray the maternal as a model of absolute sacrifice or as that which requires depassioning of its threatening sensibilities, we contend that these characterizations seek to castrate and exclude the maternal voice from the realm of the ethical, thereby, foreclosing its philosophical and ethical relevance. In response to the ethical-philosophical problems obtaining to the maternal voice, we work at the intersection of Edmund Husserl's (1962) account of internal time consciousness, Emmanuel Levinas's theory of diachronic disruption, Stanley Cavell's theory of passionate utterance, and Vladimir Vladimir Jankelevitch's (1974) theory of the irreversibility of time, in order to develop an account of maternal temporality which subverts philosophic misrepresentations of the maternal voice. we argue that maternal temporality provides us with the necessary theoretical grounding to account for a compelling ethical theory based on a conception of alterity which welcomes the possibility of disruptive surprise, while refusing to be taken hostage to an immemorial past.

According to Levinas, it is on account of the indeterminacy of the future which is encountered through the diachronous experience of the other that we have a sense of what we owe, of our responsibility; whereas, for Edmund Husserl (1962), the analysis of internal time consciousness and duration is concerned with assuring continuity between the past,





the present, and the future. For Edmund Husserl (1962), the originary meaning of the present is determined by one's elapsed intentional lived experiences; because these experiences are retained in their original fluidity by present consciousness, they can for that reason be made present again at any moment in the form of a memory. For Edmund Husserl (1962), the past is a displaced present, pushed back from the center of one's present consciousness towards its horizon by the emergence of a new lived experience, while the future is constituted by and grows out of one's memory of past experiences. As such, the protentions of the future function in the inverse way to that of past retentions.

For Levinas, however, Edmund Husserl's (1962) conception of the continuity of internal time consciousness is unable to do justice to the radical ethical sense of alterity of the other person, which Levinas foregrounds in his analysis of temporality and which ultimately provides him with the foundation for an ethics of temporal interruption. By contrast, Husserl's conception of retention and rememorative representation consists in an effort to recuperate the past, by safeguarding the continuity of the flux of intentional consciousness. In order to establish the unity of intentional consciousness and, with that, a unified sense of experience, Husserl describes what he takes to be structures of consciousness which are enriched through a sense of "a thickness of time."

Although Edmund Husserl (1962) describes his phenomenology as the only systematic version of "transcendental idealism," (Husserl, 1962) we suggest that we understand Husserl's phenomenology as an experiential idealism, wherein all existent entities are in principle experientially accessible. We argue that for Husserl the interdependency of his descriptions of the structures of consciousness and time consciousness establishes the unity of a subject's sense of experience, upon which he bases his theory of empathy. Based on a close examination of Levinas's critique of Husserl's account of intersubjectivity, we then argue that Levinas's account of the alterity of ethics is a response to Husserl's desire to establish experiential access to the intersubjective world - rather than, as is commonly accepted, as a means of establishing epistemological access. Although Husserl strived to develop a phenomenology which would provide an absolute and ideal access to meaningful experience of the perceptual and sensual world, by relying on the unifying function of internal time consciousness, Husserl anticipated the shortcomings of his strictly phenomenological method with regard to intersubjective consciousness and later presented an argument for empathy based on an intermingling of imagination and consciousness.

Levinas devotes his career to responding to Husserl's earlier problematic by developing his own phenomenology of the time of the other which is interwoven with his description of both the feminine as well as the maternal. We argue that although the central role of diachronic temporality in Levinas's description of the ethical encounter is in direct response to limits arising from Husserl's unified subjective phenomenology (both in response to internal time consciousness as well as to intersubjective apperception), Levinas does not break entirely free from Husserl's influence. As Husserl strived for an absolute and ideal experiential access to the world and to others via a unified subjectivity, Levinas privileges the absolutely demanding ethical relation to which one is held hostage to the exclusion and negligence of alternative ethical relations such as empathy and love, which We contend are critical to an ethics of the maternal.

In response to Husserl's highly theoretic description of the noetic conscious experience, Emmanuel Levinas presents a description of alterity as belonging to and originating with the absolutely other; although Emmanuel Levinas tries to distance himself from the theoretical unifying tendencies of Husserlian phenomenology, we argue that Emmanuel Levinas's differing accounts of alterity are plagued by the absolutist leanings of his Husserlian inheritance. For Emmanuel Levinas, alterity must radically interrupt the sense that I have of time belonging to myself in order to mark the experience of heterogeneity or the beginning of ethics. Rather than providing a single account of the relation between alterity and temporality, Emmanuel Levinas provides us with two somewhat conflicting accounts of the relation of temporal interruption. In Emmanuel Levinas's early works, Levinas highlights the embodied experience of the "face-to-face" relation according to which the feminine is excluded from the ethical relation and appears either as the external condition of possibility for the ethical or as that which erotically threatens the ethical (Levinas,1969). While, in his later works, Levinas develops the notion of alterity as the radically absolute "alterity of the other within the same," which is personified by the maternal experience as





suffering and self-sacrificial, as one held hostage to the other upon the threat of death (Levinas,1998). Although these accounts of alterity differ significantly, they both subscribe to an absolute separation of the self and the other and are meant to challenge Husserl's depiction of the return to the self by the self. Accordingly, Levinas's well known characterizations of alterity functions primarily to underscore the radicality of the disruption of the return of the self to itself. we show that although Levinas fails to recognize and address the theoretical tensions raised by his absolutist descriptions of alterity with regard to temporality for much of his career, he opens the door for a new way to think the temporal maternal experience in his account of the relation of "illeity" to the "third," wherein he maps out the possibility of openness to an indeterminate future.

In his later publications and in his interview with Bracha Ettinger, Levinas finally speaks to the possibility of a diachronous time of the future which not only informs a new and promising – if not late – insight to the maternal (Levinas, 1998) .No longer framing the feminine maternal as a spatially external condition for possibility of ethical disruption of the present and the past, Levinas identifies the feminine as the temporally external "category of the future....It is the possibility of believing that there is a reality without me." For Levinas, the relationship to the Face of the Other "is already a grasp of a past and is already a contact with a future...In the future, there they are: my possibilities and my impossibilities.... And time is there: in what is possible, in what is no longer possible, and in the unforgettable." Accordingly, I show that while Levinas originally argues that the diachronous time of the other is that which provides the ethical disruption which destabilizes the return of the self, – at first situated from a distance within the face-to-face relation, and then from within the close proximity of the other within the self, we find in Levinas's writings the concept of illeity according to which an openness to the indeterminacy of the future is figured as constituting human responsibility.

Although Levinas and Jankelevitch develop what might appear to be refractory accounts of temporality, this work investigates the intersection between Levinas's and Jankelevitch's thought in order to articulate a new conception of the maternal which highlights an ethical audacity and intuition which we attribute to the maternal. We argue that insofar as the maternal voice is a site of a disrupted sense of temporality coupled with a generational experience of duration, the maternal voice constitutes a productive tension within a complex lived experience which manifests itself as an ethical awakening of courage, resistance, and empathy – all critical aspects to an ethic which exceeds both ontological as well as essentialist categories. Rather than personifying the maternal voice as a voice of vulnerability, victimization or of self-sacrifice, we argue that the maternal voice has a radical, yet underestimated potency and resiliency to effectively create new situations and to awaken compelling, passionate, and unforeseen realities.

Following Bergson, Jankelevitch privileges intuition over discursive reason; according to Jankelevitch, through the process of concept formation, discursive reason parcels objects up and, therefore, fails to grasp objects as wholes. Whereas, intuition relies on spontaneous action which grasps reality as complete and whole. Accordingly, intuition occurs in what Jankelevitch refers to as an "instant." Because the instant of time is experienced intuitively as a creative experience, where one thing is going out of existence just as something new is brought into existence, Jankelevitch attributes to the instant of intuition an inherent generosity which falls outside of any category of discursive reason. The instant is likewise experienced as an irrevocable moment of moral resistance to the irreversibility of time: for Jankelevitch, it is the denial of the intuition of a past moment as "irrevocable" which prompts the "temporal illnesses" of resentment, regret, and remorse; whereas, the acceptance of the irreversibility of time is a pre-condition for the moral acts of courage, fidelity, and generosity. Hence, the irreversibility of time has the potential to act as both an "obstacle" as well as an "organ" for the vicissitudes of moral life, we therefore suggest that maternal intuition be understood as both reflecting an original site of creative empathy as well as a courageous resistance to the irreversibility of time. For Jankelevitch, humans are not essentially hybrids; nor are we static "beings." Rather, again following Bergson, Jankelevitch characterizes humanity as "essentially free" and, as such, as "becoming." As such, we find within our lived human experience an inherent resistance to the necessity of the irreversibility of time. Accordingly, we alternate between dimorphic experiences of time, between the "instant" and the "interval." As such, we suggest that maternal intuition





finds itself at a unique threshold of becoming, "pneumatically" recurring as a never-ending resistance to the irreversibility of time, all the while creating an irrevocable ethic of generosity and courage.]

6. Listening to Maternal Passions

I rely on Stanley Cavell's extension of John Langshaw Austin's theory (1975) of performative speech acts in order to develop an account of the improvisational passionate temporal register of the maternal voice. In contesting John Langshaw Austin's performative (1975) speech act theory, Cavell develops an account of "passionate utterance" as a form of language which is positioned to reflect the non-redemptive yet responsive ideals of "moral perfectionism." By foregrounding the significance of the perlocutionary effect of illocutionary speech acts, Cavell introduces a new category of speech act, namely, "passionate utterance." According to Cavell, passionate utterance is a form of language through which acknowledgment of others both exposes my vulnerability as well as the state which I am willing to make for the promise of a relationship. "A performative utterance is an offer of participation in the order of the law. And perhaps we can say: A passionate utterance is an improvisation in the disorders of desire." Cavell argues that Austin (1975), having made the revolutionary discovery that truth claims in language are bound up with how words perform, then gets caught up by convention when discussing what is done by words. In failing to account for the less predictable, unconventional aspects of language, Austin (1975) accordingly washes his hands of the expressive passionate aspects of speech. For Cavell, ignoring these aspects of language is to ignore the morally compelling dimension of perlocutionary language, namely, "passionate utterance."

While Austin (1975) provides a framework which challenges the exclusivity appropriated to normative speaking/thinking, Cavell makes room for an alternative conceptualization of language and conversation which locates that which is most ethically compelling to the passionate yet fragile register belonging to what I identify as the "Je ne sais quoi" of the maternal voice. For Cavell, passionate utterance is a form of language which reveals a self moving between a self-conforming "attained self" and a self moving against itself toward an "unattained yet attainable self." As "doubled and divided," there is an inherent failure or absence obtaining to subjectivity, which for Cavell paradoxically provides an opening for moral responsibility. Similarly, Levinas's account of the "saying" of "illeity" contributes to a moral language which aligns with Cavell's non-theistic concept of "passionate utterance" and which informs his concept of "moral perfectionism." For Levinas, illeity refers to the absent condition of an encounter with another; it is the very act of self-effacement, the act of undoing which indirectly account for responsibility.

As Cavell's account of passionate utterance reveals a self – "doubled and divided," moving between a self-conforming "attained self" and a self, moving itself toward an "unattained yet attainable self" in a movement of resistance and love, we defend the view that the successful outcome of passionate utterance requires an improvisational ability of talent which we argue is paradigmatic of maternal speech acts. Our research defends a broad notion of the perlocutionary, whose compass points to what Jankelevitch describes as the "Je ne sais quoi" of language. Accordingly, we show that the language form of passionate utterance resonates within Jankelevitch's temporal account of moral action and further that it provides us with an account of the ethical force of the maternal voice. As the tension of the maternal temporal experience is registered in a voice which moves between an empathetic attunement and the courage of resistance, we contend that we listen to the maternal voice as a paradigm for the perlocutionary fluency and moral force of passionate utterance.

Thus, by working at the intersection of Austin's performative speech act theory, Cavell's account of passionate utterance, Levinas's concept of illeity, and Husserl's concept of motivational empathy, we present an account of the maternal voice which defies the conventional characterizations of the maternal as either essentially sentimental and forgiving or as ultimately vigilant and self-sacrificing and rather finds itself alternating between these two characterizations, that is, moving between empathy and courage. Accordingly, we suggest that the intersection between empathy and courage be identified as the "virtue par excellence" of the maternal. In support of this suggestion, I bear witness to the testimony of empathetically courageous young women, who in "becoming mothers," resisted





the atrocities and horror of the war, all the while protecting and safeguarding their family's future.

7. Maternal Resonance: "Becoming Another's Mother" during the Second World War

In arguing that the lived tension between a phenomenological experience of temporal continuity tangentially coupled with an experience of unpredictable temporal disruption constitutes an affective tension which resonates within the maternal experience, I explore the ethical resistance and the exigency of decisions and actions made by mothers and those who acted like mothers of others - as reflected in maternal narratives of the Shoah. I show that such "mothers" recount similar experiences of affective tension resulting in creative improvisation and morally responsive resistance. Not only do the narratives of the war provide us with an historical testimony of the experiences of women who took on maternal roles, without necessarily being mothers themselves, these narratives moreover provide us with a critical educational model for ethical resistance and action. In order to support my analysis, I rely on personal interviews with children/adolescents who lived within the current day boundaries of Slovenia and who unpredictably and bravely acted as mothers to protect the lives of others.

During the Second World War, Slovenia found itself to be an 'in-between State,' which magnified the struggles of those living within its boundaries. Accordingly, I contend that the radical precarity of the Slovenian experience is reflected in the narratives of children survivors. In order to accomplish this, I interview, with the assistance of a translator, two living survivors of the concentration camp, a young boy saved by his nanny, and a young girl who was taken care of by a peasant mother. By amplifying the resonance and the audacity of the experience of those who acted like mothers during the horrors of the war, I articulate a new way to re-think the "tensed time" of the maternal experience. I propose that such a maternal ethics be conceived as being available not exclusively to biological or adoptive mothers. Rather, I suggest that such an ethics provides an educational model of resistance for all to combat the current rise in fascist sensibilities. In support of my articulation of the tensed time of maternal ethics, my research highlights the improvisational and intuitive timing of passionate maternal utterance and argues that the maternal voice calls upon a continual negotiation and temporal openness to an intuitive ethics of disruption while maintaining a promise of care.

7. Conclusion

In exploring the ethical exigency of the experience of the maternal voice as heard through wartime testimony, I contend that the temporal experience of the maternal emerges as a risky yet possible harbour for the competing yet irrecuperable forces, one of temporal continuity and endurance and one of disruption and rupture; as such, the maternal voice singularly commands our attention as a source for ethical creativity, resistance, and action. The precarity and unpredictability of the maternal voice contributes to its compelling quality; always on edge, the maternal stakes itself out to secure a future for the next generation, all the while providing compassion and empathy within the present extension of the past. The experience of rupture coupled with a shared sensibility of enduring temporality forces our attention and constitutes that which we argue is the uniquely compelling mark of the maternal, namely, the possibility of becoming a mother to another through an ethics of empathetic courage.

Accordingly, we propose that through maternal voicing, we might learn to unsay – or disrupt - that which has become dangerously and tacitly accepted. By listening to the voices of children who became mothers during war time, and by listening to the narratives of mothers who saved others' children, we hope to amplify the courage, generosity, audacity and resistance of these voices in an effort to articulate and nurture our common human sources of resistance, empathy, and courage.

Rather than honoring the memory of a person as a past memory intact as an identity, the testimony of the survivors which I have collected reveals a diachronic experience of time, which ruptures a conventional sense of memory, thereby, bearing witness to the vital possibilities of the next generation. Through the exigency of moral time, our research likewise honors he survivors of the concentration camp and the children victims of war who acted "as second mothers," fighting courageously for freedom, and generously risking







death to save the future lives of others. We argue that it is the unique tension between the irrevocable testimony of an immemorial past and a prophetic voice which finds itself as directed toward an unknowable future which informs the maternal testimonies of those who experienced firsthand the horrors of war. We further claim that it is the experience of temporal tension, as demonstrated through the courage and empathy of those who acted like mothers and as expressed by those who mourned and lamented the loss of their mothers, which elicits a powerful source of moral resistance and provides us with a critical antidote to the violence and debasement of the most vulnerable.

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Research

On-line Art Therapy for a Woman with Uterine Cancer: Integrating Sensorimotor Art Therapy through Guided Drawing with (TT-AT) Protocol

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Abstract:

Art therapy offers a safe space for individuals to process complex emotions and regain control over their narrative, particularly in the context of chronic illness such as cancer. In the wake of the COVID-19 pandemic there has been an increased use of virtual platforms by art therapists, allowing people with geographic and other logistical challenges access to mental health support. A 45 year old woman undergoing treatment for uterine cancer, with no access to in-person therapy, was the client in this 10-session art therapy intervention that explored the integration of Sensorimotor Art Therapy through Guided Drawing with the Trauma Treatment through Art Therapy (TT-AT) protocol, delivered via Zoom. Each session offered structured art-making along with free expression that helped the participant feel empowered, even in the virtual setting. The intervention was very effective and the benefits reported by the client such as improved sense of self, reduced anxiety, and increased hope and vitality, highlight the growing potential of integrating trauma-informed therapies and protocols to support individuals through their unique life experiences, particularly when paired with the accessibility of virtual delivery.

Key words: Art therapy; Sensorimotor; Cancer; Online therapy; Protocol





1. Introduction

1.1 Context- Cancer as Trauma

Over the past decade, the incidence of cancer has increased globally. Uterine cancer or Endometrial Cancer (EC) is the most common form of gynecological cancer and is the sixth most common cancer among women in the world. Based on FIGO guidance, this type of cancer is categorized into stage I, II, III, and IV and Mazidimoradi et al. (2024) report that survival rate is high because of early diagnosis. A review by Wang & Feng (2022) substantiates that although the mortality rate of cancer has decreased, cancer patients and survivors can experience several psychosocial challenges such as deterioration in sense of self, disturbance of body image, sexual problems and difficulties in social relationships while attempting to cope with daily life. The average age of onset of the disease is in women above 60, but Bassette & Ducie (2024) observe that there is a growing incidence of EC among women under 50. Poor psychological well-being of the patient could impact their adherence to treatment, Quality of life (QoL), and overall health outcomes, report Fereidouni et al.(2024), which can add to the global burden of care.

The diagnostic criteria of Post Traumatic Stress Disorder (PTSD) was adjusted in the Diagnostic Statistical Manual of Mental Disorders Fourth Edition (DSM-IV) to include the diagnosis and treatment of a life-threatening illness such as cancer, as a traumatic stressor that can result in PTSD (Leano et al., 2019). The Physician Data Query (PDQ), the National Cancer Institute's source of comprehensive information on cancer (PDQ Supportive and Palliative Care Editorial Board, 2023) report that no specific therapies for Post Traumatic Stress (PTS) symptoms in the cancer setting have been developed, and recommend that treatment modalities used with individuals with PTSD can be used in alleviating distress in cancer patients and survivors. Levine (2008), believes that trauma is unique to each individual and that the healing process of trauma can be a catalyst for profound awakening; a portal opening to emotional and spiritual awakening.

1.2 The Client

Mariana (pseudonym), a 45 year old woman living alone as an expatriate in Spain was diagnosed with uterine cancer FIGO grade III, and had undergone a complete hysterectomy. The parameters of confidentiality, anonymity, and privacy were thoroughly discussed, and an information sheet was emailed to the client. The client provided signed informed consent for both the treatment and the use of information, images and recorded media content for academic and research purposes, expressing her willingness to have her experience benefit others. She also shared relevant medical records regarding her diagnosis and treatment.

Complications of surgery led to severe pain in Mariana's back and the need for the use of a wheelchair for mobility. As a result she was unable to work and although her treatment was funded through government agencies, due to other expenses, she became financially unstable. Although she was offered free face to face psycho oncology, she felt it was insufficient to address her emotional distress. Her mobility and financial limitations made it difficult to find an art therapist who she felt at ease with in Spain, where she lived, so Mariana contacted the therapist living in Dubai, United Arab Emirates. A year prior to this, the client had interacted with the therapist online, in a non-therapeutic setting, and was familiar with the therapist's ability to work with hard to reach clients. She contacted the therapist and by appointment, the therapist met Mariana via Zoom. After discussing her medical history and treatment and seeing her visible distress, the therapist agreed to her request of providing art therapy pro bono (free). At the time she began Art Therapy, Mariana had completed nine months of treatment for cancer. She had not responded well to chemotherapy, had begun immunotherapy; was continuing with physiotherapy and albeit painfully, she was able to walk around her room without the aid of her walking





stick. Mariana had the support of a family member who was visiting and a community health worker who made regular home visits to assist her with healthy meal planning and preparation. She was a spiritual person and her strong religious faith and church community provided some degree of emotional support. Although she had no prior experience of art psychotherapy, Mariana enjoyed art making as a hobby. She was fluent in Spanish and English and had good computer skills. Her posture and presentation suggested underlying pain and fatigue; she had not lost her hair during chemotherapy and was of average height and weight. She expressed extreme feelings of anxiety, loneliness, vulnerability and loss of a sense of purpose and identity.

2. Approach-Theoretical Framework and Rationale

2.1 Art Therapy

Art Therapy traditionally focuses on therapeutic image-making and the cognitive or symbolic interpretation of these creations. Positive results in clinical practice have been reported in the use of Art Therapy for the treatment of cancer (Schouten et al., 2015). In addition to this, Van Lith (2014) posits that art making provides a spiritual aspect to the recovery process which she views as a rebuilding of a sense of self. She adds that art making serves to inspire meaning making, and a sense of purpose and hope. A systematic review of the effect of art therapy in women with gynecological issues (Fu et al., 2020) shows that diagnosis of cancer is traumatic and can cause psychological distress. Another review (Jiang et al., 2020) shows that art therapy reduced symptoms of anxiety significantly, had a positive effect on quality of life (QoL) and symptoms in cancer patients, and can be used as a complementary treatment for cancer patients. However, Fu et al. (2020) argue that while this may be true, the quality of the current evidence limits the efficacy of these findings. They add that research on art therapy with patients with gynecological cancer is insufficient and that more rigorous research is needed.

2.2 Online Art Therapy

The Covid-19 Pandemic forced the need for art therapists to adapt their practice and move to online delivery of art therapy with little or no support (Zubala & Hackett, 2020). Zubala et al. (2023) report that online art therapy is not only a feasible intervention, but can also potentially be a powerful and impactful one, capable of facilitating a positive change in a short period of time. They recommend exploring ways to expand and introduce new art therapy services. British Association of Art Therapists (2021) advice that clear communication parameters, protocols of safety and information sharing are set prior to the onset of online delivery.

2.3 Need for an Art Therapy Protocol and Paola Luzzatto's TT-AT Protocol

The practice of art therapy is eclectic. Treatment protocols like the Trauma Treatment through Art Therapy (TT-AT) protocol pioneered by Paola Luzzatto (Luzzatto et al.,2021) describe the steps of the art therapy interventions along with therapeutic goals simple language which make it easy to replicate or successfully adapt (Pearce et al., 2023) for diverse populations and efficient research. Although TT-AT was designed as a group intervention, Pearce et al. (2023) report success and advocate further research exploring the use of the protocol in individual settings and Luzzatto et al. (2021) suggest that additional sessions could be added to address specific symptoms that emerge.

Developed in Tanzania by a group of health professional at Muhimbli University, Dar es Salam, the protocol design followed the recommendations of the International Society of Traumatic Stress Studies (STSS), which had three basic concepts (Pearce et al., 2023; Luzzatto et al., 2021):





- 2.3.1 The treatment should address the patient's needs, more than focusing on trauma exposure which often leads to re-traumatising clients or result in clients dropping out from treatment
- 2.3.2 The treatment targets should be emotional regulation, relationship with others, relationship with self, gradual exposure to trauma and personal resources. These are described as the six 'symptom clusters' and their related needs.
- 2.3.3 The TT-AT protocol should follow a flexible sequential approach in three phases: a) self-strengthening and stabilisation b) awareness of the trauma memory and c) resources for present life. Strengthening interventions should be provided throughout the treatment.

2.4 Sensorimotor Art Therapy

Sensorimotor Art Therapy through Guided Drawing, developed by Cornelia Elbrecht, is a body based , trauma informed art therapy modality that recognizes the role of the body in storing trauma (Van der Kolk, 2014) and emphasizes the role of the body in processing traumatic memories and regulating arousal. Like Somatic Experiencing (SE) (Levine,1997), Sensorimotor Art Therapy is a Bottom-Up processing approach, that begins with mindful awareness of bodily sensations and gradually moves up towards cognitive processing. This approach allows the integration of somatic experiences disconnected from the client's conscious awareness. Trauma manifests physiologically as blocked internal movement which is commonly experienced as tension, pain or numbness, and can be accompanied by strong emotions. The focus in sensorimotor therapy is on restoring this internal movement(flow) and not on understanding the trauma story or the exposure of past trauma.

Guided Drawing uses slow, repetitive, rhythmic bilateral movement to explore the emotions and felt sensations in the body in the present. Lines and archetypal shapes in Guided Drawing have various qualities and can be used at the therapist's discretion to support affect regulation and the release of stored body memories in the client. Elbrecht (2021) advocates the use of the Lemniscate shape for the downregulation of emotions, promoting relaxation and supporting integration of dissociated aspects of the self and recommends that the Bowl shape be used to support clients when they need soothing, comforting or a need to be held.

2.5 Benefits of Integrating Approaches

Elbrecht (2021) suggests that a combination of guided movement, body awareness exercises, and creative activities, can help individuals access and process traumatic memories in a safe and controlled manner to address not only the cognitive aspects of trauma but also its somatic and emotional components. There is very little or no research available on online delivery of Guided Drawing or on integrating Sensorimotor Therapy through Guided Drawing Art Therapy with a TT-AT protocol. However, studies

(Hetherington, 2024; Hetherington & Gentile, 2022) report the success of integrating psychodynamic art therapy with SE, advocating that such a combination of psychological and physiological techniques complements each other. Cornelia Elbrecht (Elbrecht, 2021) supports this perspective, noting that body-focused art therapies can be particularly effective when paired with trauma-informed therapy.

This paper is based on an Art Therapy intervention delivered online via Zoom exploring the integration of Sensorimotor Art Therapy through Guided Drawing with a TT-AT Protocol with Mariana (pseudonym), a 45 year old woman undergoing treatment for Uterine Cancer. The structure followed for the 10 sessions was based on the TT-AT Protocol (Pearce et al., 2023; Luzzatto et al., 2021).





3. Methodology

3.1 Pre - therapy agreements via zoom (90 minutes) - a week before therapy began.

The client was receptive to the therapist's suggestion of adapting the Trauma Treatment through Art Therapy (TT-AT) protocol, pioneered by Paola Luzzatto, which had been successfully implemented with various populations across different countries. Sensorimotor Art Therapy, using Guided Drawing, would be combined with the protocol to help resource the client - supporting grounding, self-regulation, and the processing of emotions that surfaced during therapy. A brief, focused approach was agreed upon for a five-week period via Zoom, consisting of a total of 10 sessions, held twice a week on Mondays and Wednesdays (which best suited the client). Each session was set for a duration of one hour. Considering the different time zones in which the therapist and client resided, a mutually convenient time for therapy was selected. The client was free to express herself in any language during the sessions but was asked to translate into English whenever possible.

All sessions would be recorded by the therapist and the client agreed to take clear photographs of all the images made in the session and send it to the therapist via email within 24 hours of completing each session. The client would retain the hard copies safely in a file till the end of the course of therapy. To ensure proper delivery of the TT-AT protocol and prevent bias, the therapist would receive fortnightly online supervision during the course of therapy from Emma Mills- refer Pearce et al. (2023) p.8, trained by Paola Luzzatto, in the TT-AT protocol.

The table and laptop camera setup in the client's space was adjusted to ensure that the therapist would view both the client and her image-making process during sessions. The client agreed not to use a virtual background. A list of art materials, was suggested. The client, being familiar with art-making, already had a good variety of art materials available. A safety protocol was discussed, outlining what the client should do and whom she should contact in case of emotional distress between sessions. The therapist's emergency contact information was also shared. Additionally, the client was encouraged to maintain a journal throughout the course of therapy.

3.2 Structure of each session

Each 60 minute session included the following elements:

- 3.2.1. Introduction involving a brief check-in on the client's emotional state and physical health and a slow breathing exercise where both client and therapist (co-regulation) take three to five deep breaths with slow exhaling.
- 3.2.2. Psycho education: Introduction of the day's activity, the purpose of it and how it could benefit the client (**Table 1**).
- 3.2.3 Art Making using a Directive (TT-AT): The Protocol has six directives that could be used for six sessions or expanded to be delivered through more sessions based on client needs as therapy progressed. (i) The Body Outline; (ii) A Positive Memory; (iii) The Hidden Seed; (iv) The Sad /Angry Child; (v): The life line; (vi): Resources
- 3.2.4 Free Art/Image making and or Sensorimotor Art Therapy (Guided Drawing): The second part of each session would emphasize free image-making with a focus on expressing emotions that surfaced during the process of making the first image or Guided Drawing through bilateral drawing
- 3.2.5 Reflection: The final 10 minutes of each session involved reflection of the session where the client and therapist jointly looked at the images created.





Table 1- Trauma treatment through art therapy (TT-AT) (Luzzatto et al., 2021).				
	Sessions	Focus	Goal	Art therapy workshops
Phase 1	1	Emotional dysregulation	Expression and containment	"The Body Outline"
	2	Disturbed relationships	Re-activating positive relationships	"A Positive Relationship in
				Childhood"
	3	Negative self-concept	Contacting inner energy and hope	"The Hidde Seed"
Phase 2	4	Avoidance and numbing	Symbolic exposure to trauma	"The Sad/Angry Child"
	5			"My Life Line"
Phase 3	6	Present life challenges	Personal resources and coping	"Second Body Outline"; "My
			strategies	Resources"

A non interpretative and non-judgemental approach was decided upon. Note: After the three sessions of Phase 1 (**Table-1**), two sessions would be used to consolidate the surfaced awareness of emotions and strengthen the client before beginning Phase2. This would be done through Guided Drawing of the lemniscate and bowl shapes, followed by free image making (art making with no directive) in each session. After completion of all the TT-AT Protocol directives, any remaining sessions of the ten, would be used for free image making and Guided Drawing.

3.3 Data and Evaluation of Progress: The effectiveness of the intervention was assessed qualitatively and quantitatively

3.3.1 Qualitative

Verbal and, or written feedback from the client (every third session), therapist observation and supervision.

3.3.2 Quantitative: Clinical Outcomes from Routine Evaluation (CORE 10): The clinical score from the CORE 10 evaluation sheets completed in sessions 1, 6 and 10 would be compared at the end of therapy (Barkham et al., 2013)

4. The Practice - The Art Therapy Intervention

4.1 Session 1- The Body Outline

Goal: Emotional self-efficacy through expression and containment: The client was invited to: Step1- Draw a body outline on white paper and locate difficult emotions she was experiencing in her body. Step 2. Choose a colour for each of these emotions and colour the area inside the body outline where she felt that emotion. Step 3. Choose a colour that feels healing to her at the present time and trace it slowly around the body outline, symbolically creating a contained space to hold in all these difficult emotions. She was given the option to write what she felt were some positives in her life that made her feel good outside the outline. Client response: (Figure 1) Mariana (pseudonym) identified and located the emotions and wrote the name of the emotion in the colours of her choice inside the outline using sketch pens. She chose blue as the healing colour and instinctively drew 2 outlines outside the body outline instead of one. Inside the Body Outline she wrote the words (translation): tension, fear, doubts, faith, pain, loneliness, stagnation, resilience, search for peace, I want to say what I feel but I limit myself, I would like to be focused. Outside the outline, 'words of protection' were written: strength, love, faith, hope, good friends, elegant, studious, intelligent, wise, generous, ambitious.









Figure 1A: Session 1, Image 1: "Body Outline"; B: Session 1, Image 2: "My Safe Space".

Translation of **Figure 1A**: "I am afraid of being stagnant, alone, without purpose and without being able to realize my dreams. I need to learn to appreciate myself more, and be able to see the value and potential that others see in me as a treasure."

Free Image: "My Safe Space". Draw using the "healing colour". Translation of **(Figure 1B):** "Cancer is not my identity. My identity is protected and will be released."

4.2 Session 2- Memory of a Positive Relationship in Childhood

Goal: Re-activation of a positive relationship as a resource: The client was invited to create an image about a positive relationship from her childhood (**Figure 2A**). Client response. Gave the title "The two Marianas" and said there were and still are two parts of her. "It is not schizophrenia, but I know there are two separate parts."

Translation of **Figure 2A**: "Thank you for coming into my life. You are there, I am standing here and I need to find strength by myself and be brave by myself now. I have a new way, a new hope". Free Image (**Figure 2B**): The client created a collage with magazine cuttings and worked in silence. Client response: "Enjoy today. I am weak, but I am still standing. The water is calm but can have stormy waves, peaceful but turbulent. The boat is inviting me to live my belle vie (beautiful life) in this time and be free".





Figure 2A: Session 2, Image 1: "The Two Marianas"; B: Session 2, Image 2: "Free image".

4.3 Session 3 - The Hidden Seed

Goal: Getting in touch with inner energy and cultivating hope: The client was told that the session had 3 steps and she was invited to focus on her feelings. Step1 - Create an image of 'a barren landscape'. A landscape where there was no life. Step 2 - Imagine that somehow, one seed survived in that landscape, all alone. Mark a small dot to indicate where the seed was in this barren landscape (Figure 3A). Step 3 - Imagine the seed growing. What kind of plant would it grow into, and what did it need to grow and be nurtured? Illustrate this (Figure 3B).









Figure 3A: Session 3, Image 1: "Barren Landscape with seed"; B: - Session - New growth

Therapist: If the seed that was hidden could talk, what would it say to you? Client response: "No one is attacking you now" she said "I used to build walls to protect myself and people thought I was a strong woman. I was vulnerable and felt frail, but people did not know. Maybe people can't see the real me because of my walls. Maybe I should take a risk and show myself, but it is frightening".

4.4 Session 4: Review, Guided Drawing and Free Image Making

Goal: Building resource: Review: Mariana said she was comfortable with the pace of therapy and said she reviewed the images between sessions and had begun keeping a journal. Guided Drawing of lemniscate shape, (Figure 4A), using both hands in parallel and simultaneously moving the upper body creating a horizontal 8. She mimicked rocking a baby in her arms and said she felt she was being rocked like a baby. This was followed by drawing of free image (Figure 4B). Client response: "These are the four feelings I am experiencing now. Love, Patience, Resilience and Goodness". Daily practice of the bilateral drawing of the shapes introduced in the session was encouraged during the week to support grounding and downregulation between sessions.

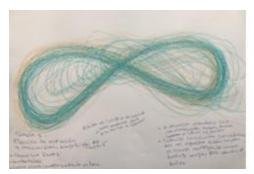




Figure 4A: Session 4, Image 1: "Lemniscate"; B: Session 4, Image 2: Free image

4.5 Session 5: Guided Drawing and Free Image Making

Goal: Building resource: Guided Drawing (Figure 5A). Client introduced the bowl shape and included horizontal lines as a base for the bowl. She engaged well with the bilateral drawing, and was encouraged to work slowly and find a rhythm. She was comfortable trying this with her eyes closed . Free image making (Figure 5B). Client response: "I was in a cave. Now I feel the walls that I have put up are cracked open and the light is coming through. I want to allow the light to come in - open wide the black area - and let more of the yellow light in."









Figure 5A: Session 5, Image 1: "Bowl"; B:Session 5, Image 2: Free image.

4.6 Session 6: Sad /Angry Child

Goal: Symbolic exposure to trauma. The client was invited to divide a white sheet of paper into 3 panels. Step 1: In the first panel, draw a boy or a girl experiencing a sad or angry feeling; Step 2: In the second panel, draw what caused the child to feel sad or angry; Step 3: Now draw how or what the child needs to be happy in the last panel. (Note: it was recommended during supervision that in TT-AT Protocol that the angry/sad child should be drawn in the middle panel and the image showing cause of sadness in the first panel, so that the child gets a central placement). First panel - A boy's birthday party (the child was missing in the picture she drew (Figure 6A)). Client response: "It is a sad event, because although cakes and gifts are present, the atmosphere was not good". Second panel - "He is sad because the family is broken. Always noise and parents fighting". Third panel was broader and she drew a big arched rainbow, a plant that looked like a child, outline of two hearts and two overlapping circles. "Love, respect, family unity and support can 'perhaps' make the child happy". Free image making (Figure 6B): "These stripes I painted are different stages of my life. Good weather (good times) will come. I am working towards that. I am not just dreaming it will happen. All this, the therapy and the art are for that. It is difficult but I am making it happen."

Client was asked by the therapist if therapy was supporting her and how. Client response: "I feel I have received an awakening, I feel I have been shaken awake, I am not alone, I have people, I am alive, I still have goals, purpose, but different ones. I have to make changes. I have tools - my education, my art. Cancer is not my identity".



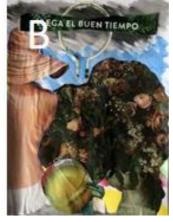


Figure 6A: Session 6, Image 1: "Sad/Angry Child"; B: Session 6, Image 2: Free image





4.7 Sessions 7 and 8 - My Life-line

Goal: Facilitating emotional distance and integration of trauma

The client was invited to draw a horizontal line from one end of the paper to another. The line could be straight or wavy.. On one end of the line she was asked to put her date of birth and to put the present time on the right side. On this line, using symbols, she was asked to mark the life events that felt most important; the positive events above the line, and the negative events below the line. She could write when this happened and what the event was next to the symbols. The drawing is shown in **Figure 7A**. There were more symbols above the line than below the line, but she said the impact of the negative experiences overshadowed the positive. There was no writing on the image and she sat looking at it silently. She spoke of some positive experiences, but was reluctant to revisit the past experiences, because she said it was over and she wanted to leave it in the past, but was willing to think about it if it was beneficial for her progress.

The client willingly engaged , saying she had time to reflect after session 7 and felt that this was her chance to get things out of her and be heard. She looked at the lifeline again (Figure 7A) and spoke graphically about past issues of neglect, deprivation, multiple forms of abuse and said that shame about these events had forced her into silence. She was tearful and spoke slowly through her tears. Towards the end of the session she recognized a feeling of overwhelm in her body and requested an activity to help her 'feel better'.

Guided Drawing-Vertical upward and outward strokes (pushing away) was suggested, followed by the now familiar lemniscate shape. The therapist gave verbal instructions and made the movements alongside the client and then gave her the freedom to continue alone, guided by what she felt she needed. The drawing is shown in **Figure 7B**. She found her rhythm and chose to engage with her eyes closed and using the palms of her hand instead of oil pastels, saying words of affirmation loudly: "I am not alone. I am not a victim, cancer is not my identity". At the end of the session she held her stomach and exclaimed: "I feel hungry"! Note: Post session, she added the words of affirmation onto the image.





Figure 7A: Session 7: Lifeline; B: Session 8: Guided drawing.

4.8 Session 9 - Resources Second Body Outline

Goal: Addressing present life challenges.

This session involved a final review of the clients therapeutic journey and discussion of strengths and resources. The client was invited to arrange all the images made in the session side by side in chronological order for a final review. She assigned titles to each image: Session 1- The beginning, Session 2 - Mariana's (pseudonym) meet, Session 3 - Sincerity, Session 4 - A truth that lulls and liberates Session 5 - An enlightened time opens up, Session 6 - Good weather arrives, Session 7 - The line that broke the silence, Session 8 - The owl of wisdom. She was then invited to create a body outline (**Figure 8**) on a piece of white paper, as she did in session 1 and compare the two images. Client response:





Translation of words written outside the body outline: There are no more fears, my loneliness is beautiful and full of wisdom, I have understood the purpose of friendship, I am free, I am alive, I exist, I feel safe, I am loved and important. Note: The client was informed that the next session would be the last and to come with any queries she had. In preparation for the next session, she was asked to identify personal interests and resources that she had access to and think of how these would support her after therapy.



Figure 8: Session 9: Body Outline.

4.9. Session 10 - What Next

Goal: Closure and building resource

Discussion of strategies moving forward: Mariana had identified swimming, yoga, meditation, art making, pilates, prayer, reading and writing. She had a desire to give back to the community in some way. She spent the rest of the session doing Guided Drawing independently with pastels and paint, using the palms of her hand and with her eyes closed. A 60 minute review session via zoom was scheduled after 4 weeks.

Table 2. CORE 10 Clinical Scores based on (Connell & Barkham, 2007)

Session	Clinical score	Interpretation
Session 1	20	20-24 Moderate to severe psychological distress
Session 6	12	11-14 Mild psychological distress
Session 10	6	6-10 Low level distress

5. Results

A CORE 10 clinical cut off score of 11 or above indicates clinically significant distress (Barkham et al., 2013). Analysis of CORE 10 **(Table 2)** shows that the client's distress has reduced significantly from a score of 20 at the start of therapy to a score of 6 at the end of therapy. Although she still experienced low level of distress, she reported an increased levels of hope and ability to cope. The integration of Trauma Treatment through Art Therapy (TT-AT) Protocol and Sensorimotor Art Therapy through Guided Drawing proved to be a very effective intervention for Mariana (pseudonym), a 45-year-old woman





undergoing treatment for uterine cancer. Throughout the sessions, the client demonstrated increased verbal emotional expression, particularly regarding feelings of disempowerment, and fear. As the sessions progressed, the client reported a growing sense of hope, empowerment, and self-worth. The virtual delivery of therapy offered easy access for the client from the comfort of home.

6. Discussion

The TT-AT protocol provided a structured but flexible framework that was easy to replicate and expand by the therapist. The protocol's three-phase structure - selfstrengthening, awareness of trauma memory, and building resources for present life - and the art-making directives facilitated the externalization of emotions and allowed her to connect with the bodily and emotional pain rooted in past trauma. Bilateral drawing techniques, such as the Lemniscate and Bowl shapes, were particularly effective in helping her regulate emotions and promote relaxation and enabled her to integrate fragmented aspects of herself. One of the key benefits of this combined approach was its facilitation of holistic healing. This integration of mind-body healing highlighted in this study aligns with the growing recognition of somatic approaches in trauma treatment, particularly for individuals managing chronic illnesses like cancer. The virtual format of therapy made this intervention particularly well-suited for the client who faced challenges accessing inperson therapy. The therapist found it comfortable to hold space via the Zoom platform, but recommends that a 12 session model beginning with Bilateral drawing techniques and free image making and building up to the integrated approach used in this study could benefit clients with complex trauma. The client's access to images fostered a sense of connection with the therapy and therapist between sessions, allowing her to feel safe and supported. During the review after four weeks the client reported that she was responding well to cancer treatment and was moving from living alone to living as a paying guest with a young family. She continues with journaling and guided drawing. Lastly, the therapeutic relationship played a critical role in ensuring Mariana's active participation. Her preparedness, willingness to engage deeply in the therapeutic process, and insightful reflection were essential factors in the success of the intervention. This underscores the importance of cultivating a strong, collaborative therapeutic alliance in facilitating healing, especially in virtual settings.

7. Conclusions

This model highlights the growing potential of integrating creative, trauma-informed therapies and protocols to support individuals through their unique and complex life experiences, particularly when paired with the accessibility and convenience offered by virtual delivery.

Institutional Review Board Statement: The client provided signed informed consent for both the treatment and the use of information, images and recorded media content for academic and research purposes.

Conflicts of Interest: The author declares no conflict of interest.

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Research

Production of Plant Biostimulants from Microalgae Grown on Biogas Digestate and Tests on Selected Crops

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Abstract:

This study investigates the potential of microalgae-based biostimulants cultivated on the liquid part of anaerobic digestate as a sustainable solution for modern agricultural chal-lenges. The liquid part of anaerobic digestate, a nutrient-rich by-product of biogas pro-duction, is used as a growth medium for the microalgae Scenedesmus sp. to illustrate the principles of circular bioeconomy by converting organic waste into valuable agricultural products. The microalgae Scenedesmus sp. were cultivated in raceway ponds, har-vested using sedimentation, microfiltration or centrifugation, and further dried in venti-lated cabinet at 35°C. Laboratory germination tests and field trials were conducted to eval-uate the efficacy of the produced algae as plant biostimulant. In laboratory tests microal-gae-derived biostimulants significantly improved seed germination and root develop-ment of garden cress at all concentrations, Proso millet gave positive response to 1 and 5 g DM/L, however cabbage showed no response. Foliar applications on Proso millet in the field trial further highlighted its potential to improve crop yields stability across the field after but not increased millet grain yield. This research highlights the viability of integrat-ing microalgae cultivation with waste management systems to produce biostimulants that can improve crop production while supporting sustainable agriculture.

Keywords: Circular bioeconomy; Sustainable agriculture; Anaerobic digestate; Nutrient utilization; Microalgae,;Plant biostimulant





1. Introduction

Microalgae-based biostimulants are emerging as innovative tools in agriculture to address critical challenges posed mainly by climate change, such as drought, extreme temperatures, irregular rainfall and soil degradation. These biostimulants are known to promote seed germination, enhance root system development and improve nutrient utilization, thereby increasing plant resilience to environmental stresses (Van Oosten et al., 2017). The biostimulant potential of microalgae is strongly influenced by algae growing conditions, with the availability of nutrients and environmental stresses playing a crucial role in determining their biochemical composition and efficacy (Petkov et al., 2009). A sustainable approach to the production of microalgae-based biostimulants involves the cultivation of microalgae on the liquid fraction of anaerobic digestate, a nutrient-rich by-product of biogas production (Barzee et al., 2022). Biogas plant digestate, derived from the microbial decomposition of organic waste, is an excellent example of the principles of the circular economy, transforming agricultural and industrial waste streams into valuable inputs for crop production. The liquid fraction of the digestate, which is produced after mechanical separation of the solid components, is rich in nitrogen, potassium, calcium, sodium, and trace elements, making it a cost-effective and nutrient-rich medium for the cultivation of microalgae such as Scenedesmus sp. (Resman et al., 2021). The use of anaerobic digestate as a growth medium offers several advantages. It supports efficient nutrient utilization by microalgae, which promotes the production of secondary metabolites, including amino acids, polysaccharides and other bioactive compounds that are critical for their biostimulatory properties (Hossain Sani et al., 2024). In addition, this approach reduces reliance on mineral fertilizers, which production is energy-intensive and has negative environmental externalities, further contributing to the sustainability of agricultural practices. Biostimulants from microalgae grown on digestate have been shown to improve plant resilience by promoting seed germination, stimulating root development and increasing plant tolerance to abiotic stress factors such as drought and salinity (Bulgari et al., 2014). Their application also benefits soil health by increasing microbial diversity, which is all in line with the principles of sustainable agriculture and regenerative farming systems (Win et al. 2018; Gonçalves et al., 2023). This study investigates the cultivation of microalgae in growing media containing water solution of anaerobic digestate as a sole source of nutrients and evaluates the effectiveness of the resulting algae biomass as plant biostimulants in laboratory and field trials. By integrating waste management and agricultural innovation, this approach offers a scalable and environmentally friendly solution to improve crop productivity and soil health, thus contributing to global sustainability goals.

2. Material and Methods

The biogas plant digestate used in this study was derived from the anaerobic digestion (AD) of food production residues (KOTO d.o.o.). The process is thermophilic (55-55.5°C) with separate hydrolysis at pH 3.5-4 and digester reactors at pH 7.8-8.0 and thus differs from conventional mesophilic processes. Mixing is continuous. Regulation of pH is only minimally regulated; soda is occasionally added (e.g. NaHCO₃, Na₂CO₃) (Lavrič, 2019). The system in this study includes a screw press for digestate separation to liquid and solid part. The solid digestate is transported for drying and the liquid digestate which is rich in essential nutrients such as nitrogen, phosphorus, potassium and trace elements can be used for microalgae production. For the growth of the green microalgae *Scenedesmus* sp. (dominant *Scenedesmus quadricauda*) a liquid part of the separated digestate was used as a source of nutrients (Table 1). The addition of digestate is regulated based on the basis of the limit values of the indicative parameters defined in previous experiments: pH 6-8, electrical conductivity EC 1000-2000 µS cm⁻¹, NH₄-N 50-150 mg L⁻¹, NO₃-N 5-50 mg L-1, NO₂-N <1 mg L⁻¹, optical density OD680 from 1 to 1.5 (Resman et al., 2023). The input of digestate is about 4 to 10 L per 1000 L of growing medium per day, depending on the cultivated microalgae and intended product. The nutrient content of the digestate and the produced microalgae biomass was analyzed, including parameters such as





electrical conductivity (EC), dry matter (DM), organic carbon (Corg), nitrogen (N), phosphorus (P), potassium (K), and the C:N ratio. Additionally, the levels of micronutrients and potentially toxic elements in microalgae grown on digestate (Pb, Cd, Zn, Cr, Cu, Ni, Hg, As) were measured and compared to regulatory limits.

The microalgae cultivation on digestate took place in an unheated greenhouse with a double PE foil roof at the Center for Algal Technologies of the Biotechnical Faculty, University of Ljubljana. The microalgae were grown in open race-way ponds, which were heated only at the bottom to prevent freezing in the winter. The effective production period was approximately 240-280 days per year. The average daily biomass production was 11±1.5 g/m² at temperature between 10 and 30 °C under optimal conditions, from March to October. The microalgae were harvested using sedimentation (for 24 hours) and later filtration of the sedentary part with a vibro-microfiltration unit (Vibro-Lab 3500, SANI Membranes A/S, DK) to obtain ca. 7.5 % DM and/or centrifugation (rpm 12 000; max. rcf 27 500; $T = 15^{\circ}C$; 10 min; V = 250 mL) to achieve a dry matter content of approximately 15 %. The obtained algae paste could be used fresh or dried at 35 °C and ground into a fine powder. Obtained powder was diluted with water to a concentration suitable for application as plant biostimulant. The biostimulants were tested for their effectiveness in promoting seed germination, root development and crop yield. Germination tests (Figure 1) were carried out in in-vitro tests with Proso millet, garden cress and cabbage seeds, with treatments including different concentrations of the biostimulant (1 g DM/L, 5 g DM/L, 10 g DM/L, 25 g DM/L, 50 g DM/L) and deionised water as control. The second germination tests were repeated with Proso millet treated with microalgae Scenedesmus sp. biostimulant at different concentrations (1 g DM/L, 5 g DM/L, 50 g DM/L) and with growth medium after Scenedesmus sp. cultivation (V-BF). Again, water was used as a control. The average root length and germination index were measured and analyzed using the ImageJ software.

$$GI \ [\%] = \frac{\text{(no. germinated seeds * root lenght)}}{\text{(no. germinates seeds in control *root lenght in control)}} \ x \ 100$$



Figure 1. Visual example of germination test (left) and evaluation of roots length with ImageJ app (right)

The field trials were conducted at the Biotechnical Faculty in Ljubljana, where Proso millet (*Panicum miliaceum*, variety 'Sonček') was sown (on June 20, 2024), 125 kg seeds/ha. The soil is eutric-brown, pseudo-gleyic, texture sandy-clay-loam to clay-loam, with a pH of about 7. In 2024, a basic fertilization with 138 kg N/ha NPK 15-15-15 was applied, followed by 75 kg N/ha KAN 27% in the BBCH 16 phase. Four treatments were studied: control (untreated seeds and no foliar application; NS-NF), biostimulant applied to seeds (AS-NF), biostimulant applied foliarly (NS-AF), twice during the growing season (first in the development phase of BBCH 59; on August 7, 2024, and the second time in the phase of BBCH 69; on August 9, 2024) (Ventura





et al., 2020), and biostimulant applied to seeds and foliarly (AS-FA), also twice during growing season. The concentration of microalgae biostimulant for the seed application was 0,5 kg/ha and for the foliar application a concentration of 1 g microalgae DM preparation/L of water (for each application a dose of 300 L/ha). The effectiveness of the biostimulation was evaluated by measuring root depth (at BBCH 13; July 3, 2024), fresh biomass, air-dry biomass and yield. Plants were harvested (on September 4, 2024) within a 1 m x 1 m frame in three replicates for each treatment for the purposes of crop analysis. We weighed the produced biomass and grain (drying of the plants was carried out in a ventilation drying cabinet at 40 °C to a constant mass). The data obtained from the germination tests and field trials were statistically analyzed using the R statistical language (R Commander).

3. Results

The liquid part of the digestate from the KOTO biogas plant had an electrical conductivity (EC) of 14.9 mS/cm and a dry matter (DM) content of 0.74%. The phosphorus (P) content was 0.53 g/kg DM and the potassium (K) content was 6.70 g/kg DM, which are crucial for the optimal growth of microalgae like *Scenedesmus* sp. The high organic carbon (Corg) content and low Corg:N ratio further support the efficient nutrient utilization by the microalgae (Table 1). The produced microalgae biomass of *Scenedesmus* sp. grown on the liquid digestate had a Corg content of 45.0 % DM, an N content of 7.5 % DM and a C:N ratio of 6.0. The nutrient profile of the microalgae biomass showed higher levels of organic carbon and phosphorus compared to the digestate, reflecting the microalgae's ability to concentrate these nutrients (**Table 1**).

Table 1. Electrical conductivity (EC), dry matter (DM) in digestate, N, P, K and C:N value in the used liquid part digestate (KOTO) and in produced microalgae biomass *Scenedesmus* sp. (Centre of Algae Technologies, Biotechnical Faculty) grown on the liquid part of the digestate.

		Liquid part of digestate	Scenedesmus sp.
		(KOTO)	(DM)
EC	[mS/cm]	14.9	/
DM	[%]	0.74	/
Corg	[% DM]	34.7	45.0
N	[% DM]	17.8	7.5
Corg:N		2.0	6.0
P	[g/kg DM]	0.53	13.0
K	[g/kg DM]	6.70	5.70

The content of potentially toxic elements in the digestate (Fuentes et al., 2004) and in the produced microalgae biomass were measured and compared with the legal limits. The levels of elements such as lead, cadmium, zinc, chromium, copper, nickel, mercury, and arsenic were all below the permissible thresholds, ensuring the safety of the biostimulants for agricultural use. Notably, the microalgae biomass had lower concentrations of these elements compared to the digestate, suggesting that the microalgae can effectively mitigate the presence of potentially harmful substances. All values of the potentially toxic elements in the digestate and in the cultivated microalgae were within the legal limits (Regulation (EU) 2019/1009) for plant biostimulants (Table 2).





Table 2. Values of potentially toxic elements according to Regulation (EU) 2019/1009 in the biogas plant digestate (KOTO) and produced microalgae biomass *Scenedesmus* sp. (Centre of Algae Technologies, Biotechnical Faculty) grown on the liquid part of the digestate.

				Scenedesmus sp. grown	
		Liquid part of digestate	Limit values for plant	on diluted digestate	
		(KOTO)	biostimulant (DM)	(DM)	
Pb	[mg/kg DM]	9.20	120	3.43	
Cd	[mg/kg DM]	0.70	1.5	0.13	
Zn	[mg/kg DM]	277	1500	114	
Cr	[mg/kg DM]	29.0	/	3.30	
Cu	[mg/kg DM]	47.2	600	20.5	
Ni	[mg/kg DM]	8.10	50	1.90	
Hg	[mg/kg DM]	0.05	1	0.009	
As	[mg/kg DM]	<0.5	40	0.50	

The roots of germinated plants were on average longer with biostimulant concentration up to 5 g DM/L. Higher biostimulant concentrations had inhibitory effect. Optimal concentration depended on the plant species itself. For millet concentration of 50 g DM/L had phytotoxic effect (**Figure 2**).

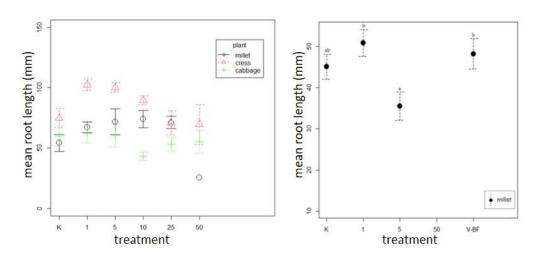


Figure 2. 1st germination tests (left figure): average **root length** of germinated plants (millet, cress and cabbage) at different treatments (1 g DM/L (1), 5 g DM/L (5), 10 g DM/L (10), 25 g DM/L (25), 50 g DM/L (50), control (K)) with standard error and 2nd germination tests (right figure): average root length of germinated Proso millet plants at different treatments (1 g DM/L (1), 5 g DM/L (5), 50 g DM/L (50), growth medium from *Scenedesmus* sp. (V-BF), control (K)) with standard error.

Additional germination tests were conducted on Proso millet seeds with different treatments of microalgae biostimulant including growth medium from *Scenedesmus* sp. This additional germination test on millet indicated that the 1 g DM/L treatment was significantly better than 5 g DM/L (p = 0.00553), with the best germination index (GI) of 113% (**Table 3**). Higher concentrations showed a decrease in germination and root development, indicating an optimal concentration for biostimulant efficacy at 1 g DM/L (**Table 3**).





Table 3. Average root length and germination index (**GI**) of Proso millet plants at different treatments (1 g DM/L (1), 5 g DM/L (5), 50 g DM/L (50), growth medium from *Scenedesmus* sp. (**V-BF**), control (**K**)) with standard error.

Treatment	Average germination [%]	Average root length [mm]	GI [%]
K	86.1	45.13	100
1	86.1	51.11	113
5	75.0	31.38	61
50	0.0	0	0
V-BF	86.1	48.90	108

Field trials were carried out with Proso millet (*Panicum miliaceum*) to evaluate the effectiveness of the biostimulant in real-world conditions. The results of biostimulant application on seeds at 0.5 kg DM/ha showed no improvement in root length at BBCH 13 (**Figure 3**). The highest plant dry biomass (dried at 35°C in a ventilated drying rack) measured at harvest was observed at foliar application (NS-FA, AS-FA) with 9.5 t/ha (the lowest at AS-NA 8.2 t/ha).

The highest average yield of dry millet grain was observed in the control (3780 kg/ha), but it showed the greatest yield variability within treatment. The lowest yield variability between repetitions was observed in the foliar application (3549 kg/ha), followed by the combined treatment with seed and foliar application (AS-FA) (3671 kg/ha). The treatment in which the biostimulant was only applied to the seed had the lowest yield (3071 kg/ha). The results indicate that the application of biostimulants can increase yield the stability across the field when applied on leaves (foliar), although the differences were not statistically significant.

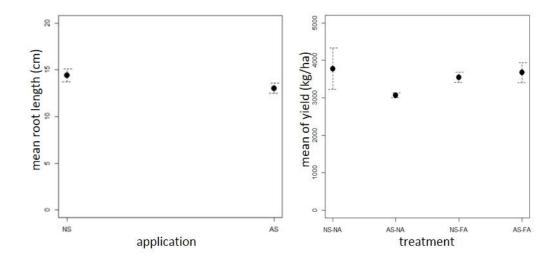


Figure 3. Field trials (Figure left): root depth [cm] of Proso millet plants in the BBCH 13 stage in untreated seed (**NS**) and in the case of seed treated with a microalgae biostimulant (**AS**) with standard error, and (Figure right): yield of dry millet grain [kg/ha] at treatments without microalgae biostimulant application (**NS-NA**), the application of a biostimulant on seed (**AS-NA**), only foliar application of the biostimulant (**NS-FA**) and the application of a biostimulant to seed and foliar (**AS-FA**) with a standard error.

4. Discussion

The results of this study demonstrate the potential of using anaerobic digestate as a growth medium for the cultivation of microalgae and the subsequent production of effective plant biostimulants. The nutrient analysis of the digestate and the produced microalgae biomass shows that the liquid fraction of the digestate provides a nutrient-rich environment that promotes the growth of microalgae *Scenedesmus* sp. The content of potentially toxic elements in the digestate and the produced microalgae biomass is within legal limits, ensuring the safety of the biostimulants for agricultural use. This is a crucial aspect for the widespread introduction of microalgae-





based biostimulants, as it minimizes concerns about the accumulation of harmful substances in soil and plants.

The germination tests showed that microalgae-based biostimulants significantly improve seed germination and root development only at the optimal concentration which was found to be 1 g DM/L for Proso millet, cabbage and garden cress. Garden cress was the most responsive test plant, but Proso millet showed the highest susceptibility; the highest germination index and average root length for millet concentrations was at 1 g DM/L. Higher concentrations resulted in a decrease in germination and root development, suggesting that there is a threshold beyond which the biostimulant effect diminishes. This result is consistent with previous studies that have determined similar optimal concentrations for biostimulant applications (Oancea et al., 2013; Alling et al., 2023; Chovanček et al., 2023). The field trials also confirmed the effectiveness of microalgae-based biostimulants in promoting plant growth and development under real-world conditions. The biostimulant treatments did not improve yield in Proso millet, although the trend towards higher yields in treated plants with foliarly applied biostimulant suggests potential benefits of biostimulant application, so the experiments should be repeated. Current challenges and opportunities in biostimulant research emphasize the need for standardized testing protocols (Roopashree et al., 2024). Future research should focus on optimizing the cultivation conditions for microalgae in anaerobic digestate to maximize the production and efficacy of biostimulants. In addition, long-term field studies are needed to assess the different environmental conditions and stressors (Khalid et al., 2024). Exploring the potential of different microalgae species and their specific biostimulant properties could further enhance the versatility and effectiveness of this sustainable agricultural practice.

5. Conclusions

In conclusion, this study highlights the potential of microalgae-based biostimulants cultivated on anaerobic digestate as a sustainable and effective solution to address the growing agricultural challenges posed by climate change and environmental protection. The reduction in reliance on chemical fertilizers combined with the transformation of waste into agricultural resources aligns with global sustainability goals and highlights the potential of this technology to support more resilient food systems. The use of the liquid fraction of digestate, a nutrient-rich by-product of anaerobic digestion, provides an efficient and cost-effective medium for the cultivation of microalgae, for species such as *Scenedesmus* sp. The results of this study show that the digestate supports microalgae growth and enables the production of biostimulants that significantly improve germination and root development of different plants. Yield increases in the field trial with Proso millet were not statistically significant, however the stability of yield across the field was improved.

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Conflicts of Interest: The authors declare no conflict of interest.

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