Review

Effect of Complexation Cyclodextrins with Phenolic Acids and Coenzyme Q_{10} on their Physico-Chemical Properties and Bioavailability

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Dedicated to the memory of the late Prof. Dr. Valentin Koloini

Abstract

This article deals with the oxidation problems of food caused by atmospheric oxygen. Two approaches to overcome these problems are involved: (i) by reducing the double bonds in unsaturated fatty acids in lipids, the so-called hydrogenation; and (ii) by introduction of some antioxidants, modified by cyclodextrins, in order to protect food against the oxidation processes. In the area of hydrogenation, an alternative method of hydrogenation was presented, catalytic transfer hydrogenation of edible oils, using sodium formate as a hydrogen donor. This method is fast, simple, safe, economical and offers good selectivity and therefore good oxidative stability of the product. In the area of inclusion between phenolic acids and coenzyme Q_{10} with cyclodextrins, increased stability, solubility and antioxidative activities of included compounds were achieved. Complex of coenzyme Q_{10} with β-cyclodextrin shows superior bioavailability over formulations based on lipid soluble coenzyme Q_{10}.

Keywords: Catalytic transfer hydrogenation, coumaric acids, coenzyme Q_{10}, cyclodextrin

1. Introduction and Objectives

With storage food quality, excluding bacterial and enzymatic deteriorates due to oxidative processes induced and propagated by atmospheric oxygen. Oxidative processes affect the appearance, texture, flavor and odor of food and may consequently induce oxidative stress in the body. However, one of the most important structural components and in the same time quite exposed to the atmospheric oxygen, are lipids. Therefore, much research has been carried out to better understand the basic processes of oxidation of polyunsaturated fatty acids and antioxidant actions of natural and synthetic antioxidants. The Laboratory for Food Chemistry at the National Institute of Chemistry of Slovenia has been involved in studying these problems for at least a few decades. Two approaches were used for solving of these problems: the first, by reducing the specific double bonds in unsaturated fatty acids in lipids, the so called hydrogenation, and by introduction of appropriate antioxidants. Hydrogenation is a process in which dissolved hydrogen in the presence of catalyst saturates fatty acids double bonds in acylglycerols of edible oils and consequently increases their oxidation stability.

This work took into consideration demand and needs of simple, quicker and better controlled hydrogenation processes of vegetable oils for the production of better products with optimal proportion of fatty acids and with negligible oxidative sensitivity. For this purpose, an alternative mode (process) of hydrogenation called catalytic transfer hydrogenation, has been developed and patented by Kobe and co-workers.[1]

This process allowed the production of hydrogenated fats, with the production of hydrogenated oils and partly hydrogenated oils with highly antioxidative stability, and with specific characteristics like solid content index, iodine value and melting points, with great stability under storage conditions at higher temperatures, and especially extremely low contents of *trans* fatty acids. Therefore, one does not have to use molecular hydrogen within the process. Instead, the hydrogen is generated from selected organic molecules in the presence of an appropriate catalyst. These organic molecules are defined as hydrogen

donors. Such novel mode of a reaction was suitable for the transformation into the desired oil modifications. The simplicity of the procedure and its ecological acceptability was clearly revealed. With this procedure, all undesirable reactants and catalyst are recycled. There are more advantages of catalytic transfer hydrogenation disclosed in this work compared to conventional methods of hydrogenation. Replacement of hazardous use of hydrogen with non-volatile donors allows the whole process to be carried out at atmospheric pressure and temperatures below 90 °C with easier control and selectivity. This reaction is performed in sodium formate solution in the presence of 10% Pd/C catalyst and stabilizing agent or emulsifier. The reaction was investigated in details. Kinetics parameters of all important reactions at the reactivity stage conditions have been determined. In this research, important significant scientific assistance and exemplary cooperation was given by Prof. Dr. Tine Koloini and his colleagues on the Faculty for Chemistry and Chemical Technology, University of Ljubljana, which resulted in several scientific papers and final products. The effects of agitation, surfactant (stabilizer or emulsifier), concentration of catalyst and concentration of donor (with or without emulsifier) on the reaction rate, were initially established. Kinetics studies in water solution revealed that different starting concentrations of donors influence primarily the reaction rates and do not disturb the mechanism of the reaction. The same statements could be applied to the attempts to accelerate the reaction in a microwave oven. It is believed that with the appropriate choice of experimental conditions (hydrogenation temperature, agitation rate, oil/water ratio, concentration of sodium formate solution, amount of catalyst and amount of emulsifier), an appropriate selectivity of the desired oil modification can be achieved. These investigations allow the interpretation of the reaction mechanism with respect to the fatty acid and explain the rule of interfacial water-oil surface. Therefore, these kinetics studies allow the preparation of various hydrogenated oils with a very low concentration of linolenic acid (about 1%) at an acceptable level of linoleic acid and with no increase in stearic acid concentration.[2–6]

It was evident at the time that phospholipid mixture of (soybean) lecithin would not possess the optimum composition for every potential application. Therefore, the catalytic transfer hydrogenation of lecithin using sodium formate as hydrogen donor seemed to be a simple and useful solution in order to modulate its chemical properties.[7]

For many years, the oxidation of edible oils during storage and during heating up to 250 °C has been studied by using various synthetic and natural antioxidants.[8] The origin, physico-chemical properties, effects, antioxidant activity and ways for further improvement of effectiveness and stability of various natural substances, especially phenolic substances, have also been studied. At the same time, every effort has been made to change these substances with physical and chemical reactions and transform them into the appropriate nutritional supplements.

It is known, that the interaction with some macromolecules: cyclodextrins (CDs), dextrins, starches, proteins etc. can lead to alteration of physico-chemical properties of guest molecules. In order to improve the pharmacological properties of active substances and drug release formulations for the therapeutic purpose, various carrier materials are being developed to deliver the necessary amount of drug to the targeted site for necessary period of time, both efficiently and precisely.[9–14] The essential information about CDs as the potential candidates for complexation or simply for functional carrier materials in pharmaceutical formulations, was mediated by Szeytli.[15–19]

Since antioxidants themselves are vulnerable to external conditions and subject to disintegration, researchers have tried to design more stable antioxidants with better antioxidant activity. The optimal solution proposed was the inclusion of antioxidant molecules into the hydrophobic cavities of CDs.

The CDs are a series of cyclic oligosaccharides consisting of six or more alfa1,4-linked d-glucopyranose rings. The cyclodextrins are named according to the number of glucopyranose rings present: α-CD has six, β-CD has seven, and γ-CD has eight. The most immediately apparent structural feature of the smaller CDs is the rigid, well-defined cavity resulting from the overall bucket-shape of the molecules. The secondary hydroxyl groups at the C2 and C3 positions of the glucopyranose residues encircle the open top of the bucket, and the primary hydroxyl groups at C6 encircle the bottom of the bucket. The interior of the bucket is lined with the oxygen atoms associated with the glycosidic linkages and the hydrogen atoms located at the C3 and C5 positions on the glucose residues. The rigid, relatively hydrophobic cavity of the cyclodextrins gives rise to their voracious appetite for molecular guests of all types. In all cases some part of the guest is bound within the cavity of the CD. The principal binding interaction in a CD-guest complex is most likely a summation of several relatively weak effects, for example van der Waals interactions, hydrophobic binding, and the release of "high-energy water" from the cavity. The consequence of these properties is that CDs can form inclusion complexes both in solution and in solid state.

The process of inclusion of antioxidant molecules into the hydrophobic cavity of host (CD) includes the mechanisms known from the nano or supramolecular chemistry, indicated as a self-assembly or a self-synthesis. This highly convergent synthetic protocol is exclusively led to the formation of supramolecular complexes by a non-covalent interaction, and can result in new products with increased oxidative stability, water solubility and bioavailability. In the short term, researchers aim the formation of non-covalent complexes with apparent high stability constants, focused on CDs as hosts. In this way, phe-

nol part of the guest (antioxidant) included in proper CD, specifically changes physical and chemical properties of the introduced entity. As it is shown further on the natural antioxidant CoQ_{10}, which is sensitive to high temperature and light, has become stabilized in the complex. Moreover, the solubility of the guest in aqueous solution is much improved by this invention, as well as its bio-acceptability and permeability. Fairly improved antioxidant activity of phenolic derivatives was observed after penetration into CD and this improvement probably depends on the depth of the inclusion into CD cavity of its aromatic part.

Research was oriented to simple phenolic acids, especially those used for the benefit of the human health. Some of them demonstrate strong antioxidant, anti-mutagenic, anti-inflammatory features and exhibit a variety of important biological effects such as protective role of atherosclerosis and coronary heart diseases or potential cancer chemo preventive effects in carcinogenesis.

Benzoic acids, cinnamic acids and many related substances have been reported to display a wide variety of pharmacological properties e.g., antioxidant, anticancer, antimutagenic and antimalarial activities. Dinkova-Kostova and her co-workers have found that there is a good correlation between their protection ability against carcinogenesis and mutagenesis and their radical scavenging capacity.[20] These compounds are ubiquitous in plant food (i.e., fruits, vegetables, coffee), and therefore a certain quantity of them is consumed in our daily diet.[21]

Some researchers have noticed that biological active substances, like phenolic substances and other poorly water-soluble substances become more effective if the organism can easily incorporate them into its biological process. This happens when they are well soluble in the body liquids. It can be assumed that the bioavailability of many active substances may be improved with the increase of their water solubility and numerous researchers have attempted to enhance their adsorption in the body by creating better water-miscible formulations.[9–14] One of the possible ways to achieve this goal is to incorporate these substances into CDs cavity. This concept showed that the most appropriate phenolic substances for studying the inclusion complexes with CDs are smaller molecules like coumaric acids (CAs) and their derivatives. The main interest of researchers is to evaluate the characteristics of pharmaceutical interest, such as solubility in aqueous media, dissolution rate, chemical stability and bioavailability, while basic researches are set to determine the stoichiometry of the complex and to obtain useful information about the complex geometry in a solution and in gas phase.[22,23]

Another important interest is a biological active lipophilic substance coenzyme Q_{10} (CoQ_{10}), which is a much larger molecule, therefore a challenge to incorporate it into the CDs, especially because Lutka and Pawlaczyk could not obtain any inclusion complexes with α-CD and β-CD.[24]

Crane discovered CoQ_{10} in 1957.[25] It is a lipophilic substance with an essential role in the cellular respiratory chain.[26,27] CoQ_{10} is also an effective antioxidant due to its role in biochemistry of oxidative stress, protecting against lipid peroxidation, DNA and protein oxidation.[28] Recently, interest in the CoQ_{10} has been increased significantly due to its role in longevity, adjunctive therapy in cardiovascular diseases, and partial prevention of age related diseases. [29–31] The main source of CoQ_{10} in an organism is biosynthesis, while some CoQ_{10} may be introduced into the body by food. An exogenous intake of CoQ_{10} becomes important with age, because soon after the age of twenty, the endogenous level of CoQ_{10} begins to decrease.

CoQ_{10} is classified as lipophilic compound, while it is practically insoluble in aqueous solutions. Due to its high molecular weight and poor water solubility, it is absorbed from gastrointestinal tract, poorly and slowly.[32] On the market, mainly soft and hard gel capsules are available. These capsules are filled with powder or CoQ_{10} dispersed in sesame or soybean oil. Therefore, the formulation of CoQ_{10} becomes important and provokes chemists to develop a formulation for oral administration with better water-solubility and therefore better bioavailability.

First attempt to include the CoQ_{10} into the β-CD cavity was displayed in a patent.[33] It was a difficult task due to its chemical structure. With special procedure, the water soluble CoQ_{10}, which was successfully implicated into 19 new food products in the last five years was obtained. The licence was imparted to Slovenian enterprise, which trades patented products in Slovenia and in thirteen other countries in Europe and Asia.

Development of water-soluble form of CoQ_{10}, which can be used as a food additive, is a contribution into the elimination of oxidative stress in humans and animals. Enhanced bioavailability was obtained with originally developed water-soluble CoQ_{10} in a form of patented water-soluble paste, based on molecular encapsulation of CoQ_{10} into the β-CD's lipophilic cavity. Industrial experiments showed that the mentioned product could be mixed with many food products. Additionally, when functional food is placed into a medium with pH value below 3, the inclusion complex is disintegrated and CoQ_{10} is easily and uniformly released from the β-CD carrier in its natural form. Results proved that this water-soluble substance in a form of paste with 7–10% of CoQ_{10} is very convenient product suitable for humans and animals, that dislike or cannot swallow relatively big capsules.[33,34]

For solubility, determination the complexes of CA isomers, their methyl esters and CoQ_{10} with β-CD, and CoQ_{10} with γ-CD were prepared using the most widely used method, the co-precipitation method. In these cases, CD was dissolved in water at room or elevated temperature, and the guest in solid or liquid phase was added during the stirring the reaction solution. The reaction conditions were chosen in the way that the solubility will be excee-

ded, while the reaction of complexation proceeds or while cooling is applied. Such conditions enable precipitation of the complex, which can be collected by decanting, centrifugation or filtration.[35]

The ultimate characterisation of CD complexes requires the use of most representative spectroscopic methods like mass and NMR spectrometry.[22,23]

One of the most informative methods for determining of CD inclusion complexes in aqueous solutions is NMR spectrometry. Proton NMR is used to distinguish the capabilities of CDs to form an inclusion complex, by including the whole guest molecule or rather some nonpolar part of it inside its cavity, to determine the stoichiometry and stability (association constant-K_a) of the complex, and to obtain useful information about the complex geometry in aqueous solution. This technique coupled with a molecular modeling gives us a useful tool of establishing the possible geometry of the investigated system. In spite of their simple structure, these substances are specific and of particular interest from the point of view of penetrating steric dependency and therefore represent a good groundwork for the study of other phenol based antioxidants, which will be used to work with in the future.

In the following chapters, we present a few findings, which are the results of complexation of some chosen substances with cyclodextrins.

2. Solubility

Solubility tests were designed to determine the apparent total molar solubility of the coumaric acid isomers, their methyl esters and CoQ_{10} as a function of total concentration of β-CD, and γ-CD in the case of CoQ_{10}.

2.1. Solubility of Coumaric Acids and their Methyl Esters

Solubility studies of Stražišar and co-workers and Milivojevic-Fir and co-workers showed linear relationships between the amount of solubilised ligands (CAs and CoQ_{10}) and the concentration of CD in solution (A_L diagrams).[34,35] In all cases, the solubility of ligands have increased with the increasing concentration of CD. According to the theory of Higuchi and Connors, that may contribute to the formation of soluble complexes between ligands and CD with stoichiometric ratio 1:1.[36]

The apparent binding constant of the resulted complexes was calculated from the slope (k) and intercept (n) of the straight lines of the phase solubility diagram according to the relation Ks = k/n(1-k) of Higuchi and Connors. Calculated apparent stability constants, for coumaric acids are 0.39×10^3 M^{-1}, 2.81×10^3 M^{-1} and 49.25×10^3 M^{-1} for *o*-CA, *m*-CA and *p*-CA, respectively, and 0.39×10^3 M^{-1}, 0.19×10^3 M^{-1} and 0.43×10^3 M^{-1} for their appurtenant methyl esters. Calculated stability constants for complexes of CoQ_{10} with β-CD and γ-CD are 4.32×10^2 M^{-1} and 2.21×10^3 M^{-1}, respectively.

In previous paper, it was proven that dioxan could be used as a selective solvent for adsorbed or free and included CA in the CA/β-CD systems. In all cases, inclusion of the coumaric acid or methyl coumarat into CD cavity increased its solubility in water. *o*-CA increased 3.3-times (from 0.49 to 1.61 g/L), *m*-CA for 1.7-times (from 1.54 to 2.58 g/L), and *p*-CA for 2.4-times (from 2.86 to 2.03 g/L), while for CH_3*o*-CA the solubility increased from 0.24 to 0.46 g/L, for CH_3 *m*-CA from 1.29 to 1.1.2 g/L and for CH_3 *p*-CA from 0.5 to 1.02 g/L.[35]

2. 2. Solubility of CoQ_{10}

Milivojevic-Fir and co-workers indicate that solubility of pure CoQ_{10} at pH 6.5 is 8.03×10^{-2} mg/L, and increases significantly in relation to CD concentration in water.[33] The increase of CoQ_{10} solubility depends on the type of CD. The solubility of CoQ_{10} at room temperature is 13.88 and 19.26 mg/L, and at 95 °C 57.03 and 109.36 mg/L in the presence of β-CD and γ-CD, respectively. They found that the solubility of complexes is also pH dependent. There is relatively high solubility of CoQ_{10} in water at pH 6.5 and 37 °C (body temperature) in the form of CoQ_{10}/β-CD (23.05 mg of CoQ_{10}/L) or CoQ_{10}/γ-CD (39.56 mg of CoQ_{10}/L), and insignificant concentration at pH 2.5 (0.29 mg and < 0.0005 mg of CoQ_{10}/L, respectively). It has to be mentioned that physical mixtures show very low water solubility of CoQ_{10}, which indicates weaker interactions between CoQ_{10} and CDs. Lower solubility of CoQ_{10} at lower pH depends on disintegration of complexes due to hydrolytic cleavage of CD glycosidic bonds. This is of a paramount importance in the development of pharmaceuticals. The enhancement of oral bioavailability by CD complexation is usually based on an increase in the dissolution rate of drugs, which is finely dispersed in the gastrointestinal fluids.[31] In the case of the complex CD and CoQ_{10} (CoQ_{10}/CD), relatively high water solubility and low acid stability were achieved. Therefore, it is expected that oral bioavailability of CoQ_{10} in the form of CoQ_{10}/CD will be enhanced.

3. Stability

3. 1. Stability of CoQ_{10}

The guarantee of chemical and mechanical stability for a minimum of 2 years for pharmaceutical products is of a key importance for marketing the product. The active component in various pharmaceutical formulations may decrease significantly due to different processes, such as decomposition by heating, volatilization, sublimation, oxidation, hydrolysis, and reactions with other pharmaceutical ingredients. Nowadays, various materials, especially CDs are used for stabilization of unstable substan-

ces, because they can increase the stability of chemically and mechanically unstable molecules.[37–40] CoQ_{10} can be used in the form of CD complex as a food additive in various food products, such as milk, yogurt, fruit syrup etc. Most of the food is pasteurized directly prior to their packaging. Pasteurization is conducted for a few minutes at temperatures above 80 °C, and therefore stability tests at the mentioned temperature are needed. Experiments which were performed by Milivojevic Fir and co-workers show, that heating at high temperature in the dark does not significantly affect the stability of CoQ_{10}.[34] After 92 hour of monitoring at 80 °C, 96.3% of CoQ_{10} remain intact. They observed even smaller influence of heat in the case of inclusion complexes, where after 92 hours at 80 °C less than 1% of CoQ_{10} was degraded, both as β-CD complex and as γ-CD complex. At lower temperatures, the heat effects are even smaller or practically negligible. After preserving the samples for 92 hours at room temperature in the dark, more than 99.9% of CoQ_{10} was determined.[34]

Greater influence of temperature on the stability of CoQ_{10} in various forms would most likely be seen during longer period. Kommuru and his associates were monitoring the stability of CoQ_{10} for 16 months at various temperatures; 37, 45 and 55 °C.[41] They observed degradation largely at 45 and 55 °C, while CoQ_{10} was relatively stable at 37 °C. Using the Arrhenius plot of logK versus T^{-1}, they predicted shelf life at room temperature to be about 6.3 years. The above-mentioned results suggest that the use of CDs as protective agents would prolong the shelf life of CoQ_{10} even more, which was determined by Kommuru.

Since higher temperatures do not have a great influence on the stability of inclusion complexes of CoQ_{10} with CDs, the complexes are suitable to be added to food during the manufacturing process or packaging.

Further, Milivojevic-Fir and co-workers established that heat and the type of CD also affect the photolytic degradation of CoQ_{10}. Neither temperature nor UV irradiation has an important effect on CoQ_{10} stability, but the combination of both shows synergistic effect on CoQ_{10} stability. Pure CoQ_{10} readily decomposes when it is exposed to the UV light (254 nm) at room temperature and specially at elevated temperatures, while almost no degradation occurred in concurrent presence of β-CD or γ-CD at room temperature. However, β-CD and γ-CD offer relatively high protection also at 80 °C. The stability study also provides the information about the nature of the complex (adsorption or inclusion).[34] Uekama and co-workers explain that in order to attain a photo stability enhancement by CDs, the photosensitive moiety of the guest molecule has to be located inside the CD cavity.[42] Therefore, photo stability of CoQ_{10} would not be enhanced by adsorption on the outside of the CD surface. As to the fact that lypophilic polyisoprenoid chain is the most unstable part of CoQ_{10}, that suggests that this part of CoQ_{10} is included into the CD cavity.

4. Antioxidant Activity

4. 1. Antioxidant Activity of Coumaric Esters and their Methyl Esters

The antioxidant capacity of phenols is generally tested using the reaction with oxidants where resonance-stabilized phenoxyl radicals occurred. The antioxidant activity of phenolic compounds depends on the position and degree of hydroxylation as well as the nature of radicals of the ring structure. Antioxidative activity is intensified by the presence of a second hydroxyl group, as in caffeic and protocatehuic acids, through the formation of an intramolecular hydrogen bond.[43] The presence of the –CH=CH–COOH group ensures increased antioxidative efficiency of CAs derivatives participating in stabilization of the phenoxyl radicals by resonance in comparison to –COOH and corresponding hydroxybenzoic acid.[44]

During the study of physico-chemical properties of the complexes between flavonoles and CD, Calabro and his co-workers found that the antioxidative activity of flavonols was improved.[45]

The hypothesis of Stražišar and coworkers was that in the CD inclusion complexes of *o*-CA and *m*-CA the distance between secondary hydroxyl groups of βCD and –OH on the aromatic ring of *o*-CA and *m*-CA is approximately the same as the distance of –OH groups in the caffeic acid molecule (< 3 Å). Therefore, the formation of »intermolecular« hydrogen bond of the inclusion complex is possible, consequently an increase of antioxidant capacity can be expected.[35]

Antioxidant activity of free CAs, their methyl esters, CA/β-CD, and CH_3CA/β-CD was measured in terms of hydrogen donating or radical scavenging ability, using the modified method according to Brand-Williams and coworkers.[46]

Antioxidant activity of the tested substances, decreased in the order: CH_3 *o*-CA/β-CD > *p*-CA > CH_3 *m*-CA/β-CD > *o*-CA/β-CD > *p*-CA/β-CD > *o*-CA > CH_3 *p*-CA > CH_3 *o*-CA > CH_3 *p*-CA/*β*-CD > *m*-CA/*β*-CD > CH_3 *m*-CA > *m*-CA.

Findings of Stražišar and coworkers confirmed the statement of Marinova and Yanishlieva, that *ortho* and *para* isomers are better antioxidants due to electron donating effects of the COOH – CH = CH – group on the aromatic ring, participating in the resonance stabilization of the phenoxyl radicals.[44] Complex of *o*-CA and its methyl ester with β-CD showed the improvement of elimination ability of DPPH radical from 14.5 up to 26.4% and from 13.8 to 60.5%, respectively. In the case of *meta* isomer of CA, antioxidative activity increased approximately 2.5 times, while in the case of their methyl ester its antioxidative activity increased 5 times, from 6.3 to 29.7%. On the other hand, antioxidative activity of free and complexed *p*-CA and its methyl ester did not

differ significantly. These differences among *ortho, meta* and *para* isomers can be explained by the distances of CA's OH groups from the β-CD's OH groups in the complex itself. It could be that the –OH at *ortho* and *meta* positions of CA molecules is close enough to –OH groups of β-CD to form the hydrogen bonds and contribute to antioxidant activity. On the other hand, the distance of OH group in *para* position is too far away from the secondary OH groups of β-CD to re-establish the hydrogen bonds.

4. 2. Antioxidant Activity of CoQ_{10}

Besides the well recognized vital role of ubiquinone in energy transduction and oxidative phosphorilation, considerable evidence exists that CoQ_{10} functions as lipid soluble antioxidant in biological tissue. Antiperoxidative effect of CoQ_{10} has been checked in lyposomes exposed to a radical insult and in micelles of unsaturated fatty acids subjected to the thermal autooxidation.[28,47]

The antioxidant activity of CoQ_{10} and their corresponding complexes was estimated with a slightly modified spectrophotometric method published by Blois.[48] The equimolar solutions of CoQ_{10} and CoQ_{10}/β-CD were prepared, and antioxidant activity was tested with free radical DPPH. This method is widely used to test the ability of compounds to act as free radical scavengers or hydrogen donors, and to evaluate antioxidant activity of plant extracts.[49,50]

In nature, the oxidized form of CoQ_{10} is in equilibrium with its reduced form ubiquinol. The reduced form of CoQ_{10} possesses a hydroxyl group on quinone ring that can reduce and stabilize free oxygen radicals, and protect lipids, proteins, and DNA against the oxidation process.

The radical-scavenging activity against DPPH radical for CoQ_{10} was determined to be 1.8%; CDQ_{10} shows significantly higher antioxidant activity than pure CoQ_{10}. Complexation of CoQ_{10} with β-CD and γ-CD resulted in a 26% and 21% increase of antioxidant activity respectively, meaning that such cyclodextrin has small effect on antioxidant activity. [51]

5. Bioavailability

Several studies have been performed to examine the plasma CoQ_{10} response to per orally ingested CoQ_{10} formulations as an indicator of the compound bioavailability of these products.[52,53] Due to its insolubility in water (< 0.1μg/L) relatively high molecular weight (M = 863.34 g/mol), CoQ_{10} is very poorly absorbed *in vivo*. Therefore, search for the formulation with increased absorption has become important.[10,52,54] Since the increased absorption of CoQ_{10} has been reported for some solubilised formulations, this was also expected for water-soluble paste formulation.[52,55,56] To confirm the expectations two bioequivalence studies were performed. In the first study, relative bioavailability was investigated for two forms, water-soluble CoQ_{10} formulation and commercially available oil-based CoQ_{10} in the form of soft-gel capsules. The bioavailability was determined by measuring plasma CoQ_{10} levels at precise time intervals, after administration it to a group of beagle dogs. Mean value for base line plasma concentrations, maximum plasma concentrations (C_{max}), time of maximum plasma concentration (T_{max}), area under the plasma concentration $AUC_{(0–48h)}$, elimination half life ($t_{1/2}$) for both formulations were determined. The results of the experiment show the advantage of water-soluble CoQ_{10} in comparison with commercially available soft-gel capsules. It is seen in nearly three times higher $AUC_{(0–48h)}$, nearly two times higher C_{max}, and shortened T_{max} from 6 to 4h, where $AUC_{(0–48h)}$ represents area under the plasma concentration, C_{max} maximum plasma concentration and T_{max} elimination half life.[57]

Second bioequivalence study was also performed for two formulations, the novel CoQ_{10} paste with increased water-solubility in comparison to soft-gel capsules with CoQ_{10} in soybean oil, but this time in human. This single-dose bioequivalence study once again revealed, that oral absorption and bioavailability of CoQ_{10} can be significantly affected by increasing the water solubility with the formation of CoQ_{10}/β-CD complex, because it demonstrates the superior bioavailability over the soft-gel capsules.[58]

6. Conclusions

Catalytic transfer hydrogenation of edible oils with a solution of sodium formate as hydrogen donor presented in this review is a simple and useful alternative to hydrogenation of vegetable oils. Besides good selectivity, short reaction time and ecological acceptability, mild reaction conditions (low temperatures, under 90 °C and atmospheric pressure) allow a safe and easy handling method.

Inclusion of some antioxidants (coumaric acids, their methyl esters and coenzyme Q_{10}) into β-cyclodextrin cavity results in beneficial effects on their basic properties. The main goal was achieved. The solubility of coumaric acids and their derivatives was increased due to their inclusion into β-CD cavity. Besides better solubility, also better antioxidant activity was achieved at *o*- and *m*-CA but not at *p*-CA.

The complexation of CoQ_{10} with cyclodextrins (β and γ) also increases aqueous solubility, thermo- and photo stability of CoQ_{10}, while the increase depends on the CD type. Finally yet importantly, the formation of water soluble CoQ_{10}/β-CD complex demonstrates superior bioavailability over some lipid soluble formulations.

7. References

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Povzetek

Prispevek obravnava preprečevanje oksidacije hrane povzročene z atmosferskim kisikom z dvema pristopoma: (a) z nasičenjem dvojnih vezi nenasičenih maščobnih kislin v lipidih in (b) z uporabo nekaterih antioksidantov modificiranih s ciklodekstrini, ki lahko zaščitijo hrano pred oksidacijskimi procesi. Na področju hidrogenacije je predstavljena alternativna katalizna transfer hidrogenacija, kjer se kot donor vodika uporablja natrijev format. Ta metoda je hitra, enostavna, varna in ekonomična ter ponuja dobro selektivnost in zato dobro oksidativno stabilnost produkta. Z vključevanjem fenolnih kislin in koencima Q_{10} v ciklodekstrine so se izboljšale stabilnost, topnost in antioksidativna aktivnost vključenih substanc. Dokazana je tudi večja biorazpoložljivost koencima Q_{10} iz β-ciklodekstrinskega kompleksa glede na koencim Q_{10}, ki je raztopljen v olju.