Hojka Kraigher

CONSERVATION OF FOREST GENETIC RESOURCES WITH FOREST REPRODUCTIVE MATERIAL MANAGEMENT GUIDELINES

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PREFACE

Large-scale disturbances, such as extreme weather, wildfires, and pests, have weakened forests across Europe, reducing their ability to regenerate and adapt to climate change. To address this, professional seed and nursery management practices are essential to maintain forest health and resilience.

This textbook and included guidelines were developed with the Slovenian environment in mind but are applicable across Europe. Since 2000, we have emphasized the importance of forest reproductive material and tree nursery activities for successful, close-to-nature, and multifunctional forest management, which forms a cornerstone of forest management practices. Following 2001, long-lasting droughts became more frequent, and in 2014, glaze ice affected 40% of Slovenian forests. This accelerated bark beetle infestations, while already weakened forests were further damaged by severe windthrows in the subsequent years.

Forests affected by such disturbances can recover naturally, but restoring commercially viable forests is a lengthy process. During this recovery, issues like weed infestation and overpopulation of herbivorous wildlife often arise. Large-scale disturbances have also impaired the natural regeneration and genetic diversity of young forest stands, making them less adaptable to future environmental changes.

This highlights the growing recognition of the need for professional seed and nursery management, based on local knowledge and forest nursery capacities. Although natural regeneration remains common in close-to-nature managed forests, it is no longer the primary method of regeneration. A shift in strategies, planning, and support for this critical aspect of forestry is necessary to address these emerging challenges.

Large-scale disturbances have also affected the natural regeneration capacity and genetic diversity of natural young forest stands, which will be even less able to adapt to a changing environment in the coming decades. This is why awareness of the importance of professionally appropriate seed and nursery production, based on local knowledge and capacities of forest tree nurseries in the Slovenian territory, is returning to the forestry sector. Natural regeneration still prevails in our close-to-nature managed forests, but it is no longer the main regeneration practice in the Slovenian Forestry School; what is needed is a change in thinking, strategies, planning and support for this long-neglected sector of forestry in Slovenia.

Within the context of several targeted developmental projects (CRP), we have set ourselves the task of preparing forest reproductive material management guidelines with a focus on the conservation of forest genetic resources as a basis for the sustainable management of multifunctional forests in times of rapid environmental change, increased demand for the use of forest reproductive material and for maintaining the adaptive potential of future populations of forest trees to a changing environment. Consequently, conserving the genetic diversity of forest trees is in line with the Slovenian Forest Genetic Resources Programme (SIFORGEN), which is also part of the European Forest Genetic Resources Programme (EUFORGEN).

The draft of the *Seed Practicum – Forest Reproductive Material Management Guidelines* was distributed as working material (in five spiral-bound copies) at the Public Forestry Service workshop for the needs of the Slovenia Forest Service and the forest seed and nursery sector in 2019. I would hereby like to thank Professor Emeritus Franc Batič and Academic Ivan Kreft for providing peer reviews of this draft. However, it has taken five years to complete and finalise the material, to accompany it with original illustrations, and to propose it to the University of Maribor for review as a textbook entitled *Conservation of Forest Genetic Resources with Forest Reproductive Material Management Guidelines (Seed Practicum);* Prof. E. Franc Batič, retired professor of botany and plant ecology, and Prof. Dr. Mitja Kaligarič, full professor of botany at the University of Maribor, were nominated as reviewers. I take pride in quoting the reviewers:

"The work comprehensively presents all the elements relating to forest genetic resources. It gives a historical overview of the issue and its management in the EU and Slovenia. The biological and genetic basis of forest genetic resources, silvicultural measures for the selection and management of forest seed resources, as well as the related essential basics concerning forest seed husbandry and nursery activities, are presented in an appropriate manner.

/…/

I consider this work to be of high priority, particularly at a time of great environmental change, which is not sparing our forests. For the first time in a long period, the state of forest resource management in our country and in the EU is described and collected in one place, with all the professional, technological and management basics. As such, the work will serve as an indispensable resource for everyone interested in sustainable forest utilisation (forest owners, the Slovenia Forest Service (SFS), the Chamber of Commerce and Industry of Slovenia (CCIS), the Ministry of Agriculture, Forestry and Food of the Republic of Slovenia, students, teachers and researchers in the field of forestry, etc.), as well as for all other parties interested in nature and environmental issues."

/…/

Franc Batič, Professor Emeritus, retired full professor of Botany and Plant Ecology

"A highly complex subject matter is covered here, ranging from the basics of morphology, germination, genetics, forestry and legislation. There is also an emphasis on the applied aspect ("Seed Practicum – Forest Reproductive Material Management Guidelines") - the management of forest seed resources, including forest seed and nursery activities.

/…/

Therefore, the *Conservation of Forest Genetic Resources with Forest Reproductive Material Management Guidelines (Seed Practicum)* certainly covers more areas than its title promises, as it deals comprehensively with all aspects of forest genetic resources, while providing a useful recapitulation of the biological and forestry basics. Almost two-thirds of Slovenia's national territory is covered by forest (including overgrown farmland), which makes the topic relevant and topical, including in the light of major climate changes (droughts and bark beetle infestation) and land use changes (reclamation of farmland and grazing meadows). This monograph is intended not only for foresters and forest professionals, but also for students of forestry, biology and ecology (the latter courses are also taught at the University of Maribor), as well as anyone who is in any way interested in forests, their management and conservation."

Prof. Dr. Mitja Kaligarič, Full Professor of Botany at the University of Maribor

I would hereby also like to thank all the contributors to this textbook:

I would like to thank my colleagues at the Slovenian Forestry Institute (SFI): Dr. Marjana Westergren, Dr. Gregor Božič and Dr. Peter Železnik for discussions, for the transmission of our commonly produced guidelines for seeds and seedling manipulation thanks go to Andrej Breznikar. MSc, from the Slovenia Forest Service (SFS), and to Marina Herman Planinšek and Vlado Planinšek from the Omorika Nursery for their nursery practices overview. We have also used our own workshop materials and the Forest Genetic Monitoring Handbook, which was developed within the framework of the *LIFE for European Forest Genetic Monitoring System* (LIFEEGENMON) project (2014-2020), which is partly based on the EUFORGEN Programme joint reports, particularly regarding the issues of genetic diversity conservation in forest seed and tree nursery production (Gömöry et al. 2021), and on the charts developed within the framework of the CRP, LIFE SySTEMiC (2019-2024) and other projects.

Thanks also go to colleagues at the Department of Forest Physiology and Genetics at the Slovenian Forestry Institute (GIS), who contributed photographs; special thanks go to Melita Hrenko, to everyone who participated in the design and selection of the drawings, especially Katja Kavčič Sonnenschein, MSc, and to all authors of the drawings, to Dr. Maja Peteh for her help in editing the references and classifying the work, the Slovenian language proofreader Teja Kačar, the layout designer Lenka Trdina, and the editor of the Forestry Journal for permission to publish the abstract of the Systemic Problems of Reforestation meeting (from 2017).

Special thanks go to our co-funders: the Ministry of Agriculture, Forestry and Food of the Republic of Slovenia, the Slovenian Agency for Scientific Research and Innovation (ARIS), the Ministry of the Environment, Climate and Energy, the Slovenian Forestry Institute, the LIFE Programme for co-financing LIFE projects, targeted research projects (V4-1616, V4-1819, V4-2015, V4-2222), the research programme P4-0107 and the Public Forestry Service (JGS).

1
INTRODUCTION

1.1 Conservation of Forest Genetic Resources and the SIFORGEN Programme

Forest trees are characterised by their size, long lifespan and, in most cases, large genetic base, as domestication and cultivation processes are relatively new, starting mostly in the 1950s (Nanson 2004). In natural populations of forest trees, genetic variability within populations is generally higher than between populations, and it is this high variability within the populations that allows them to maintain the adaptive capacity (of future generations) of forest trees to changing environmental conditions. Therefore, the primary task of conserving forest genetic resources (FGR) is precisely to preserve as much genetic diversity as possible in all forest governance and management practices, in particular in terms of silvicultural measures, which should be based on "genetic forest protection", i.e. forest protection based on the conservation of genetic diversity. Measures for the conservation of forest genetic resources include:

- **Silvicultural measures, which take into account the importance and potential for the conservation of genetic diversity** of the populations concerned, i.e. forest tree stands and plantations.
- **Silvicultural measures** in **support of the minority species and populations of forest trees at the edge of their area** of distribution, and in support of particular populations of forest trees that need support for long-term survival in a particular site.
- **Reforestation measures**, which include:
  - ◊ *Natural regeneration of forests* at times of abundant production, in populations with an adequate number of mother trees – seed trees with appropriately developed reproductive parts of their crowns.
  - ◊ *Regeneration by planting and sowing in areas:*
    - where natural reforestation is inadequate in terms of species and genetic diversity; or
    - where it fails to meet the objectives of sustainable forest development in an optimal time-scale; or
    - where large areas of forests have been cleared as a result of large-scale disturbances (fires, windthrows, glaze ice, etc.), sanitary measures due to infestation of pests and diseases, or due to inadequately implemented felling operations; or
    - where the reproductive parts of crowns of trees have been damaged by past disturbances (e.g. glaze ice or snow break), reducing the genetic diversity of naturally occurring young forest regeneration centres; or
    - where spatial plans define a change of land use to forest plantations and forest.
  - ◊ *Regeneration by enrichment planting and sowing:*
    - where the quality of natural saplings is not sufficient to achieve the long-term objectives of conserving forest genetic resources and the sustainable/viable development of multifunctional forests; or
    - where the aim is to increase the species and genetic diversity of existing young forest stands by enrichment planting and sowing; or
    - where we want to include additional provenances of individual forest tree species as a pre-preparation of forests for (un)expected climate change.

Conservation of the FGR based on the implementation of the above measures, the dissemination of knowledge on the importance of genetic diversity for the sustainable development of forests as we know them today, communication with stakeholders at different levels, with the target and the general public, and concrete measures for the conservation of the FGR of individual forest tree species and tracing the effectiveness of the implemented measures through the establishment and implementation of the genetic monitoring of forests are

SIFORGEN
is the Slovenian Forest Genetic Resources Programme.

EUFORGEN
Is the European Forest Genetic Resources Programme, which has been operating since 1994 on the basis of resolutions adopted by the Ministerial Conferences for the Protection of Forests in Europe (now Forest Europe). Slovenia has been a participant since the first meeting of national coordinators in 1995.

EUFORGEN

the basis of the **Slovenian Forest Genetic Resources Programme,** or **SIFORGEN** for short.

These measures are based on international strategies and are formulated jointly by with the **European Forest Genetic Resources Programme, EUFORGEN**, of which Slovenia has been a member since its inception: a report on the Slovenian Forestry School and the forest genetic resources in Slovenia was already presented at the first Steering Committee (SC) meeting of EUFORGEN in Sopron, Hungary, in 1995, and Slovenia formally signed the membership agreement in 1997. The country also participated in the first European project to organise a joint European database on Dynamic Conservation Units (GCUs), EUFGIS, and in several working groups during all the phases of the EUFORGEN Programme, which entered its sixth operation phase in 2020. Experts and the face-to-face meetings within the context of the programme also contributed to the effective design of the new legislation on forest reproductive material (FRM) in Slovenia, which has been in place since 2002, when the **Forest Reproductive Material Act (ZGRM 2002)**, was adopted, the European Directive on the marketing of forest reproductive material (EC/105/1999) was harmonised, and the list of approved forest seed facilities (FSFs, which, after approval, are permitted for producing FRM of different categories) is also published annually in the European FOREMATIS database (http://ec.europa.eu/forematis/). Since 2018, we have been participating in the meetings of the OECD Working Group on FRM, and are a member of the Working Group on Forest Seeds and Propagating Materials since 2022, and we have previously attended the regular annual meetings of the European Working Group on Forest Reproductive Material within DG SANTE. Since Slovenia's accession to the EU, the Ministry has been sending a representative to the regular annual meetings of the Working Group in Brussels or Paris.

Photo 1: Mast beechnut production: beechnuts hang from all the branches throughout the crown (photo by Hojka Kraigher).

1.2 Phenology

PHENOLOGY
defines the growth and development of forest trees.

The growth and development of forest trees is significantly defined by **phenology** – the onset of the developmental stages within the context of the vegetative growth-development cycle and of the generative growth-development cycle, as well as lignification and senescence, which have an important impact on the winter survival of woody plants in temperate climates.

In the warm temperate climate zone of the northern hemisphere, the year starts with a dormancy phase in January and February, mainly conditioned by low temperatures and, in the case of some alien species, a short photoperiod. In March (equinox), the dormancy is no longer affected by the short photoperiod, while in April and May **vegetative shoots start to grow and develop**, which depends on the increase in ambient temperature. This is also the period when height (axial) and thickness (radial) growth begins, which ends in July to August for most tree species. Height growth ends in the summer months, between late June (pines) and late September (larch), except in individual species where favourable conditions (humidity or fertilisation) after the end of growth may stimulate further growth in August. However, for most species, **thickness growth** ends in September. Although the shortening of the photoperiod (autumnal equinox) affects the onset of dormancy in a different manner depending on species and ecotype, it includes bud development, an increase in osmotic pressure and dry matter content, the cessation of stem growth and lignification, and the storage of nutrients.

Development of generative organs begins with the **induction of flower stems in the year before flowering,** in June to early July for most species (Wareing 1958, in Nanson 2004). This is stimulated by a warm period of dry and sunny days. In addition, the trees must have sufficient reserve substances that can be used at the time of flowering (therefore, for example, oaks and beeches do not form flower stems if there was a high mast production the previous year). The induction of flower stems can be influenced by a variety of factors, ranging from hormonal regulation in natural systems (including interactions with symbiont organisms) to the stimulation of flowering in seed plantations, e.g. by inducing stress (drought stress or the removal of part of the crown tissue) or by the use of plant hormones.

Usually, forest trees **bloom** in the year after flower stem induction and mostly from April to May, in many species before the start of leafing and the growth of vegetative shoots, which facilitates pollen dissemination and pollination. Early flowering also makes them more susceptible to late spring frosts. In a given forest tree population, the onset of flowering of different trees may extend over several weeks, and there may also be differences in different parts of the crown of the same tree. Self-pollination is reduced because of the different maturation times of male (**protandry** – male flowers mature first) and female flowers (**protogyny** – female flowers mature first). **Flowering leads to** pollination **either by wind** (anemophily) **or insects** (entomophily), which takes place over one to two weeks in dry weather, interrupted by rainfall. **In most tree species, pollination is immediately followed by fertilisation,** though with others it starts the year after pollination (pines and oaks) or in the third year after pollination (cedars). **The fruits and cones usually reach their final size in July, followed by a period of maturation** that most species reach between September and December. This is followed by the **dispersal and dissemination** of the seed, usually in the autumn, though it can last until the following spring.

Mature seeds can be **dormant** (e.g. larch, ash and Douglas fir), and breaking of dormancy requires **stratification,** i.e. a period of exposure to moisture and cold, or in some species a period of alternating cold and warm periods (warm, cold or alternating stratification), or the mechanical or chemical treatment of the seed – **scarification.**

Forest tree species can be **monoecious** where the female and male flowers grow on the same plant (flowers may also be hermaphroditic) or **dioecious** where only male or only female flowers are present on some individual specimens. There are also transitions between the two forms, and monoecic and diecic forms can appear in the same species, or even **triecious** trees with hermaphrodite flowers, e.g. in ash trees; dioecious male trees usually grow better than female trees because all the assimilates are used for the growth of the tree alone and not for the production of the seeds.

In the following sections, we look at the flower, seed and fruit, as well as the flowering and mast production processes in more detail.

There are **monoecious, dioecious** and **triecious** tree species.

2

FROM SEED TO SEEDLING

2.1 Flower, Seed and Fruit

The contents are mainly taken from Mala flora Slovenije (Little Flora of Slovenia) (2007).

A **flower** is a short, rosette-type, limited-growth shoot with transformed leaves for sexual reproduction. In the case of hermaphrodite flowers the stamens and pistil(s) develop in the same flower. Unisexual flowers only have stamens or only pistil(s) developed. Plants with male and female flowers on the same plant are monoclinous or monoecious (hazels, black alders, spruces, firs, oaks, beeches and hornbeams). Plants that only have female flowers on one plant and male flowers on the other are diclinous or dioecious (hops, yews, willows, poplars and junipers). Plants whose populations only have unisexual (either gynoecious or androecious), female and hermaphrodite, male and hermaphrodite or only hermaphrodite flowers on the same specimens are **polygamous** (such as horse chestnut). Triecious plants have only female, only male or only hermaphroditic flowers on individual specimens (Little Flora provides the example of the European ash, but the occurrence of different sex combinations of flowers can be much more complex within this species). The flowers of gymnosperms are mostly unisexual, the plants are mostly monoecious.

The transfer of the pollen grain to the stigma of the pistil of angiosperms and to the ovule of gymnosperms is called **pollination**. **Self-pollination (autogamy)** is pollination with pollen grains from the same genotype. **Allogamy** is pollination by external pollen, **zoophily** is the transfer of pollen grains by insects, and **anemophily** is the transfer of pollen grains by the wind.

The pollen grain on the stigma of the pistil germinates and grows directly to the ovules. **After fertilisation,** the ovule starts to develop into a seed. **The formation of an embryo without fertilisation is called agamospermy.** In the case of angiosperms, the seed develops in the ovary, while in the case of gymnosperms, the seed develops on the surface of the carpels. **Parthenocarpy** is the development of fruit without seeds.

A **seed** is a young plant, a living organism in which metabolic processes are slowed down or suppressed. The seed is enclosed in a seed coat (testa). This is particularly strong in the case of seeds that do not remain in the fruit. The hilum is the part of the seed where the funiculus (funiculus cord) was attached, by which the seed was connected to the placenta in the ovary (the carpel). Inside the seed is the initial stage of the developing plant germ (embryo) and often also the nutritive tissue (endosperm). Depending on the method of fertilisation, the endosperm is haploid (gymnosperms), diploid or triploid (secondary endosperm). Conifer seeds have an endosperm, while the seeds of our deciduous trees often do not. If a seed does not have it, the spare nutrients are stored in the cotyledons, which fill most of the space in the seed.

Important concepts in forest seed production include:

- embryo (consisting of the radicle, plumule and cotyledons),
- endosperm (nutrient reserve, often very thin or non-existent),
- seed coat.

SEED
is a young plant, a living organism in which metabolic processes are slowed down or suppressed.

After fertilisation, the pistil, and sometimes also some other parts of the flower (insertion point, bracts, etc.) develop into the fruit (i.e. a pericarp), which consists of an exocarp, mesocarp and an endocarp, which could be quite different in structure). The fruit contains seed and it develops only in angiosperms. This can be simple, when it is formed mainly from the ovary of pistil (beechnuts, acorns, samaras of maple, elm, hornbeam, linden trees, etc.), or drupelets (berries), when it is formed mainly from the receptacle, on which there are several pistils (e.g. apple, raspberry and strawberry fruits, etc.); composite fruits develop from the entire inflorescence – a large number of blossoms (mulberry, fig, etc.). The fruit is succulent (fleshy) when the pericarp is fully or partially fleshy and dry when the pericarp is dry. A dehiscent fruit opens for the seeds to be released, while a closed fruit (indehiscent) falls off as a whole (achenes). The fruits are extremely diverse in their formation and structure.

FRUIT
consists of seed(s) and pericarp.

1) **Simple fruits** develop from one or more mature pistils.
   - **Simple fleshy fruits.**
   - **Fleshy indehiscent fruits:** fall off complete when ripe.

     Berries: the fruit is fully fleshy and produced from one (barberry) or several carpels (bilberry).

     Drupaceous (stony) fruits: only the outer part of the pericarp (exocarp and mesocarp) is fleshy and the inner part is woody (stone – endocarp); they are formed from one (cherry) or several mature carpels (walnut, elder and olive).
   - **Fleshy dehiscent fruits:** are fleshy capsules (spindle tree – *Euonymus* sp., touch-me-not - *Impatiens* sp.).
   - **Simple dry fruits.**
   - **Dehiscent dry fruits:** at maturity, the fruits open and the seed is shaken out.

     The follicle develops from a single carpel. When ripe, it opens along the abdominal suture (hellebore, larkspur (*Delphinium* sp.) and peony). The pod is a specially shaped follicle, and when ripe it opens along the abdominal and dorsal sutures (pea, bean, golden chain (*Laburnum* sp.) and black locust).

     The capsule develops from a pistil, which is made up of two or multiple carpels, with either a single or multiple compartments. When ripe, it opens with elongated flaps (horse chestnut); or with calyx teeth at the top (primrose) or with the cap falling off (black henbane); or the seeds shoot out through holes (poppy).

     The siliqua is a distinctive capsule that develops from two carpels. If the siliqua is less than three times as long as it is wide, it is a silicle. When ripe, it opens in two flaps and the seeds disperse from the central partition or septatum (crucifer).
   - **Indehiscent dry fruits:** their pericarp does not open and the seed is therefore not disseminated alone but together with the pericarp.

     The nut is a single-seeded fruit with a dry pericarp. Syncarpous nuts (from two or more carpels) may be wing nuts or samaras (maple, ash, birch, alder and elm) or have no wings (hazelnut, beechnut, acorn, buckwheat and hemp nuts). Nut-like form of the fruit is also the caryopsis of some cultivated grasses (wheat, rye and maize) and the glumaceous fruit of many grasses and some grains (oats and barley), and the achene of composites (sunflower and dandelion), the seed in caryopsisis grown with the seed coat but not in achene. In the case of a glumaceous fruit, the fruit is accompanied by the palea and sometimes also the lemma.

     Schizocarps or splitting fruits are fruits that can split into as many fruitlets as there are carpels (umbelliferous plants and maple). The fruitlets are often connected by a petiole (the carpophore in umbelliferous plants) or have appendages that aid in their dispersal (maple seed wings).

     Some other schizocarpic fruit are multi-seeded closed fruit that breaks into single-seeded fruitlets. These include the loment – consisting of a single carpel that breaks transversely into single-seeded fruitlets (crownvetch), the lomentose siliqua – consisting of two or four carpels that break transversely (radish), and the schizocarpic nutlet (labiate and boraginaceous plants), which breaks into four single-seeded fruitlets at maturity.

2) **Aggregate fruits** develop from flowers with several separate carpels that mature into individual fruitlets aggregated in fruit which is spread as a whole (raspberry, blackberry, apple) or they fall off separately (buttercup, cinquefoils). The fruitlets can be drupaceous (raspberry or blackberry) and apple (apple or pear tree). Aggregate nutlets may be fully dry (buttercup and cinquefoil) or partially fleshy, in which case they may be either on the fleshy receptacle (strawberry) or inside it (rosehip).
3) **Composite fruits** develop from inflorescences that give the appearance of a single fruit (fig, mulberry, pineapple and hop strobiles). Conifer cones are also considered as composite fruits (with the caveat that the seeds develop on the surface of the carpels) but as the pistil is not developed they are not proper fruit. Spruce, pine and fir cones have woody seed scales, while juniper berries have fleshy scales.

DISSEMINATION OF SEEDS:
- **Autochory** – the plant has a mechanism for dislodging seeds and fruit.
- **Allochory** – is carried out with the help of external factors (among types of allochory: water - hydrochory, wind - anemochory, animals and man - zoochory).

2.2 Seed and Seedling Growth and Development

Growth and development are how plants grow in size and develop different shapes and patterns of leaves, flowers and roots. This consists of:

- **Morphogenesis** – the development of cell and organ shape. It depends mainly on the regulation of the direction of cellular enlargement and the control of the plane of cell division.
- **Differentiation** – the process by which cells undergo biochemical and structural changes for the performance of certain functions. The daughter cell attains metabolic, structural and functional properties that are different from the parent cell.

Plant growth and development are the common response of a plant, **the result of interactions between genetic composition and the environment** that are based on physiological processes during growth. The heredity of plants is mainly stored in DNA in chromosomes, which regulates the synthesis of proteins and enzymes providing feedback control of the structure of cells and the response of plants through transfer RNA. Genetic variability in plant growth and development can be caused by gene mutations, recombination and gene migration.

Plant growth and development are the common responses of a plant, the result of interactions between heredity and the environment that are based on physiological processes during growth.

Stress (Larcher 1995) is defined as a significant change in environmental conditions relative to optimal conditions for growth. It causes changes and responses at all functional levels of a given organism. Quantifying the impact of specific environmental stressors is difficult because:

1) some stress factors affect plants permanently, others only intermittently,
2) the importance of individual stress factors varies at different stages of development,
3) the correlations between changes in environmental stressors and plant growth do not necessarily have a clear cause-and-effect relationship,
4) the growth response to stress may only become apparent after a long period of time, when physiological processes are altered to the extent that they affect growth rates or developmental processes,
5) the growth responses depend on prior adaptation (pre-conditioning) to changes in the environment,
6) the effects of stress on growth depend on the vitality of the plant,
7) the changes in plant morphology that occur over time interact with the physiological processes that regulate growth.

Individual environmental factors can be analysed under controlled laboratory conditions, which greatly reduce the variability of results and these are easily reproducible. It should be noted that not all environmental stressors are harmful to the plant. For example, mild drought stress can reduce subsequent damage from transplanting and gaseous pollutants, affect fruit and wood quality, stimulate flowering and induce frost resistance. Storing the fruit under controlled conditions, e.g. low temperature or increased CO_2 concentration, affects their quality and extends the time period of marketing.

Woody plants are most susceptible to dieback during the dormant embryo stage in the seed and during the development of cotyledons in seedlings. Natural regeneration of forests is therefore more dependent on the environmental conditions that are suitable for maintenance of seeds and cotyledons in a suitable physiological state. Even very brief and mild stress at this time will stimulate abnormalities in the physiology of the plant, drastically affecting growth and development during this critical period and causing seed and seedling dieback.

The embryo is an essential component of the seed, therefore much of the care in handling and storage is devoted to maintaining conditions that will keep the embryo vital and ready to germinate at sowing or planting. The start of embryo growth and its development into a seedling involves most of the life processes that are important for a plant: water uptake, respiration, conversion of nutrients into soluble forms, synthesis of enzymes and hormones, metabolism of nitrogen and phosphorus, translocation of carbohydrates, hormones, water and mineral nutrients into the meristem, and the consumption of nutrients for the development of plant tissues.

2.2.1 Anatomy and Chemical Composition of Woody Plant Seeds

The seeds of woody plants vary greatly in size, shape, colour and structure. The smallest ones are barely visible, while the largest ones can reach a few kilograms in weight. Their surfaces can range from smooth to wrinkled. The seeds may have appendages such as wings, fleshy sheaths – arils, thorny vestures, papillas, and hairs. A true seed is a fertilised matured ovule that contains an embryo, a nutrient reserve and a protective seed coat. In practice, the term seed can be used in a broader sense as a functional unit of dissemination. In this sense, it can be a dry fruit with one or more seeds, as well as a true seed.

In our seed production practice, we often confuse the fruit with the seed. For example, an acorn is not just a seed but also a fruit, except that the carpel sheath is thin, woody and does not open. The same is true, for example, of beechnut (Figure 1) and elm samaras.

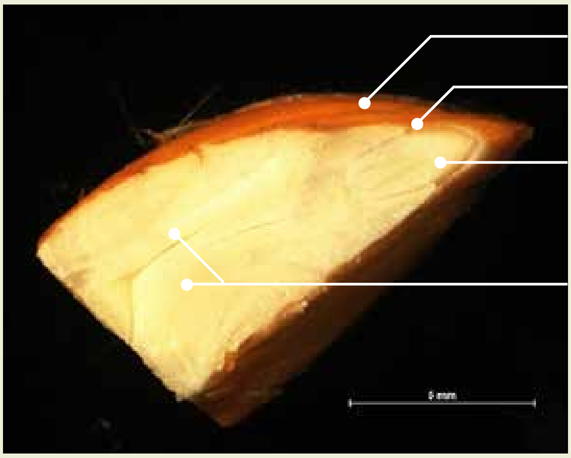

Figure 1: Beech nut in cross-section (photo by Melita Hrenko)

The seeds contain different quantities of nutrients in the form of carbohydrates, fats and proteins. Starch is stored in starch granules in the secondary endosperm of angiosperms, the megagametophyte of gymnosperms and the cotyledons of many angiosperms. Starch is the most abundant reserve carbohydrate, but some gymnosperms store it in small quantities. The most abundant reserve sugar is sucrose, while trehalose is found in beechnut, and stachyose and raffinose in legume seeds.

Fats are found in fat bodies in the form of oils or fats, depending on the ratio of saturated to unsaturated fatty acids.

At least three-quarters of the proteins in the seed are stored in aleurone grains, organelles 1-20 µm in diameter that are surrounded by a single membrane. In the cotyledons of beech seeds, the reserve proteins originate from a series of divisions of vacuoles in which the reserve proteins are deposited. In addition to protein, protein bodies may also contain mineral nutrients and crystals. Some proteins are distributed in the nucleus, mitochondria, proplastids, microsomes and cytosol. Most proteins are metabolically active, but some are not. In addition to protein, seeds contain other nitrogen compounds, amino acids and amides. Other components of seeds are minerals, phosphorus compounds, nucleic acids, alkaloids, organic acids, phytosterols, pigments, phenols, vitamins and plant hormones.

2.2.2 Germination

Germination is the initiation of embryo growth, which causes the seed or carpel coat to burst and a young plant to grow through it. Embryo growth requires cell division and their growth. When germinating, the radicle develops into the seed root, the plumule into the shoot, the apical meristem, which produces the stem. With **hypogeic germination**, subterranean and near-ground germination, the hypocotyl is not extended and the cotyledons remain underground (oak, walnut, chestnut). With **epigeic**, aboveground germination (gymnosperms, beech, hornbeam, ash, most maples, black locust), the hypocotyl elongates and the cotyledons develop above ground into green leaves or needles, which can be very different in shape from the true green leaves, but are capable of photosynthesis. After emergence, not all cells divide as they did in the embryo, but division is restricted to the meristematic zones of the apical meristems of the stem and root.

Rapid seed germination is usually highly desirable because the seed is exposed for a shorter time to infestation by insects, fungi, adverse weather conditions and the feeding habits of birds and rodents. However, in temperate zones, it is often better for a species' conservation and dissemination strategy not to germinate immediately, but to produces large quantities of seed as a seed bank. The most important environmental conditions that affect seed germination are water, temperature, light, oxygen, various chemicals and the interactions between these factors. The environmental conditions needed for germination are often more critical than the factors needed for subsequent seedling growth.

Heavy-seed plants usually have large nutrient reserves, therefore they can survive post-germination stress more easily than plants with small seed and limited nutrient reserves. Some of the environmental conditions that influence seed germination are discussed in more detail below.

Water and drought stress: Non-dormant seeds must take up, or imbibe, a certain amount of water before the physiological processes involved in germination can begin. The absolute amount of water needed is usually small, equal to two to three times the weight of the seed. After germination, water must be available at all times and its required quantity increases with growth and transpiration. Virtually all vital seeds are able to take up sufficient water from the soil at the field water holding capacity of the soil. The effect of soil moisture on seed germination is species-specific. It often depends on the prevailing temperature. Seeds that germinate in the optimum temperature range are less sensitive to water stress.

Flooding stress: The start of the biochemical processes necessary for germination depends on the availability of water and oxygen. The soil oxygen content decreases when water fills large pores in the soil. In general, immersing seeds under water for a short time stimulates germination, while long-term immersion in water causes a decline in seed vitality. Underwater seed survival is also species-specific.

Temperature: Dormant seed may need a low temperature to break dormancy, after which it needs a much higher temperature to germinate. Non-dormant seeds can germinate at low temperatures, but take much longer to do so. The minimum, optimum and maximum temperatures required for germination are species-specific. They also vary between different seed lots. The range of optimum germination temperatures may vary widely for some species, others have a narrow range, while some can germinate in one temperature regime, others need day/night temperature changes.

Light: The species and its origin (tropical species, temperate species, etc.) determine the necessary light, dark or light-dark regime. Most seedlings (of both gymno- and angiosperms), which need light to germinate, germinate fastest when they have a light period of 8 to 12 hours. The wavelength of light is important for some species of ash, birch, elm and some gymnosperms. The wavelength is detected by plants through the phytochrome (a pigment complex that detects mainly red light and, to a lesser extent, blue light, which is better detected by plants through other receptors). As a rule, red light (red, R, 650 nm) stimulates germination, while long-wavelength red light (far red, FR, 730 nm) stops it. The wavelength of the last light flash is important when changing between the two wavelengths. In forest stands under the tree crown, the ratio of R to FR is small. Additionally, the litterfall mainly transmits FR light, which can inhibit germination. Germination is influenced by the physiological state of the seeds – imbibed seeds at the right temperature can germinate even at a lower R:FR ratio.

ENVIRONMENTAL FACTORS AFFECTING GERMINATION:

- water availability
- drought stress or the opposite, flooding stress
- temperature
- light
- oxygen availability
- substrate suitability
- phytotoxic chemicals
- allelopathy

Oxygen: The early stages of germination are characterised by intense respiration. Therefore, the seed usually needs higher concentrations of oxygen to germinate than the seedling does to grow. The relatively high oxygen demand originates from the seed coat, which acts as an oxygen diffusion barrier. Oxygen plays an important role in the respiratory chain, but it can also act as an inactivator of some inhibitors of germination.

Germination substrate: Germination substrates vary widely in their physical characteristics, temperature and availability of water and mineral nutrients. Mineral soils are generally good substrates because they have a high capacity for infiltration, aeration and close contact with water molecules. Peat and decomposing woody debris are suitable germination substrates, probably because of their water-holding capacity. The suitability of the litterfall depends on the type of plant, the amount and type of litterfall and the prevailing environmental conditions. The litterfall affects the plant community both directly (through impact on seed germination and seedling survival) and indirectly (through effects on the availability of light, water and mineral nutrients).

Chemicals: The phytotoxicity of various herbicides, fungicides, insecticides, growth regulators, secondary metabolites, fertilisers and salts can inhibit seed germination or have toxic effects on young seedlings. Phytotoxicity depends on the chemical used, its concentration, the plant species, environmental factors and the method of application. Absorption in the soil can cause long-term effects on plants, resulting in abnormal growth and mortality of seedlings.

Allelopathy: Seed germination and seedling growth are often suppressed by natural chemicals secreted by other plants. Toxic compounds can be extracted from roots or aboveground parts by gasification, leaching, root exudation and decomposition of plant tissues. They contain various phenylpropanoids (phenols, coumarins, quinones, tannins, etc.), terpenes (resins, saps, essential oils, etc.), alkaloids and organic cyanides.

2.2.3 Germination Physiology

Germination includes:

1) imbibition of water in the seed,
2) increased respiration,
3) accelerated enzyme synthesis and degradation,
4) conversion of energy reserves into adenosine triphosphate,
5) altered activity and ratios of plant hormones,
6) decomposition of reserve nutrients and transport of soluble products to the embryo, where cell components are synthesised,
7) accelerated cell division and cellular enlargement,
8) differentiation of cells into tissues and organs.

The order of these changes in the seed may vary and may overlap. But the first stage is always water uptake. Some of the changes in the seed are explained in more detail below.

Imbibition: The seed must first take up water in order to increase the hydration of the protoplasm and start the series of metabolic processes associated with germination. Imbibition also softens the seed coat/carpel sheath, the seed extends so that it bursts and the seed root can grow out of it. Imbibition takes place in three phases:

1) rapid imbibition due to physical hydration of the protoplasm, cell walls and colloids occurs in dormant, non-dormant, vital and non-vital seeds;
2) this is followed by a congestion phase in imbibition as the water potential of the seed increases (water potential depends on osmotic potential, turgor and matric potential), during which the metabolic activity of the seed increases dramatically;
3) the third phase occurs only in germinating seeds, first due to an increase in the cell volume of the seed root and later due to changes in water potential.

Respiration: Respiration involves the oxidative degradation of organic compounds, mainly sugars, starch, fatty acids and triglycerides, to produce the energy in the form of ATP (adenosine triphosphate) that is required for germination. ATP is used to synthesise new cellular components in the seedling and to synthesise enzymes to break down nutritional substances. The oxygen consumption of dry seeds is very low, but quickly increases sharply with imbibition and also varies during the three phases of imbibition listed above. High energy consumption is also required when dormancy of the seed is broken.

Enzyme synthesis and degradation: The enzymes are present in dormant seed, but there is a large increase in their activity at the start of germination. This is caused by enzymes changing from an inactive to an active form, new enzymes being synthesised after imbibition, and enzymes being degraded and new enzymes synthesised at the same time.

Phosphorus metabolism: FThe phosphorus compounds in seeds are nucleotides, nucleic acids, phosphate esters of sugars and phytin. They are important for energy conversion and the activation of enzymes in metabolism.

Reserve nutrient metabolism: Germination is associated with major changes in the metabolic processes of the nutrient tissues. Cells that originally synthesised insoluble starch, proteins and lipids now hydrolyse them. During seed development, the seed is transported to the reserve tissues; during germination, it is transported from these tissues to the cotyledon meristems. The sources and sinks change.

IMBIBITION – WATER ABSORPTION
The seed is moistened – it takes up water by diffusion.

2.2.4 Seed aging

The lifespan of a seed varies greatly, from a few days to several centuries. The survival of seeds in the soil is important from an ecological point of view, in particular for successions and the emergence of weeds. Short-lived seed is seed with a high water content, e.g. in the genera *Taxus*, *Populus*, *Ulmus*, *Salix*, *Quercus*, *Betula* and *Aesculus*. Depending on their dessication capacity, seeds are divided into orthodox (capable of being dried) and recalcitrant (do not survive drying).

Seed that does not survive drying cannot be dried under a relatively high level of humidity without rapidly losing its vitality. Even in a perfectly hydrated state, it loses its vitality rapidly (within weeks or months). These include, for example, oak acorns, walnuts, hazelnuts, chestnuts, horse chestnuts.

Seed capable of drying can be dried to a very low moisture content (normally up to 5%, but possibly up to 2% of fresh weight). For a given genotype, the loss of vitality of seed capable of drying, which is stored for a longer period, depends on time, humidity and temperature. General storage instructions for this seed therefore state:

1) for every 1% decrease in the moisture content of the seed, the storage time is increased by a factor of 2;
2) for every 5.6 °C lower storage temperature, the storage time is increased by a factor of 2;
3) the sum of the storage temperature (in F) and the relative humidity (%) should not be greater than 100 if the share of temperature is less than half.

High-vitality seed tends to germinate quickly, seedlings grow rapidly in nature. Expressing the vitality of the seed depends on genetic composition, development, production, storage conditions and the environment during germination. As the seed ages, its vitality declines: germination and resistance to micro-organisms are reduced, the seed root is shorter during germination, the cotyledons fail to grow outside the seed coat and finally the seed withers. Similarly, structural, biochemical and genetic changes are taking place. The latter include changes in DNA, cytoplasm, RNA and chromosomes (e.g. chromosome cleavage).

2.2.5 Seed Dormancy

Most seeds of woody plants from temperate climates have some degree of dormancy (resting seeds). This ensures that the seed does not germinate, regardless of favourable external conditions.

Seeds that do not germinate when harvested are characterised by **primary dormancy.** Those that are able to germinate when harvested, but lose this ability later, develop **secondary dormancy.** This can be triggered by specific environmental conditions, e.g. very high or low temperatures, burial in the ground, anaerobic conditions and immersion in water.

Dormancy can be an advantage for the seed, as the longer periods of time at low temperatures needed to break dormancy can prevent germination in temperate climates before spring. The seed of some species survives in the soil for several years, allowing the species to survive even in the event of late frosts or droughts in previous years. In places with a dry and wet season, dormancy prevents germination during the short wet season. If germination inhibitors are present in the testa, they are washed away by rainfall, thus allowing germination to take place in the wet season. At the same time, dormancy is also a nuisance for nursery farmers. It is therefore of both physiological and practical relevance.

Causes of dormancy include:

- an immature embryo,
- testa impermeability,
- mechanical resistance of testa against embryo growth,
- metabolic causes in the embryo,
- a combination of the aforementioned causes,
- secondary dormancy.

Some of the causes of dormancy and how they are intertwined are shown below.

Testa dormancy: Causes can be water and gas impermeability of the testa, mechanical barrier of the seed root, retention of inhibitors in the embryo and inhibitor production. It is common in legumes and also in some pines and apple trees.

Embryo dormancy: Sometimes the embryo is immature and needs to mature (e.g. *Viburnum*, *Ilex*, *Ginkgo*). More often, it is morphologically mature but not capable of regrowth and germination (apple, oak, chestnut, pear, sycamore maple, some pines, cypresses, larch, fir, juniper). Physiological dormancy develops in two stages: for example, an ash seed can germinate if it is collected before it desiccates naturally on the tree. The double dormancy is the result of a mechanical barrier of the testa and inhibitors: blackthorn, juniper, yew, holly, Swiss stone pine, etc. In the genus *Acer* some species exhibit testa dormancy (e.g. sycamore maple), while in others (e.g. Norway maple) embryos exhibit all stages in between the two types of dormancy.

The complexity of dormancy occurring in seeds is evident, for example, in the case of ash seeds. The ash (European and narrowleaf ash) has a dry fruit, the samara, in which the seed is dormant, and germination usually starts at least one season after the seed has fallen. The embryo is morphologically perfect, but must grow to at least twice its size during imbibition before germination can take place. However, the growth of the embryo is inhibited by the testa, which hinders access of oxygen. Growth is therefore inhibited until the outer seed coats start to decay. Even a fully grown embryo is dormant and cannot grow from seed until it has been exposed to frost for a few months. Therefore, the first spring germination is inhibited by the growth of the embryo and by the hindered access of oxygen. The frost exposure requirement is reached in nature in the second spring and germination can start in the second spring after the seed has fallen. Germination may also be slower, delayed by slower testa decomposition and insufficient freezing in the second winter.

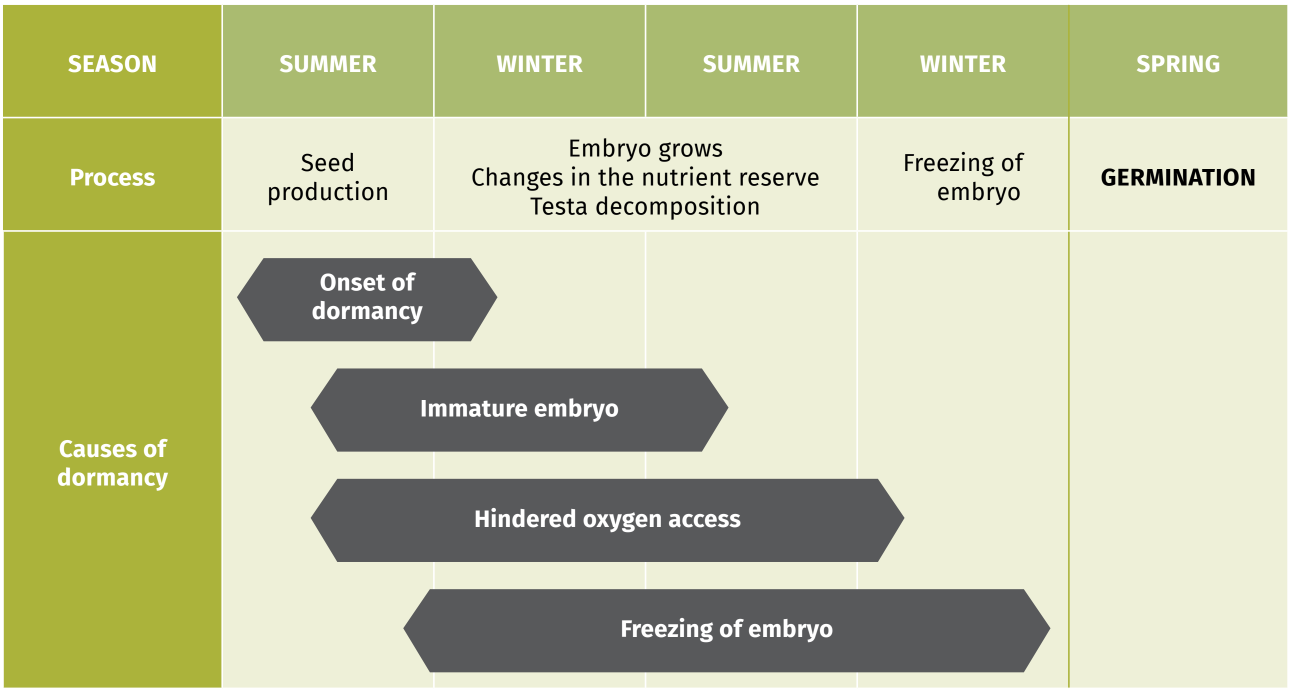

Diagram 1: Combination of dormancy types in European ash seeds (Kozlowski and Pallardy 1997)

Hormones and seed dormancy: In the past, seed dormancy was attributed solely to plant hormones, but the mechanisms are more complex. The ABA concentration (abscisic acid, a stress hormone associated with water stress, senescence and growth inhibition) is correlated with the dormancy rate of apple seeds. The ABA concentration often drops during stratification, but this is not necessarily related to the breaking of dormancy. Gibberellins, which have also been linked to dormancy, are probably mainly involved in metabolism during germination and growth. Cytokinin levels usually increase during germination, but this is mainly due to meristem activity. Auxins are also not associated with dormancy. There are obviously complex interactions between hormones and other endogenous factors in the tissues around and inside the embryo, as well as environmental factors.

Breaking of dormancy: Dormancy can be broken by procedures that affect the metabolism of the embryo, the oxygen and water permeability of the testa and/or reduce the mechanical resistance of the testa to embryo growth. Success rate depends on the type or combination of dormancy types, but for some seeds dormancy cannot be broken.

Procedures:

- seed ripening (interplay of light and temperature),
- stratification at low (or alternately low–high) temperatures in a humid medium or environment,
- with chemicals (hormones, peroxide),
- scarification (in concentrated sulphuric acid or mechanical testa treatment),
- with heat.

WHAT INFLUENCES THE BREAKING OF DORMANCY:

- **seed ripening**
- **stratification** (low or high temperatures in a humid medium or changing temperature conditions)
- **chemicals** (hormones, peroxide)
- **scarification** (mechanical or "acid-softened" testa)

2.2.6 Seed Quality Analyses

On representative samples of the seed, the following analyses are carried out:

- purity (% of pure seed, abnormally small seed, mechanically damaged seed, insect-damaged seed, rodent-damaged seed, fungus-infested seed, empty seed, seed of other species, impurities resulting from cleaning of the seed (e.g. wings, parts of flowers, twigs, etc.), impurities of mineral origin (e.g. sand), other impurities),
- mass of 1000 seeds,
- moisture content (%),
- vitality (%, for dormant seed: cut seed test, isolated embryo test, seed X-rays images, colour tests: indigo carmine, tetrazolium – TTC),
- germination test (performance depends on the species; germination is expressed in %, and the germination curve over the time interval and the time taken for 50% germination provide additional information on the physiology and, in particular, the ageing of the seed). The results of a germination test can be expressed in different ways, including germination percentage, germinative energy (refers to the percentage of seeds in the sample that have germinated in a test up to the time when the number of seeds germinating per day reaches its peak), and germination capacity (total number of seeds in the sample that have germinated in a test, plus the number of seeds remaining ungerminated but still sound at the end of the test, in %).

Internationally comparable methods are mainly based on the International Seed Testing Organisation (ISTA) method. They are usually carried out in four replications of 100 seeds each.

For the purposes of inspection and professional control, the maximum number of seedlings produced per kilogram of seed (depending on the % germination or % vitality and the % purity of the seed) may also be calculated.

2.2.7 Emergence of Seedlings

The cotyledons are crucial for the growth and development of the seedling, as they store reserve nutrients and minerals, photosynthesise actively and transport the substances needed for growth to the apical meristems.

Hypogeal seedlings: Cotyledons remain underground or on the earth's surface, and are mainly storage organs for nutrient reserves (Figure 2). The reserves are sufficient for development until the first leaves are fully developed. These take over the photosynthetic function until the true green leaves are fully developed. Leaf development and the number of leaf shoots per year depend on the environmental conditions.

Epigeal seedlings: The physiology of epigeal seedlings is more species-specific. The above-ground cotyledons of some species can store significant amounts of spare carbohydrates. For example, pines accumulate only small amounts, but very quickly start photosynthetic activity. After germination, the pine seedling is a sink for carbohydrates. The synthesis site shifts continuously from the cotyledons to the first needles and then to the second needles, and reserves are drawn first from the cotyledons

GERMINATION CAN BE:

- **HYPOGEAL:** Cotyledons remain underground or on the earth's surface, and are mainly storage organs for nutrient reserves.
- **EPIGEAL:** In epigeal germination, hypocotyl grows faster than epicotyl. However, even in species with epigeal germination, the aboveground cotyledons of some species can store significant amounts of reserve nutrients.

and then from the first needles for the development of the second needles. During the cotyledon stage, seedlings are particularly susceptible to environmental stress. The growth of primary needles depends on the availability of assimilates from the cotyledons. Low temperatures or low light intensities during the cotyledon stage can prevent the normal development of primary pine needles. After transferring the seedlings from a cold environment to a suitable temperature, primordia of primary needles immediately develop and grow.

Species without endosperm have cotyledons adapted for both nutrient storage and photosynthesis (e.g. black locust). Cotyledons of species that have an endosperm also transfer nutrients from the endosperm to the growing tissues. Early seedling development can be divided into three stages (Figure 3):

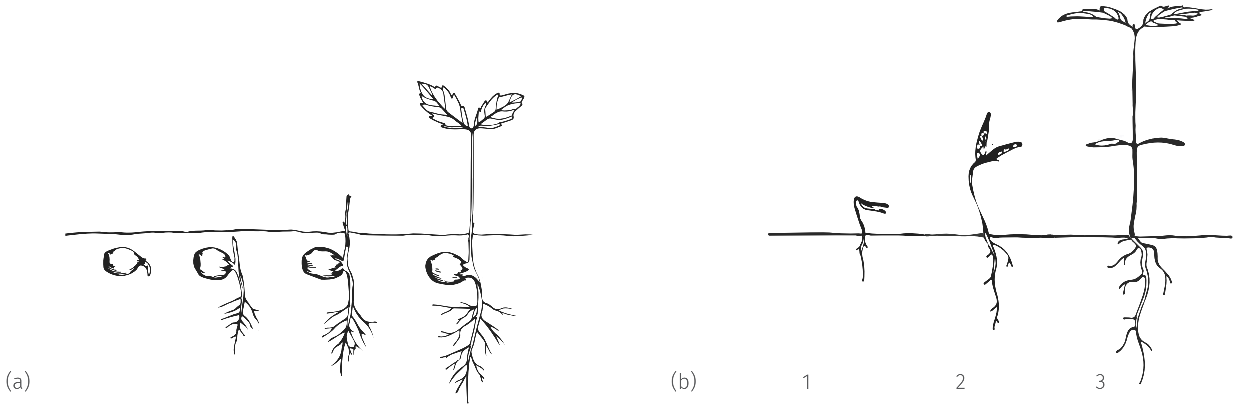

Figure 2: Schematic representation of (a) subterranean (hypogeal) and (b) aboveground (epigeal) germination (after Kozlowski and Pallardy 1997) (1, 2, 3: emergence phases)

1) in the first phase, from the time the cotyledons emerge from the soil, expand and develop, assimilates are transported from them mainly to the hypocotyl and the root;
2) in the second phase of development, i.e. until leaf development, the assimilates move from the cotyledons mainly to the apical meristem of the stem and the young leaves;
3) in the third phase, as the cotyledons age, the function of supplying assimilates to the roots is transferred to the first true leaf.

The development of cotyledons from storage, transfer, photosynthesis and ageing functions requires major changes in enzyme activity. Cotyledons of different species also store, use and take up mineral nutrients differently. In the next phase, they grow to their full-grown size, using up all nutrient reserves for meristematic zones, cell differentiation and chlorophyll synthesis. During this time, the cotyledons undergo positive net photosynthesis. As they age, they begin to yellow and finally fall off. Their lifespan varies depending on the species, and is usually shorter in the case of gymnosperms than in the case of angiosperms.

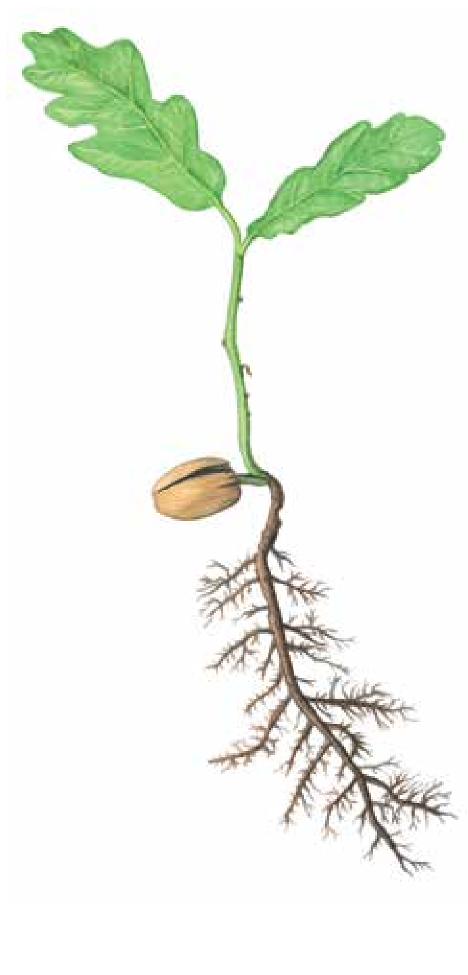

Figure 3: One-year oak sapling (drawing by Eva Margon)

2.3 Tree Nursery Basics

The growth and survival of seedlings depend on their quality, which in turn depends on the quality of the seed, tree nursery practices and the management of the seedlings during manipulation from the tree nursery to planting in the forest.

Seedling quality is the result of several physiological and morphological characteristics. Individual signs include. dormancy status, water and nutrient ratios, morphology, root growth potential, and frost and drought resistance. Individual signs are difficult to evaluate in different ways. Seedling size and thickness at the root collar are very useful, but in specific growing conditions, e.g. on arid sites, a small seedling with a small transpiration area may perform better than a large seedling.

Tree nursery practices should be aimed at preserving those physiological processes of the seedling that will help it to grow well and survive in the forest. For example, the loss of a large number of fine roots during transplanting can cause dehydration (desiccation) of the transplanted seedlings. Successful transplanting and planting in the wild requires an appropriate root to aboveground part ratio and rapid root growth in a large soil volume that allows rapid absorption of water and mineral nutrients. Nutrient reserves are important because plants can only regain positive net photosynthesis several weeks after transplanting. Producing quality seedlings requires attention to all phases of tree nursery work: soil and bed preparation, planting method and density, fertiliser application, irrigation and disease and pest control. In addition, root pruning is sometimes necessary (if this does not damage the oblique roots, does not open up areas for pathogens to enter, and minimises the impact on genetic diversity caused by possible root damage to faster- and deeper-rooting seedlings), as well as colonisation with mycorrhizal fungi.

Storage of seedlings often requires adequate cold storage facilities where seedlings can wait in a dormant stage until they are ready to be planted in the wild. They must not dry out during storage, so a humidity of over 85% is required in cold storage facilities. As such conditions stimulate mould proliferation, they need to be inhibited by low temperatures. Suitable storage temperatures range from +3 to -6 °C. Attention should also be paid to the photoperiod, as there is a practice of storing seedlings in constant light. The date of extraction, the start and the total time in cold storage facility are also important. The longest the seedlings can be stored is eight months in the High North. During storage, carbohydrate reserves are used up, with consumption rates of 0.4 to 0.6 mg per gram per day measured for some conifers.

Handling seedlings: Transplanting of "bare-root" seedlings is only allowed during the dormant stage of the seedlings. The roots should remain moist at all times and the air temperature should be low, even in the areas where they are buried in the ground and sorted. Frail seedlings or those that have suffered insect-damage should be removed. Exposure of the roots can cause growth inhibition and affect seedling survival in the short term. Even after planting in the wild, roots are often exposed to water loss because they grow too slowly to keep up with transpiration losses. Transplant shock can take a long time, up to several years.

Growth potential of seedlings after planting in the wild: The growth of the seedlings depends on the species and genotype of the plants. Many gymnosperms grow much more slowly than angiosperms. The differences probably occur because of species-specific photosynthesis, leaf area index, leaf longevity and the distribution of assimilates in the plant. Growth is also affected by competition with other plants, e.g. grasses strongly inhibit the growth of conifer seedlings.

LEAFLET:
The protocol for handling of forest tree seedlin-gs is described in a leaflet published under the LIFE project LIFEGENMON (LIFE ENV/SI/000148) (in Annex 1).

3

THE PHYSIOLOGY OF FLOWERING AND SEED PRODUCTION IN FOREST TREES

Studies of flowering and fruit production in forest trees are much more difficult compared to herbaceous plants. Trees are mostly too tall to study the physiology of flowering under controlled conditions, have a long juvenile period before flowering, long reproductive cycles, complex flower bud composition and an unpredictable pattern of flower initiation.

3.1 Flowering Periodicity

Trees go through a relatively long juvenile period during their development, which can range from one to more than 40 years in different species (Table 1).

There is a young (juvenile, vegetative) stage and an adult (mature, reproductive) stage, also known as the reproductive stage, when the tree is in “full bloom”. The transition to this phase is species-specific and depends on the cultivating conditions. After the transition to the reproductive phase, the tree retains the ability to flower, and the flowering periodicity (Table 1) varies between and also within species, depending on the genetic makeup of the individual tree. Differences in the periodicity of the flowering are particularly large in deciduous trees, but smaller in coniferous trees. The formation of cones, fruits and seeds is a climax phase during long reproductive cycles that can be influenced by many different factors. Although the periodicity of flowering is well documented, its causes are not sufficiently explained due to the complex interactions of several endogenous and exogenous factors.

Many tree species do not flower every year - the flowering periodicity is both hereditary and dependent on environmental conditions

Table 1: Onset of flowering age and its periodicity in some tree species (adapted from Owens 1991 and USDA 2008**)

TREE SPECIES	AGE AT THE ONSET OF SEED PRODUCTION PERIOD (years)	INTERVALS BETWEEN SEED YEARS (years)
Acer platanoides	25–30	1–3/1**
Alnus glutinosa	15–20/6–7**	2–3
Betula pendula	15	1–3/2–3**
Fagus sylvatica	50–60/60–80**	5–15/3–20**
Fraxinus excelsior	25–30/15**	3–5/1–2**
Populus nigra*	8–12	1
Populus tremula*	8–10	–
Quercus petraea	40–50/40**	2–5/5–7**
Quercus robur*	20	2–4
Abies alba*	25–30	2–3 (4–6)*
Abies grandis	40–45/20**	3–5/2–3**
Larix decidua	25–30/10**	3–5/3–10**
Picea abies	30–35/40–60**	3–5/4–13**
Pseudotsuga menziesii	30–35	5–7
var. *glauca**	20	3–10
var. *menziesii**	7–10 (in case of production 20–25)	2–11

* At higher altitudes.
** According to USDA 2008.

3.2 Reproductive Cycles

Three reproductive cycles are known for most tree species from the temperate climates:

A) The most common cycle is two years from the ovule development through a period of winter dormancy, to seed release. Reproductive buds are developed in summer and pollination occurs the following spring. Usually, it takes only a few weeks between pollination and fertilisation. Embryo and seed development is rapid and uninterrupted. Mature seeds can be released as early as late summer in the year of pollination. Seed retention over this time is usually determined by climatic or biotic requirements, which are species-specific and depend on how the seed of that species is disseminated.

B) The second reproductive cycle is similar to the first, except that usually it takes a year between pollination and fertilisation.

C) The third reproductive cycle is also similar to the first, except that after the embryo and seed development starts it is interrupted in late summer or autumn. The immature seed overwinters and development continues the following spring.

3.3 Initiation and Stimulation of Flowering

The initiation of flowering involves the transition of an indeterminate vegetative terminal or axillary apical meristem (apex) to a determinate reproductive apical meristem, which develops into a flower or flower shoot in gymnosperms or a cone in conifers. The classical explanation of flowering initiation in herbaceous plants is that flowering is stimulated in different parts of the plant, from where it is transferred within a few hours or days to the apex, where it passes from the vegetative to the floral meristem. Most plants blossom depending on environmental factors such as photoperiod and temperature. In woody plants, however, this "simple" classical model does not hold, because flowering depends on a series of developmental processes, which are sequentially determined by the hormonal and/or mineral balance in the substrate, and modified by environmental factors acting on different plant organs.

Most measures to improve flowering involve changes in environmental factors: the number of possible combinations is large and their effects are often unreliable or contradictory. Direct applications of plant growth regulators in combination with other measures (fertilisation, pruning, thinning, etc.) are also used.

FLOWERING INITIATION
Most plants blossom in accordance with factors in their environment, such as photoperiod and temperature. In woody plants, however, this "simple" classical model does not hold, as flowering depends on a whole series of genetically conditioned developmental processes.

The morphology and anatomy of the reproductive buds are species-specific, and the timing and location of the flowers are relatively uniform within each species. The environmental factors of particular interest include the following:

A) **Temperature.** High summer temperatures tend to stimulate flowering, which was already described for beech by Linnaeus in 1751. Similar impacts have been described for birch, pine, fir, spruce and Douglas fir. The effects of high summer temperatures, often combined with the application of certain hormones (mainly gibberellins) and drought stress, have, for example, been studied in spruce seedlings cultivated in a greenhouse.

B) **Light intensity and photoperiod.** Most studies show that the effects of light intensity are indirect, depending on crown illumination, terrain gradient, shading and the genetically conditioned distribution of flower buds in the crown. Branches that are more exposed to high levels of light tend to flower more abundantly. Thinning has stimulated flowering in some pines, Douglas-fir and some fruit-producing species. In the case of maple, canopy density influenced the distribution of male and female flowers. In tree species, photoperiod does not have as direct an effect on flowering as in herbaceous species. For three pine species, flowering was not dependent on photoperiod, while in spruce it affected the development and number of cones. In pot experiments, where the effects of light and photoperiod can be separated from those of temperature, it has been found that birch, spruce and pine seedlings grown in continuous light or long daylight flower faster than control seedlings (at 10-12 months compared to 4-5 years), and also much faster than trees grown in the wild (15-20 years) (data compiled by Owens 1991). They concluded that the transition to the reproductive phase in these tree species depends more on reaching a certain size than on the number of annual growth cycles.

C) **Water stress and roots.** In pine, spruce, fir, Douglas fir and beech, a positive correlation was found between increased cone/beechnut production and low rainfall. However, roots also play a more direct role in stimulating flowering, not just an indirect one through the uptake of water and nutrients. This role is mainly in the production of plant hormones, cytokinins and gibberellins, which are synthesised in the roots and transported to the aboveground shoots. More direct results on the impacts of drought stress on flowering can only be obtained in pot experiments. They found that a drought stress of -1.4 to -2.0 MPa before dawn has an effect on higher cone production in Douglas fir. Root pruning, or even flooding of the root system, can impact the root activity cycle and hence the flowering.

D) **Mineral nutrients.** In otherwise equal conditions, trees growing on better soils produce more seeds than trees on poorer growing sites. That is why the use of fertilisers is one of the oldest measures to stimulate flowering. In conifers, the main nutrient that impacts flowering is nitrogen. The form of nitrogen (ammoniacal or nitrate) depends on the tree species. In tree species with symbiotic nitrogen fixation capacity (alders, legumes), the use of phosphate fertilisers has been tested with different effects.

E) **Other stress factors.** Any damage to the tree can stimulate flowering, therefore various tree girdling, tying and strangulation techniques are used to accelerate flowering. These procedures are intended to increase carbohydrate concentrations in the crown by inhibiting transport towards the roots. The theoretical starting point is to promote a high C/N ratio, which favours the development of flower buds and inhibits the development of vegetative buds. Opinions on these methods vary. Induction of flowering can also be the result of pruning of branches, grafting, bending, wounding, resin accumulation, defoliation and other biotic influences (pathogens and pests), cold damage and damage to the root system, but there is no simple explanation for the effects of all these stress factors.

The lack of positive relationships between environmental factors and flowering may be related to endogenous factors in the tree. A good seed year is usually followed by a year with no or poor seed production. The growth and development of reproductive organs consume a lot of metabolites, therefore it may take a year or more for the tree to regain a condition suitable for the development of reproductive shoots. The appropriate internal conditions must be matched by external environmental factors, which is why the periodicity of flowering and seed production is relatively infrequent in many tree species.

Controlling flowering using **growth regulators** serves three purposes:

1) it prevents early flowering so that most of the energy can be used for vegetative growth and development;
2) it stimulates flowering of mature trees to increase fructification;
3) it regulates flowering and fructification over shorter periods.

Various gibberellins are mostly used as stimulators of flowering in conifers, while these can inhibit flowering in angiosperms. The timing of gibberellin application is important and should precede the development of reproductive buds. Combinations of other plant hormones, e.g. auxins, cytokinins, abscisic acid and ethylene, can either stimulate or modify the plant's response to gibberellins, but on their own they are usually without effect. Hormonal regulation of flowering and endogenous plant hormone content in buds, shoots and roots at different stages of development remains one of the most interesting problems in basic studies of plant physiology. However, after hormone treatment, if appropriately timed and short-lived, the seeds should be of the same quality as normal seed.

From Section 3.1, it can be summarised that the onset of fructification is mainly related to the transition from the vegetative to the reproductive phase, flowering period, hormonal regulation and environmental factors. Measures to promote flowering can include naturally adapted thinning of stands, but most other measures can only be implemented in the artificial environment of seed orchards. These include grafted vines, which can help plants transfer more quickly into the reproductive phase, pruning of the crown and roots, girdling, heat or drought stress, spraying with hormones or injecting hormones or hormone inhibitors, and the use of various fertilisers. Depending on the species-specific responses of forest trees, flowering can be stimulated by a combination of hormonal and environmental stress-based measures. However, the effects of symbionts and parasites on flowering are still understudied, not least because of the multi-hormonal effects on the growth and development of the plants.

Due to the high maintenance costs of conventional seed orchards, the risk of pollination with unselected basic material (naturally occurring trees of the same species in the vicinity of the plantation), and the much better possibilities of manipulation and modification of environmental factors, the development of seed orchards has also moved to potted orchards in greenhouses. For example, the hybrid larch *(Larix x eurolepis)* orchard at the Forest Research Centre in Graupa near Dresden may be completely covered by netting during pollination, but otherwise it is exposed to external influences or modified in terms of temperature, light and water regimes in an uncovered or partially covered greenhouse.

Photo 2: Spruce (*Picea abies* subsp. *obovata)* cones (photo by Hojka Kraigher)

4

BASICS OF FOREST GENETICS

4.1 Mendel's and Morgan's Laws

Genetics studies heredity in organisms and variability of genes. Each gene in the DNA of an organism has a specific site **(locus)** on a particular chromosome. In sexual reproduction recombination of the genes of two parents occurs, resulting in genetically different offspring. Sexual reproduction is the main source of genetic diversity (variation) in offspring; the second is mutations. Adaptation to environmental conditions during the process of evolution is based on genetic variation in the population. Genetic diversity within a population is the basis for evolution by natural selection.

GENETIC DIVERSITY - THE BASIC LEVEL OF BIODIVERSITY
Adaptation to environmental conditions during the process of evolution is based on genetic variation in the population. Genetic diversity within a population is the basis for evolution by natural selection.

In 1860, **Gregor Mendel** proved that parents pass on genes to their offspring that retain their identity over a series of generations. He observed hereditary traits that showed different characteristics in pea varieties that could undergo true cultivation, i.e. the offspring were all of the same variety after self-fertilisation of the parents. The traits tested in crossing of two varieties were observed from the parent generation (P) through the first generation of offspring (F1), then by self-fertilisation of these into the next generation of offspring (F2, etc.): 75% of the F1 generation offspring showed **dominant** traits, 25% recessive traits, i.e. a ratio of 3:1. Mendel explained the law by the existence of two alternative forms of the same gene, two (or more) **alleles,** with each offspring inheriting only one gene (one form or one allele) from its parents. During meiosis, two homologous chromosomes of the two parents cross and diverge, and the second division produces four offspring, each carrying one pair of chromosomes and one series of alleles on each chromosome. The segregation of alleles for a particular trait from two parents into different gametes – the offspring – is explained by the law of segregation. In development, only the dominant allele is expressed, or only when two recessive alleles are distributed the expression of a recessive trait takes place. If two alleles of a certain gene are different after fertilisation, only the dominant trait is expressed, but not the recessive trait. **Homozygotes** have identical alleles for a particular trait, while **heterozygotes** have two different alleles for the same trait.

According to **the law on independent assortment** each pair of alleles is independently distributed (segregated) into gametes. Mendel explained the law by observing the inheritance of two independent traits (the colour and shape of the seeds of the pea, the pods and the size of the peas). The F2 generation has four possible phenotypes in a 9:3:3:1 ratio. Mendel's laws of inheritance express rules for the random distribution of independent trait characteristics.

However, the relationships between genotype and phenotype are usually not so simple. In incomplete dominance, the heterozygote shows an intermediate phenotype between the two types of homozygotes. In codominance, a heterozygote expresses the trait characteristics of both its alleles. In a population, many genes exist in a large number of alleles. The ability of a single gene to affect multiple (apparently independent) phenotypic traits is called **pleiotropy. Epistasis** means that one gene influences the expression of another. Some of the characteristics are **quantitative** and vary continuously. This suggests that there is a multi-gene inheritance, for the **additive effect** of two or **multiple genes** on a single phenotypic characteristic. Quantitative characteristics, which are also influenced by the environment, are **multifactorial.**

The theory of inheritance was further developed by **Thomas H. Morgan,** who located genes on chromosomes. **Linked genes** are inherited together, not independently, because they are located on the same chromosome (and often physically adjacent). Each chromosome consists of hundreds of thousands of genes. These genes are linked and are not inherited independently. Some genes are linked to sex chromosomes and their inheritance does not follow the rules of Mendelian genetics. The phenotypic effects of some sex-linked genes depend on whether they originate from the mother or the father.

WHICH GENES DO NOT FOLLOW MENDEL'S LAWS:
- linked, sex-linked, chloroplast and mitochondrial genes

Also the genes that are not linked to chromosomes in the nucleus do not follow the rules of Mendelian genetics: mitochondria and chloroplasts have their own genes. Where the cytoplasm in the zygote originates from the egg cell alone, some of the phenotypic traits of the offspring depend exclusively on maternal cytoplasmic genes.

4.2 Population Genetics, Evolution and Speciation

For the most part, each population produces many more offspring than can survive in the environment. These offspring are different and carry different hereditary traits. Between 1840 and 1858 **Charles Darwin** conceived the theory of the evolution of species (evolution through modification) based on natural selection. Natural selection is based on varying survival and reproductive success in a population, leading to the evolution of species adapted to different niches in the environment. Environmental influences on gene expression can additionally lead to (also heritable) epigenetic modifications (Nanson 2004).

A population is a limited group of specimens of the same species. **A species** is (in a simplified manner) a group of populations whose individual specimens can reproduce among themselves and produce fertile offspring (under natural conditions). Each species has a geographic range, in which individuals are unevenly distributed, mostly clustered in a few populations. A population may be isolated from others, so that genetic material is exchanged only occasionally. Alternatively, a population may directly pass into another in a transition zone, but even in these populations, individual specimens in the centre of the range are more likely to interbreed than those at the edge of the populations. Therefore, they are generally more similar to each other in the centre of their distribution than to individual specimens from other populations. The entire aggregate of genes in a population at a given time is called the **genetic pool.** It consists of all alleles at all gene loci in all individual specimens of that population. Diploid species have each locus represented twice in the genome of an individual, which can be either homozygote or heterozygote for that homologous locus. If all individual specimens of a population are homozygotes for the same allele, the last one is **fixed** in the genetic pool of that population. Usually, two or more alleles of the same gene are represented in the genetic pool at different relative frequencies. Allele frequencies are reflected in genotype frequency. The frequency of alleles (and, in a derivative variant, genotypes) in a population is reflected by the term **genetic structure** of the population.

A population that is consistent has a gene pool described by **Hardy-Weinberg equilibrium** or **equation.** The simplified form, which describes only two alleles, indicated by *p and q,* is:

$$p^2 + 2pq + q^2 = 1$$

The equation allows us to calculate the frequency of individual alleles in a population's genetic pool if we know the frequency of the underlying genotype, and vice versa. The Hardy-Weinberg equilibrium is maintained in the population::

- if the population is very large;
- if it is isolated from the remaining populations;
- in the absence of inheritance of mutations;
- if there is random sexual reproduction;
- in the absence of natural selection.

Microevolution is the change in the frequency of alleles (or genotypes) in a population from one generation to the next. Causes of microevolution include:

- **genetic drift** – random variations in allele frequency in the genetic pool (due to random sampling of individual specimens), variations because of a bottleneck – an extreme reduction in population size, or variations because of the impact of forming a new population with a very small number of starting specimens;
- **gene flow** – a population gains or loses alleles through the migration of fertile specimens or gametes between different populations;
- **mutations** – changes in the DNA of a particular specimen;
- **non-random sexual reproduction** – individual specimens reproduce more frequently by mixing the genetic material of adjacent specimens than with others, which leads to inbreeding; another form is selective reproduction when individual specimens carefully select a reproductive partner, e.g. one as similar to themselves as possible;

- **natural selection as a mechanism of adaptive evolution** – the influence of selection on different characteristics can act as a stabiliser, oriented either towards aligning or division of species.

Of all the causes of microevolution, natural selection is the only one that leads a population to adapt to its environment. Other causes can act positively, negatively or neutrally on adaptations to environmental conditions. Natural selection is made possible by **genetic diversity** within and between populations – **variation.** Within a population, it is characterised by **polymorphism** for a particular characteristic. **Measures of genetic diversity** are the proportion (%) of loci (genes) that have two or more alleles in a population; the average proportion of loci, i.e. heterozygotes among individual specimens in a population; the number of alleles (or allele diversity) per locus; heterozygosity and nucleotide diversity.

Most species show some **geographic diversity** representing differences in genetic structure between populations. In a geographically widespread species that thrives in several ecological zones, differences between populations can arise within a few generations because of different **selection pressures** which, over large areas, can lead to the formation of **geographical races** and within these **ecotypes** adapted to particular environments, and within these to populations, which in forestry are called **provenances.** Thus, a provenance is the population of forest trees – the forest stand in a particular geographically restricted area; such **adaptive polymorphism** also corresponds to genetic **polymorphism** (Nanson 2004). Besides natural selection, genetic drift can also contribute to polymorphism. A specific form of geographical diversity is **clinal variation,** in which the transition from one ecological zone and geographical race to another is continuous. Genetic diversity is maintained by diploidy and balanced polymorphism – genes are still exchanged between ecotypes and geographical races, maintaining a single – continuous – **genetic pool** of a species. **Marginal (edge) populations** of forest trees thrive under restrictive environmental conditions for a particular species, therefore they become rapidly specialised through natural selection, losing genetic diversity in the process. In these populations, species can form gradually, i.e. **speciation** occurs.

Thus, the adaptive polymorphism of a species over a large geographical area corresponds to the overall adaptive capacity of the species over its entire distribution area, due to speciation of peripheral races and flexibility in gene transfer between races. Long-distance gene transfer has proven to be very important, particularly in maintaining the species' resilience to climate change and other large-scale disturbances.

GENETIC DRIFT:
Random variations in allele frequency in the genetic pool (due to random sampling of individual specimens), variations because of a bottleneck - an extreme reduction in population size, or variations because of the impact of forming a new population with a very small number of starting specimens.

It should be borne in mind that adaptation involves the entire biological cycle, vegetative and generative, of a population, but it is often the reproductive phase that limits the population's ability to thrive and adapt to changes in the environment (Nanson 2004). The current distribution area is largely the result of several factors, such as migration routes from ice-age refugia, where the genetic pool of the species may have been diverse, often very narrow. Therefore, both ecological and historical sources of variability need to be taken into account when assessing the performance and adaptive capacity of a species in a given area. Hence, the ecological conditions in each **region of provenance** do not reflect all the potential threats to the thriving of tree populations, and natural forest rejuvenation is not always optimal, nor the most productive, nor the best adapted to the current conditions in that area. The results of provenance experiments and simulations of possible changes in environmental conditions should also always be taken into account

A biological species is a population or group of populations whose individual specimens can interbreed in nature and produce fertile offspring, but cannot produce fertile offspring with individuals from other species. The evolution of the species is affected by prezygotic barriers such as habitat isolation, isolation due to different behaviour/phenology, temporal, mechanical isolation and gamete isolation, reduced hybrid vitality, reduced hybrid fecundity and hybrid collapse. The concept of a biological species is not perfect; there are exceptions. This has led to the development of other concepts of species: morphological, cognitive, cohesion, ecological, evolutionary, etc.

The origin of species includes:

- geographical isolation, leading to **allopatric speciation;**
- geographical overlap of species, leading to **sympatric speciation;**
- genetic changes in populations that can include:
  - ◊ interspecies hybridisation,
  - ◊ agamospermia,
  - ◊ self-pollination.

PROVENANCE
is a population of forest trees - a forest stand in a specific geographically limited area.

Conversely, introgression can allow separate or nearly separate species to merge into one in areas of introgression where **natural hybridisation** occurs. Such a hybrid species is. e.g.. *Abies borisii-regis* in the introgression zone between *Abies alba* and *Abies cephalonica*. The difficulty in identifying oaks and oak acorns arises because of natural hybrids between pedunculate and sessile oaks, Downy oak, sessile and pedunculate oaks, etc.

Where the areas of related species overlap, **NATURAL HYBRIDISATION** can occur between them.

5

FOREST REPRODUCTIVE MATERIAL PRODUCTION AND TREE NURSERY TECHNOLOGY

5.1 Fructification in Forest Tree Species

Onset of fructification: A practical rule emphasises that light-seeded trees, such as birch and alder, start producing fruits at 10-15 years of age, and heavy-seeded species, such as beech and oak, at 50-60 years of age. The timing of the onset of seed production is also influenced by external factors, so that solitary trees start producing fruits much earlier than those in the forest.

Periodicity of fructification: The tree seed production is not the same every year, but is mostly periodic. In mast seed years, which follow each other every few years in certain species, fructification is stronger, with little or no seed production in between.

Fructification intensity: For the conservation of forest genetic diversity and the economics of seed husbandry, it is important to assess the intensity of fructification. From the seed harvest practice, based on ocular inspection, the characteristics and ranking of the fructification are derived, as shown in the following table. When assessing the ranks of the fructification intensity, the first and fifth ranks are the most significant, while the remaining ranks can shift continuously from the second to the third and from the third to the fourth.

Table 2: Assessment of the fructification intensity

Fructification rank	Description of rank	Characteristics	Proportion of branches with seeds (%)	Suitability for harvesting
1	no fructification	healthy mature trees do not bear fruit or only individual specimens and/or individual branches bear seeds	0–10 %	**harvesting is not possible**
2	poor fructification	poor fructification, mainly of marginal and dominant trees, very small quantities, poor quality	11–40 %	**insufficient for harvesting**
3	medium fructification	good fructification, mainly of dominant and marginal trees, poor fructification of the remaining trees, poor quality	41–70 %	**conditionally acceptable for harvesting**
4	Strong fructification	very good fructification of most of the fruit-producing branches of the marginal and dominant trees, good fructification of the remaining trees, good quantity and quality	71–90 %	**economically viable for harvesting**
5	Massive fructification	virtually all branches in the crown of all trees of the selected species show high fructification, ideal for obtaining FRM of high genetic diversity	91–100 %	**recommended for harvesting**

There are also more precise methods that can be used for estimating fructification intensity, by:

- analysing the number of fruits per branch from 10-20 trees in a stand;
- counting the number of visible fruits/seeds/cones per branch/crown in a given time (e.g. 30 seconds);
- collecting the seed in gauges of known dimensions, distributed evenly throughout the stand (at least 5 gauges), calculating the amount of seed collected per hectare (useful for wind-dispersed seed);
- the experimental harvesting method is used to determine fructification, particularly in conifers: a few cones from a few trees are harvested, the cones are cut and the proportion of full and empty seed is determined;
- the average method data on the optimum seed production per hectare for a given species, compared with the experimental seed collection per tree or per specific area in a given year.

Light-demanding tree species, light-seeded species and solitary trees produce fruits more often, while shade-tolerant species, heavy-seeded species and trees growing in stands or shade produce seeds less often. It is important to collect the seed when fructification is strong and save it for years when it is poor, or to grow larger quantities of seedlings.

Forecasting flowering and fructification: It is useful to be able to predict the year of full fructification in advance. The first, but very unreliable, prediction can be made as early as autumn if we are dealing with a tree species in which the flower buds are different from the leaf buds. A higher number of flower buds can indirectly predict a stronger flowering. This is possible, for example, in the wild cherry, where the flower buds are distinctly rounded compared to the leaf buds, but is less certain, for example, in the yew, where the female buds are quite similar to the leaf buds.

Later estimates of fructification intensity based on flower abundance may be more reliable. It is important to bear in mind that pollination (even with pollen from incompatible species) is often sufficient for the initial growth of the fruit, while fertilisation must actually take place for durable fruit growth. Even at this stage, these are still estimates, as the actual fructification is highly dependent on a number of external factors that can significantly affect the (lack of) success of the fertilisation or damage the already fertilised flower to such an extent that it does not develop into a fruit. The external factors that most often cause poor fructification despite strong flowering are low temperatures, prolonged drought during flowering or early fruit development, and infestation by plant diseases or insects. Therefore, a more reliable estimate is mostly only possible in our species when the fruits are well developed, or 15-30 days before ripening.

Time of harvest: The timing of harvest can have a decisive influence on the subsequent germination. It is therefore necessary to know the state of full ripeness of the seeds for each species and the mean and median maturation time of the seeds of that species in a particular area.

The fruits can also be important food for forest animals, therefore it is useful for seed production practice to collect them as soon as they are sufficiently ripe. For example, for wild cherry in June or July, for yew and rowans (Sorbus) in September or October, and for wild pear in October or November.

Seed variability: Seeds differ with respect to:

- location in the crown or cone (topophysis),
- age of the stand (cyclophysis),
- site characteristics (periphysis).

5.2 Collecting, Cleaning and Sorting of Seeds

When collecting seed, the first thing to be determined is the quantity needed, the ripeness, the duration of production and the method of harvesting. Immediately after finishing collection in the forest, it is advisable to clean the seed before transporting it for final processing for sowing or storage.

Collecting seeds from the ground: Collecting from the ground is the simplest method to produce seeds. This is mainly used to collect heavy seeds/fruits, e.g. acorns, beechnuts, chestnuts, walnuts, apples, pears. The work can be performed manually, by raking/sweeping or using special vacuum cleaners. It is also possible to track and use seed stocks stored by various rodents.

Hand-picking of oak acorns has been quite successful. Choosing the right time to collect is important because the empty and grubby ones are the first to fall off before the healthy ones. The harvesting interval depends on the climatic conditions, temperature and humidity, as the seeds germinate quickly in good weather (in particular in the case of sessile oak). It is also permissible to collect fruit with seed roots up to 1 cm long. If the fruit is heavily infested by insect larvae (mainly in the case of the *Balaninus* species), the required quality is achieved by immersing the fruit in water. It is best to sow the harvested acorns immediately in the tree nursery, as prolonged storage is problematic.

Hand-picking of beechnuts is a time-consuming process. The triangular shape of the beechnut makes them difficult to grip with the fingers, therefore collectors clear the ground under the seed trees in stands before they fall off. When the fruits fall off, they are swept up, large sieves are used to separate the beechnuts from other miscellaneous impurities and the clean, full seeds (fruits) are obtained by immersing them in water; the problem with immersing them in water is the sand that is left at the bottom together with the full seeds. Air-dry beechnuts, which still have around 30% moisture, are almost fully cleaned with a windmill. It is stored on the ground in a ventilated area in a layer no more than 10 cm high until shipping. Daily stirring is necessary, otherwise the beechnuts will get mouldy quickly. Immediate sowing is best.

Tree picking: The seeds on the tree can be collected by shaking the tree, climbing, shaking and cutting the branches, or the tree can be felled for the purpose of collecting.

There are various aids for climbing: ladders, foot rungs or crampon-like climbing irons, tree bicycle (Ger. Baumvelo), a safety harness, a climbing rope with a pulley system for ascending and descending the rope, appropriate clothing and personal protective equipment, and perhaps a crossbow or a manual swinging mechanism for attaching the rope to the crown. In Slovenia, ladders are usually used to climb tree trunks without branches, and climbers are secured with a safety harness during collecting. Picking from standing trees is one of the most hazardous jobs in forestry. Individual climbing equipment manufacturers also specialise in tree climbing equipment, which is also promoted in various competitions.

Maple and ash fruits (winged nuts – samaras) are collected by shaking the tree, climbing, knocking down and cutting branches with fruit or from felled trees. It is best to collect from standing trees. The pickers spread tarpaulins under the tree, and a person climbs up the tree and knocks them down from the branches with a stick. On the ground, they separate the leaves from the fruit by hand and further clean them using screens. These fruits are ready for sowing. For storage, they are dried and stored in plastic or jute bags.

The fruits of the wild cherry, which must be pitted the same day or they will ferment, destroying the germinability, are also picked by knocking the branches. They are then soaked twice in water to remove empty and grubby fruits. As the stone still retains a lot of moisture, the seed is dried in a shady and ventilated place for at least three weeks if it is to be stored, otherwise it is immediately transported to the tree nursery after soaking in water.

The seeds of conifers (spruce, fir, black pine, larch) are in the cones, which have to be plucked from the branches. Picking from standing trees is one of the most difficult jobs in forestry. Climbers use climbing irons, firefighters' safety harnesses and lightweight collapsible aluminium ladders to aid them in climbing. In the crown, the picker ties a safety harness around the trunk, plucks the cones with his hands and stores them in nets or throws them on the ground. His assistant picks them up in sacks and carries them to the truck road, where they are stored until they are transported to the drying plant. Afterwards there may be problems with removing the resin from the skin.

Water surface collection: Alder seeds can be collected from the water surface, especially in lowland floodplain forests, using nets and sieves.

The effect of seed collection: PAverage daily quantities of seed when collecting from standing conifer trees: larch 10-15 kg of cones, pine in selected seed stands 15-50 kg, fir 30-80 kg, spruce 40-100 kg, and from cut trees and individual trees with a low crown 40-60 kg for larch, black pine 80-100 kg, Scots pine 30-50 kg, fir 80-100 kg and spruce 100-150 kg of cones. For deciduous trees, the quantities are as follows: pedenculate and sessile oak 30–80 kg, beech 20–50 kg, sweet chestnut 20–40 kg, walnut 20–40 kg, ash 30–70 kg of samaras, sycamore 15–35 kg, black alder 30–80 kg of cones, grey alder 10–15 kg of cones, birch 25–50 kg, elm 4–10 kg, hornbeam 15–25 kg, linden 10–25 kg of ripe or 2–5 kg of green fruits and cherry 10–20 kg of stones.

Some difficulties occurring in seed collection: The biggest problem is obtaining and verifying information on fructification of each species. This leads to a lack of time or poor coordination in selecting stands for approval and collection, particularly in autumn when seed of several species is collected. The problems with collecting conifer seed are the dangers of the work and the relatively few strong fructification years. Seeds can be stored for longer periods, particularly in the case of larch, spruce and pines, but fir seeds can also be stored for several years; however occasional shortages may occur.

Much greater difficulties arise when collecting seed of deciduous trees. For species where the seed is collected from the ground, the main problems are the abundant understorey, the shrub layer and the stony character of the soil. Another issue is the past management practices of the seed stands or their selection, which has mainly been based on the appearance of the tree and the stand, which is often too dense and the crown can be poorly developed and produces few seeds. This is most noticeable in the case of seeds collected by knocking at the branches. In such a stand, it is virtually impossible to get to the seed.

Seed collection by cranes and mechanised shaking is not yet an option in Slovenia, due to the high investment costs and the small amount of seed that is collected. However, nets or vacuum cleaners to collect acorns and beechnuts from under the trees are being increasingly used.

LEAFLET: The protocol for the collection of seed material, plant parts and plants from natural regeneration is described in a leaflet published under the LIFE project LIFEGENMON (LIFE ENV/SI/000148).

Collecting seed when trees are felled requires a lot of coordination, from the owner's agreement to cut the appropriate number of trees, to the time of felling when the seed is ripe, to the presence of collectors at the site (taking into consideration occupational safety), and to the willingness of the team to collect the seed immediately after felling, as it can be completely picked overnight from various deciduous trees by wildlife.

Seed extraction: Seed extraction can be done from cones or fruits. It is necessary to facilitate immediate processing and storage, to reduce spoilage and for appropriate manipulation before and after sowing.

Extraction by drying: The cones or fruits can be air-dried or dried in a drying chamber. Individual conifer species can be dried in the sun and at relatively high temperatures, whereby special covered screens are used in which the cones open (with daily turning) and the seed falls through the grille in a relatively short period of 4-5 sunny days. After drying, the seed is cleaned, if necessary, to remove cones, wings and other miscellaneous impurities. Drying in a fan drying oven is possible at 45-55 °C for conifers, but seed from a large number of deciduous trees should not be dried at a temperature higher than 20 °C. The wind speed must also be regulated to ensure that light seed is not blown out of the drying oven. Larch cones that are full of resin need to be further rasped after drying, as otherwise about a third of the seed remains trapped.

Extraction by cracking the fruits: This is used for walnut, hornbeam and various alien species.

Extraction by removing the fleshy outer part of the fruits: For sowing, the small fleshy fruits can be dried and used whole. Very fleshy and dehiscent fruits should be pitted immediately to prevent fermentation and reduce germination, for example, the seeds of cherry, apple, rowan, pear, sorb tree, birch, hawthorn, yew, etc. These fruits can be crushed or carefully ground first, but only enough to avoid damaging the seed. They are then rinsed vigorously with water. Care must be taken to avoid anaerobic fermentations in the pulp or in the water with the soaked fruit. The water-cleaned fruit is also already properly prepared.

Cleaning and sorting the seed: This includes all the procedures for removing impurities and sizing the seeds. Specific procedures are used for removing the wings, sowing and sorting the seed. The seed is usually distributed by size using vibrating nets and sieves of different sizes. Water rinsing and immersion in water are commonly used to remove empty seeds.

5.3 Seed Drying

The seed is a living organism and any error in its manipulation can affect changes in vigour, germination and dormancy. Each individual extraction and storage process has to be studied under laboratory conditions and developed under quantitatively completely different conditions in practice. In addition to the species, the physiology of the seeds from different seed lots, different accessions, collected in different seed facilities in different years and with different quality of the crop, also differs. As regards the time and methods of collecting the seed, with particular attention being paid to ensuring that the parent stock is not damaged, cleaning, the various ways of pre-treating the seed and other finishing procedures, particular mention should be made of the drying process.

SEED
is a living organism that emerges from a flower after fertilisation and is used to spread through the area.

Drying is one of the most demanding processes in seed manipulation. For storage, it is advisable to dry the seed as much as possible, and the degree of drying should be determined on a species-by-species basis. The drier seeds can be stored at a lower temperature and for a longer period of time.

Drying must be started immediately after cleaning or removal of the carpel sheath, at low temperatures, preferably between 15 and 20 °C, with air flow, at a low relative humidity (ideally below 10% relative humidity (RH)) in the room, in thin layers, up to the specified percentage of moisture. For example, for long-term storage, e.g. for 10 years according to the literature, beech seed is optimally stored at 8-9% humidity and at -5 to -10 °C. A difference of 1% in humidity may result in a disproportionately large change in the germination of the seed after storage, or a 1% increase in humidity may require a storage temperature of about 1 °C higher.

The following formula quantifies the relationships between seed survival, storage time, temperature and moisture content:

$$v = K_i - p \;/\; 10\; K_E - C_W \log_{10} m - C_H t - C_Q t^2,$$

where v is the probable percentage survival of the seed after *p days* of storage at *m%* seed moisture and *t* °C, K_i is the seed lot-dependent constant, and K_E, C_W, C_H and C_Q are the seed type-dependent survival constants. This equation shows how the survival of orthodox seed species in air-dry storage depends on the environmental conditions in which they are stored.

In practice, the moisture content of the seed can be tested during drying or weighed to determine whether it has reached the desired dryness (DM%). The initial moisture content (M%) must be known and is determined by weighing, drying at 103 °C for 17 + 1 hours and re-weighing (according to ISTA 2000 protocols). The control of drying to the desired humidity is based on the following formula:

$$\text{seed weight (g) at the desired moisture content DM \%} = \frac{(100 - \text{initial } M\ \%)}{(100 - \text{DM }\%)} \times \text{initial seed weight } (g)$$

Example: 125.3 g of seed with a moisture content of 52.1% are to be dried to a moisture content of 15%.
Calculation: weight of seed at 15% humidity: ((100 – 52.1)%) / ((100 – 15)%) × 125.3 g = 70.61 g

For example, the drying of acorns is very problematic. They are dried by temporarily storing them in a dry, covered area with air exchange. The layer of acorns must be no thicker than 15 cm. If it is too thick or there is no air exchange, inflammation can occur. Because of high humidity, the acorn respires intensely when it is collected, releasing heat. If it is not drained quickly enough, the temperature in the acorn can rise up to 70°C, leading to inflammation and then loss of vitality. The acorns need to be stirred frequently to dissipate heat and dry more efficiently. Before permanent storage, it is dried to a moisture content of around 45%, but the actual moisture content depends on the seed lot; the moisture content of the acorns at the time of dropping from the trees varies clinally from west to east, where it is generally a few % lower than in the west, and storage is therefore also possible at a lower moisture content of approximately 40.5% (Žitnik 2003).

The process of rehydrating the seed at the end of storage is also important, as it can be mechanically damaged by imbibition (water absorption into the seed). Therefore, the conditions prescribe a process in 100% RH, but not in direct contact with water.

However, each process needs to be specifically developed in the laboratory and subsequently refined to be put into practice for the processing and storage of large quantities of seed.

5.4 Seed Storage

Seed storage is one of the most problematic areas in seed production. Seeds can be stored in the short term from harvesting to sowing, or in the long term for years or even decades. Short-term storage is mainly over one winter. For smaller quantities, autumn sowing or storage in the wild may also be appropriate, as the investment in equipment could be far greater than the potential savings. For larger seedlings, storage under controlled conditions is recommended, as this reduces losses, increases viability after storage and eliminates dormancy more successfully. However, long-term (multi-year) storage only makes sense under artificial conditions. It is used to provide seed in years when there is no fruiting, as most trees produce irregularly over a few years. Due to the negative impact of humans on nature and climate change, it is also becoming increasingly important to juxtapose and maintain seed banks for use in seed production and forest gene bank components, e.g. seed banks, in which long-term storage plays a key role.

In the past, spruce seed was the main product stored in Slovenia, and it can easily be stored for several decades. In recent decades, the increasing share of deciduous trees in regeneration through planting has led to a growing need to save their seeds. However, this is problematic. In the short term, a possible solution is to wait until the next sufficient crop to sow or plant, but in the long term, effective storage methods need to be developed that are also simple and inexpensive, as the quantity of seed collected and used in Slovenia means that we cannot afford complex and expensive methods.

Seeds are divided into two groups according to their ability to be stored for a long time. The first includes orthodox seeds or seeds capable of being dried, which have a significant reduction in moisture content during the final ripening phase. After harvesting, they can be further dried under controlled conditions to a moisture content of a few percent and then stored at temperatures well below 0 °C. Under these conditions, the development of pathogenic fungi and the biochemical activity in the seeds is almost completely stopped, so they can be stored for years or decades without any significant loss of vitality. To this group belongs spruce seed.

The second group consists of recalcitrant seeds or seeds that cannot be dried, which do not experience a significant decrease in moisture content during ripening. As a result, they have a high moisture content after falling, which can be more than 40%. Under artificial conditions, we cannot reduce their moisture content without significantly reducing their vitality. Therefore, they cannot be stored at temperatures well below 0 °C because the water in them would freeze and the water crystals would damage the tissue. These seeds are therefore stored at a temperature of around 0 °C. Under these conditions, the development of pathogenic fungi and biochemical activity is only slowed down, so seeds can usually only be stored for a few months, or until the following spring, without any significant loss of vitality.

Seeds are also divided into dormant or resting seeds and non-dormant or non-resting seeds. Non-resting (non- dormant) seeds germinate immediately under favourable conditions (heat, humidity), while resting (dormant) seeds germinate under favourable conditions only when the cause of dormancy (impermeability of the seed coat, immaturity of the embryo, presence of growth inhibitors, etc.) has been eliminated.

More than 50% of forest tree and shrub species have dormant seeds. In nature, dormancy is eliminated under the right conditions. The removal under controlled conditions is called **stratification,** and the removal of the mechanical dormancy of the testa is called **scarification.** Dormancy can be eliminated in different ways: by collecting immature seed, which is then immediately stratified (ash, hornbeam, small-leaved linden), and by stratifying mature seed before, during or after storage. In a simplified form, stratification is carried out by sowing the seed and, if conditions are favourable, nature will eliminate dormancy. Better efficiency is achieved by stratification under controlled conditions, which is more suitable for larger quantities of seed because of the investment in equipment. There is cold stratification, which takes place at lower temperatures, and warm stratification, which takes place at higher temperatures, both in a moist medium.

There are all possible combinations of these seed groups:

- capable of drying (orthodox) + non-resting (non-dormant),
- capable of drying (orthodox) + resting (dormant),
- non-drying (recalcitrant) + resting (dormant),
- non-drying (recalcitrant) + non-resting (non-dormant).

Storage complexity is increasing

This makes developing optimal seed storage methods a very complex and difficult task. At the Slovenian Forestry Institute, we have contributed, among other things, to the development of some methods of seed storage and dormancy removal: the importance of phytic acid in relation to available phosphorus in the soil, sugars and starch in long-term storage of acorns, storage of acorns at low temperatures down to -9°C (Žitnik 2003), and multi-year storage of spruce seed (several experiments).


Figure 4: Black poplar: stages of male flower development (drawing by Marina Gabor)

5.5 Work Procedures for Seedling Cultivation in Nurseries

Cultivation of forest tree seedlings is a multi-year process, with successive stages of work:

- soil preparation,
- sowing,
- transplanting of perennial seedlings and care thereof,
- excavating and preparing the seedlings for collection.

Soil preparation includes:

- sowing of green manure crops,
- autumn ploughing,
- fertilisation,
- soil harrowing or tillage,
- bed layout planning,
- row layout planning,
- pesticide spraying,
- watering.

Sowing includes:

- analysis of seed purity and germination,
- dewinging,
- soaking in water,
- disinfection and protection against rodents and birds,
- stratification,
- sowing, covering and rolling the seed,
- watering and spraying with fungicides,
- hoeing,
- dressing with fertiliser,
- preparation for winter or overwintering.

Transplanting of perennial seedlings and their care:

In the next year of the seedling cultivation the following is carried out:

- spring fertilisation with artificial fertilisers;
- weeding is less frequent as the seedlings are dense enough to cover the area;
- water seedlings only as needed during prolonged dry periods;
- regular protection of seedlings with fungicides against fungal diseases.

Seedlings are transplanted in spring, except for spruce, which takes place mainly in summer. When transplanting, we need to know what the target growth form of the seedlings is. The number of plants per unit area depends on the number of plants per unit area. 1 + 1, 1 + 2, 2 + 2, 2 + 3, 1/2 or 1 + 2 are the symbols that tell us the age of the seedling: one year and two years. This means that the seedling has grown as a seedling for one year and as a transplant for two years, giving a total age of three years. Conifers such as spruce, pine, Douglas fir, etc., are transplanted after two years because the seedlings are tall enough at that age. Fast-growing species, such as larch, should be transplanted after the first year. These species are grown as transplants for two years. During this time, the root ball is enriched and the seedlings are tall enough to be planted in the field. Deciduous trees are transplanted after the first year. For most species, e.g. mountain maple, ash, beech, pear, hazel, rowan etc., 1 + 2 is the best form of training, but for cherry and black alder 1 + 1 is more suitable.

Excavating the seedlings and preparing them for collection:

- mechanical or manual;
- different forms of growth and root development;
- grading according to size and quality, with poor ones being discarded;
- pruning seedlings, tying them into bunches;
- storage in a humid burial or cold store (bags, +2°C);

When digging up seedlings, we pay particular attention to:

- the time from digging to planting is as short as possible, which is particularly important during the dry spring months;
- the seedlings are buried as soon as possible after digging and sorting in the ground;
- the seedlings are protected from drying out during transport.

SFS, in cooperation with SFI, has prepared a **Protocol for the manipulation of forest tree seedlings from the nursery to the planting in the forest in cases where the seedlings are provided by the Slovenia Forest Service funded by the budget of the Republic of Slovenia** (Annex 1).

A good quality seedling is suitable for the particular site in terms of its origin and form, is healthy and vigorous, and may be colonised with suitable root symbionts in special cases. The appropriate size of the seedling may vary depending on the site; the combination of species in planting is a particular issue; the method of planting should be adapted to both soil and vegetation conditions. Greater attention will also need to be paid to the protection and cost-effectiveness of protecting seedlings from wildlife browsing.

5.6 Container Seedling Cultivation

Due to the ease of handling the seedlings during transport from the nursery to the forest and the possibility of planting during the growing season, a certain proportion of seedlings are grown in container production. Containers are restrictive in terms of space for root growth. There is less chance of excessive wetting or drying out of the root systems and of attack by pathogens, since production, at least until the first transplanting of the seedlings, is carried out under as controlled conditions as possible in glasshouses and greenhouses, which means that production costs are higher than for bare-root seedlings. Therefore, such production requires:

- building suitable facilities, usually greenhouses made of plastic or glass, with elaborate ventilation, cooling and sun protection systems;
- the construction of container stands, which should be at least 10 cm off the ground;
- the construction of an irrigation or misting system, preferably with automatic irrigation metering and dosing, and a water demineralisation system;
- planning and protecting seedlings against pests and for fertilisation;
- the development of suitable container substrates that are able to retain moisture, can be easily removed from the roots when transplanting seedlings, provide optimum nutrition and are not too favourable for the development of pathogens;
- the development of appropriately sized containers for the seedlings, which allow the roots to be 'air pruned' and prevent them from twisting, which contributes to the proper development of the root system; this is because root systems often twist in ordinary containers, which, when transplanted into the forest, leads to improper root development and reduced standing capacity of such trees, as well as to reduced stability of the future stand. Planting in the forest can also lead to a reduction in the number of trees that can be transplanted into the forest.

For the reasons mentioned above, it is relatively expensive to raise seedlings in containers, so nursery workers try to maximise the germination of the seed in containers. In most cases, two or three pre-prepared seeds are sown in each “cell”, which should have as high a germination rate as possible, close to 100%, be as uniform in size as possible and have a timed germination. Therefore, for the purposes of container breeding, size grading of seed, various pre-treatment procedures and the removal of seed of poorer quality from the majority seed are used, all of which lead to a reduction in the genetic diversity of the forest reproductive material thus cultivated.

Containerised seedling production requires seed size grading, certain pre-preparation procedures and the extraction of seed of a different quality from the majority, all of which lead to a reduction in the genetic diversity of the forest reproductive material thus produced.

5.7 Mycorrhizal Seedling Cultivation

Mycorrhiza is the symbiosis between the fungus and the plant root and acts as the organ for water and nutrient uptake (Frank 1885, e.g. in Kraigher 1996). In ectomycorrhiza, in which the fine roots of tree species (e.g. spruce, fir, larch, pine (ectendomycorrhiza can also occur in pine and larch), beech, oak, birch, and to a lesser extent poplar, willow and alder, where other forms of root symbioses have also developed) are completely surrounded by the fungus, the fungus takes over the function of taking up the plant's intake of water and nutrients. Seedlings with roots colonised by ectomycorrhizal fungi appropriate to the tree species, provenance and site are more successful when transplanted into the forest; mycorrhizae replace the initial fertilisation and accelerate growth in the first year after transplanting. It is therefore useful, from the point of view of the success of planting in the forest, to identify which combinations of fungal species and strains and forest tree species and populations are most compatible and suitable for particular sites, and to undertake the isolation and rearing of suitable fungi, the multiplication of their mycelia, and the colonisation of the roots of seedlings of forest tree species.

The fungi involved in ectomycorrhizal symbiosis belong mainly to the ascomycetes and basidiomycetes, and some can be isolated and multiplied as mycelia or their spores isolated for colonisation of forest reproductive material. Some species of these fungi are also of commercial interest as edible fungi, e.g. some species of truffles, boletes, chanterelles and other edible and culinarily interesting mycorrhizal fungi.

Colonisation – Mycorrhization of seedlings can take place in several ways:

- mycorrhizal fungal spores can be used to "coat" forest tree seeds that are included in special nursery sowing films; this process is used, for example, in forest nurseries in the Czech Republic and Slovakia;
- spores of mycorrhizal fungal species can be incorporated into the muddy slurry in which the roots of forest tree seedlings are soaked;
- spores or mycelium of mycorrhizal fungi species can be added to the soil substrate in the nursery or in containers;
- It is also possible to use a soil inoculum, i.e. a naturally occurring mixture of mycelium and fungal spores present in natural soil substrates that have not been treated with fungicides or heat-treated.

In Slovenian tree nurseries, most bare-root seedlings are naturally mycorrhized. For example, the Omorika Muta tree nursery naturally contains several species of mycorrhizal fungi, among which the predominant species is *Thelephora terrestris* Ehrh., which is not particularly useful for forest tree seedlings, but does not harm them either. It would therefore also be appropriate here to take a planned approach to raising seedlings colonised with selected species and strains of mycorrhizal fungi.

5.8 Effects of Nursery Practice on Genetic Diversity – from Stand to Seedling, Genetic Diversity Decreases

Within the framework of the EUFORGEN programme (European Forest Genetic Resources Programme), the Working Group on Forest Reproductive Material has prepared a Report of the impact of seed and nursery activities on the genetic diversity of FRM. The guiding principle is that the initial genetic diversity captured in the seed lot at the time of its collection in the stand can only be reduced through the processes of processing, storage, seedling cultivation and planting back into the forest (Diagram 2).

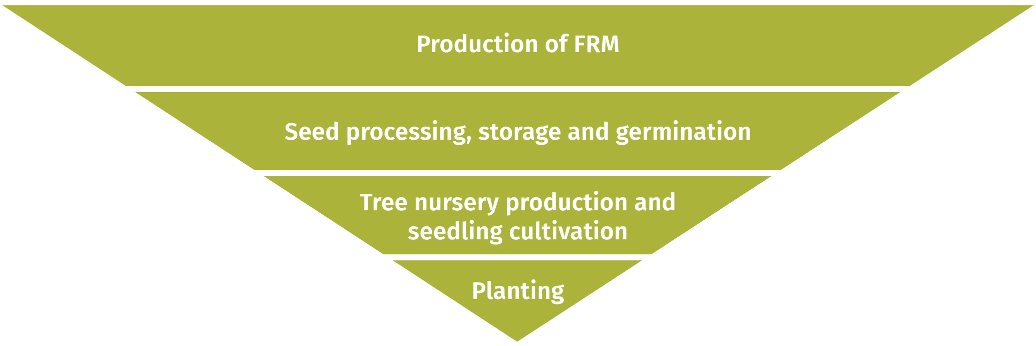

Diagram 2: The genetic diversity of FRM decreases from stand to planting back into the forest.

Forest seed and tree nursery practices that reduce genetic diversity, an excerpt from the EUFORGEN Working Group Report (Gömöry et al., 2021):

- Seed Production:

◊ collection (selection of non-related trees), timing (physiologically mature - immature seed) and method of collection (from the whole crown, accessible from the ground, side of the crown, etc.).

- Seed processing and germination processes:
  - extraction, thermotherapy/chemotherapy/encapsulation, drying, size selection;
  - storage, stratification/scarification, germination procedures (in germinators, in the bed);
  - germination rate (elimination of later-germinating genotypes).
- Exclusion/selection/standardisation of seed and/or seedlings:
  - seed unsuitable for seed producers (species purity in the EC Directive);
  - for tree nursery operators (standardisation required by the EC Directive only for Mediterranean oak species and poplar cuttings).

- Transplanting and pruning to optimise the ratio between the shoot and root system:
  - ◊ uniformity of the root system – seedlings of species with deep primary (oblique) roots are more damaged and affected.
- – Fertilisation and protection against diseases and pests:
  - ◊ hereditary resistance of individual provenances and clones to diseases and pests;
  - ◊ experiments with different N fertilisation regimes – high and optimal N concentrations with differences between families, low concentrations with no differences (several pine species);
  - ◊ differences in growth completion and frost resistance – linked to fertilisation regimes.
- Mycorrhization:
  - ◊ the use of mycorrhizal beech and oak seedlings in Germany – as a replacement fertiliser (Kottke et al. 1987);
  - ◊ dependent on the fertilisation regime, the use of pesticides and the naturally occurring species and strains of fungi in the nursery;
  - ◊ coexistence efficiency, depending on the fungal species and strain and the species and provenance of the forest tree (Gianinazzi-Pearson et al. 1984, Hazard et al. 2017),
  - ◊ use in forest and non-forest plantations (forest by-products; Grebenc et al., forthcoming).
- Storage of seedlings:
  - ◊ the period, duration and storage regime;
  - ◊ In Turkish semi-desert conditions, the presence of heart roots and induced drought stress prior to harvesting increase post-planting survival – impact on increased genetic diversity after successful planting,
  - ◊ growth completion and frost tolerance are hereditary – inappropriate timing of excavation can reduce genetic diversity.
- Vegetative reproduction:
  - ◊ cuttings, somatic embryogenesis (SE), micropropagation.
- Containerised seedlings:
  - ◊ extreme standardisation of seeds and seedlings;
  - ◊ the possibility of planting in different seasons and conditions;
  - ◊ the cost of production – the speed and price of planting;
  - ◊ experience 2017/2018 at Postojna OU: mice.
- Manipulation and monitoring of stand quality after planting (traceability):
  - ◊ a protocol for manipulating seedlings from nursery to forest;
  - ◊ planting methods and timing (conifer-conifer, container seedlings, etc.);
  - ◊ protection of seedlings (game, rodents, use of chemical products, etc.);
  - ◊ planting planning,
  - ◊ monitoring the success of planting;
  - ◊ monitoring the effects of management on the quality of future stands.

Figure 5: Black poplar: stages of female flower development (drawing by Marina Gabor)

6

FOREST REPRODUCTIVE MATERIAL ACT

A PREREQUISITE FOR FOREST STABILITY
is regeneration with site-adapted forest reproductive material (for natural rejuvenation or reforestation by planting and sowing).

Regeneration is one of the most crucial phases in forest development. It is where the hereditary patterns of the future forest are formed, required for the stability of future stands. The prerequisite for forest stability is regeneration with site-adapted forest reproductive material (for natural rejuvenation or reforestation by planting and sowing). The need for forest reproductive material to be adapted to the site is reflected in the demarcation of regions of provenance and the rules on the use of forest reproductive material in them, which are being adapted to the necessary measures for the genetic conservation of forests in a time of rapid climate change.

EUROPEAN DIRECTIVE ON THE MARKETING OF FOREST REPRODUCTIVE MATERIAL
is a common instruction that prescribes professional supervision and facilitates free trade within the European Union. The basic requirements are aligned with the OECD Forest Seed and Plant Scheme.

The European Directive on the marketing of forest reproductive material within the European Union, adopted in December 1999 (EC/105/1999), is a common guideline that prescribes the necessary professional supervision and allows a free market. Each Member State shall, on the basis of the minimum criteria set out in this Directive, determine in its national legislation and regulations the responsible operators and procedures for professional supervision, the demarcation of regions of provenance and the suitability of the use of forest reproductive material therein, and the means of maintaining stability and biodiversity in its forests. These procedures may include rules on seed reserves, seed storage, seed banks and the wider forest gene bank in each country. In this chapter we summarise the essential articles, terminology and procedures of the Forest Reproductive Material Act, the requirements for registration (approval) of basic material (forest seed facilities) and the procedures for certification of FRM.

FOREST REPRODUCTIVE MATERIAL ACT (ZGRM 2002)
includes:

- the requirement to conserve forest genetic resources (FGR) in forests and plantations outside forests;
- the characteristics of traditional and sustainable multifunctional forest management as prescribed by the Forest Act (ZOG 1993).

6.1 Interpretation of Terminology According to the Forest Reproductive Material Act (2002)

Terminology is mostly based on the European Directive on the marketing of forest reproductive material (EC/105/1999).

Reproductive material includes:

1) Seed material: seeds, cones, fruits and co-products intended for the production of planting material.
2) Plant parts: cuttings, cutting material, explants or embryos for micropropagation, buds, layers, roots, substrates, grafts and any parts of plants intended for the production of planting material.
3) Planting material: plants grown from seed (seedlings), plant parts and plants from natural regeneration (plants from natural rejuvenation).

Forest seed facilities is the basic material for the production of reproductive material, which may be a group of seed trees, a stand, a seed orchard, parents of family, a clone or a mixture of clones:

- A group of seed trees are trees within a given area from which seed is collected.
- A stand is a spatially restricted population of trees with a uniform composition.
- A seed orchard is a plantation of selected clones or families which is isolated or managed in such a way as to prevent or reduce pollination from external genetic sources and in which the management method allows frequent and abundant cropping and easy seed production.
- The parents of family are trees intended to produce offspring by controlled or free pollination, with one known parent as the mother tree and the pollen of another parent (sibling family) or group of parents (half-sibling family).
- Clones are a group of individuals (ramets) derived originally from a single individual (ortet) by vegetative propagation, for example by cuttings, micropropagation, grafts, layers or divisions;
- A clonal mixture is a mixture of identified clones in defined proportions.

Reproductive material categories:

1) "Source identified" Reproductive material of known origin derived from basic material which may be either a seed source or stand located within a single region of provenance and which meets the prescribed minimum requirements.
2) "Selected" Reproductive material derived from basic material which shall be a stand located within a single region of provenance, which has been phenotypically selected at the population level and which meets the defined requirements.
3) "Qualified" Reproductive material derived from basic material which shall be seed orchards, parents of families, clones or clonal mixtures, the components of which have been phenotypically selected at the individual level, and which meets the requirements prepared based on this Act. Testing need not necessarily have been undertaken or completed.
4) "Tested" Reproductive material derived from basic material which shall consist of stands, seed orchards, parents of families, clones or clonal mixtures. The superiority of the reproductive material must have been demonstrated by comparative testing or an estimate of the superiority of the reproductive material calculated from the genetic evaluation of the components of the basic material.

Table 3: The categories in which each type of reproductive material can be marketed, depending on the basic material from which it was produced.

Type of Basic Material	CATEGORY OF FOREST REPRODUCTIVE MATERIAL			
	Source Identified	Selected	Qualified	Tested
Seed Source	X			
Stand	X	X		X
Seed Orchard			X	X
Parents of Family(-Ies)			X	X
Clone			X	X
Clonal Mixture			X	X

Reproductive material that is marketed must be accompanied by a **document.** If this is coloured, then yellow indicates source identified, green indicates selected, pink indicates qualified and blue indicates tested FRM.

Autochthonous stand or seed source: An autochthonous stand or seed source is one which normally has been continuously regenerated by natural regeneration. The stand or seed source may be regenerated artificially from reproductive material collected in the same stand or seed source or autochthonous stands or seed sources within close proximity.

Indigenous stand or seed source: An indigenous stand or seed source is an autochthonous stand or seed source or is a stand or seed source raised artificially from seed, the origin of which is situated in the same region of provenance.

Origin: For an autochthonous stand or seed source, the origin is the place in which the trees are growing. For a nonautochthonous stand or seed source, the origin is the place from which the seed or plants were originally introduced. The origin of a stand or seed source may be unknown.

A **provenance** is a locally defined site of any stand of forest trees.

For a species or sub-species, the **region of provenance** is the area or group of areas subject to sufficiently uniform ecological conditions in which stands or seed sources showing similar phenotypic or genetic characters are found, taking into account altitudinal boundaries where appropriate.

Production means any stage in the extraction and finishing of seed material into seed and the raising of planting material from seed or plant parts (such as: collecting, harvesting and other extraction of seed material, plant parts, direct-use shoots and buds from the starting material, drying, dehulling, cleaning, calibrating, storing, packing, labelling, preparing for planting, determining quality elements, etc.).

A **lot** is a well-defined and physically limited quantity of reproductive material produced in a given year and in a given forest seed facilities (basic material).

Marketing means selling or delivering to other persons, including delivery under a contract for services, displaying with a view to sale, offering for sale.

A **supplier (professional operator)** is any legal or natural person carrying out a production activity with a view to marketing or importing reproductive material.

The production of seed and “pullings” (naturally regenerated saplings collected in approved forest seed facilities for further nursing in nurseries for planting in forests) in forests is a **function of the production of other forest goods** under the forest regulations.

The **seed storage** is a store of seed material intended for use in planting and sowing in the event of a shortage of reproductive material in forests and constitutes a compulsory reserve of reproductive material for the needs of sustainable forest management in the Republic of Slovenia.

The **Slovenian Forest Genebank** is a controlled or cultivated population of forest woody plants managed for the conservation of species and their gene pools. It consists of forest seed facilities, including forest gene reserves and genetic monitoring stands, special specimens or populations of forest trees, living archives of forest tree species, test plantations, a seed bank and other biological materials.

A **seed bank** is a long-term collection of representative samples of seed material from a seed storage and other sources.

Export is any exit of a consignment of reproductive material from the customs territory of the European Union (valid for the territory of the Republic of Slovenia) and includes re-export and temporary export.

Import is any introduction of a consignment of reproductive material into the customs territory of the European Union (valid for the territory of the Republic of Slovenia), irrespective of the purpose of the introduction, with the exception of introduction for transit.

Transit is any movement of a consignment of reproductive material through the customs territory of the European Union (valid for the territory of the Republic of Slovenia).

The **master certificate of origin** is the document certifying the origin of the reproductive material.

The **accompanying document** is the document that accompanies the reproductive material when it is marketed.

A **plant passport** is a document that accompanies plants or parts of plants whenever they move within the EU, in accordance with the provisions of plant protection legislation.

A **comparative test** is a statistically designed experiment in which reproductive material is evaluated against one or more predetermined standards.

A **standard** is a stand, plantation or plant that shows better characteristics than the average in a given area and is established as a benchmark before the comparative test is started.

A **PROVENANCE**
is a locally defined site of any stand of forest trees.
A **REGION OF PROVENANCE**
for a species or subspecies is an area or group of areas with similar ecological conditions in which stands or groups of seedlings have similar phenotypic or genetic characteristics, taking into account elevational zones.

6.2 Minimum Requirements for the Production of "Source Identified" Forest Reproductive Material

The basic material must be a group of seed trees or a stand growing in one region of provenance, approved according to a prescribed procedure, during which the proposed stand is evaluated against criteria 1 to 7 of Directive EC/105/1999 and the ZGRM (see Section 6.4 for explanations).

The minimum number of trees from which FRM is harvested is 25 for most tree species, and optimally at least 50 trees growing at least one and preferably two crown heights apart. For minority species, the minimum number of trees is 10, and preferably more than 25, spaced at an appropriate distance from each other to ensure that they are not natural ramets of the same clone.

Phenotypic criteria are taken into account in the selection:

- for conifers, general adaptation to site conditions, straight growth and appropriate crown shape;
- for deciduous trees, in addition, twisted growth must be minimised; forking, if present, is only allowed in the crown.

> Requirements for seed facilities for forest reproductive material production in the category "source identified" in Slovenia include stricter criteria than those laid down in European Directive EC/105/1999.

The following information must be known and provided:

- region of provenance;
- a location plotted on a map;
- altitude/elevational range;
- the bedrock and possibly the plant community;
- the regional unit, possibly the local unit, the forest management unit, the cadastral municipality, the forest department and its section;
- origin, which may be indigenous, non-indigenous, natural or unknown.

In the case of non-native or non-natural origin, the origin of the basic material, if known, must be given.

6.3 Minimum Requirements for "Selected" Seed Stands

The stand is assessed according to the specific purpose for which the reproductive material will be used. In this respect, the requirements of points 1 to 10 shall be reasonably taken into account. The purpose of the use shall be indicated in the Register of Forest Seed Facilities of the Republic of Slovenia.

1) **Origin:** It must be established, on the basis of historical sources or by other appropriate means, whether the selected stand is of indigenous/native, allochthonous/non-indigenous or unknown origin. For allochthonous/non-native, the origin must be given, if known.
2) **Isolation:** Stands shall be sufficiently distant from stands of inferior quality of the same tree species or variety which may form hybrids with the tree species in the selected stand. This is particularly important when the surrounding vegetation is of non-native or unknown origin.
3) **Population size:** The stands must be composed of one or more groups of trees with an appropriate distribution and in sufficient numbers to allow adequate cross-pollination. The number and density of trees in a stand should be sufficient to eliminate the undesirable effects of inbreeding.
4) **Age and development phase:** The stands must be of such an age and stage of development that the selection criteria can be assessed unambiguously.
5) **Uniformity:** Trees in stands should show a normal degree of variability in morphological characters. If necessary, poorer quality trees shall be removed.
6) **Adaptability:** The stands must show adequate adaptation to the ecological conditions in their area of origin. This is expressed by the ability of the population to thrive permanently on its site, i.e. to rejuvenate successfully, either generatively or vegetatively; generative rejuvenation means that flowering, fruiting (respecting periodicity), natural rejuvenation (seed germination) and survival of the young saplings are observed in the stand. For non-native species, the potential invasiveness of the species or the forest tree population being assessed is recorded under this criterion.

7) **Health condition and immunity:** Trees in stands must generally be free from pests and diseases and must be resistant to adverse climatic and soil conditions (pollution is excluded) on their site. On particularly unfavourable sites, specific characteristics can also be identified regarding the resilience of the forest tree population to such conditions, e.g. lead contamination of the soil, etc.
8) **Volumetric increment:** In general, the volumetric increment of a stand should be greater than the average of the other stands growing in similar ecological conditions and that are managed in a similar way.
9) **Wood quality:** The quality of the wood must be taken into account when selecting stands; under certain conditions, it can also become a decisive criterion.
10) **Growth form:** Trees in stands should show good morphological signs of growth, in particular trunk elongation and symmetry, adequate canopy growth, thinness of branches and their adequate natural die-back. In addition, the proportion of forking trees and trees with spirally twisted timber should be low.

6.4 Forest Seed Facilities for the Production of the "Qualified" and "Tested" Categories of FRM

FRM of the category "qualified" is produced from seed or clonal orchards. For the purposes of designing such a plantation, a register of plus trees should be prepared for each tree species, corresponding to the phenotypic characteristics determined at the tree level. In Slovenia, the criteria for the approval of plus trees for wild cherry and black poplar have been elaborated, but the register is still relatively deficient, as the formal requirements for approval of each tree are the same as for the approval of the whole stand. In addition, agreements must be reached for the acquisition of grafts or cuttings from the individual trees (ortets) for the production of planting material for the design of a seed or clonal plantation (number of clones – ramets per each plus tree – ortet).

For the design of seed and clonal orchards, it is important to determine the distance from stands of the same species that could affect the mixing of genetic material, the organisation of the work and the planting design, i.e. the minimum number of clones and replications of the same clone in the plantation, the spatial distribution, the tending and thinning system, and the flowering and pollination control system. The plan with all the necessary components is then approved by the Institute and an expert opinion is issued as the basis for the plantation design. Once planted and before FRM production begins, this seed facility must be approved, verifying the adequacy of the planting, the number of clones and the replication of clones in the plantation. At the same time, the maximum number of seeds or planting stock allowed from each orchards shall also be fixed.

FRM of the category "tested" may originate from *in situ* (selected) or *ex situ* (qualified) seed facilities that **meet predefined standards** or have **been tested in progeny tests.** This category of forest seed facility has not yet been registered in Slovenia.

6.5 Production of Forest Reproductive Material

The sequence of operations necessary to initiate and control the production of forest reproductive material is shown below.

1) **Recording** forests with a production function for forest reproductive material is carried out by the Slovenian Forest Service (hereinafter referred to as the Service).
2) **Only a forest seed facility approved by a decree** may be used for production of forest reproductive material intended for marketing. The procedure for the approval of a forest seed facility is conducted by the Slovenian Forestry Institute (hereinafter referred to as the Institute) under an administrative procedure based on an inspection by a committee formed by the authorized experts from the Institute and the responsible personnel from the Service.
3) **The procedure** for the approval of a forest seed facility **shall be initiated on the basis of an application from the owner** or other formally acknowledged forest user forests owned by the Republic of Slovenia is submitted by the Slovenian State Forests company (SiDG). The forest owner or user may authorise the Service to represent him in

the procedure for approval of a forest seed facility in his forest.

4) The Institute decides by a written **Decree** on the approval of the forest seed facility. The decree specifies the type of approved forest seed facility and the category of reproductive material to be produced therein, and shall prescribe the conditions for the production of seed material and plants from natural regeneration in the forest seed facility.

For approved forest seed facilities to be used for the production of reproductive material in the categories "selected", "quantified" and "tested", the institute shall draw up tending guidelines which are a mandatory content of forest management and detailed silvicultural plans.

5) For approved forest seed facilities to be used for the production of reproductive material in the categories "selected", "quantified" and "tested", the institute shall, upon approval, write guidelines for tending which are a mandatory content of forest management and silvicultural plans. The Service plans the management of forest seed facilities and advises their owners on the function of production of seed and pullings. An approved forest seed facility intended for the production of selected reproductive material shall be inspected by the Service at least once a year; seed facilities for the production of FRM in the 'qualified' and 'tested' categories shall be inspected by the Institute at the time of flowering (seed orchard) or before the start of production.

6) For the purpose of approving forest seed facilities intended for the production of reproductive material in the categories 'source identified' and 'selected', the regions of provenance shall be established. The Regulation on the Delineation of Regions of Provenance, of which the Provenance Regions Map is a component, also contains more detailed recommendations for the use of reproductive material in the individual provenance regions and at elevational zones.

7) For forest seed facilities, in addition to the data required under the forest regulations, the Service also keeps records on the status of these stands and groups of trees, and on flowering and fructification, and reports on these regularly to the Institute. The Institute keeps records of the quantity, type and quality of forest reproductive material produced.

8) The Institute shall establish and manage a register of forest seed facilities. Entry in the register shall be *ex officio* on the basis of the approval decrees issued.

The Institute shall establish and manage a register of forest seed facilities (basic material).

Figure 6: Black poplar: female flowers and female catkins with seed capsules (long white silky hairs giving a fluffy and cotton-like appearance) (drawing by Marina Gabor)

6.6 Master Certificate for Forest Reproductive Material

If the reproductive material is produced in a forest seed facility under the provisions of the ZGRM, a certificate of origin is issued.

Suppliers must notify the Service (for *in situ* seed facilities) **or the Institute** (for *ex situ* seed facilities) in good time, but at the latest one week before production is due to start at the forest seed facility, in order to secure the issue of a master certificate of provenance. They submit an **application for FRM production** to the responsible SFS local unit, which also includes the **consent of the owner** of the seed facility for production of the FRM.

During production, **the Service monitors the progress** of the production in the forest seed facility and issues a **confirmation of FRM production.** Before the end of production at the forest seed facility, the prescribed quantity of plant material (alive twig with three buds, seeds, fruits or cones) from each tree from which FRM has been obtained shall be sent to the Institute (by the Service or the professional Operator), as well as the confirmation with the quantities of collected seeds or pullings (e.g. cones or fruit obtained and a record on the extracted seeds) and a representative sample of the extracted seed, together with a copy of the confirmation to the institute for analysis and inclusion in the active part of the Slovenian Forest Genebank, which is used for FRM certification, for any possible future inspection and for inclusion in the forest genebank (extracted DNA for the DNA library or extracted seed for the seed bank).

On the basis of the confirmation of the Slovenia Forest Service and the report on the extraction of seed by the professional operator, **the Institute** shall issue a **master certificate of origin** to the seed dealer – professional operator no later than one week after the end of production in the forest seed production facility or one week after receipt of the record of the seed extraction from the FRM producer, provided that the conditions laid down in the guidelines written at the time of approval of the facility have been met and that the origin clearly corresponds to that recorded in the SFS confirmation.

In order to ensure uniform records, the owner shall notify the time and quantity of the production of reproductive material for his own needs to the Service no later than one week before the start of production.

MASTER CERTIFICATE FOR FOREST REPRODUCTIVE MATERIAL
If the reproductive material is produced in a forest seed facility under the provisions of the Forest Reproductive Material Act, a certificate of origin is issued.

6.7 Forest Reproductive Material Lot

At all stages of production, the reproductive material must be separated by lots, each identified by the following information:

1) certificate of origin number and country code;
2) botanical name of a species or hybrid of species;
3) category;
4) purpose;
5) type of forest seed facility;
6) the registration number of the forest seed facility in the register of forest seed facilities;
7) the provenance region – for source identified and selected forest reproductive material, and for the other categories where appropriate;
8) the origin of the reproductive material (indigenous, natural, known (indication of origin), unknown);
9) the year of seed production;
10) the age and type of planting material (seedlings, plants for natural regeneration (pullings), cuttings);
11) genetic modification.

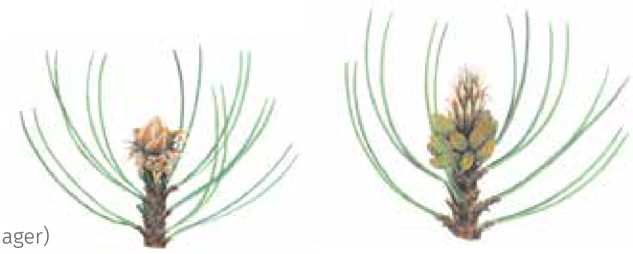

Figure 7: Black pine: stages of needle development (drawing by Klara Jager)

6.8 Reserves of Forest Reproductive Material

In order to prevent a shortage of a certain species and category of seeds and other reproductive material that would endanger the stability of forests, the environment, genetic resources and biodiversity in the territory of the Republic of Slovenia, SEED STORAGE shall be organised within the framework of the public forestry service.

In order to prevent a shortage of a certain species and category of seeds and other reproductive material that would endanger the stability of forests, the environment, genetic resources and biodiversity in the territory of the Republic of Slovenia, Seed Storage shall be organised within the framework of the public forestry service.

Samples of seed material stored in the Seed Storage are also obligatorily included in the seed bank, which is an integral part of the Slovenian Forest Genebank, managed by the Slovenian Forestry Institute.

The establishment and use of stocks of reproductive material are carried out by the Service, but individual tasks may also be carried out by concessionaires.

The Institute provides guidance for the development of the Seed Storage and the Slovenian Forest Genebank and carries out expert tasks regarding quality control and provenance of the seed material.

The minimum quantity and type of reproductive material to be stored in the seed bank shall be planned by the Service in the forest investment programme and is determined by the Minister by regulation.

The Service stores the compulsory reserve of reproductive material not covered by the concession as an increased commercial stock with major suppliers on the basis of contracts on the purchase, storage and ongoing renewal of reproductive material. If it is not possible to store reproductive material in this way, the Service stores it itself in its own or rented warehouses.

The supplier or the person entrusted with the reproductive material from the Seed Storage under a concession or storage contract may not pass on or otherwise dispose of the material without the authorisation of the Service.

The use of the reproductive material reserves is decided by the Service through the Forest Investment Programme. It specifies the type of reproductive material, the purpose and manner of use, the time limit and manner of replacement of the used reserves, if any, and, at the time of sale, the price of the reproductive material and the portion of it to be used to finance the reserves.

According to the Service's programme, planting material grown from the seed reserves is intended for use in forest planting in the territory of the Republic of Slovenia.

Funds for the creation and maintenance of the Seed Storage and for the purchase and storage of the compulsory reserve of reproductive material are provided in the budget of the Republic of Slovenia according to the forest investment programme drawn up by the Service. EU funding from the Rural Development Programme can also be used to help with the rehabilitation of large-scale disturbances.

Samples of seed material stored in the Seed Storage are also obligatorily included in the **SEED BANK,** which is an integral part of the **SLOVENIAN FOREST GENEBANK,** managed by the Slovenian Forestry Institute.

Figure 8: Black pine: stages of needle development (drawing by Klara Jager)

6.9 Comparison of the Competences of National Legislation and EU requirements

The EC Directive on the marketing of forest reproductive material (EC/105/1999) establishes a basic level of professional control and exchange of information on the production and marketing of forest reproductive material, which each EU Member State can build upon with its own national legislation.

The European Commission's Directive requests:

- control of forest reproductive material of forest tree species listed in the Directive and used for forestry purposes,
- marketing only the prescribed categories of forest re-productive material,
- traceability of forest reproductive material at all stages of production and marketing,
- a clear map delineation of the provenance regions and their description,
- the possibility to freely transfer and market forest reproductive material throughout the EU.

It depends on the EU country:

- how it will design the system of supervision and authorise organisations for professional supervision;
- which additional forest tree species it will include in its national lists;
- whether it will set stricter criteria for the granting of a decision for approval of forest seed facilities intended for the production of reproductive material in the category "source identified";
- how it will design the delimitation system and demarcate regions of provenance;
- which provenances of forest trees it will designate for use in forests/provenance regions in its territory;
- whether it will prescribe ways to maintain the stability and biodiversity of forests in its territory by making it compulsory to purchase the planned quantities of forest reproductive material from seed facilities in its territory in order to set up a seed storage;
- whether it will also include in its legislation in this area additional content that has nothing to do with the Directive, e.g. content relevant to the system for the conservation of forest genetic resources, the content and functioning of the Slovenian Forest Genebank, forest gene reserves, etc

The demarcation between the European and national legislation is shown schematically in Figure 5. However, Slovenian legislation is also based on the requirement to conserve forest genetic resources (under Article 2 of the Forest Reproductive Material Act), from which the by-laws also derive.

Diagram 3: Delimitation of the European and national content of legislation on the marketing of forest reproductive material (FRM) and the conservation of forest genetic resources

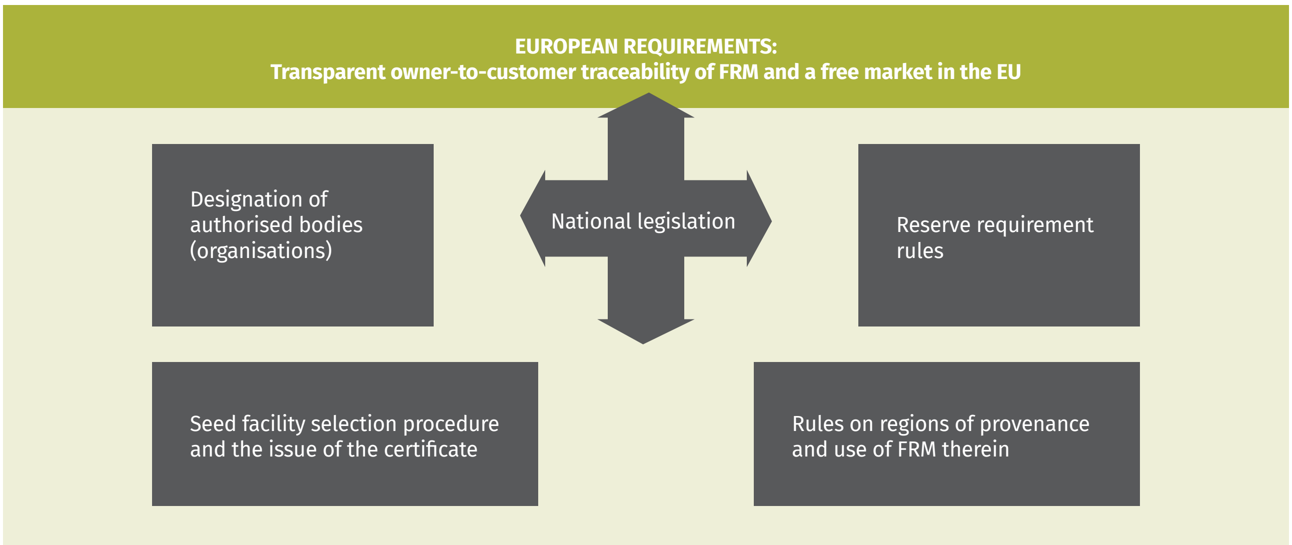

Diagram 4: The Slovenian Forest Reproductive Material Act is also based on the requirement to conserve forest genetic resources, which is set out in detail in sub-legislative acts.

6.10 Meaning, Competences and Sequence of the Rules

Reforestation is one of the most crucial phases in the lifetime of a forest and one of the most important activities affecting its development. The basic condition for stability is regeneration with site-adapted forest reproductive material (for natural rejuvenation or for regeneration by planting and sowing). The timing of regeneration depends on silvicultural planning or a series of silvicultural measures. This is followed by quality regeneration by planting/sowing, which primarily depends on the physiology and technology of storage, cultivation and planting of forest reproductive material. The complex of professional guidance is directly linked to and dependent on professional supervision of all phases of the work, i.e. supervision of the origin and quality of forest reproductive material and, in the case of forest nurseries, health checks. The expert guidance and supervision process includes:

1) reforestation with site-adapted forest reproductive material, i.e. a regulation delimiting provenance regions and recommendations for the use of FRM in them,
2) timing, which for seed and seedlings means in particular the timing of the final cutting and reforestation of stands, and the planning of the appropriate quantity and quality of planting material, including measures in the seed facilities, which are part of the forest genebank, and the management of the seed storage,
3) transparent professional control of forest reproductive material based on nationally proven procedures and institutions, including inspection and international cooperation on the transfer of seed and seedlings across national borders.

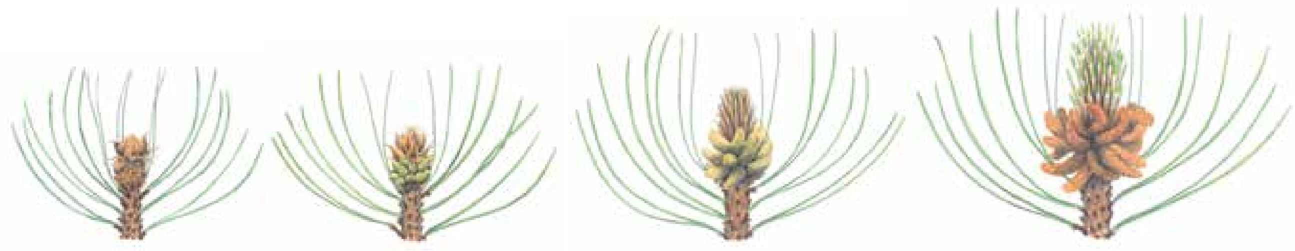

Figure 9: Black pine: stages of male flower development (drawing by Klara Jager)

7

REGULATORY PROVISIONS CONCERNING FOREST REPRODUCTIVE MATERIAL

Tree populations adapt to local environmental conditions through natural selection. Selection influences the adaptation of populations to local conditions at the genetic level, and the outward expression of differently expressed genetic composition is reflected in the phenotype. Therefore, the genetic variability between individual populations of trees must be taken into consideration when planting or sowing. To maintain the natural stability of stands, the use of reproductive material with a genetic makeup similar to that of autochthonous stands is the most appropriate for planting or sowing. This also prevents the introduction of alien genetic material which, through cross-pollination, could affect indigenous genetic material and weaken the natural stability of stands. To meet this requirement, we define regions of provenance that are geographically distinct and within which populations of a given tree species have a similar genetic makeup. It is therefore recommended that forest reproductive material collected in a particular region of provenance be used only in that area.

7.1 Demarcation of Regions of Provenance

The best methods for delimitation of regions of provenances are provenance trials and DNA analyses, as these methods can determine differences in genetic makeup between individual populations of trees. However, methods of provenance trials are time-consuming and logistically challenging, and DNA analyses are not yet available for all species and populations within the territory of the Republic of Slovenia. The current and provisional solution is to select regions of provenances based on natural factors (ground rock, soil, topography, vegetation, local climate, etc.) that are assumed to impact the genetic diversity of individual tree populations. Regions of provenances can be subsequently updated and verified based on the results of provenance trials and DNA analyses. The boundaries of the regions of provenance are also generally aligned with the administrative boundaries, which are preferably marked in the field, to facilitate the identification of the regions of provenance in the field, to meet the practical needs of the growers in the individual regional SFS units in their planning, and to facilitate the supervision of planting or sowing. The whole of Slovenia is divided into altitudinal zones, which, even more than the regions of provenance, define the suitability for utilisation of FRM.

7.1.1 Seed Districts (1951-1986)

The first division of Slovenia into seven regions of provenances, named seed districts,[1] was made- by Maks Wraber in 1950. He stressed the biological basis of forest seed and tree nursery production and the need for a systematic, guided and supervised seed service, in particular the provenance and selection of forest seeds. On the basis of geographical, geological-petrographical, climatic and vegetation zones defined on the basis of phytocoenology, Slovenia was divided into seven forest seed districts: the Triglav, Kamniško-Savinjska, Pohorsko-Kozjaška, Podravsko-Pomurska (sub-Pannonian), Posavsko-Dolenjska, Postojnsko-Kočevska (mountain forest karst) and Karst Region seed districts. With the cooperation of the Expert Group for Seed Production and Forest Nursery Activities, the Forest Management Authority and the Slovenian Forestry Institute, a large number of forest seed stands intended for the permanent production of quality seed were selected in the field, and seed areas were identified where quality seed of native forest species was collected. At the same time, it was also foreseen where the collected seed would be used for sowing in the field and in tree nurseries. A system of a large number of forest nurseries has been designed, distributed across geographical areas and altitudinal vegetation zones. The selection of seed stands was based on phytocoenological (phytosociological), biological and ecological, genetic, systematic, technological and economic criteria. The division, based on vegetation units, represents the basic vegetation division of Slovenia and thus also reflects climatic impacts well. In the following decade, until 1961, Miran Brinar also set out detailed principles and methods for the selection of seed stands and drew up a detailed map for the demarcation of seed districts (Figure 4), and later the seed-production districts were further subdivided into altitudinal zones.

The division of Slovenia into seed-production districts was also given legal effect in the Seeds and Seedlings Act (1973): "Forest seeds and seedlings may be used only within the altitudinal zones and seed-growing districts where the forest seeds were produced."

1 The terms seed and seed-growing district were used. For practical purposes of translating Slovenian legislation into English language, the current proposals use the related term regions of provenance, which is more clear.

Figure 10: Map of forest seed-growing districts of Slovenia from 1971 (Register, 1971)

7.1.2 Seed Units (1986-2002)

At the time of the first revision of seed stands in 1982-1985, Marjanca Pavle introduced the division of Slovenia into seed-production units for the purposes of seed production. A seed-production unit is a group of similar forest communities on the same bedrock (carbonate, siliceous) and in the same altitudinal zones (0-399 m, 400-699 m, 700-999 m, > 1000 m). Reproductive material collected within a particular seed-production unit may only be used within that unit. Seed-production units are not geographically coherent units, but are fragmented across Slovenia. They were determined for each harvesting or planting and sowing site separately. They were designated per tree species or group of tree species. The division into seed-production units was applied in practice until 2002, and a second revision of the seed stands was performed on this basis, which was also led by Pavle until 1997. Unfortunately, the distribution of the seed-production units was inadequate for the mapping of the regions of provenance and difficult due to the high level of fragmentation in the field, therefore the demarcation of the regions of provenances had to be redefined after the adoption of the ZGRM in 2002, when the seed-production legislation had to be brought into line with European requirements.

7.1.3 Demarcation of Slovenia into Regions of Provenances (2002-)

In accordance with Directive EC/105/1999, the territory of Slovenia was delineated into regions of provenance (Kutnar et al. 2002). These are delineated to continue in the direction outlined by Wraber in 1950 and improved by Pavle in 1987. The improvement is based on the new phytogeographical division of Slovenia as proposed by Mitja Zupančič and Peter Žagar in 1995. For individual species, they are also delineated on population genetic studies. A similar division based on ecologically related areas is adopted in some other European countries, e.g. Germany (BML 1999).

The basis for the demarcation are the broad ecological zones, which are demarcated in more detail in the field by the boundaries between.the regional units – the forest management regions and the administrative boundaries – the cadastral municipalities, allowing a detailed overview of the production and recommended use of forest reproductive material in the individual forest management regions and forest management units Article 7 of the Rules on determining regions of provenance (2003) includes the following guidelines for use:

> The Rules on determining regions of provenance include recommendations for the use of FRM.

» (1) To guide the utilisation of FRM, the following suitability scale is used:

1. Most suitable: FRM from a seed facility within the same subregion of provenance and altitude zone;
2. Very suitable: FRM from a seed facility within the same region of provenance and altitude zone;
3. Suitable: FRM from a seed facility in the adjacent region of provenance and the same altitude zone;
4. Less suitable: FRM from the same region of provenance and altitude zone, but from different seed facilities;
5. Exceptionally suitable: FRM from other regions of provenance and the adjacent altitude zone in the same regions of provenance and altitude zone.

(2) If the most suitable or very suitable FRM is unavailable in the seed facilities of a specific region of provenance and altitude zone, and it is not available in the seed vault, FRM for suitable or less suitable utilisation may be stored or utilised, up to a maximum of one year.

(3) If FRM for less suitable utilisation is unavailable for more than 10 years, FRM for exceptionally suitable utilisation may also be stored or utilised, up to a maximum of one year.

(4) Regardless of the provisions of the preceding paragraphs, for the purpose of preserving forest genetic resources, only the use of FRM from the Šavrin sub-region of provenance shall be permitted."

This article was subsequently amended several times, so that, following the latest harmonisation, the utilisation of FRM from certain regions of provenance from neighbouring countries (Austria, Croatia and Hungary) in certain areas and altitudes in Slovenia is also permitted, subject to the prior expert opinion of the Institute.

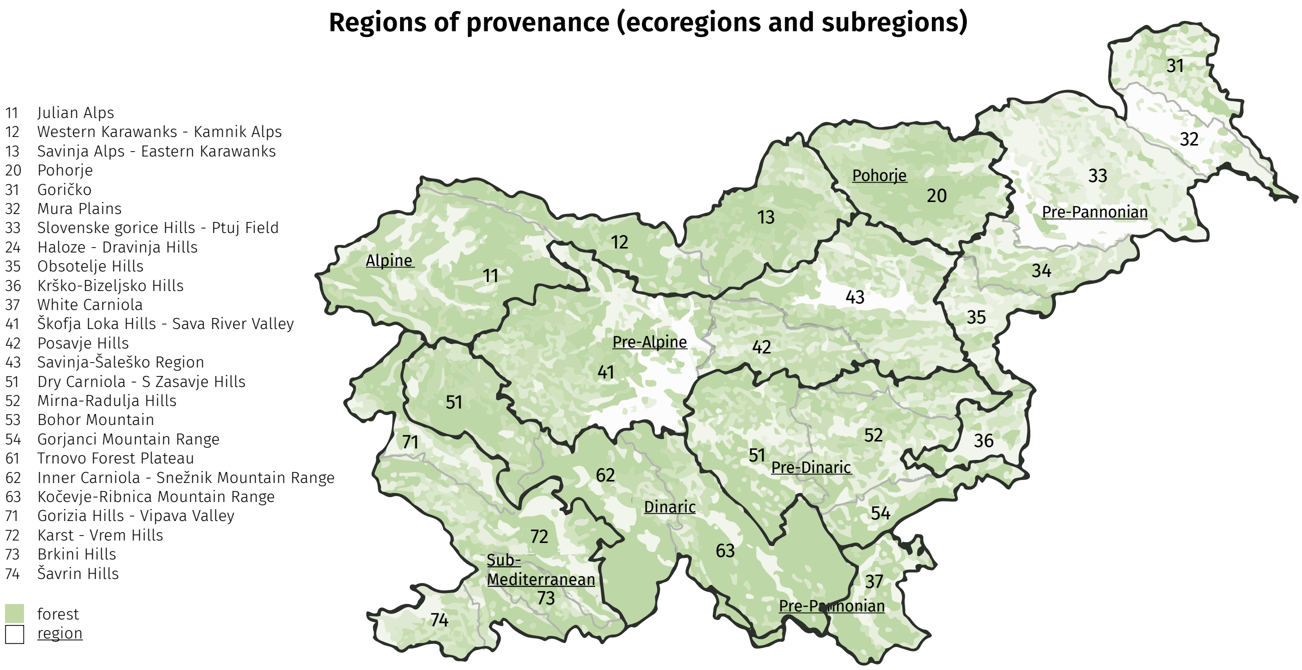

Figure 11: Demarcation of regions of provenance in Slovenia

7.2 List of Species to Which the Forest Reproductive Material Act (ZGRM) Applies

Directive EC/105/1999 lists 48 species and artificial hybrids subject to the guidelines of the Directive, which must be transposed into national law; it is permissible to supplement the rules set by this Directive, which may be stricter than those listed, but for each exception an informed application must be made to the EC for a waiver of the specific requirement. The latter was used by Slovenia in 2005, when the EC granted Slovenia and Denmark a decision to remove individual alien and economically non-viable species from the national list of species subject to the ZGRM.

From the principle of conservation of forest genetic resources, it is in the interest of the State to regulate all forest tree species used for planting in forests and permanent forest plantations, including windbreaks, park forests in urban areas, etc. For this reason, Slovenia has already included some additional species in the first species list, in 2010 it added all indigenous tree species to the list, and since 2019 efforts have been underway to include a larger number of shrub species that are important for the stability of forest ecosystems and whose funding for use would allow them to be included in the list of species subject to the ZGRM.

Table 4: List of species subject to the ZGRM; for clarity purposes, authors of the latin names have been omitted.

Abies alba	*Larix kaempferi*	*Prunus avium*	*Acer campestre*	*Phillyrea latifolia*
Abies cephalonica	*Larix x eurolepis*	*Pseudotsuga menziesii*	*Acer monspenssulanum*	*Pinus mugo*
Abies grandis	*Larix sibirica*	*Pyrus pyraster*	*Acer obtusatum*	*Pistacia terebinthus*
Abies pinsapo	*Malus sylvestris*	*Quercus cerris*	*Acer tataricum*	*Populus alba*
Acer platanoides	*Picea abies*	*Quercus ilex*	*Alnus viridis*	*Populus nigra*
Acer pseudoplatanus	*Picea sitchensis*	*Quercus petraea*	*Carpinus orientalis*	*Populus tremula*
Alnus glutinosa	*Pinus brutia*	*Quercus pubescens*	*Celtis australis*	*Prunus mahaleb*
Alnus incana	*Pinus canariensis*	*Quercus robur*	*Cercis siliquastrum*	*Prunus padus*
Betula pendula	*Pinus cembra*	*Quercus rubra*	*Ficus carica*	*Pyrus amygdaliformis*
Betula pubescens	*Pinus contorta*	*Quercus suber*	*Fraxinus ornus*	*Quercus crenata*
Carpinus betulus	*Pinus halepensis*	*Robinia pseudoacacia*	*Ilex aquifolium*	*Salix* x spp.
Castanea sativa	*Pinus leucodermis*	*Sorbus aria*	*Laburnum alpinum*	*Taxus baccata*
Cedrus atlantica	*Pinus nigra*	*Sorbus aucuparia*	*Laburnum alschingeri*	*Ulmus laevis*
Cedrus libani	*Pinus pinaster*	*Sorbus domestica*	*Laburnum anagyroides*	*Ulmus minor*
Fagus sylvatica	*Pinus pinea*	*Sorbus torminalis*	*Laurus nobilis*	*Quercus crenata*
Fraxinus angustifolia	*Pinus radiata*	*Tilia cordata*	*Mespilus germanica*	
Juglans regia	*Pinus sylvestris*	*Tilia platyphyllos*	*Olea europaea*	
Larix decidua	*Populus* x spp.	*Ulmus glabra*	*Ostrya carpinifolia*	

Species deleted by EC Order 2005

Alien tree species in the current Order

Species on the national list, most of them added by SI Order 2010

Species prescribed in the European Directive

7.3 Procedures for the Approval of Forest Seed Facilities

According to the Forest Reproductive Material Act (2002, including subsequent amendments and supplements), forest seed facilities (FSF) can include:

- seed trees or stands of forest trees for the production of forest reproductive material of the "source identified" category;
- stands of forest trees for the production of forest reproductive material of the "selected" category;
- tree orchards for the production of the forest reproductive material of the "qualified" category;
- stands or plantations of trees for the production of forest reproductive material of the "tested" category.

Approval of FSF for the "source identified" and "selected" categories is initiated on the basis of an application received from the owner of the seed facility; in the case of a demonstrated need to obtain forest reproductive material, SFS registers presumably suitable seed facilities and initiates the process of obtaining an application for approval from the owner – communication with the owner depends mainly on the competent district foresters or other locally or regionally responsible SFS experts. After the date of the visit has been coordinated between the owner, SFS representatives and those responsible for conducting the commission inspection with SFI, the SFS representative prepares the following documentation: descriptions of the stands and mapping material: an overview location map at a scale of 1:25,000 and a TP at a scale of 1:5000, with the boundaries and numbers of the land plots, divisions and sections plotted, as well as the information on the owner/-s of the land plots where the seed facility is located.

The commission inspection, led by SFI, shall be attended by authorised representatives of SFI, SFS and the owner or the owner's or manager's representative; if necessary, other experts, dendrologists, phytopathologists, and seed or nursery personnel shall also be invited to participate in the field inspection. During the field inspection, the stand is assessed according to the requirements on the information document, the stand/facility information, the access route to the facility, and the guidelines agreed between the SFI, the SFS and the owner/manager or representative for the tending of the stand and the collection of FRM are entered in the assessment sheet; these guidelines form a mandatory part of the unit's forest management plan (FMP). On the basis of the quantification of the criteria in the assessment sheet, at the end of the evaluation of the site and in comparison with the quality of other stands of the same species in the wider area, a decision is made by the committee as to whether the proposed site is approved and in which category ("source identified" or "selected").

7.4 FRM Certification Procedures

Confirmation of the SFS for *in situ* production of FRM: At least seven days prior to the intended production, the registered supplier must notify the local SFS unit (or, in the case of production in a seed plantation, the Institute) of their intention to do so by submitting an application for an FRM master certificate (Annex 6); the application shall guarantee the consent of the owner or manager of the FSF or his/her legal representative to the production of FRM on the property in question. NB: Since the main cost of FRM production is the collection, processing and storage of the FRM, the arrangement is usually verbal, often based on a commodity exchange agreement, e.g. delivery of a few fruit tree seedlings, etc.

If the production of FRM is in accordance with the medium-term reforestation needs of the territory of Slovenia, the application is eligible for exemption of the payment of administrative fees. The SFS shall review the application, accept it, provide the supplier with a detailed presentation of the approved forest seed facility and the requirements for the production of FRM in the stand or on a map, and shall specify the process of the SFS expert supervision for obtaining the SFS confirmation for the *in situ* production of FRM in FSF. If the production of the FRM was not carried out under the supervision of the SFS, the person in charge of the SFS regional unit shall, upon completion of the production, write on the SFS confirmation "Not under the supervision of the SFS".

The amount of FRM produced is signed daily on the SFS form by the person in charge in the field, usually the district forester, and the total amount is certified by the regional head of silviculture. During the production process, a sample (plant tissue – twig with three dormant shoots or seed/fruits/cones) in the prescribed quantities (in accordance with the Rules on Certificates) shall be taken from each tree from which the FRM has been produced, by the SFS expert officer (district officer or head of silviculture) or by the supplier producing the FRM in the FSF, from each of the trees from which the FRM has been produced (in accordance with the Rules on Certificates), stored in a paper bag and sent immediately to the SFI, together with a copy of the SFS certificate, for the purpose of checks on the origin at the time of issue of the master certificate or subsequent identification of the FRM. The supplier shall also deliver to the SFI a representative sample of the entire batch (lot) of processed seed, together with a processing record showing the initial quantity of FRM produced in the stand and the quantity of FRM processed.

Providing all information is adequate, and the directives for collection of FRM in FSF have been taken into account, the SFI shall issue a Master Certificate of Origin (Annex 7) no later than 14 days after receipt of the dossier. If requested by the supplier or the SFS, the SFI may also produce a seed quality analysis and then a quality report (Annex 8). The quality analysis can also be carried out by the supplier themselves according to an internationally recognised methodology (ISTA).

7.5 Overview of the Series of Procedures for Approval of FSF and FRM Certification

1) **APPROVAL OF SEED facilities under the General Administrative Procedure Act (ZUP)**

1. Stranka The customer submits an application on the prescribed form to the SFI.
   Fee: application – 50 points (tariff code 1), decision – 200 points (tariff code 3). The client is:
   - the owner;
   - a supplier to whom the owner has assigned the production of reproductive material for a fixed or indefinite period of time, by means of a notarised written declaration;
   - any other person authorised to represent the owner in the approval procedure.
2. The application shall be accompanied by:
   - a notarised declaration of renunciation of the production of reproductive material;
   - power of representation of the owner;
   - qualified: management plan;
   - tested: test results;
   - genetically modified: authorisation for release into the environment.
3. If the application is deficient, the SFI has 3 days to request additional information from the Ministry of Agriculture, Forestry and Food (MKGP) from the official records, which must be received within 15 days.
4. In the categories "source identified" and "selected", the SFI obtains an information document from the SFS. The cost of compiling the information document is borne by the SFS – Public Forest Service (PFS).
5. SFI prepares:
   - tending guidelines: for stands (for the category "selected");
   - guidelines for production: seed tree groups, stands;
6. SFI issues the approval decision with a written decree. Time limits for issuing a decision:
   - shortened declaratory proceedings: one month,
   - specific declaratory proceedings: two months.
7. The SFI enters the facility in the register.
8. The costs incurred in the approval process shall be charged to the client by the SFI. The SFI shall decide on the costs in a decision, whereby in practice all costs are covered by PFS.

2) **OVERVIEW OF SEED FACILITIES**

1. SFS inspects facilities of "source identified" and "selected" once a year. Costs: PFS.
2. The SFI reviews the "qualified" and "tested" categories once a year. Costs: PFS.
3. At the request of the owner, SFS/SFI inspects the facility. Costs: owner.

3) **DELETION OF SEED FACILITIES (ZUP)**

1. *Ex officio,* if the facility does not comply with the requirements. Fee: decision – 200 points (tariff code 3).
2. At the request of the facility owner.
   The costs of the deletion procedure are borne by the owner and decided by the SFI in a decision.
   Fee: application – 50 points (tariff code 1), decision – 200 points (tariff code 3).

4) **PRODUCTION OF REPRODUCTIVE MATERIAL (ZUP)**

1. Seven days before the start of the production of reproductive material (*in situ*), the supplier shall submit to the SFS a written application from the supplier for obtaining a master certificate from the Institute.
   For seed and clonal orchards, SFI is included instead of SFS.
   Fee: application – 50 points (tariff code 1), certificate – 50 points (tariff code 6), certificate – 30 points (tariff code 6).
2. If the supplier is not the owner, the application must be accompanied by:
   - a notarised declaration of renunciation of the production of reproductive material;
   - a written declaration of a one-off renunciation of the reproductive material;
   - power of representation of the owner.
3. During production, the SFS (SFI) periodically monitors the harvesting:
   - an inspection report shall be drawn up;
   - a sample of the reproductive material, which is forwarded to SFI, is taken
   - the supplier enters the quantity of reproductive material harvested on a daily basis on the application form
4. SFS issues the application as a confirmation to the supplier and sends a copy of the confirmation to SFI. The costs incurred in the procedure are charged to the supplier by the SFS by decision (price list established by the Ministry of Agriculture and Rural Development).
5. The SFI issues the master certificate no later than seven days after the completion of the production or completion of the processing. Depending on what the supplier has requested in the application, the SFI issues a Master Certificate:
   - as soon as they have received the certificate and the sample from the SFS after the certificate has been issued, a sample of the processed seed is taken for the genebank;
   - after processing at the seed establishment, where SFI checks the quantity and takes a sample of the processed seed for the genebank.
6. The SFI charges the costs incurred in the procedure to the supplier by decision (price list to be established by the Ministry of Agriculture and Rural Development).
   If all the procedures are for the approval of seed facilities and the certification of FRM for the purposes of medium-term planning of the need for FRM in Slovenia in accordance with the plans of the SFS, the procedures are exempt from fees, and the costs of the approval are part of the tasks of the public forestry service.

Figure 12: European ash: leaf-out phases (vegetative bud burst phenological phases) (drawing by Metka Kladnik)

8

FOREST SEED FACILITIES

Forest seed facilities are approved by the Slovenian Forestry Institute under a modified administrative procedure on the basis of an application from the owner/s and/or operator/s of the facility and a commission inspection to assess whether the facility meets the regulatory criteria. It is assigned an identification number by the SFI, the first number of which indicates the region of provenance (1 to 7, or 0 if the region of provenance covers the entire territory of Slovenia), and is entered in the Register of Forest Seed facilities published in the Official Gazette of the Republic of Slovenia in January each year and added to the European FOREMATIS list. In 2019, a total of 380 seed facilities were approved in Slovenia, of which 104 for the production of FRM "not for use in forestry". Among the others, the majority are in the "selected" category for FRM production, fewer are in the "source identified" category, only one seed orchard is approved for FRM "qualified", and there is no registered seed facility in Slovenia for the "tested" category.

It should be noted that the purpose for which the FRM is produced in a particular seed facility may be indicated by:

1 – "for multifunctional forestry";

2 – "not for use in forestry" or "of limited wood production significance" – such a seed facility is for use in forestry, but is primarily intended for the conservation of forest genetic resources.

The purpose for which the FRM is produced may be
1 – "for multifunctional forestry";
2 – "not for use in forestry";
A footnote indicates if it is a forest genetic reserve or a genetic monitoring plot.

8.1 Seed trees or stands for the "source identified" FRM category

Due to the need to conserve forest genetic resources and the scarcity of FRM, an increasing number of forest seed facilities have been approved in Slovenia for the production of FRM in the "source identified" category. However, these FSF must also meet the criteria for approval as written for FSF of the "selected" category, except that criteria 8, 9 and 10 are not taken into account for approval (see 6.4); the proportion of trees with major defects can be up to 40%. Such a seed facility may be a stand or a group of seed trees; for autochthonous tree species it shall normally be of natural origin, except that for spruce and Austrian (black) pine it may be non-autochthonous or of unknown origin; for seed facilities that are not of natural origin, a note shall be added to the effect that the stand originates from FRM of local origin or from a local tree nursery.

8.2 "Selected" Seed Stands

Seed material for reforestation by planting in Slovenia is mainly produced from selected seed stands. They represent the best part of the populations of a tree species in terms of the characteristics that are important for the future development and yield of that tree species in a harvested forest. The objective of seed stand management is tailored to the role of seed production and includes the production of quality seed with excellent genetic composition, while at the same time meeting other forest management objectives. Seed from our seed stands is categorised as "selected" in the European Forest Reproductive Material Categorisation Scheme. The provenance, climatic conditions in the area and a series of other data are known, and the starting material (the seed stand) is selected on the basis of the phenotypic characteristics of the whole population of trees in the stand.

The practical benefits of selected seed stands were already identified by Wraber (1951):

1. ZReliable provenance and good quality forest seed are guaranteed.
" 2. It is possible to effectively control the collection and distribution of seed, which must be rational in terms of use within and outside the seed district.
3. All the racial characteristics of forest trees contained in certain forest stands are preserved and strengthened by the correct grouping of the seed districts.
4. The demarcation of forest seed districts and the isolation of seed stands is the scientific and practical basis for the selection of forest seed, i.e.

for the intensification of good growth characteristics and for the cultivation of as pure a species as possible, with an established hereditary basis or a high vital force.

5. The grouping of the seed districts determines the appropriate horizontal and vertical distribution of the forest nurseries and allows for an efficient utilisation of forest seedlings.
6. Seed districts provide a realistic basis for forest seed planning, both in terms of absolute quantity and the quantitative relationship between tree species.
7. Adherence to the previous criteria will increase the quality of forest seeds and forest seedlings and, of course, the quality and quantity of forest production.
8. Accurate information on the provenance and selection of forest seeds will greatly increase their value when sold abroad."

Sustainable, multifunctional and co-natural forest management requires strict adherence to the origin of the seed and continuous selection of seed material when resowing and replanting. The seedlings, which are formed through reforestation by planting or sowing, must have the genetic makeup to be able to meet all the forest management objectives of the future forest. The type of measures in the selected seed stands is therefore forest tree cultivation. It aims to improve the genetic makeup of future forest tree populations in line with the intended objectives, while maintaining a broad genetic diversity that will provide the population and the species with security in the face of unpredictable changes in the environment.

8.2.1 Description and Tending of Selected Seed Stands

The description and analysis of the condition form the basis for planning care measures in seed stands. A detailed analysis of the forest stand condition is already carried out in the process of selecting seed stands of individual tree species. Situation analysis involves evaluation and assessment of:

- the need for selected seed stands of a given species in the region of provenance and altitudinal zone under consideration;
- information on the site and stand;
- the stand based on the phenotypic characteristics of all specimens;
- stands of the tree species concerned in the area adjacent to the seed stand.

The basic information includes information on the site, forest community, stand size, timber stock, increment, number of trees of the tree species under consideration, stand structure, stand composition, tree species mix, etc. This information helps us to formulate an appropriate long-term silvicultural objective for the seed stand and a list of silvicultural measures to achieve this objective. The necessary information is given in the seed stand information document, which forms part of the documentation for each seed stand. This information is also the basis for the silvicultural plan, which is somewhat more complex and requires more information than the harvested forest, where the seed production function is not as emphasised to such an extent.

A qualitative analysis of the stand based on the phenotype of all individuals is the basis for planning measures to improve the genetic composition of the stand or for cultivating a particular forest tree species. The objectives of cultivation of a particular tree species are defined by the traits that we want to improve in that species, while the cultivation programmes define the methods, procedures and selection criteria that will be used to achieve genetic improvement. Measures in seed stands are an important part of a species' cultivation programme. They do not destroy the genetic structure of populations, but ensure a continuous process of improvement. For this reason, it is considered that seed stand management is a major component of the sustainable breeding of forest tree populations and an integral part of "cultivation without cultivation" (El Kassaby and Lstibůrek 2009).

Morphological and phenological trait characteristics, which are the criteria for selection in oaks, may be more or less genetically conditioned. The environment and silvicultural measures can influence the expression of these trait characteristics to a greater or lesser extent. The long lifespan, and therefore the long lead times for provenance tests, and the difficulty of organising progeny tests that best demonstrate genetic composition of the traits under consideration because of linked inheritance and trait characteristics that result from the expression of a large number of genes, make it difficult to distinguish accurately between hereditary genetic traits and environmental influences. The greater or lesser influence of heredity on a particular trait characteristic is shown in the following table.

Table 5: Relative effects of heritability and environment on trait expression in beech and oak and silvicultural measures in selected seed stands

TRAIT	Hereditary influences	Environmental influences	Effects of tending	Measure
straightness of the trunk	low	great	great	note
tree forking	great	possible	low	remove
trunk twisting	great	low	low	remove
adventitious shoots	low	great	great	value
vitality	great	great	medium	note
trunk cracks – dry	medium	great	medium	note
trunk cracks – wet	low	great	medium	remove
late onset of leaves/needles	great	low	low	support
thickness of branches	great	medium	medium	note
approach to cylindrical form	medium	great	great	support
crown length	medium	great	great	note
shape of the crown	great	great	great	support
damage caused by diseases and pests	medium	great	medium	note

The list of selection criteria may be extended. In France, it includes the physical and chemical properties of the wood – the thickness of the conductive elements, the shrinkage and tension of the wood fibres in different directions, the thickness of the annual ring, the colour of the wood, the tannin content, etc.

In selection for breeding, it is necessary to decide on a single criterion or a small group of selection criteria, and it is therefore advisable to rank them, to determine the rank of each criterion for all the trees in the selected seed stand, and, in the case of silvicultural measures, to eliminate mainly the trees with the worst characteristics, which are predominantly hereditary.

A long-term silvicultural objective determines the future state of the seed stand that will meet our needs. It should be borne in mind that a seed stand fulfils more functions than a normal harvested forest. Thus, the long-term silvicultural objective consists of several components:

- the production of seed material with an excellent genetic makeup, maximising genetic diversity;
- the production of large quantities of quality seed over a given period of time;
- ensuring conditions for seed collection (adequate stand structure);
- fulfilling the wood-production function of the stand (seed stands are stands with a particular quality of wood mass, and the importance of this objective is therefore emphasised);
- the fulfilment of all other forest functions, such as protective and social functions (their relative importance is the result of their evaluation in a wider context).

Silvicultural measures (thinning) in seed stands are mainly aimed at:

- selection for specific target traits (removing individuals with undesirable traits, especially those that are more dependent on genetic composition);
- increasing seed yield (crown release);
- maintaining adequate stand structure (distribution of trees, stability, persistence of the filler layer, slowing down of natural rejuvenation);
- protecting genetic diversity (the size of the population that cross-pollinates);
- increasing the value increment of the stand (the wood production function is not less important, only the harvesting of quality trees is delayed for the time when the stand will be used for the production of quality seed).

8.2.2 Some Difficulties in the Selection and Tending of Selected Seed Stands

The selected seed stands have historically been represented mainly in conifers, with most of the seed stands from the time of the first registers and their revisions being approved for spruce. With the establishment of the Slovenia Forest Service, the selection of seedlings for planting has shifted to broadleaved trees, and the number of seed facilities for these has been greatly increased. However, it still falls short of meeting all needs for all species in all provenance regions and altitudinal zones. The selected seed stands are often too small, so the number of trees pollinating each other is too small. Using seed material from such stands, it would be possible to initiate major changes in the genetic structure of future stands (the bottleneck effect from the chapter on genetics).

In the past, intervention in selected seed stands has been very limited due to breeders/silviculturists' fear of losing the positive characteristics of a phenotypically superior stand. As a result, these stands are often too thin, untended, the canopy is cramped and, due to their age, often no longer able to respond to possible release. This results in poor fructification even during a massive seed year for a particular tree species in the vicinity of the selected stand.

In the past, not enough attention has been paid to the development phase of the selected stand. Often they have been selected during the rejuvenation phase, so that they have developed a thick layer of young seedlings or saplings that prevents the seed from being collected from the ground.

Since 2002, we have also been including the late pole stage forests in the selection of seed stands, with relatively intensive measures depending on the structure and age of the stand and the expected time of seed production of the "selected" category in each stand. In particular, we try to approve as a seed stand as large an area of forest as possible (optimally around 100 ha for the majority species or a larger number of groups of minority tree species), representing the same population of forest trees (inter-pollinating), in which it is possible to apply an irregular shelterwood system, leading to a mosaic structure of the parts of such a stand suitable for FRM production.

Photo 3: A branch with cones during the mass fructification of spruce (photo by Hojka Kraigher)

8.3 Organisation of Seed and Seedling Supply

Without a comprehensive system in place to ensure a sustainable supply of seeds and seedlings, it is not feasible to sustainably guide forest development through planting and sowing to complement natural regeneration. As a rule, seedlings are raised over several years. Therefore, a medium-term programme (5-10 years) of seedling needs and seed collection is needed, as a basis for planned seedling production and, if necessary, for sowing in the forest. The medium-term programme needs to be updated annually. On the basis of forest management plans, the annual reforestation programmes and the medium-term programme of seedling and seed requirements, annual silvicultural plans and seed collection plans are drawn up, as well as seedling cultivation programmes by tree species, quantity and provenance. To ensure that tree nurseries have an uninterrupted supply of seed, it is necessary to have transitional stocks available for years when there is no seed crop. This role is performed by the Seed Storage. However, because of the above problems with seed production in selected seed stands, we also approve, if necessary, ordinary seed stands or groups of seed trees ("source identified") for the production of a limited quantity of seed in a given year. As an example of the only FRM production in Slovenia, we provide data on the main certificates issued (Table 6) and the SFS data (Table 7) for the FRM produced in Slovenia for the years 1998 and 2018.

Table 6: Seed collection in selected seed stands, ordinary stands and seed trees in Slovenia in 1998/1999 (individual lots collected and used by Slovenia Forest Service (SFS) in the same area) and 2018.

TREE SPECIES	Seed quantity (kg)	Quantities needed for SFS in 1999	Quantities by certificate issued in 2018
Abies alba	69	69	92
Fagus sylvatica		1.316	538
Fagus sylvatica – pullings from natural regeneration			200.000
Quercus petraea	600	1.429	4.950
Quercus robur	600	1.151	7.620
Acer pseudoplatanus	136	414	90
Acer platanoides			160
Fraxinus excelsior	98	537	0
Fraxinus ornus		10	0
Prunus avium	94	30	540,5
Carpinus betulus		3	4.200
Ostrya carpinifolia		10	0
Alnus glutinosa		1	2,8
Sorbus torminalis	1	1	0
Pseudotsuga menziesii			1,3
Castanea sativa			2.600
Malus sylvestris			1,3
Pyrus pyraster			1,3

Table 7: Planting and sowing of seeds and seedlings in Slovenia in 1998 (only for reforestation after regular felling, sanitary felling is not taken into account; sanitary logging may require up to 1.3 million additional seedlings per year) and in 2018.

TREE SPECIES	Seed sown (kg) in 1998/2018	Seedlings (in 1000) in 1998	Seedlings (%) in 1998	Seedlings in 2018	Seedlings (%) in 2018
Picea abies	24	789	46	488.233	48
Abies alba	4	18	1	10.298	1
Pinus silvestris	5	41	2	5.865	1
Pinus nigra	414/32		3	1.200	< 1
Larix decidua		51		21.400	2
Other conifers		4		2.083	< 1
Fagus sylvatica	4	229	13	309.598	30
Quercus petrea	20/5	152	9	15.580	2
Quercus robur	724/1.460			68.225	7
Noble Broadleaves		353	21		
Other Hardwoods		11	1		
Fast-growing broadleaves		61	4		
Other Broadleaves	31	5			
Fraxinus excelsior	N. Z./3				
Acer pseudoplatanus				49.110	5
Prunus avium				21.695	2
Alnus glutinosa	N. Z./0,5			17.000	2
Carpinus betulus				3.730	< 1
Populus spp.				2.805	< 1
Tilia platyphyllos				2,420	< 1
Sorbus aucuparia				842	< 1
Crataegus spp.				650	< 1
Malus sylvestris				595	< 1
Acer platanoides				575	< 1
Juglans regia				551	< 1
Castanea sativa				527	< 1
Ulmus minor				400	< 1
Pyrus pyraster				382	< 1
Sorbus domestica				38	< 1
Sorbus torminalis				27	< 1
Sorbus aria				1	< 1
TOTAL		**1.710**	**100**	**1.023.830**	**100**

8.4 Systemic Problems in Forest Reproductive Material Production and Tree Nursery Activities (2017 Summary)

We summarise the conclusions of the meeting Systemic Problems of Forest Regeneration, which took place in November 2016 at the Slovenian Academy of Sciences and Arts (SAZU). The conclusions were published in the Forestry Journal in spring 2017.

Figure 13: Cover of the Forestry Journal, which published the papers from the meeting Forest and Wood: Systemic Problems of Forest Regeneration.

Summary and conclusions of the Forest and Wood Scientific Meeting: Systemic Problems of Forest Regeneration.

(published with permission of the Forestry Journal and proofread)

On 24 November 2016, the 4th Class of Natural Sciences of the Slovenian Academy of Sciences and Arts (SAZU), the Council for Environmental Protection of SAZU and the Slovenian Forestry Institute organised the third traditional scientific meeting FOREST and WOOD, this time with the title: Systemic Problems of Forest Regeneration. In addition to academics, the meeting was attended by representatives of all key stakeholders in Slovenian forestry, representatives of the Ministry, scientific and educational institutions, planners and managers, forest owners, the Chamber of Agriculture and Forestry, the forestry cluster and forest nursery representatives.

Forests are increasingly threatened by rapid climate change, resulting in extreme weather events, such as the large-scale glaze ice in February 2014, a shift in rainfall patterns and a consequent increase in pests and diseases, such as bark beetles in 2015 and 2016. It is therefore necessary to supplement the forest management system which in Slovenia has been among the most advanced – close-to-nature – in the world for decades. Unfortunately, in a context of large areas being cleared, problems of decline of individual tree species, and crown damage in areas where seed development takes place, in the reproductive part, natural regeneration cannot always guarantee successful regeneration to support all forest functions. The role of forests in maintaining biodiversity is not affected by the gradual transition through different successional phases, e.g. through the so-called silvicultural phase, but the development of wood production and some other functions is postponed to a more distant future. For forest owners, the timber industry and society as a whole, there is therefore a need to complement the existing doctrine of sustainable forest management, based on the predominance of natural regeneration, with regeneration supported by planting and sowing at a higher rate than the current 3% of annual forest regeneration. This can help to speed up the transition to the wood-producing phase; the use of different tree species, which are in a minority in existing communities and do not provide adequate regeneration centres, can greatly reduce the risk of future disturbances; and the use of genetically diverse forest reproductive material can help to maintain the adaptive potential of future forests to climate change and other factors that threaten the development and the multiple roles of forests. The productive function of forests is also highlighted by representatives of forest managers, planners and owners, as well as by the wood processing industry, which has experienced a revival in recent years, as wood has traditionally been Slovenia's main renewable natural resource for boosting the economy.

At the panel entitled Systemic Problems of Forest Regeneration, lecturers and other participants of the November 2016 meeting FOREST and WOOD at SAZU critically addressed the state of forest seed and nursery production in Slovenia, which, due to the transition to predominantly (more than 90%) natural regeneration, has been losing traditional knowledge over the last two decades, as nurseries are being closed down and the production of planting material has dropped to one tenth since the time before Slovenia's independence. In the wake of problems of individual tree species (ash decline, oak, alder and the already existing diseases of elm and chestnut, the expansion of bark beetle on spruce, which has been markedly accentuated in the last two years), there is a need to assess the suitability of other species and provenances of forest trees, potentially including the use of alien species (in a limited and prudent way), as well as the use of a greater number of species in reforestation to disperse the risks.

Forest regeneration planning is carried out by the Slovenia Forest Service (SFS) in close cooperation with forest owners. The Director of SFS, D. Oražem, therefore stressed that climate change has placed forests and forestry in a fundamentally different situation from the one prevailing until recently. The dilemmas are not only faced by forest owners, the timber industry, forest policy and the broadest range of forest users, but also by the Public Forest Service. The latter, through appropriate targeting of restoration works and networking, is one of the key conditions for a sustainable future forest without too many risks and for the benefit of the forest as an ecosystem, its owners, users and society as a whole. As part of forest management planning, the expert services prepare an overview of medium-term seed and seedling requirements for each tree species by provenance and altitude zone. Seed production depends on the biology of the species, as they do not produce every year, and seed storage on their biology. For example, the seed of oak trees cannot be stored for more than over one winter to maintain its germinability. The germination of our conifer seeds can be stored for decades. However, the time for raising seedlings of any tree species is always several years. Therefore, medium-term planning is important both for the adequate storage of seed stocks in the SFS seed storage and for the availability of suitable seedling forms for planting in forests. In addition to understanding the effects of the environment during seed development and seedling rearing on the physiology, phenology and success of the young forest in the later years after planting (gene expression studies – the effects of epigenetics on tree physiology), it is important to obtain seed at the time of massive fructification, when the seed is of good quality and genetically diverse, and to raise seedlings in locations that are as close as possible to the climatic conditions of the stands in which they are to be planted. Planting should be carried out in a manner that is appropriate to the climatic conditions of the forest stands in which they are to be planted. This means that we urgently need to support local or at least regionally distributed nurseries, and it is worth noting that, as Dr N. Ogris of SFI mentioned in his paper, there were at least 45 forest nurseries in Slovenia in the past, there were 16 three decades ago, and today there are only three functioning forest nurseries, while the largest forest seed-production organisation collapsed a few years ago. It is worth noting here that individual forest nurseries have for many years also financed their activities for forestry purposes from their horticultural activities, as V. Planinšek from the Omorika Tree Nursery in Muta presented with an overview of their operations.

Appropriate forest seed and tree nursery planning is based on long-term and medium-term forest regeneration plans, using seeds and seedlings of as many forest tree species as possible, suitable for use in different provenance regions and altitudinal zones. The irregularity of fructification requires flexible financing for seed production, processing and storage, and the multiannual system of seedling rearing means that medium-term plans must provide for renewal at least five years in advance, taking account of the increasing need for reforestation following sanitary felling in the context of regular regeneration by planting and sowing. To implement these plans, the procurement system needs to be adapted for the entire plan period, and planning needs to provide for minimum, optimum and maximum levels of regeneration through planting and sowing (within the financial envelope foreseen or increased).

Over the last 15 years, 25-35 different species of forest trees have been used in the context of replanting and sowing, but only a few species dominate in terms of quantity, and spruce still accounts for around 40% of all seedlings. The decision to support and plant tree species must take into account the trends in the impact of climate change on species success: T. Levanič, SFI, pointed out that the trend is negative in the lowlands and that changes in temperature and precipitation regimes are expected to lead to the collapse of lowland forest ecosystems, while in the highlands the effect is at least transiently positive. When selecting species, R. Brus and L. Kutnar pointed out that pure, large-area, single-species stands should be avoided, and as many different tree species as possible should be used to spread the risk. It is appropriate to use mainly species with which we already have experience, while at the same time starting to try out new species, which may be alien, in a more bold approach. Where there is no possibility for natural seeding, where large areas of forest are damaged, where there is a risk of erosion processes developing, or where silvicultural objectives are not achievable due to disturbances in rejuvenation, where there is a desire to replace existing tree composition that is inappropriate to the site, or to increase the biodiversity of the stand, reforestation by planting and sowing is a necessary complement to natural regeneration, as pointed out by M. Westergren et al. (2018). Maintaining forest seed and nursery knowledge and flexible funding are important within this context. At the same time, the whole chain from seed to well established seedling planted in the forest must be improved, or, as phytopathologists N. Ogris and D. Jurc point out, in addition to ensuring healthy seedlings, it is necessary to ensure quality in a systematic way, to define the required characteristics of the seedlings, the way they are dug, transported, handled before planting, the way they are planted and the way they are cared for after planting by means of a standard or a quality code, as the success of the artificial reforestation by means of seedling planting is determined by all of the above requirements. In addition, regular testing for the presence of hidden, latent and cryptic pests, with a focus on *Phytophthora*, should be ensured in forest tree nurseries.

The dilemmas are faced not only by the public forest service, which is moving towards complementing predominantly sustainable forest management, but also by the demands of forest owners, the timber industry, forest policy and the broadest range of users, especially nature conservation, which has traditionally been the primary focus of forest management planning. J. Krč from the Forestry Department of the Biotechnical Faculty (BFG) pointed out that careful and professional forest production (reforestation of areas affected by disturbances) has a positive impact on the success of stand rejuvenation – both in terms of the composition of mosses, herbaceous and shrub species, and the structure (composition, density, vigour and height of individual tree species) of the future stand. It is therefore necessary to create the conditions (a quality assurance system) to ensure that as much as possible of the quality reforestation of the areas affected by the disturbances is achieved. M. Humar from the Department of Wood Science at the Biotechnical Faculty (BFL) pointed out that proper planning of wood use and interdisciplinary cooperation can significantly reduce the economic damage caused by bark beetle and wood staining. If the bark beetles are removed from the forest in time, the mechanical properties of the wood will not deteriorate. On the other hand, blue stain fungi have a positive effect on permeability, making it easier to impregnate this wood with biocidal wood preservatives. In some countries, blue-stained wood is particularly valued and sought after for higher added-value products, so the emphasis in the use and valuation of such wood is on its innovative use and promotion.

In addition to the biological problems, it is important to think about better organisation of the owners' associations combining their efforts in the market and in the organisation of work in forests, given the very unfavourable ownership structure, as pointed out by State Secretary M. Podgoršek of the Ministry of Agriculture, Forestry and Food. The forest and its biodiversity do not need forestry intervention, but, as the Director of the Directorate for Forestry, Hunting and Fisheries, Mr Jakša, pointed out, in the face of such major disturbances as Slovenian forestry has faced in the previous years, planned management cannot be based solely on natural regeneration of the forest, but must help nature by planting appropriate tree species. We need to ensure the productive function of the forest for the future. Forest owners cannot wait decades for natural regeneration and a century or more for an adequate composition of economically interesting forest tree species.

At a time when we are celebrating the 30th anniversary of the IUFRO World Congress in Ljubljana, where Prof. D. Mlinšek introduced the principles of sustainable forest management to the global forestry professionals and scientists, the forestry profession is facing new challenges of climate-driven changes in forest ecosystems, which necessarily lead to action and the further development of the forest management doctrine. We are entering a phase where natural regeneration is still the *'alpha'*, but no longer the *'omega'* of forest management in Slovenia. We urgently need to act before we lose our traditional knowledge, while it is still possible to revive Slovenian seed and tree nurseries, and thus protect forest genetic resources, forest ecosystems and all functions of forests in Slovenia.

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key notes from the presentations were provided by the lecturers and the representatives of forest tree nurseries.

9

SEED AND SEEDLING DATA FOR SELECTED TREE SPECIES

9.1 European Beech (*Fagus sylvatica* L.)

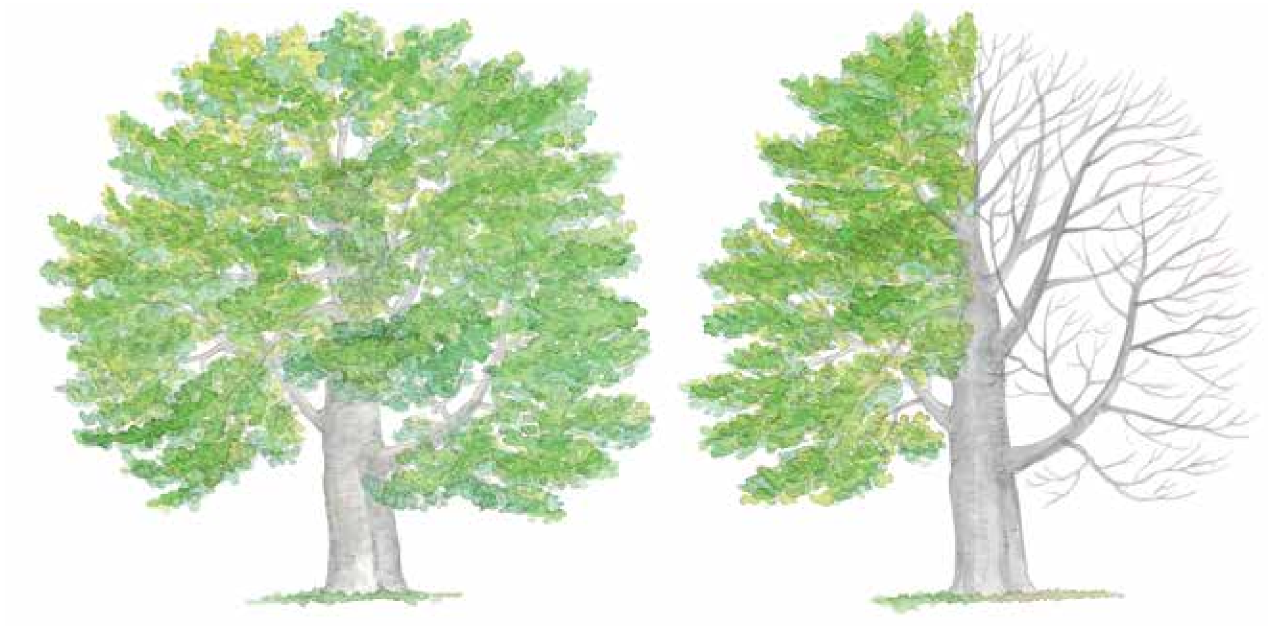

Figure 14: European beech: habitus (drawing by Marija Prelog)

Beech is the most widespread naturally occurring forest tree species in Slovenia. According to the Forest Development Programme of the Republic of Slovenia, in the total growing stock of Slovenian forests it could potentially comprise 58%, but in fact it comprises 29% of the total growing stock. According to the forest inventory conducted 30 years ago (1990), it is distributed over approx. 900,000 ha (in 75% of all forest departments and sections), roughly evenly across all altitudes, 84% on carbonate and 14% on non-carbonate bedrock, in relation to the total area of forests in Slovenia. Its natural distribution in Slovenia is the result of natural expansion from glacial refugia and the relatively small proportion of planting in the history of Slovenian forestry before the establishment of the SFS.

Research on the genetics, morphology, physiology and silviculture of beech in Slovenia was mainly led by Dr M. Brinar between the years 1950 and 1973. Interesting features of the research at that time were the possibility of the occurrence of a taxonomically not well defined species of Balkan beech or Moesian beech *(Fagus moesiaca* (Domin, Maly) Czecz.) in Slovenia, whose characteristics are related to, among other things, the rooting ability of cuttings; he also described a beech population with oak-like bark, and another beech population with birch-like bark. Within the context of seed and germination physiology, the emphasis was then on research into the effects of cholines on germination and alternation of forest tree species, and the effects of ecotypes and site-based races on the quality of beechnut. In the last period, a thesis on high-frequency electrophotography and seed vitality, especially of beech, was written, an internship thesis on testing seed vitality with tetrazolium (Čater 1995), a PhD thesis on genetic variability of beech in Slovenia (Brus 1999), and later several studies on the effects of silvicultural measures on genetic diversity (Westergren et al. 2015) and the identification of the origin of seed lots from the 2016 seed year (Westergren et al 2017), as well as an expert study on the processing, storage and germination of beechnuts (Finžgar et al. 2016).

Planting of beech seedlings accounts for an increasing share of replanting and sowing in Slovenia. In 1997, for example, approximately 250,000 beech seedlings and 64,000 pullings from natural regeneration were used for regular planting, and more than 500,000 beech seedlings were used for sanitary planting; in 2018, nearly half a tonne of beechnuts were used to grow seedlings, and 200,000 pullings from natural regeneration were planted; in total, beech seedlings accounted for almost one-third of all seedlings planted. Reforestation by planting and sowing beech is constrained by irregular harvesting and the high cost of storing the beechnuts for a long period, which affects the difficulty or delay in implementing silvicultural plans. Below are some basics on the physiology of flowering and fructification of beech trees, as well as the problems of storage, dormancy and the planning of a coordinated germination of beechnuts.

9.1.1 Flowering and Fructification

Beech trees reach reproductive maturity between the ages of 60 and 80, and solitary trees between the ages of 40 and 50. It blossoms at about the same time as it develops foliage, from early April to mid-May. The flowers are monoecious; separate male and female flowers grow on the same tree. They are sensitive to frost. The beech is an anemophilous plant, where each pollinated female flower produces a single fruit, the beech fruit, where 2–3 (4) together in a prickly cupula form a beechnut.

The beech seed (a fruit, in fact) ripens in September and October. beechnuts continue to fall down till the end of November. Wind and rain accelerate the release of the nuts. The small triangular-shaped pyramids are about 1.5 cm long. The pericarp is relatively thick, smooth and shiny. As the seeds dry out, they change colour from dark to light brown. Ripe beechnuts have a moisture content of 20-30%, depending on weather conditions. It does not have any endosperm. The contents of the seed are filled by two twisted cotyledons and an embryo axis. The seeds are edible.

Beechnut production starts in stands usually at the age of 70/80 years and can last up to more than 200 years. Seed production is irregular, depending on microclimatic conditions. A full crop can be expected every 5-10 years and an incomplete crop every 3-5 years. In the intermediate years, either there are no seeds on the tree, or individual seeds develop, but they are mostly empty. The first seed that falls in September is usually of poor quality, empty or contains parasites – insects. Some of the characteristics of beech mast are shown in the following table.

Table 8: Characteristics of beechnuts ([1] - taken from Suszka, Muller, Bonnet-Masimbert 1996, [2] - taken from Regent 1980)

	Number of beechnuts per kg
– when harvesting (humidity 25%)[1]	3,000–5,000
– after drying (humidity 8%)[1]	3,500–5,800
– when harvesting (humidity 22-30%)[2]	3,600–6,800 (AVG. 4,600)
Weight of 1,000 nuts[1]	150–300 g (avg. 250 g)
Weight of 1 hectolitre of nuts[1]	approx. 50 kg of fresh beechnuts, 39-45 kg of dry beechnuts

Figure 15: European beech: stages of female and male flowering and the fruit of the European beech tree (drawing by Marija Prelog)

9.1.2 Harvesting and Processing of beechnuts

Beechnut is almost always harvested from the ground. If closed fruits are collected from fallen trees, they should be spread out in a thin layer in a dry place until they open, then the individual beechnuts are separated by shaking and sifting. It is recommended to harvest immediately after they fall off to minimise losses to rodents and birds and to reduce the development of diseases. Clearing the ground under the trees improves the conditions for picking. It is not recommended to harvest the beechnuts that fall off first.

Hand-picking is the most widespread method. Other methods include the use of nets and punctured foils, sweeping under trees and vacuuming with portable vacuum cleaners. In each case, a mixture of fruits, beechnuts, twigs, leaves and stones are collected. The first cleaning can be done in the field, removing stones, sand and most of the foliage. The beechnuts are then spread in a thin layer for a few weeks near the harvesting site. Each lot is spread separately on the ground to partially dry. During this time, the beech mast is hand-mixed on a daily basis. Another way of sorting and cleaning is by using water settling tanks, but in this case immediate partial drying of the beechnuts is necessary.

The mixture of beechnuts and impurities is transported in jute sacks or nets to the processing site. In Slovenia, processing was carried out at Semesadike Mengeš until their closure, and later at the Omorika Tree Nursery in Muta and at the Slovenian Forestry Institute for the needs of the seed storage of the Slovenian Forest Service. On arrival, the moisture content of the beechnut is usually between 25-32%, therefore it is dried first (48 hours in the air or in an oven at 18-20 °C). It then goes on to mechanical cleaning, depending on the type and amount of impurities:

- in a water settling tank to remove empty seeds;
- in the air blower to remove light impurities;
- in an air screen, in which different mesh sizes (e.g. upper with 9 mm openings, lower with 8 mm openings) are used to remove stones, small beechnuts and light impurities in jets of air.

By foreign standards, the acceptable purity of a lot of beechnuts is 85-95%.

Beechnut is an orthodox seed, which means that its moisture content can be greatly reduced before storage without affecting its vitality. The degree of drying depends on the intended storage time and the drying method used, but in any case the temperature during drying should not exceed 18-20°C.

When stored over one winter, the beechnuts may be dried to 20-25% moisture and stored at 3°C in a dry substrate (peat or vermiculite) or dried to 12% moisture and then stored at 3°C until the end of the winter, when a moist pre-treatment at the same temperature is started before germination. When storing for more than one year, it is essential to reduce the humidity level to 8-9% before the start of storage, which takes place at -5 to -10 °C in hermetically sealed containers. It can be dried at 18–20 °C with a fan, a procedure that can be interrupted by drying without blowing. Drying can take from a few hours to a few days, depending on the relative humidity and moisture content of the seed lot. Drying capacity can be improved by using dry air by condensing the moisture on the surface of the condenser where the cold water circulates or on the surface of the cooling system in the refrigerator. After drying, the beechnuts must be cleaned again.

9.1.3 Storage of Beechnuts, Dormancy Breaking and Germination

There is a distinction between short-term storage over one winter (until spring sowing) and long-term storage due to irregular periodicity of fructification (up to five or six years).

Short-term storage can take place with or without medium/substrate. The choice of method depends on how the dormant beechnut is removed before sowing. Traditional storage methods are based on maintaining humidity above 25% during the storage period, possibly with the help of water sprinkling nozzles and by including a cold period (but without freezing). Thus, beechnuts (without substrate) can be spread regularly on bare forest floor and covered with a layer of straw or leaves, and later with a blanket of snow. The gradual melting of snow causes imbibition of the seeds, which is necessary to break dormancy.

Beechnuts can also be stored in a cold pit, but in this case a large amount of water must be added three weeks before sowing and then the beechnuts must be stirred daily until sowing. Sowing follows when the seed root exceeds 1–2 mm in length.

Beechnuts that desiccate below 20–25% can also be stored in a dry substrate (vermiculite or sand). The mixture is then stored in a refrigerator (3–4 °C) and the substrate is moistened several times before sowing to break dormancy. In addition, in France, beechnuts are stored at 12% moisture in hermetically sealed containers at 3 °C, followed by pre-treatment with or without substrate.

It is difficult to determine the degree of dormancy and therefore the pre-treatment time for all these processes, as a lower dormancy develops during the storage period, especially when the moisture content is above 20%. It is therefore advisable to use the same method for short-term storage as for long-term storage, as this makes it easier to determine the degree of dormancy and the pre-treatment time of the seed.

Long-term storage is necessary due to the irregular beechnut production. Successful methods have been developed in a collaboration between France and Poland, including storage at elevated $C0_2$ concentrations.

Determination of seed dormancy (X) for beech: On a sample of 25 seeds (for each individual lot), the dormancy rate (X) is determined by a germination test on 10% vital seed (seed root development) at 3 °C (on filter paper or wet vermiculite in a cold chamber) at a humidity of 30-32% (never up to 34%!); the vitality of the seed is determined by a TTC test. The X time can vary from four to a maximum of 20 weeks, depending on the seed lot (provenance, year of harvest, method and time of harvesting, etc.).

Cold stratification of beechnuts

A) Cold stratification of the seed in the medium:
- the seed is treated dry with a fungicide (against the fungus Rhizoctonia solani J.G. Kühn);
- then it is fully moistened by contact with a moist medium (not in excessive wetness);
- then it is stored in a moist medium at 3 °C until the seed root appears in 10% of all vital seeds; depending on the X, this period may last from 1 to 3 months; the time of stratification cannot be extended, although it may last for approx. 50 days for all the seeds to germinate.

B) Removing dormancy without medium by pre-cooling:
- the seed is moistened to 30-32% (never 34%!) at 3 °C;
- followed by a fungicide treatment (wet);
- Then it is stored at 3 °C at a humidity of 30-32% (never 34%!) for X + 2 (or X + 4) weeks; the X time depends on the degree of dormancy of the seeds and is important for further storage (the X time can vary from 4 to a maximum of 20 weeks, depending on the seed lot); a constant time of 6-10 weeks can also be used;
- since the humidity level is sufficient to break dormancy but not to germinate, it is possible to completely break dormancy of all seeds before germination in the tree nursery or when conducting a germination test.

In each case, the seed is placed in small covered trays on which the date of the start of the process and the estimated date of completion of the process are written. The trays are placed in a cold chamber and periodically checked for moisture and infection (once every one to two weeks, at least once a week at the end of stratification). In case of infections, the infected seed should be removed and the remaining seed washed and placed in a new medium.

Seed quality analyses: Seed testing is carried out according to the general ISTA protocols. The size of the sample taken is 1000 g and the size of the working sample is 500 g. Due to dormancy, the vitality test usually replaces the germination test. Analyses of cut seed can be used, with the TTC test being the most common one, while radiography gives less information about the seed. After dormancy has broken, tests of emergence of germlings are used in addition to germination tests to provide better information on the germination capacity of the seed lot.

Sowing in the tree nursery: For forestry planting, 40-80 cm tall seedlings are used, which require 3-5 years of cultivation in a tree nursery (1 + 2 or 1 + 3). In France and Poland, non-dormant seed is usually sown at the end of April or beginning of May, 5 to 7.5 kg of seed per 100 m^2 bed in rows, whereby they cultivate 8,000 to 32,000 seedlings per 100 m^2. The covering layer is 2-3 cm thick. Fungi of the genera *Phytophthora, Pythium* and *Rhizoctonia* usually need to be suppressed. By sowing in tunnels, it is possible to cultivate usable seedlings in just one year. In any case, the soil quality needs to be adequate to allow good root development.

THE SOURCE OF THE ILLUSTRATIONS:
Manual for Forest Genetic Monitoring
Bajc, M., Aravanopoulos, F., Westergren, M., Fussi, B., Kavaliauskas, D., Alizoti, P., … Kraigher, H. (ed.). 2020. Priročnik za gozdni genetski monitoring. / Manual for Forest Genetic Monitoring. Ljubljana: Slovenian Forestry Institute, Silva Slovenica Publishing House. http://doi.org/10.20315/SFS.168

9.2 Oaks: Pedunculate Oak (*Quercus robur* L.) and Sessile Oak (*Quercus petraea* Liebl.)

Figure 16: Oaks: habitus of pedunculate (left) and sessile oak (right) (drawing by Eva Margon)

In Slovenian forests, oaks account for 8% of the growing stock. Six or seven species occur naturally: *Q. robur* L. (7% of the total oak timber stock), *Q. petraea* (Matt.) Liebl. (82 %), *Q. pubescens* Willd. (2%), *Q. cerris* L. (8%), individual *Q. ilex* L. and *Q. crenata* Lam., and there are also records of *Q. virgiliana,* which later turned out to be a hybrid of downy oak with sessile and peduniculate oaks (Jerše and Batič in Gozd V 2007). The introduced species are *Q. rubra* L. and *Q. palustris.* The annual harvest of all oaks is in the order of magnitude of 90.000 m^3.

Lowland oak forests are among the most altered forest ecosystems in Slovenia. Most of them have been converted into farmland or built up. The oaks' plot is thus highly fragmented, with small, isolated populations, often under the constant influence of pollution and changes in groundwater levels. The largest complex of pedunculate oak forests is in the area of the intermittently flooded Krakov Forest, which covers about 3,000 ha. Sessile oak also thrives on more arid sites, mostly up to 700 m above sea level, occasionally above 1000 m, but this should also be checked due to possible mistakes with pedunculate oak done by inventory-takers.

The group of systematically related oak species – pedunculate oak, sessile oak and downy oak – in Slovenia is characterised by high genetic and morphological diversity, frequent interspecific hybridisation with introgression of genes from one species into another, and undefined ecological niches of individual oak species. The source of species diversity in oaks is their genetic system. In evolutionary terms, the development of the oak trees studied today is at different stages. In some environments, the differentiation of pedunculate, sessile and downy oaks is complete, resulting in separate and morphologically distinct populations of all three species. Elsewhere, especially on intermediate sites between oak species optima, the differentiation process is not complete, resulting in a multitude of intermediate, hybrid oak ecotypes. The occurrence of hybrids limits the accuracy of the forest inventory data, so that a knowledgeable forester can also record the thriving of pedunculate oak at high altitudes and on hill stands (above 1000 m), and hybrids and single pure specimens of sessile oak can be observed on transitional sites in the Krakov Forest.

The annual number of planted oak seedlings is 150,000 for the two major species (about 9% of the total), and the quantity of acorns varies from about 300 kg to several tonnes per year for each species. Around 100 to 300 kg of Turkey oak and downy oak seeds have been collected and sown to restore the Karst burned areas.

Figure 17: Oak: leaves and fruits of the pedunculate oak (drawing by Eva Margon)

Pedunculate and sessile oaks reach reproductive maturity in the stand around the age of 50-60, and as solitary trees at around 30-40 years. The optimum age for producing quality seed is between 100 and 220 years. The oaks are monoecious and pollination is by wind. They blossom in May and June. Of particular interest in the case of pedunculate oak are the late-flushing individuals and populations, which are of interest from the point of view of resistance to spring frost and pests. Flowering can be relatively regular every few years, but seed production is highly dependent on weather conditions, so a good fructification is not common.

The acorn grows to its full size by the end of the autumn. The first acorns start to drop in the summer in case of drought, and in August and September there is usually a high proportion of empty seeds and seeds that become home to pests. Acorns of both species usually reach physiological maturity and adequate quality in October. The following table provides information on acorns.

Figure 18: Oak: leaves and fruits of the sessile oak (drawing by Eva Margon)

Table 9: Characteristics of pedunculate and sessile oak acorns (1 - taken from Suszka, Muller, Bonnet-Masimbert 1996)

	Pedunculate oak[1] (France, Poland)	Pedunculate oak (Krakov Forest)	Sessile oak[1] (France, Poland)
Number of acorns per kg	145–500 (avg. 250)		130–650 (avg. 375)
Weight of 1,000 acorns	2–7 kg (avg. 4 kg)	4,6–7 kg (avg. 6 kg)	1,5–5 kg (avg. 3.1 kg)
Weight of pure acorns per hectolitre	60–80 kg		50–80 kg
Acorn mass per tree (120 years, crown with 120 m^2, strong mast production)		~ 20 kg	
Acorn weight per ha (good yield)		1–2 t/ha	

Because of the natural cross-fertilisation between the pedunculate and the sessile oak, it is advisable to carry out morphometric analyses of the foliage in the litterfall. By multivariate analysis of at least five leaf characteristics, it is possible to determine the taxonomic affiliation of each tree with relative precision, and the population mean. In principle, only a pure stand of one species may be approved as a selected stand.

Acorns are harvested by hand from the ground in October. An individual worker can collect between 20 and 50 kg per day. Nets can be spread in the stand before harvesting, or they can try to find the caches of squirrels that collect the top-quality acorn. Acorns are sensitive to impact shocks and care must be taken when collecting and handling them. When harvested, the seed is mostly 90% pure. It is stored and transported in thin layers and jute bags.

The seed is not capable of drying (recalcitrant) and its moisture content must not fall below 40-42%. Because of the high physiological activity, the temperature of the seed can rise rapidly during storage and transport, while the humidity drops. The acorn often carries spores of the pathogenic fungus *Ciboria batschiana* Buchw., which mummify it. For this reason, the seed is usually sprayed with benomyl and similar preparations (0,4 g/kg acorn) and, in larger trees, thermotherapy (two to two-and-a-half hours in water at a temperature of exactly 41 °C) is also applied before storage. Seed is difficult to store, and usually remains vital at high humidity (40-45%) at around -1 °C for only one winter or up to a maximum of 18 months (up to three winters in small quantities in the laboratory). Several research groups in Europe are working on developing a method to store acorns for longer, but so far without success. Acorns are not dormant and can sprout in autumn. Postmaturation and precalcification of the acorn are part of the research on the development of a method to store young seedlings or isolated embryos.

Seed quality analyses: The sample size is 2.5 kg for the test sample, 1 kg for the working sample, including approx. 500 acorns For germination tests, 1/3 of the acorn is cut off and the testa removed using the ISTA method.

Tree nurseries in France produce around 7,000 seedlings per 100 m^2 bed from around 35 kg of acorns. The sowing density in France is around 25 acorns per linear metre. In Poland, about 55 kg of acorns are sown in rows in the same area in the nursery, and 250-300 kg are sown undistributed in the bed. It is covered with a 3-5 cm thick layer of soil or other material. The germination rate in the tree nursery is lower than in the tests: the normal germination rate for acorns of the 44% humidity period is up to 100%, with 37% humidity it is 89%, while acorns from the Krakov Forest germinated at 82% at 37% humidity. The average germination rate in an open nursery in Poland is around 50% for pedunculate oak and 70% for sessile oak.

Figure 19: Oaks: stages of male flowering (drawing by Eva Margon)

9.3 European ash (*Fraxinus excelsior* L.)

The European ash is a widespread species that thrives from lowlands to altitudes above 1000 m. It thrives in approximately 120,000 ha of forests, mostly in small groups or individually. For reforestation by planting and seeding until the appearance of the ash canker (*Chalara fraxinea*, *Hymenoscyphus fraxineus* (Baral et al. 2014), *Chalara fraxinea* (Kowalski et al. 2006, *Hymenoscyphus pseudoalbidus* (Queloz et al. 2011) an average of 200-400 kg of seed was collected annually and around 100,000 seedlings were planted, or 5% of the total number of seedlings to be planted during regular cultivation work. After 2010, due to the recommendations of phytopathologists, ash seedlings are practically not used in tree nurseries and for planting in forests, although it would be appropriate for the conservation of the species to systematically propagate more disease-resistant specimens and to support the increased planting of European ash seedlings with financial incentives, as this would be the only way to conserve this species (and the related species) in our forests.

The biology of flowering is complex, as usually one of the two sexes of flowers on a tree dies off, resulting in male or female trees, which may also differ in growth phenotype. The ash reaches reproductive maturity at the age between 30 and 40 years. It flowers regularly every two to three years.

Figure 20: European ash: habitus (drawing by Metka Kladnik)

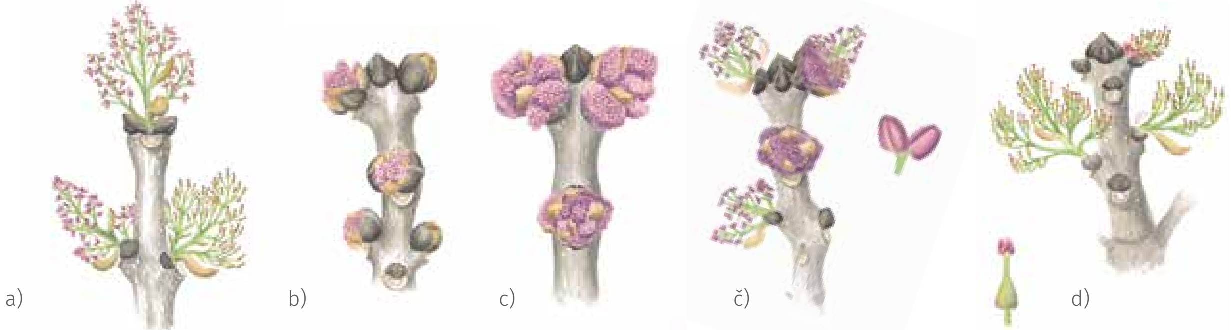

Figure 21: European ash: stages of flowering, female and male flowers. a) bisexual (top), male (left), female (right), b, c, d, e) male flowers, d) female flowers (drawing: Metka Kladnik).

The samaras, which are attached in clusters, gradually develop until the end of September or October. They are still green in August, then gradually dessicate on the tree and turn brown. Samaras are about 4 cm long and 6-8 mm wide, each containing a single seed. The embryo develops in the middle of the endosperm to about half its final size at seed maturity.

Characteristics of the large ash samaras: the weight of 1,000 samaras is 65-100 g, the number of samaras per kg is 10,500-15,500 and 1 hectolitre of samaras weighs up to 15-20 kg.

The harvesting of the mature seed usually takes place in October and November, while the harvesting of the green seed takes place at the end of August. Green samaras cannot be stored, they are immediately used in sowing. Ash needs a few months of warm stratification followed by a few months of cool stratification. Samaras sown while they are still green have adequate time to produce embryos before the onset of the cold season. The timing of the first warm period is very critical; if it is too short, the success of sowing green samaras is poor, the seed will survive and germinate only in the second spring. It is therefore better to collect brown seeds. Seeds collected in October and November should be stored in the tree nursery and sown after pre-treatment in the second spring after collection.

During harvesting, the samaras can have a humidity of around 50-60%, therefore they must be dried at a temperature below 20 °C in a thin layer to a humidity of 8-10%. The seeds can be dried and can be stored for a long time at -3 to -5 °C for up to 10 years. Shorter storage is possible in a ventilated, shaded shed in jute sacks.

Breaking of dormancy: The complex form of dormancy requires both warm and cold stratification, in or out of the medium.

In the media: The seeds are completely moistened in contact with the medium (peat:sand in a 1:1 ratio); if necessary, they may be treated with a fungicide (in France, copper oxyquinoleate); the moisture content of the medium has to be checked weekly.

No medium: The seed is gradually moistened by spraying to 55-60% moisture (at this moisture dormancy is terminated and the seed has not yet started to germinate). It is then treated with a fungicide and stored in a plastic container which must not be hermetically sealed (limited air circulation must be permitted). The humidity level is determined weekly by weighing.

Procedure: First, the seed is stratified in a warm place for 6-16 weeks (usually 16 weeks) at 15 °C or 20 °C (embryo growth time in the seed until it fills 80-90% of the seed length; it can be inspected by in longitudinal section under a magnifying glass), then 16 weeks at 3 °C (removal of embryo dormancy).

Seed storage: After stratification, the seed can either be:

- immediately sown (or germination is tested by the ISTA method or according to the French method at 5-15 °C (14 h + 10 h) and the Polish method at 3-20 °C (16 h + 8 h) for 8 weeks on filter paper;
- stored damp, non-dormant at -3 °C for up to 8 weeks;
- gradually dessicated at around 20 °C to 8-10% moisture and stored non-dormant for two years or more at a temperature of -3 °C to -5 °C.

Seed quality analyses: The ISTA test sample weighs 400 g, the working sample 200 g. For dormancy reasons, only vitality tests shall be carried out:

- in longitudinal section after soaking in water for 18 hours; used for % of full, empty, parasitised and dead seeds, usually for a quick test before harvesting; vitality is indicated by a white embryo in a white endosperm, but must be verified with vitality staining methods;
- TTC: the seed is removed from the samara and soaked in water for 18 hours; it is then cut 0.5 mm along both sides and soaked again for 8 to 24 hours at a temperature of 30 °C in the dark in 1% TTC at pH 6.5-7.5 (fresh seed is soaked for only 8-10 hours); after rinsing in water, the endosperm is cut in half and the embryo is examined, which must be completely stained; only minor necrosis in the endosperm far from the embryo is allowed;
- indigo-carmine and X-radiography are less frequently used for ash.

In the tree nursery, green samaras are often sown in late summer, but success rate can vary widely. It is recommended to sow the stored seed from March to April after using various dormancy-breaking methods. It is important that the warm period lasts about 16 weeks. Cover the seed with 1-2 cm of sand. In Poland, 2-3 kg of samaras are sown in rows in 100 m^2 beds, producing about 8,000 seedlings or up to 30,000 seedlings per bed. The sowing rate of vital seed per m^2 is 40-60, with a yield of 30-50 seedlings. 160-200 vital seeds are sown in plastic tunnels for cultivating up to 80 seedlings.

Figure 22: European ash: leaves and seeds (samaras) (drawing by Metka Kladnik)

9.4 Norway Maple (*Acer platanoides* L.)

Norway maple is widespread at different altitudes, growing singly or in small groups. It is rarely planted in forestry, so it is shown mainly for comparison between orthodox and recalcitrant seed of the same genus.

It reaches reproductive age at 40 years in the stand and 20 years outdoors. It blossoms in April and May; its flowers are bisexual, pollinated by bees. The seeds consist of two samaras, which are grown at an obtuse angle of more than 90°. The two samaras separate before they are dispersed. The seed is brown at maturity, has no endosperm, and the embryo is completely green even when dormant. The seeds ripen in September and the samaras are dispersed in October and November by the wind. The seed year is either every year or at least every two to three years.

Characteristics of Norway maple samaras: weight per 1000 samaras is 100-400 g (avg. 140 g), number of samaras per kg is 2,800-10,300 (avg. 6300).

Seed vitality is usually in the order of magnitude of 75%. Harvesting takes place in September, before the samaras turn brown. After this time, they are easily dispersed by the wind. A single tree can produce up to 15 kg of samaras in a seed year. The weight of seeds that a worker can collect per day from a single tree is 2.5-5 kg. In the stand, harvesting is much more tedious.

<u>Drying:</u> Green seed (early October) usually has a moisture content of 49-59%; brown seed (late October/early November) 32-41%. The seeds are dried at a temperature of 17-19 °C in a dry place for about one week to reduce the humidity from an average of 35% to 11.5%. After the second week of drying, it drops to 8%.

Storage: The samaras of Norway maples are rarely stored because the seed years are quite frequent. If stored, they are kept at a humidity of 8-10%, in closed-off containers, at a temperature of -3 to -10 °C. The seeds are resistant to low temperatures even at high humidity.

Removing dormancy: Cold stratification at a temperature 1 or 3 °C for 6-14 weeks in a moist medium is used; in this case, the seed germinates best after the end of stratification, also at a temperature of 3 °C. Cold-warm stratification and cold-warm germination are also possible, but in this case germination may be greatly reduced with minimal irregularity; freezing the seed at a temperature of -3 °C for 0-20 weeks prior to germination is also recommended, with the temperatures gradually rising from 1-3 °C, 1-5 °C, 1-10 °C, 1-15 °C, 1-20 °C to a constant temperature of 20 °C.

Figure 23: Norway maple: seed (drawing by Teja Milavec)

<u>Vitality test:</u>

1. By cutting (only for fresh seed material):
   1) the seed is removed from the coat;
   2) empty and damaged seeds are counted;
   3) the rest of the seeds are cut; the problem is green cotyledons, which can be green even if the seed is dead;
   4) a live seed is more likely to be one whose cut surface glistens, the cotyledons are imbibed and the seed is not damaged in the slightest;
   5) if this test is carried out on dry seed or seed that has been stored for a longer period of time, the results can only be expressed as % of full seeds.
2. TTC:
   1) 4 × 50 or 4 × 100 seeds are soaked in water for 18 hours;
   2) the pericarp is removed;
   3) a small part of the seed coat is removed and the seed is once again soaked in water for a few hours until it is completely imbibed;

   1) up to 3): dry seed or seed that was stored for a longer period should instead be cold stratified at a temperature between 3 and 5 °C for 10-14 days;

4) the naked embryos are then soaked for 5-8 hours at a temperature of 30 °C in a 1% TTC solution in phosphate buffer (pH 6.5-7.5);
5) vital seed is seed that is fully coloured or where the radicle is not coloured or where there is minor necrosis on the distal part of the cotyledons;
6) non-vital seeds are those with visible necrosis near the embryonic axis.

Germination test according to the ISTA method (1996):

- cold stratification (freezing) in a moist medium (sand and fine peat) at a temperature between 1 and 5 °C for two months (seed wings are cut off); conduct weekly inspection;
- then germination test on sand or paper for 21 days at a temperature of 20 °C.

In the tree nursery: The seedlings of Norway maple can be sown immediately after harvesting (between 15 September and 15 October) in the tree nursery, or after short-term storage of partially dried seeds, or after drying and short- or long-term storage at low controlled temperatures. After sowing, they are covered with 2-3 cm of soil or a mixture of sawdust and sand. Sow 4 kg of samaras in rows per 100 m^2 bed or twice that amount over the entire bed. Germination is usually in the range of 50-90%. This bed can grow 10,000-30,000 seedlings.

Samaras can also be stored in the forest over winter, covered with leaves or in pits, mixed with sand. However, this can lead to premature germination in mild and wet winters. If the samaras are dried, they must be stratified in a cool place from May to autumn, the humidity of the medium is increased at the end of autumn, and the seed is sown in spring of the following year. Seeds often start to germinate before sowing and even a drop in temperature below 0 °C does not stop this process.

Storing the samaras at approximately 10% humidity in closed containers at a temperature of –3 to –10 °C is recommended. After thawing, they may be stratified at a temperature of +3 °C for a period to be determined on the basis of a control sample in stratification. The sowing time can be calculated from the information in the control sample. In this process, premature germination can be stopped by lowering the temperature to -3 °C for up to 8 weeks. All stages of this seed processing are carried out under controlled conditions, independent of external factors.

9.5 Sycamore (*Acer pseudoplatanus* L.)

Sycamore is a widespread species. It grows at all altitudes singly or in groups in mixed stands. In Slovenia, it is most common on carbonate soils, although it also thrives elsewhere, especially in the altitudinal zone from 400 to 1000 m.

It blossoms from April to May, before and during leaf-out. The flowers form clustered inflorescences that can be 13-20 cm long. The flowers are bisexual, but usually either the stamens or the pistils deteriorate, so that they can only function as male or female flowers in different proportions of the two sexes. In the inflorescence, the basal and central flowers are usually female and the terminal flowers male. Monoecious inflorescences are rare, as are monoecious trees. Flowers of different sexes mature at different times, with a flowering period of 7-15 days. Pollination is mainly by insects.

The fruits are clustered in clusters up to 29 cm long, with the number of fruits per cluster varying from a few to more than 30. Each fruit consists of two samaras, grown at an angle of 60-90 °C. The samaras are 36-55 mm long and each contains one pea-sized seed without endosperm. Samaras ripen in autumn, when the wings turn light brown exhibiting darker veins.

Seeding of sycamore occurs regularly every two to three years, it is abundant, from the age of 20 in the outdoors and from the age of 40 if grown in a stand.

Characteristics of sycamore samaras: weight per 1000 winged samaras is 66-180 g (average 120 g), number of winged samaras per kg is 5,500-16,000 (average 11,000) samaras.

Collecting: On dispersal, the two samaras separate into independent fliers. The wind can blow all the samaras off a single tree in a single day.

Harvesting takes place before the windy season, from September to mid-October, when the samaras change colour from green to yellow. They need to be sown or stratified immediately and may germinate prematurely. Seeds can be collected on the ground or from the tree, by shaking or climbing, or from felled trees.

The leaves and stems of the clusters are removed by preliminary cleaning in the field. They are transported in bags with large openings to prevent the development of mould. The final cleaning is done by sieving or blowing. The wings may remain on the seed.

Drying: The moisture content of samaras during harvesting is 42-55% of the fresh weight. Sycamore seed cannot be dried (recalcitrant) and the moisture content must not fall below 24% (for whole samaras). When storing for one winter, drying is not necessary, but for longer periods, the seed with wings is dried to 24-32% moisture or the pure seed to 30-42% moisture with air circulation at a temperature between 18 and 20 °C. Drying under natural conditions is difficult in October and November because of the fluffy layer that insulates the pericarp seed.

Storage: The samaras can be stored for one winter in moist peat at a temperature of 3 °C and 40-50% seed and air humidity. If the seed starts to germinate, it is stored in peat at a temperature of -3 °C.

For longer storage, the seed is dried to 24-32% moisture, stored in closed containers at a temperature of 0 °C for one day, then at a temperature between -3 and -5 °C for the entire storage period.

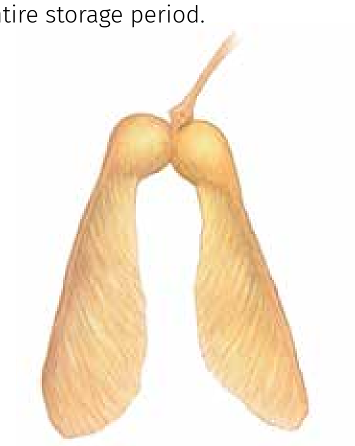

Figure 24: Sycamore: seed (drawing by Teja Milavec)

Removing dormancy:

- In the medium (5-14 weeks, exceptionally up to 20 weeks):
  - ◊ after fungicide treatment, the seed is moistened in a moist medium (peat, sand or a 1:1 mixture);
  - ◊ then stored at a temperature of +3 °C until 10% of the seeds have germinated (usually for about 14 weeks); such seeds can be sown immediately or stored at a temperature of -3 °C until germination;
  - ◊ at a temperature of +3 °C germination is very slow (8-12 weeks).
- No medium:
  - ◊ the seed is gradually moistened to 44-50% (whole seed) or 50-58% (without wings);
  - ◊ this is followed by fungicide treatment;
  - ◊ the seed is then stratified in a thin layer at a temperature of +3 °C in plastic containers, lightly covered with plastic film (not sealed); weekly humidity inspection;
  - ◊ the time for stratification is determined by the dormancy rate (X time – same as for beech);
  - ◊ after stratification without medium, the seed can be stored for two winters at a humidity of 24–32% at a temperature of -5 °C in closed containers.

Vitality test:

- By cutting (3 × 50 or 3 × 100 seeds); the problem is green cotyledons, which can be green even if the seed is dead; the result can only be expressed as % of full, empty and parasitised seeds.
- TTC:
  1) the seed is cold stratified between temperatures of 3 and 5 °C for 10-14 days;
  2) the pericarp and part of the seed are removed;
  3) the seed is then once again soaked in water for a few hours;
  4) the naked embryos are then soaked for 5-8 hours at a temperature of 30 °C in a 1% TTC solution in phosphate buffer (pH 6.5-7.5);
  5) vital seed is that which is fully coloured or in which the radicle is not coloured or there are minor necroses
  6) at the distal part of the cotyledons;
  7) non-vital seeds are those with visible necrosis near the embryonic axis.

Germination test according to the ISTA method (1996):

- cold stratification (freezing) in a moist medium (sand and fine peat) at a temperature between 1 and 5 °C for two

months (seed wings are cut off); conduct weekly inspection;
- then germination test on sand or paper for 21 days at a temperature of 20 °C.

Improvements of the germination analysis:
- after cold stratification (7-20 weeks), it is better to carry out the germination test at a temperature of +3 °C;
- after cold stratification, germination can also be determined by altering the temperature during the germination test at temperatures between 5 and 15 °C (3-4 weeks is sufficient for complete germination of the seed);
- the emergence test conducted in the tree nursery: 4 x 50 seeds pre-treated in medium until germination; the wings are then cut off and the seed is planted in containers with moist medium at temperatures between 3 and 20 °C (16 h + 8 h day) for 12 weeks (emergence is observed on a weekly basis).

Sowing in the tree nursery: Sowing fresh seeds can be done immediately after harvesting in autumn. Stratification is necessary when sowing stored seeds.

In Poland, 3.5 kg of samaras are sown per 100 m^2 bed in rows or 8-10 kg over the entire area. This can produce 10,000-30,000 seedlings. The sowing density is 160-300 samaras per m. The crop needs to be protected from birds and rodents, sometimes against frost, and should usually be kept shaded. In France, seedlings are grown in plastic tunnels on fertilised peat for a year. The sowing density is about 160 samaras per m^2, with an average sowing success rate of 50%, which results in about 80 seedlings per m^2. In Poland, the figures are twice as high.

9.6 Black Alder (*Alnus glutinosa* (L.) Gaertn.)

Black alder grows in Slovenia mainly in the lowlands and up to 700 m, but it rarely occurs at an altitude above 1000 m. They are more abundant on carbonate soils, especially along watercourses, and on wet soils.

The flower buds burst in the first year, followed by flowering, pollination, fertilisation and fruit ripening in the following year. Plants are monoecious. They bloom before leaf-out, between February and April. The male flowers hang from the ends of the branches like catkins, and the female flowers are clustered in two to three cones that develop at the base of the twigs, where they grow alongside the previous year's cones. The new strobili are characterised by a distinct stalk, 1.5-2.5 cm long. Pollination is by wind, in spring, but fertilisation only occurs in summer.

The seeds ripen in "cones" in September or October. The strobili become woody and can remain on the tree for a long time. They open after the first hard frost of winter, and the seeds are dispersed until spring. The fruit is a winged nut containing a seed without endosperm. Seed germination is highest when harvested in October, but declines when harvested later.

Trees in stands start to set seed at 30 years, in the open at 15. They can go to seed every year, but only every two to three years will they produce a strong crop. Seed set is strongly influenced by the weather at flowering time and also a year earlier, when the flower buds are forming.

Figure 25: Black alder: cones (drawing by Teja Milavec)

Black alder seed characteristics: 1000 seeds weigh 0.7-1.5 g, 500,000-780,000 seeds per kg, the weight of pure seed in 1 hectolitre of cones is 1.5 kg, the weight of 1 hectolitre of cones is 30 kg, the weight of seed from 100 kg of cones from a seed plantation in Germany was 9-19 kg.

Mature fresh seed has a moisture content of 8-9%. It can be carried by the wind in a radius of 30-60 m around the tree, but it can also be carried by the water, because its wings allow it to float on the water. It can germinate even after 12 months.

Natural rejuvenation is successful on bare soils or in a humus layer, free of competitive weeds and other vegetation, under favourable moisture and light conditions. Germination is on average around 40%, reduced by broken cone scales and fungal infestations. The cones are harvested in November and December. Around 15-20 kg of cones need to be collected to obtain 1 kg of seed. In a good harvest, one worker can harvest up to six kg of cones. Because of the high humidity, the cones have to be quickly transported to the seed factory for processing.

The fresh cones are placed in a dry and well-ventilated place where they are gradually opened over the next few weeks. Opening is accelerated by processing in a conifer oven at temperatures between 27-38 °C. Drying at 20 °C takes 20-25 days, At 40-45 °C, only two days. The seeds remaining in the cones are extracted by shaking them in a rotating drum. If it is excreted in running water on the screen, storage is not possible.

For cleaning, screens with apertures of 3 mm (for larger impurities) and 1.5 mm (for dust, etc.) are used. But it is difficult to tell the difference between a full and an empty seed. For example, in Germany, the purity was stated as 60% and the germination rate as 40%.

The seeds have a moisture content of 8-9% when they emerge from the cones and can therefore be stored for up to 2-3 years. For long-term storage, it must be dried to a humidity of 5-7% by drying at a temperature of 30 °C for 48 hours and can be stored for five years.

Seeds with a moisture content of around 10% respire intensely at room temperature, even at temperatures between 2 and 4 °C. If the germination % does not drop, the germination energy of the seed drops, indicating ageing. It should therefore be dried as much as possible (the seed is capable of drying, orthodox). Seeds with 7% humidity can be stored for several years at temperatures between -4 and -10 °C. For longer storage, drying at 3% humidity is recommended.

Seeds can germinate immediately in the wild under favourable light and temperature conditions. After drying, however, the germination is greatly reduced and can be influenced by stratifying fresh seed in a nutrient medium at temperatures between 1 and 5 °C for 180 days. Even better, after this stratification, the seed should be frozen at a temperature of -20 °C for 3 days. It is generally agreed that the secondary dormancy of alder can be removed or the germination of dry seed can be improved by stratification in snow, sand or a mixture of sand and peat at temperatures between 1 and 5 °C for 30-60 days.

The weight of the sample taken for quality analyses is 8 g and the weight of the working sample is 4 g. The vitality analysis can be carried out by means of seed X-radiography, which provides information on full, empty, parasitised and damaged seed. Only full seeds can then be selected for germination analysis. The germination test is carried out in a Jacobsen germinator, where the temperature is raised to a temperature of 27 °C once a day for two hours after field protocols and for the remaining day at a temperature of 20 °C. The germination rate and germination energy are calculated after seven and 14 days. ISTA protocols prescribe temperatures between 20 and 30 °C (16 hours + 8 hours day). The first count is after seven days, the last on the 21st day. A germinated seed is a seed in which the seed root has grown to at least half the length of the seed. At each count, the germinated and rotten seeds are removed. Germination is expressed as the average of the germination of four seed samples.

For planting in the forest, seedlings of the 1 + 0 form are usually used. Sowing is usually carried out between March and April. Use 0.5 to 1 kg of seed per 100 m^2. The cover layer is approximately 5 mm thick. Less seed is sown in the rows and double the amount is sown over the entire bed area. This allows them to grow 12,000-35,000 seedlings per bed. However, if the quality of the seed is high, it is possible to grow 20,000 seedlings by sowing only 150-200 g of seed per bed. Germination in the tree is usually low (5-15%). To cultivate quality seedlings, an appropriate sowing density is needed: for 1 + 0 seedlings, a density of 60 seedlings per m^2 is appropriate, while for 2 + 0 seedlings, only 15-20 seedlings per m^2 is appropriate.

Soil moisture is a very important factor for sowing success rate. Sowing alder as soon as the snow melts is recommended. In March and April, the optimum soil moisture is in the top 10-30 cm. The seedlings can also tolerate occasional flooding. At a later point seedlings thrive better in drier soils.

Seedlings can also be grown in pots. They are sown in 7 cm diameter pots and transplanted into 8 litre pots in May. When growing seedlings in pots, inoculation with symbiotic bacteria of the genus Frankia, which fix nitrogen from the air in the root nodules, is important.

Alder seedlings are very susceptible to drought when being transplanted and excavated for planting in the forest, and after just a few hours survival is greatly reduced. Bare roots should be immediately covered with soil. During transport, the root system must be protected against desiccation.

9.7 Wild Cherry (*Prunus avium* L.)

Wild cherry thrives singly or in groups in mixed stands from lowlands to 700 m, rarely occurring at an altitude above 1,000 m. It grows more often on carbonate soils.

The bisexual flowers bloom in April and May. Pollination is with bees. The flowers are very sensitive to frost. Pollination is not always followed by fertilisation due to allele incompatibility. This leads to inter-sterile groups that need to be taken into account in the approval of seed facilities and the layout of seed plantations.

The cherries ripen in June, and at higher altitudes also in July and August. There are two wild forms, the sweet cherry and the black cherry – the wild black cherry. Each fruit contains a single drupe (endocarp) with a small elongated seed covered by a seed sheath covering an endosperm of a single layer of cells thickened around the radicle and the two outer surfaces of the cotyledons. A drupe is a woody endocarp, incorrectly named a seed. At maturity, much of the crop can be plucked by birds. Ripe cherries fall to the ground and can be spread by birds, other animals and humans. The seeds can survive one or more winters under fall and snow. Germination depends on dormancy being established under favourable conditions: high humidity, aeration and a certain sequence of changes in temperature.

Cherry trees can bear in the nursery from the 15th year onwards, and outdoors after six years. Although they flower every year, the crop is irregular because it depends on spring frosts, bad weather for bee pollination, etc. In the early cherry, which ripens within 60 days, the embryo is incompletely developed and does not germinate. Late cherry forms, which ripen in 80 days, usually have a germination rate of around 100%. Characteristics of cherry seeds and fruits: number of pure pits per kg 6,000-8,100 (avg. 6,700); weight per 1000 pits is 125-166 g (avg. 150 g); weight of pits per 100 kg of fruit 12-18 kg.

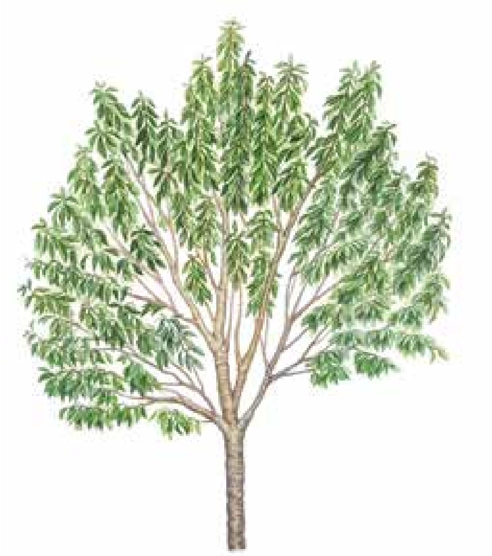

Figure 27: Wild cherry: habitus (drawing by Teja Milavec)

Cherries are picked at maturity, by hand from the tree, by shaking the tree or by gathering them in a net under the tree at the time of fall. The cherries must be protected from fermentation. They are best transported in barrels of water. Small quantities can be pitted by hand or with a home-made destoner. The stones must be rinsed well with water. Larger quantities are purified with rotating macerators and water over sieves. Between the endocarp and the seed there can be a bubble of air, which makes even a full seed float on water. Therefore, the floating stones, which are supposed to be empty, must be broken in a sample to check the quality of the seed.

Figure 26: Wild cherry: flowering stages (drawing by Teja Milavec)

Cherry pits are never sown directly after harvesting. If it is desirable to partially remove dormancy by warm stratification, the seed is sown at the beginning of October. This also gives the seed the necessary cold stratification after the warm period. While waiting for sowing, the seed can be partially dried or prepared for long-term storage.

The cherry is a drying seed and can be dried to a moisture content of 9-10%. After rinsing with water the seed is spread in a thin layer on the ground, stirring occasionally. After the surface has dried, the seed is left to dry at a temperature of 20 °C in a ventilated place for about 10 days. Drying can be accelerated by blowing at the same temperature. The moisture content of the whole stone is not the same as the moisture content of the seed; a moisture content of 9-11% for the stone means a seed moisture content of 6-8%. Storage in the wild is possible for 2-3 years. Dry seeds can be stored at a low temperature of -3 °C for three years or at a temperature of -10 °C for long-term storage. After the end of storage, the seed should be gradually thawed.

Cherry pits have a high degree of dormancy. Only a seed that has had its dormancy completely removed is capable of normal growth. Isolated embryos, for example, grow into physiological dwarfs.

Breaking of dormancy occurs in several varieties, all of them very long. Typically, the seed is stratified:

Two weeks at a temperature of 25 °C, two weeks at a temperature of 3 °C, two weeks at a temperature of 25 °C, 12-16 weeks at a temperature of 3 °C (or until germination). Alternatively, it may be preceded by six weeks at 3 °C or preceded by two weeks at a temperature of 20 °C. Stratification may take place with or without medium.

For seed quality analyses, a sample of 900 g is taken and a working sample of 450 g of stones. Moisture content is determined on the whole stone or separately for the stone and the seed. Viability is determined by cutting, TTC, indigo-carmine or X-radiography. The germination test requires about six months of hot-cold stratification.

Stratified seed is used for sowing in the nursery, as frost, snow or too much standing water can interrupt the dormancy removal process. Non-normal seed germinates 50-70% in the nursery. In Poland, 4 kg of stones are sown per 100 m^2 bed in rows or holes and 10-13 kg per entire bed. This produces 13,000-25,000 seedlings per bed. Under cover, it is possible to grow 80-100 seedlings per m^2, corresponding to 160-200 germinating seeds per m^2.

Figure 28: Wild cherry: fruit development (drawing by Teja Milavec)

9.8 Selected Conifers

9.8.1 European Silver Fir (*Abies alba* Mill.)

The fir tree reaches the reproductive stage at 60 years of age, with full mast production every 4-6 years and partial mast production every 2-3 years. It flowers in May and ripens in September. As the cones decay on the tree, they should be picked by climbing on the trees before ripening in September. Cones are worth harvesting if at least 50% of full seeds are visible in cross-section. A litre of fresh seed usually weighs around 400 g. Fresh seed has a moisture content between 8 and 11%. There are 15-30 seeds in 1 kg of cones and 1 kg of seed in 14,000-23,000 wingless seeds. There are 260-290 seeds in 1 cone. Longer storage is possible at different moisture contents and at different freezer temperatures, preferably between 8 and 10% moisture at temperatures between -10 and -15 °C in hermetically sealed containers. The fir embryo is dormant, therefore dormancy should be removed by cold stratification for 3-7 weeks before sowing.

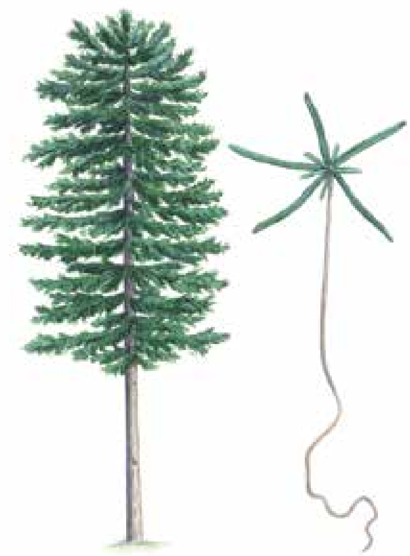

Figure 30: European silver fir: habitus and germination (drawing by Anja Rupar)

Figure 29: European silver fir: development of female (left) and male (right) flowers (drawing by Anja Rupar)

9.8.2 European Larch (*Larix decidua* Mill.)

European larch reaches the reproductive stage at 25-30 years, and in the outdoors at 10-15 years. Mast production occurs every 5-10 years. It flowers from March to May, and the cones ripen from September to November of the same year. The seeds fall from September to spring. Cones are collected from standing or fallen trees from January to May. It is also possible to spread nets on snow under large areas in stands. The cones are dried in the sun and mechanically opened in drums with scrapers when they reach 15% moisture. There are 200,000-270,000 seeds in 1 kg of seed. 100 kg of cones contain 4–7 kg of wingless seeds. 1 kg of winged seeds contains 750-800 g of wingless seeds. There are 220–280 of these in 1 kg of cones. Each cone contains up to 80 seeds. Around 60% of seeds can be empty. Seeds with a moisture content between 6% and 7% can be stored for long periods at temperatures between 0 and 10 °C. Germination is slow due to the dormant embryo. Cold stratification for a few to six weeks is recommended.

Figure 31: European larch: cone (drawing by Teja Milavec)

9.8.3 Norway Spruce (*Picea abies* Karst)

Spruce trees bear fruit from 30 to 50 years of age, optimally at the age of 100 years. Spruce flowers from April to June, the cones mature from October to December and drop whole from October to April. They are harvested by climbing, dried in the sun and decompose when they reach 25% moisture. On average, there are 9.4 kg of winged seeds in 100 kg of cones, 550 g of wingless seeds in 1 kg of winged seeds, there are approximately 32 seeds in 1 kg of cones, and about 400 seeds in a single cone. Freshly harvested cones in October have a moisture content of approximately 30-40%. After sun-drying, the moisture content of the cones was 16% and that of the seeds 10%. Seeds with a lower % moisture retain their germination for longer. In the seed bank of the Slovenian Forest Genebank, seed from the 1965 harvest still had 70% germination in 1995.

Figure 32: Spruce: cone (drawing by Teja Milavec)

9.8.4 Pines (*Pinus* spp.)

Black pines bear fruit from the age of 30, Scots pines 10 to 20 years earlier. The seed year is every 3-4 years for black pine, every 3-10 years for Scots pine. It flowers from May to June, with cones maturing from September to October for black pine and from November to December for Scots pine, in both cases the following year. The seeds fall from March to April in the third year. Cones are collected by climbing trees. There are 140,000 seeds per kg of cones for black pine, and 175,000 seeds per kg for Scots pine. Black pine has 2-4 kg of pure seed in 100 kg of cones, 800 g of pure seed in 1 kg of winged seeds, and up to 50 seeds in 1 kg of cones. Scots pine has 1-2 kg of pure seed in 100 kg of cones, 700 g of pure seed in 1 kg of winged seeds, and up to 160 kg of cones in 1 kg. Seeds dried at 8% moisture can be stored for longer periods. Scots pine seeds do not need stratification before sowing. Black pine seeds should be stratified for 30-60 days in a moist medium at a temperature of +5 °C.

Figure 33: Black pine: habitus (drawing by Klara Jager)

Figure 34: Black pine: a winged and wingless seed (left), mature open cone (centre) and branch with male strobili and female immature first-year cones (right) (drawing: Klara Jager)

9.8.5 Douglas Fir (*Pseudotsuga menziesii* (Mirb.) Franco)

Of the seven species of the genus *Pseudotsuga* Carr. the most widespread is the Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco), which occurs naturally from western northern Mexico (19° N, 97° N) to eastern British Columbia (55° N, 128° N) (Stein and Owston 2008). Fossil remains have been found in North America from the Early Tertiary onwards, and later also in Japan and Europe (ibid.). Because of its commercial use, it is planted outside its current area, in Europe, Chile, New Zealand and Australia.

Two varieties have been described, i.e. coastal (var. *menziesii* or *viridis*) and continental (var. *glauca*), which cross-breed in the interior of British Columbia. It thrives at different altitudes (275-3260 m above sea level) and in different growing conditions, which is why its range in North America has been divided into six seed zones, which replace the regions of provenance. Clinical genetic variability was found for several traits in coastal and continental variants of Douglas fir. Traits include survival, growth, growth form, phenology, disease and pest resistance, low-temperature resistance, technological characteristics and chemical structure of the wood, and breeding programmes were initiated 60 years ago (ibid.). For both varieties, drought tolerance increases from north to south and from west to east (Westergren et al. 2018).

It was first planted in Europe in the 19th century, with most plantations in France, the UK and Germany (http://www.euforgen.org/species/pseudotsuga-menziesii/). In southern Europe, coastal provenances from Oregon have performed better, while provenances from the transition zone between the two varieties, sometimes referred to as var. *caesia,* grow best at higher altitudes (Westergren et al. 2018). In Slovenia, the largest concentrated areas are in the Inner Carniola region, which also has the largest approved seed stand for the production of forest reproduction material in the »selected« category. Natural rejuvenation of Duglasia has been recorded in Slovenia at several locations, in the Inner Carniola, Kočevje, Novo Mesto, Celje and Pohorje regions. On the basis of molecular genetic analysis of 215 trees from seven stands and a (as yet unapproved) seed plantation in the Novo Mesto region, it was found that the overall genetic diversity in Slovenia is lower than in the natural range, and the heterozygosity is similar, while the latter in German and Austrian stands is also mostly lower than in the natural range. Therefore, in line with recommendations from Central Europe, we recommend collecting seed from at least 20 Douglas fir trees; and in the seed plantation, once approved, collecting seed only if all clones have flowered. On the basis of the majority of trees of one group or the other and the likely epigenetic effects of external conditions on the physiology of seeds and young, we recommend dividing Slovenia into two provenance areas for Douglas fir: provenance area 1 should contain the Pohorje and Alpine regions, and provenance area 2 should contain the remaining ecological regions (ibid.).

Douglas fir can grow to more than 100 m in height, 4 m in diameter and live for 1300 years. It starts to blossom at 7 to 10 (var. *menziesii*) or 20 years (var. *glauca*). The time between high mast production is between 2 and 11 years. The male and female cones start to sprout in late winter or at a young age, about a year after the buds are conceived. Flowering takes place from March to June, cones ripen from late July to early September, and seed is released (by gravity and wind) from August to March, and mostly in September and October (USDA 2008).

The male "cones" cover most of the crown at the proximal part of the annual branches, drooping at about 2 cm; they are yellow to dark red in colour. Female cones develop more distally on annual shoots, mainly in the upper part of the crown. They are still erect at the time of pollen release, about 3 cm long, and vary in colour from dark green to dark red. The cones on each tree are the same colour, but the colours of the male and female strobiles differ. Pollination takes place when at least half of the cone has grown from the bud scales and can take 6 to 10 days. Fertilisation occurs about 10 weeks later (Allen and Owens 1972, in USDA 2008). Mature cones are characterised by triple bractlets visible outside the cone scales. There are two seeds on each. The number of full seeds per cone varies greatly. For var. *glauca,* about 20 to 30 per cone from the same location, and for var. *menziesii,* 4 to 54 per cone (Olson and Silen 1975, in USDA 2008). The average production of pure seed in a stand is about 0.45 kg per tree. The abundance of the mast production can only be confirmed two months before seed dispersal. In addition to the periodicity of flowering, the causes that affect it are poor pollination, spring frosts, cone dieback, insects and other pests on the cones (at least nine common species in S. America) and other factors (Owens et al. 1991, in USDA 2008). However, the potential for fructification can be estimated as early as 12 months in advance from the number of female buds or 17 months in advance from the number of male buds.

Figure 35: Douglas fir: twig and cone (drawing by Teja Milavec)

The cones start to be harvested 3-4 weeks before seed set, from August onwards. Maturity is best assessed by cross-sectioning the cone – the seed should be light to dark brown, the wings light brown, the embryo should fill most of the volume and be yellow-green. The economic viability of seed production is assessed by counting the number of full seeds – at least 5 per cross-section is recommended; to estimate the number of seeds per cone, the number of seeds per longitudinal cross-section is multiplied by a factor of 4.5 (USDA 2008). Proper storage of the cones in a dry and well-ventilated place at temperatures between 7 and 10 °C for 2-4 months can contribute to seed quality. Cones open at a loss of 35-51% of wet weight. Drying takes place from 4 to 60 days in the air or from 2 to 48 days in an oven at 32-43 °C. When extracting, it must be borne in mind that higher temperatures and rough handling (robust extraction machines) are very damaging to the seed. The seeds are stored at a humidity of 5-9% (on a wet weight basis). High germination (85-87%) was still observed in coastal Douglas fir after 25 years of storage at -18 °C, but declined within a few years at temperatures between 0 and 5 °C.

In most cases, Douglas-fir seed should be stratified or pre-germinated by treatments that also minimise damage from diseases and pests (e.g. rinsing with cold or hot water, fungicides, bleach, peroxide, ethanol, polyethylene glycol or ethylene (ibid.)). Stratification is mostly seed without medium after imbibition in water for 24 hours for 3-6 weeks at 2-5°C. Germination is often not equal to established viability (TTC); depending on the position of the seed at germination. Seed pre-preparation and stratification time must be determined on a lot-by-lot basis.

In nursery practice, bare-root seedlings of 1 + 0, 3 + 0 and 1 + 1, 1 + 2, 2 + 1, 2 + 2 or one-year container seedlings are used. Nursery practices must take into account measures to preserve the genetic diversity of the seedlings. Stratified seed is mostly sown in late winter or early spring to avoid damage over winter. After imbibition for 24 to 48 hours, it is refrigerated (usually in 2 kg bags) at -5°C for 28 to 60, sometimes up to 90 days. The stratified seed can be stored for later resistance at 2°C for 3 months or dried to a humidity of 7-15% at -7 to +3°C for 9 months or more.

For the production of bare-root seedlings, the seed is sown at a depth of 3-6 mm: for 2 + 0 seedlings, the density of 1-year-old seedlings is 161-323/m^2, and for 1 + 1 seedlings 538-754/m2. The recommended soil pH is 5.0-6.5. Fertilisation is mostly finished in July or late August. Container seedlings can be prepared for planting within one year, the recommended pH of the substrate is 4.5-6. To improve quality, seedlings can be grown in containers in the first year and in a bed in the second year.

Vegetative propagation by cuttings can also be used to increase production. They take root in the first year, are transplanted in autumn and are ready for planting the following year. Cuttings from juvenile wood root better and have less plagiotropic properties than cuttings from older wood. Juvenile wood can be preserved by pruning the parent tree.

Properly woody seedlings can be stored for a considerable period of time at temperatures around zero. Planting takes place mainly from late autumn to spring.

10
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11

TABLE OF ANNEXES

Annex 1: Protocol for the management of forest tree seedlings from the time they are extracted from the nursery to the time they are planted, in cases where seedlings are provided by the Slovenian Forest Service from the Slovenian budget funding

Date: 16 September 2019

Protocol on the management of forest tree seedlings from the time of excavation in the tree nursery to the time of planting in the forest in cases where the seedlings are provided by the Slovenian Forest Service from the budget of the Republic of Slovenia

1. Purpose of the Protocol

In addition to the quality of the seedlings, the success of reforestation through the planting of forest tree seedlings depends to a large extent on how they are handled from the time they are dug up in the nursery to the time they are planted in the forest. Improper handling causes desiccation and drought stress, root rot, pest infestation, moulds and diseases, thus reducing the vitality of the seedlings. The ability of the roots to regenerate (which is even more important than the proportion of fine roots or hairs in the critical period after planting) depends mainly on the water balance of the seedlings.

Forest seedlings should be exposed to dry air, strong winds or direct sunlight for as short a time as possible before planting.

Desiccation can occur:

- during excavation in the nursery;
- in the nursery while waiting for transport;
- during transport;
- after transport is completed, due to late and poor backfilling;
- during planting, due to the seedlings being carried sloppily around the site.

The purpose of this protocol for the management of forest tree seedlings is to ensure that the work in these phases is properly organised to ensure that regeneration by planting is successful.

3. Description of the procedures for each project stage

1ST STAGE	**FROM SEEDLING EXTRACTION IN THE NURSERY TO THE CUSTOMER'S ACCEPTANCE (SFS)**
	As a general rule, the sooner the seedlings are planted in the nursery after digging, the better their chances of survival.
	The nursery or supplier should keep the seedlings in a dark, cool place or cold store until transport to the worksite and ensure that they are properly cared for.
	During transport, the seedlings must be protected from wind and sun to avoid physiological desiccation. Transport is only possible with a protective tarpaulin or in an enclosed means of transport.

2ND STAGE	**CLIENT'S (SFS) COLLECTION OF SEEDLINGS**
	The seedlings are taken over by the purchaser (SFS) and the delivery is checked for quantity, quality, breeding form, origin and the adequacy of the supplier's document as defined in Article 15 of the Forest Reproductive Material Act.
	The quality and quantity control of the growing forms and heights shall take account of the quality requirements of the seedlings as laid down in the supply contracts concluded with the suppliers.
	If the seedlings delivered correspond to the order, the delivery note is validated.

3RD STAGE	**THE FOREST OWNER TAKES OVER THE SEEDLINGS FROM THE SFS**
	The seedlings are taken over by the forest owner or manager, who checks that the quantity and quality by tree species are in accordance with the SFS decision.
	If the quantity and quality of the seedlings comply with the SFS decision, the forest owner or manager signs (acknowledges) the receipt certificate prepared by the SFS. In the case of partial deliveries of seedlings, partial certificates will be issued.

4TH STAGE	**TRANSPORT OF SEEDLINGS TO THE WORK SITE**
	During transport, the seedlings must be protected from wind and sun to avoid physiological desiccation. Transport is only possible with a protective tarpaulin or in an enclosed means of transport.

5TH STAGE	**STORING SEEDLINGS ON A WORKSITE IN THE FOREST**
	Planting must be carried out immediately after delivery to the forest work site, or within 10 days at the latest. This time limit is set by a decision of the SFS.
	If the seedlings are stored on the site for a short period, they must be adequately protected against desiccation. Two methods are known: storage in backfill and storage under metallised, reflective foil or tarpaulins.
	When storing in backfill, the bunches of seedlings should be untied and moist soil (not humus) should be spread over the roots so that they are all in contact with it. For large numbers of seedlings, the trenches have to be dug by machine.
	Watering should keep the soil in the backfill moist at all times, and the seedlings should not be soaked or watered by the roots.
	Do not store seedlings in PVC bags or under PVC foil, as the air in or under the bags becomes very hot in sunny weather, which is harmful to the seedlings and promotes the development of mould and disease.

6TH STAGE	**PLANTING SEEDLINGS**
	For planting, you will need a bag or sack to carry the seedlings; a tunnel, a pick or a planter to dig the hole (container seedlings).

6TH STAGE	The area to be planted must be well prepared. The cuttings must be removed or piled in rows so as not to interfere with planting. Where necessary, the foliage is also removed (partially or completely), shrubs and other vegetation on the area to be restored. In this task, care must be taken to create microclimatic conditions that are favourable to the further development of the seedlings. In particular, we try to preserve shrubs and trees that can mitigate against sunburn, drought, frost, etc., bearing in mind that these specimens may compete with, or damage, the seedlings planted in the future if they are removed at a later date.
	The seedlings must be well protected from desiccation and mechanical damage during transport around the site and must be transported in canvas bags.
	The best way to plant conventionally grown seedlings is in planting holes. Before digging the hole, remove the top layer of thatch, litter or grass. The planting hole should be wide enough to allow the root ball to spread out at the bottom, or deep enough to avoid damaging the taproot (e.g. in oaks). The seedling should be planted as deep as in the nursery. When excavating, we sort the soil, separating the upper humic layer from the lighter layer below.
	When planting container-grown seedlings, the planter is used to make a hole in the soil by pushing it all the way in (the hoe) and rotating it at least 180° to the left and right. The use of a pick for planting is only allowed on karst terrain. Then lift the resulting stopper with your fingers. Never plant in overgrown areas. The seedlings must be planted as deep as in the containers; those planted too shallow tend to dry out. The soil around the planted seedlings is tamped down (even firm pressure with the hands or foot is enough), allowing the soil in the container to adhere to the soil in which it is planted.
	When marking seedlings with stakes, they are driven into the planting hole in such a way that the roots are not damaged.
	A humus layer of excavated soil is placed at the bottom of the planting hole and the seedling's root ball is spread on it in its natural position. If the roots are very loose and tangled, the root ball should be loosened slightly, but care must be taken not to damage it. Then cover the roots with soil (first the darker humic soil, then the lighter soil below), gently pressing it down with your hands. There must be no empty spaces at the root. Then fill the hole all the way with the remaining soil and gently push it down with your foot to avoid damaging the roots.
	After planting, cover the ground around the seedling with dry leaves and grass to retain moisture and protect the roots from drying out.

Responsibilities/supervision

Responsibilities for each stage of the management of forest tree seedlings are:

PHASES OF WORK	ACCOUNTABILITY	SUPERVISION
1ST STAGE	supplier	Forestry Inspection Service
2ND STAGE	SFS	Forestry Inspection Service
3RD STAGE	forest owner/manager	SFS
4TH STAGE	forest owner/manager	SFS
5TH STAGE	forest owner/manager	SFS
6TH STAGE	forest owner/manager	SFS, Forest Inspection Service

Annex 2: Documents for the approval of *in situ* seed facilities:

- application for approval
- informative document
- assessment sheet

Form A: Applications for approval of a forest seed facility

. . . .
. . . .
(name and address or name and registered office of the applicant)

space for stamp

Gozdarski inštitut Slovenije
Večna pot 2
Ljubljana

SUBJECT: Application for approval of a forest seed facility

I request approval of a forest seed production facility for the production of forest reproductive material in the category:

☐ source identified

☐ selected

The use of harvested forest reproductive material in the "selected" category will be:

☐ multifunctional

☐ for planting in forests with limited timber production potential.

In the event that the forest seed facility does not meet the conditions for approval under the category "selected",

☐ I agree

☐ I disagree

that the approval procedure is conducted under the category "source identified".

Annexes:

☐ Information on the owner of the forest seed facility and on the location of the forest seed facility

☐ Declaration of ownership

☐ Power of representation of the owner of a forest seed facility in the approval procedure

Place and date.................................... Signature ....

Form B: Information on the location and owner of the forest seed facility

Information on the forest seed facility

Municipality: ....

Cadastral municipality: ....

Land plot number: ....

Tree species name: ....

Information on the owner of the forest seed facility

Name or company: ....

Street and house No.: .... Place:

Mail address .... Postal code □□□□ Municipality: ....

Personal ID No.: /to be filled in by a natural person and sole proprietor/ □□□□□□□□□□□□□
Company reg. No.: /filled in by sole proprietor and legal person/ □□□□□□□ □□□
company reg. No.
unit of a

Tax ID No.: □□□□□□□□

Responsible person of the legal person:

Name and surname: ....

Street and house No.: .... Place:

Mail address .... Postal code Municipality: ....

Personal ID No.: /to be filled in by a natural person and sole proprietor/ □□□□□□□□□□□□□
Company reg. No.: /filled in by sole proprietor and legal person/ □□□□□□□ □□□
company reg. No.
unit of a

Tax ID No.: □□□□□□□□

Description form

To be completed by SFI

Application number:	Owner code:
Municipality:	
Cadastral municipality:	
Land plot No.:	
Botanical name of the species:	

To be completed by SFS

location:	forest management unit:			
	division, section:			
	provenance:			
	region of provenance:			
	latitude:			
	longitude:			
	the competent local SFS unit:			
description of the forest:	occurrence:			
	surface area:	ha	age:	
	increment:	m^3/ha/year	timber stock:	m^3/ha
	region of provenance:			
	1.	%	4.	%
	2.	%	5.	%
	3.	%	6.	%
economic class	increment:	m^3/ha/year	growing stock	m^3/ha
site:	community:		site index:	
	altitude:	m	slope	%
	exposure:		soil	
access:				
comments:				
compiled by:	first name last name:		place and date:	
	signature:			

Assessment form

Application number:	Owner code:

Type:

☐ stand
☐ seed tree group

Population size **Isolation**

Surface area:	Distance to the nearest stand of the same species, which is:
Number of ☐ trees/ ☐ groups of trees:	Significantly poorer quality:
Distance between ☐ trees/ ☐ groups of trees:	Non-indigenous/source unidentified:

Uniformity

Degree of variability of morphological characters:	☐ low	☐ normal	☐ high

Adaptability[1]:

Stand ☐ adapted/ ☐ not adapted to the site conditions

Notes:

Health condition and immunity

Signs of disease/pests present:
Resilience assessment:

Stand productivity[2]:	☐ average	☐ above average	☐ below average

Wood quality[3]:	☐ below average	☐ average	☐ above average

Form and manner of growth

Proportion of trees exhibiting major defects:

Place and date:

Present:	Name and surname:	Signature:
Members of the commission:	1.	
	2.	
	3.	
Others invited:		

to be completed by the SFI after the decision has been issued:

forest seed facility:

☐ is approved in the "selected" category and has been assigned a number in the register of forest seed facilities:

☐ is approved in the "source identified" category and has been assigned a number in the register of forest seed facilities:

☐ not approved

Form for drawing up the list

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	R	S	T
Ref. No.	Country	Type	Category	ID No.	Provenance	Latitude	Longitude	Altitude	Type	Surface area	Source	Origin – in more detail	Purpose	Notes	Ownership	Municipality	Cadastral municipality	SFS regional unit	SFS local unit

Annex 3: Reporting form for the harvesting of forest reproductive material for the purpose of obtaining a master certificate

(name and address of the applicant) (name and address of the competent unit of the Slovenian Forest Service)	space for stamp

Pursuant to Article 14 of the Forest Reproductive Material Act (Official Gazette of the Republic of Slovenia, No 58/02 and 85/02 – corr.), is hereby notified of the procedure for obtaining forest reproductive material from the forest seed facility described below and requests the issue of a certificate of origin for the forest reproductive material:

forest seed facility registration number:	category:	botanical name:
detailed description the place of production:		
name and address, or company name and registered office of the supplier:		
the name and surname of the person responsible:		
the owner of the forest seed facility:		
estimated time of production:	from:	to:

It is intended to produce: Seed material ☐ plants for natural regeneration ☐ plant parts ☐

The master certificate of identity of forest reproductive material is sought:
☐ seed; more precise data on the quantity of pure seed produced will be reported to the Slovenian Forestry Institute at a later stage, after the completion of the processing process on the premises;
☐ forest reproductive material produced from a forest seed facility.

Place and date: Signature:

Certificate of origin of forest reproductive material

SFS ☐☐/ ☐☐/ ☐☐
SFI ☐☐/ ☐☐

Information on the quantity produced:

date										total
hl/kg/pcs										
signature										

We certify that the forest reproductive material described above was obtained under the supervision of ☒ the Slovenia Forest Service/ ☐ Slovenian Forestry Institute in accordance with the Forest Regulations and the Regulations on Forest Reproductive Material.

Date:	Name and surname of the authorised person:
Stamp:	Signature:

Annex 4: Master Certificate of Forest Reproductive Material (Specimen)

A MASTER CERTIFICATE OF IDENTITY FOR FOREST REPRODUCTIVE MATERIAL
issued in accordance with Directive 1999/105/EC

SLOVENIA	SI/22-010

We hereby certify that the forest reproductive material described below has been produced:
☐ ✓ In accordance with the EC Directive;
☐ In accordance with the OECD Forest Seed and Plant Scheme.

1. Botanical name: *Acer monspenssulanum* L.

2. Oblika:	**3. Kategorija:**	**4. Tip gozdnega objekta:**
☐ ✓ semenski material ☐ deli rastlin ☐ sadilni material	☐ ✓ znano poreklo ☐ izbran ☐ testiran	☐ skupina semenjakov ☐ ✓ sestoj

5. Purpose: 1 for multipurpose forestry

6. Reference number or statement of identity of the starting material with regard to the national register: . . 7.0425 . . .

Mixture: . . . .

7. Source authochthonous
non-indigenous, namely
unknown

8. Source of the starting material (non-autochthonous if source is identified: . . . .

9. Country and region of provenance of the starting material: Slovenia, Submediterranean

Provenance (short name): Povir - Tabor

10. The altitude or altitude range of the location of the starting material: 400m/m

11. Year of seed ripening/production of plants for natural regeneration: 2022 **12. Quantity: 5.1kg**

13. Is the lot for which this certificate is issued the result of a division of a larger lot for which an EC certificate has been issued?
☐ Yes ☐ ✓ No
The number of the previous certificate: . . . . Quantity in the previous lot:

14. Duration of tree nursery cultivation:

15. Is the planting material cultivated from seed and subsequently vegetatively propagated?
☐ Da ☐ ✓ Ne
Mode of reproduction: Number of reproductive cycles:

16.Other important information:

. . . .

17. Name and address of the supplier
Zavod za gozdove Slovenije
CE, Večna pot 2, 1001 Ljubljana

Name and address of the supplier **Zavod za gozdove Slovenije** **CE, Večna pot 2, 1001 Ljubljana, Slovenia**	**Stamp** **Date: 5 December 2022**	**Name and surname of the person responsible:** **Hojka Kraigher** **signature**

Annex 5: The certificate of the Inspectorate of Agriculture, Forestry, Hunting and Fishing (IRSKGLR) on the mixing of forest reproductive material

(name and address of the applicant) (name and address of the competent unit of the Slovenian Forest Service)	space for stamp

Pursuant to Articles 12 and 13 of the Forest Reproductive Material Act (Official Gazette of the Republic of Slovenia Nos. 58/02 and 85/02 – corr.), a master certificate of identity for forest reproductive material obtained by mixing is requested:

region of provenance/provenance:	
category:	botanical name:
detailed description the place of production:	
name and address, or company name and registered office of the supplier:	

Certificate on the control of the mixing of forest reproductive material No.

Mixture information:

Master certificate No.:	year of mast production:	forest seed facility:	quantity

We hereby certify that the forest reproductive material described above was produced under the supervision of the Forestry Inspection Service, in accordance with the regulations on forest reproductive material.

Date: Name and surname of the competent forestry inspector:

Stamp: Signature:

Annex 6: Seed Quality Certificate issued by the Slovenian Forestry Institute

Slovenian Forestry Institute Slovenian Forestry Institute
Večna pot 2, SI-1000 Ljubljana

Izvid o kvaliteti semena št. (Certificate of quality of seed No.)

Ime in naslov dobavitelja (Name and address of supplier): SFS
Slovensko in botanično ime (English and botanical name):
Registrska številka semenskega objekta (Registration number of seed facility):
Številka glavnega spričevala (Number of Master Certificate):
Kategorija reprodukcijskega materiala (Category of reproductive material):
Namen uporabe (Usage purpose):
Provencienčno območje (Region of provenance):
Provenienca (Provenance):
Leto v katerem so semena dozorela (Year of seed ripening):
Teža partije (Lot weight):
Vzorčenje opravil (Sampling done by):

Število embalaž Number of packages	Datum vzorčenja Date of sampling	Datum sprejema vzorca Date of admission of the sample	Datum zaključka testiranja Date of conclusion of testing	Številka testa Number of test

Analizni rezultati (Results of the analysis): velikost vzorca (size of sample):

ČISTOST PURITY			KALIVOST GERMINATION						DELEŽ VLAGE MOISTURE CONTENT
Uteži % Weight %				Številčni % Number %					% sveže teže fresh weight
Čisto seme Clean seed	Drugo seme Other seed	Inertni material Inert material	Št. dni Number of days	Normalne klice Normal seedlings	Nenormalne klice Abnormal seedlings	Sveže seme Fresh seeds	Trdo seme Hard seeds	Mrtvo seme Dead seeds	

Drugo seme (Other seed): /
Opis inertnega materiala (Description of inert material): /

Druge analizne metode (Other methods of analysis):

Vitalnost [Številčni %] Vitality[Number %]	Živi embrii Live embryos	Sumljivi embrii Suspicious embryos	Mrtvi embrii Dead embryos	Gluho seme Empty seed
TTC (Tetrazolium test)				
Rentgen (X-ray)				
Izolirani embrio (Isolated embryos)				
Drugo (Other)				

Uporabnost (Applicability):

Kalivost / vitalnost [Št./1 kg semen] Germination / vitality [No./1 kg seed]	Teža 1000 semen [g] Weight of 1000 seeds [g]
	7,9

Zdravstveno stanje (Health condition):

Tip poškodbe/okužbe (Type of damage / infection)	Poškodba/okužba z (Damage / Infection by) - slovensko in latinsko ime (English and Latin name):	Številčni delež poškodovanih/okuženih semen (Number percentage of damaged / infected seeds):

Opombe (Notes):/
Veljavnost certifikata (Validaity of the cetificate): 365 dni po dnevu izstavitve (365 days after issue)

Analitik (Analyst): Jana Janša

Pooblaščena strokovna oseba
Responsible Officer

Hojka Kraigher

Datum (Date):

Izvid je izdan v skladu z Zakonom o gozdnem reprodukcijskem materialu (Ur. 1. RS, št. 58/2002 in 85/2002) in Pravilnikom o ugotavljanju podatkov za seme gozdnega drevja (Ur. 1. RS, št. 127/2003)
Certificate is issud in accordance with Forest Reproductive Material Act (Ur. 1. RS, št. 58/2002 in 85/2002) and Rules for determing data on forest seeds (Ur. 1. RS, št. 127/2003)

Conservation of Forest Genetic Resources with Forest Reproductive Material Management Guidelines

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The textbook is aimed at students and practitioners in the field of biodiversity conservation, with an emphasis on the conservation of genetic diversity, forest breeders, forest seeders and arborists. Presentation of the basics of forest genetics, the importance and theoretical background for the conservation of forest genetic resources, the Strategy for the Conservation of Forest Genetic Resources in Europe and the EUFORGEN and SIFORGEN (Slovenian Programme for the Conservation of Forest Genetic Resources) programmes. Introduction to the botanical basics of seed structure and germination physiology. Technologies for the production, processing, storage and germination of seeds of forest tree species. The basics of seedling rearing in forest nurseries. European and Slovenian legislation on forest reproductive material. Examples of documents used for the approval of forest seed facilities and certification of forest reproductive material.

Keywords:
genetic diversity, conservation of forest genetic resources, forest reproductive material, legislation, certification

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I consider this work to be of high priority, particularly at a time of great environmental change, which is not sparing our forests. For the first time in a long period, the state of forest resource management in our country and in the EU is described and collected in one place, with all the professional, technological and management basics. As such, this work will serve as an indispensable resource for all those interested in sustainable forest utilisation (forest owners, the Slovenia Forest Service (SFS), the Slovenian Chamber of Commerce and Industry of Slovenia (CCIS). the Ministry of Agriculture, Forestry and Food of the Republic of Slovenia, students, teachers and researchers in the field of forestry, etc.), as well as for all other parties interested in nature and environmental issues.

I consider this work to be suitable for use at all levels of teaching where this issue is involved, both professionally and terminologically.

***Franc Batič**, Professor Emeritus, retired full professor of Botany and Plant Ecology*

Almost two-thirds of Slovenia's national territory is covered by forest (including overgrown farmland), therefore the content is also important and relevant in the light of major climate change (droughts and bark beetle) and land use changes (overgrowth). This monograph is intended not only for foresters and forest professionals, but also for students of forestry, biology and ecology (the latter courses are also taught at the University of Maribor), as well as anyone who is in any way interested in forests, their management and conservation.

***Prof. Dr. Mitja Kaligarič**, Full Professor of Botany at the University of Maribor*

The work "Conservation of Forest Genetic Resources with Forest Reproductive Material Management Guidelines" presents the history of organised development concerning forest reproductive material in Slovenia, the foundations of flowering biology, seed physiology and germination, the basics of forest genetics and the conservation of forest genetic resources in Slovenia and Europe. It presents the current legal bases and codes in the field of development concerning forest reproductive material and tree nurseries, the delimitation of regions of provenance and the list of species covered by the Forest Reproductive Material Act, as well as the procedures for the approval and traceability of the origin of forest reproductive material. Recommendations for seed collection, processing and storage, as well as procedures for the germination of seeds of selected forest tree species are also briefly summarised.

The work is an important aid for studying forestry and for working in silviculture, seed production and nursery practice. It will contribute to planning the needs for forest seed objects, seeds and seedlings for use in forests in Slovenia and to the importance of the field in Slovenian forestry at a time when large-scale disasters are increasingly preventing the successful and high-quality natural regeneration of forests.

***Academic Ivan Kreft**, Professor of Plant Genetics, when reviewing the 2019 draft of the work*

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