RESEARCH AND PRACTICE IN FOREST ECOLOGY

INTERNATIONAL CONFERENCE

BOOK OF ABSTRACTS

Kórnik, 8–12 May 2024

Research and Practice in Forest Ecology

Research and Practice in Forest Ecology

International Scientific Conference for Young Scientists
Institute of Dendrology, Polish Academy of Sciences

8–12 May 2024, Kórnik, Poland

Book of abstracts

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The Institute of Dendrology, Polish Academy of Sciences,
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entitled "Research and Practice in Forest Ecology" (KONF/SP/0200/2023/01)
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Contents

Introduction

Since the beginning of time, we, human beings, have been attempting to understand the world around us. This is a beautiful quality – we are curious about the incomprehensible. We are constantly refining our methods of perceiving the reality around us, and interpreting it more and more deeply. We pose questions, find answers, and then sometimes challenge them by posing further questions. In this way, we all participate in the extremely important process of building our knowledge of the world and broadening our horizons.

Scientists who try to describe and understand the natural world face ever new challenges. We live at a time when the human impact on the environment is becoming increasingly apparent, with global climate change and loss of biodiversity being particularly serious consequences. In the functioning of forests, too, we are seeing many changes due to ongoing environmental changes, and our knowledge of their functioning needs to be constantly improved and deepened. The efforts of scientists are not only geared to acquiring new knowledge, but also to applying it to forest management and nature conservation. For us to manage natural resources wisely, we must have a thorough knowledge of the functioning of forest ecosystems.

Forests form some of the most dynamic and complex terrestrial ecosystems. Although the biology of the numerous species that determine the biodiversity of forest ecosystems is relatively well understood, the interactions between them are much less so. Knowledge concerning the ecology of woody plants, which determine the physiognomy of forests – in conjunction with knowledge of the complexity of broadly-defined phytocenoses, zoocenoses and mycocenoses, especially considering the abiotic conditions of their functioning – is also of great practical importance, as it supports forest management and nature conservation activities. These groups of species do not function under constant conditions, and changing environmental parameters force them to react in different ways. The theoretical underpinnings of forest management are of crucial importance for forestry practice, especially as, in the face of rapid climatic (and more broadly, habitat) change, we often cannot rely on the knowledge from 200 or 100 years ago, which we had once assumed to be fundamental. The effects of these changes are already being observed, and they affect not only the naturally and economically important tree species in Poland and Europe, but also related species of fungi, other plants and animals.

The aim of the conference "Research and Practice in Forest Ecology" is to discuss the results of research in forest ecology, taking into account the transformation of forest ecosystems in local, regional and global terms; to outline directions and perspectives for new research, including interdisciplinary research; and to establish cooperation between scientific centers and practitioners. Our conference is aimed at young scientists, PhD students and students working in the broad field of forest ecology.

We hope that the conference will enable researchers to broaden their knowledge with up-to-date scientific and practical information on tree ecology and forest functioning in the light of currently observed and predicted environmental changes. Moreover, we hope that it will enable its participants to familiarize themselves with the methodology of research conducted at the Institute of Dendrology of the Polish Academy of Sciences in the field of broadly-understood forest ecosystem ecology. Participation in the conference will also provide an excellent opportunity to develop the necessary skills for presenting one's own research results. We are delighted that so many young scientists from various national and international scientific centers have expressed their willingness to participate in the conference.

In addition to the 10 keynote lectures and numerous oral presentations submitted by conference participants, we have also planned workshops to be conducted by researchers from the Institute of Dendrology of the Polish Academy of Sciences, covering the following topics: (1) Ectomycorrhizal fungi – from fruiting body to DNA sequencing, (2) *In vitro* propagation of plants, (3) Population genetics of forest trees, and (4) Woody plant species recognition in the Kórnik Arboretum. As part of the conference, we have also organized a field excursion to the Stołowe Mountains National Park. Employees of our Institute have been conducting scientific research in the area of this National Park for many years, and their results are not only a valuable addition to the knowledge of the biodiversity of this protected area, but are also used in activities aimed at improving the conservation of native wildlife. The scientific results obtained by the Institute indicate, among other things, that the species richness of the National Park is much greater than the previously documented research carried out in the area had shown. In recent years, our research has focused on the National Park's two most important watercourses (the Czerwona Woda River, and the Kudowski Potok River), and their results confirm the significant impact of dwindling water resources on the biota of the National Park. Familiarizing the conference participants with the results of these studies in a protected site will not only deepen their knowledge, but will also show how the results of scientific research can be applied in the active protection of natural resources.

We hope that the stay at the Institute of Dendrology of the Polish Academy of Sciences and the active participation in the conference "Research and Practice in Forest Ecology" will satisfy at least part of the curiosity of our guests – young scientists.

Kórnik, May 8th, 2024

Director of the Institute

prof. Andrzej M. Jagodziński

Plenary session

Institute of Dendrology, Polish Academy of Sciences (1933–2023): a history in brief

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Nineteenth-century owners of the Kórnik estate – Counts Tytus and Jan Działyński – showed great interest in dendrology, and it was thanks to them that a very rich collection of woody plants was gathered in the Kórnik castle garden: Tytus (1796–1864) initiated the creation of the woody plant species collection, and Jan (1829–1880) greatly extended the work started by his father. Jan Działyński amassed a collection in Kórnik that included around 1,500 species and varieties of trees and shrubs. The last owner of the Kórnik estate, Count Władysław Zamoyski (1853–1924), nephew of Count Jan, successfully implemented and developed the intentions of his predecessors (fig. 1). According to the last will of Count Zamoyski, in 1925 the Zakłady Kórnickie Foundation was established and the Zamoyski estate was handed over to the Polish State. Zamoyski's will was

Fig. 1. Commemorative plaque dedicated to the founders of the Arboretum of the Institute of Dendrology, Polish Academy of Sciences

to "establish and maintain a research center, both on the mountain slopes and on the plains, for the study of everything that falls within the scope of breeding, living, protection and proper exploitation of all kinds of trees, both domestic in the country and foreign ones, that may be usable for the country; forest, garden, usable, fruit and ornamental trees, their wood, fruits, leaves, juices. Care for the Kórnik gardens".

Unfortunately, the scientific institution in Kórnik was not established immediately, although many members of the Zakłady Kórnickie Foundation Board supported this initiative with their authority. The most important role in establishing the scientific institution in Kórnik was played by Professors Władysław Szafer (1886–1970) and Stanisław Sokołowski (1865–1942), who developed the first outline of the research programme of the research centre, supplemented by a number of outstanding specialists. The subject matter was extremely broad, even by today's standards.

After some perturbations, on July 1st, 1933, a scientific institution was established in Kórnik – the Centre (Department) for Research of Trees and Forest, although the establishment of only one of the four planned organizational divisions was successful. The first director of the Centre for Research of Trees and Forest was Antoni Wróblewski (1881–1944), who had managed the Kórnik Gardens of the Zakłady Kórnickie Foundation since 1926.

Antoni Wróblewski was an outstanding dendrologist (fig. 2). During his time, the collections of woody plants were significantly expanded and their complete documentation was kept. Wróblewski developed nurseries in Kórnik, established cooperation with numerous foreign institutions of a similar nature, conducted numerous experiments in the arboretum and nurseries, and published the results of these studies, as he understood the need to disseminate them. Antoni Wróblewski brought to the Kórnik Arboretum many plants, including over two thousand fruit trees and shrubs, greatly extending the arboretum's collection of woody plants. The development of the Centre for Research of Trees and Forest was halted by the Second World War. During this time, the collection of woody plants gathered in the arboretum suffered significantly, although if it had not been for Antoni Wróblewski and his colleagues, the losses would have been significantly greater.

After World War II, the management of the scientific institution in Kórnik was taken over by Eng. Stefan Białobok (1909–1992), later a Professor at our Institute. There is no doubt that Prof. Białobok was the creator of the Institute of Dendrology of the Polish Academy of Sciences in Kórnik (fig. 3). He headed our institution from 1945, as director of the Department of Trees and Forest Research, until 1979, when he finished his work at the Institute. The merits of Prof. Stefan Białobok are difficult to overestimate. Just after the war, Prof. Białobok struggled with many difficulties when organizing scientific work from scratch. Everything was missing. At the time, the Department had two buildings at its disposal, and their usefulness for scientific purposes was not great.

Rapid development of the Department took place in 1952, when the Department of Dendrology and Pomology was incorporated into the structure of the newly established Polish Academy of Sciences. Prof. Białobok assessed this event as follows: "The year 1952, when the Department of Dendrology and Pomology was taken over by the Polish Academy of Sciences, should be considered a particularly important date in the life of this institution. From now on, the Department receives resources for development and can begin to move past many years of neglect". The assets of the Department at the time were very extensive and included: a research unit in Kórnik, a research station in Turew, a farm in Kórnik, nurseries and the "Zwierzyniec" Experimental Forest (since 1958). At the time, only 20 research employees worked at the Department.

In the 1950s, Prof. Białobok made intensive efforts to build a new building for the Department. These efforts were very fruitful, and this is how the headquarters of the Department was established. During that time, the Department of Dendrology and Pomology of the Polish Academy of Sciences carried out very extensive research, which was expanded in subsequent years. In the 1950s, the main issues that employees dealt with were: (1) pomological experiments (selection of rootstocks), (2) principles of cultivation, reproduction and selection of poplars (*Populus*), (3) natural variability, systematics and distribution of poplar species (*Populus*), (4) genetics of forest trees – the importance of natural selection, (5) introduction and acclimatization of alien tree and shrub species, and (6) systematics and chorology of woody plants growing in the wild.

Then the first experiments were established in the "Zwierzyniec" Experimental Forest. Particularly valuable experimental sites located in the area include provenance experiments of Scots pine, Norway spruce, Douglas fir, black alder,

Fig. 2. Memorial plaque dedicated to Antoni Wróblewski

silver fir and grand fir, native oak species, as well as archives of pine, spruce and larch clones. Some of these experiments are part of international networks of experimental sites established in different countries under the supervision of IUFRO. As of January 1th, 2020, there were 56 experimental plots in the "Zwierzyniec" Experimental Forest, covering a total of ca. 38.5 ha. It is worth mentioning that the Department was the precursor of genetic research on forest trees in Poland. Moreover, it was with the participation of many employees of the Department that the first textbook on dendrology in Poland was created, published in 1955.

In the 1960s, the Department carried out research covering five main thematic areas: the physiology of growth and development, seed physiology, genetics, introduction and acclimatization, and the systematics of trees and shrubs. Of particular importance during this period was the research initiated by Prof. Kazimierz Browicz (1925–2009), which included studies on the woody flora of southwestern Asia and the eastern Mediterranean. Cooperation with scientists and scientific publishers from other countries was also established, thanks to which the Institute's employees made a significant contribution to the development of such works as: "Flora Europaea", "Flora of Turkey", "Flora Iranica" and "Mountain Flora of Greece". Kórnik became the center of systematic and chorological research, which was continued in the following years by our outstanding Professors: Jerzy Zieliński, Adam Boratyński, and Krystyna Boratyńska. The results of numerous scientific expeditions were not only publications, but also a significant expansion of the herbarium collections gathered at the Department. In Kórnik, as the first scientific institution of this kind in our country, extensive research was undertaken in the field of forest tree genetics, covering the main forest-forming species, and the results of these studies were used in forestry practice. Thanks to contacts with foreign centers and scientists, the latest scientific trends concerning the genetics of forest trees came to Kórnik. The development of genetic research was greatly influenced by the employment of young scientists, later professors: Prof. Tadeusz Przybylski (1929–2010), Prof. Maciej Giertych, Prof. Leon Mejnartowicz, Prof. Władysław Chałupka, and Prof. Andrzej Lewandowski. The publication by Prof. Białobok entitled "Establishment of forest tree seed plantations", summarizes the involvement of the Department's employees in the development of forest tree genetics in the 1960s.

In the early 1960s, the first phytotron in Poland was built at our institution, which led to pioneering work on understanding the mechanisms determining the dormancy and germination of seeds of many species of trees and shrubs, not only those important for forest management. We owe the initiation of seed biology research and its subsequent development at many different levels to Prof. Bolesław Suszka (1925–2020). Many years later, Prof. Suszka significantly contributed to the creation of the Kostrzyca Forest Gene Bank, as he developed the program assumptions of this unit of the State Forests. Research in the field of physiology of trees and shrubs was significantly deepened in the 1960s by Prof. Mirosław Tomaszewski (1925–1985), a chemist employed in Kórnik in 1951, who, after returning from a research internship at Harvard University, developed a research group dealing with the broadly understood physiology and biochemistry of plants, and initiated research on the mechanisms regulating the formation of mycorrhizal associations of trees. These studies were carried out by a very broad team (in various groups within the changing internal structure of the Department), including: Prof. Stanisława Pukacka, Prof. Paweł M. Pukacki, Prof. Barbara Kieliszewska-Rokicka, Prof. Maria Rudawska, and Prof. Zofia Szczotka. It is worth adding that we express gratitude to Prof. Tomaszewski for blazing the trail to such an important journal as “Nature”.

The end of the 1960s and the beginning of the 1970s was a period of intense efforts to raise the rank of our institution from a Department to an Institute. Prof. Białobok then claimed that the research centre in Kórnik should “focus primarily on tree biology rather than forest biology” because “research in the field of tree biology is not yet very advanced in the world, so developing it in our country would give Polish dendrology a chance to take on a good position in the broadly understood European science of dendrology”. The activities of Prof. Białobok brought about the intended effect. In 1974, in recognition of the level of research we conducted, the Department was elevated to the rank of the Institute of Dendrology of the Polish Academy of Sciences and uses this name to this day.

In the mid-1970s, there were four research departments at the Institute: (1) Department of Systematics and Geography, (2) Department of Introduction and Acclimatization, (3) Department of Genetics, and (4) Department of Tree and Shrub Physiology. The research topics were really very broad. At that time, research was carried out on the physiology of growth and development, seed physiology, genetics, anatomy, introduction and acclimatization, as well as systematics

Fig. 3. Memorial plaque dedicated to Prof. Stefan Białobok (Photo: Kinga Nowak)

and geography. A very important topic discussed by the Institute's employees was the impact of the degradation of the natural environment, especially as a result of industrial activity, on woody plants and the ecological systems they co-create. These studies were carried out, among others, by: Prof. Gabriela Lorenc-Plucińska – from 1973, Prof. Piotr Karolewski – from 1974, and Prof. Jacek Oleksyn – from 1976. A specialized laboratory was established at the Institute, enabling research to be conducted under controlled conditions, and numerous experimental sites were established in areas contaminated by industry. The 1970s were also an era of extraordinary publishing success. In 1970, a series of books began to be published under the common title "Our Forest Trees". These books still occupy an important place in the homes of many foresters – practitioners and others.

At the beginning of the 1980s, the management of the Institute was entrusted to Prof. Władysław Bugała (1924–2008; fig. 4). Employees conducted research covering the following issues: industrial pollution, taxonomic and chorological research, seed biology, phytopathology, forest tree mycorrhizas and physiology. One of the symposia organized in Kórnik resulted in the preparation of a book entitled "The life of trees in a contaminated environment", which was published in 1989 by the National Scientific Publishing House as part of the series of popular science monographs "Our Forest Trees". In 1986, thanks to the efforts of Prof. Bugała, a new building of the Institute was put into use, which, in addition to the laboratories, housed library collections and the herbarium. At the end of the 1980s, the Institute was assessed by Prof. Jan Kornaś (1923–1994), an outstanding botanist, plant geographer, phytosociologist and taxonomist from the Jagiellonian University and the Władysław Szafer Institute of Botany of the Polish Academy of Sciences in Kraków. Prof. Kornaś then stated that the subject of the Institute's research "is of great importance, both from the point of view of basic and applied research, and does not overlap with the subject matter of any other scientific institution in the country, and also on the scale of Europe, this type of institution is something unique and many times was taken as a model for the creation of new dendrological research centers". The Professor concluded: "I believe that the Institute of Dendrology of the Polish Academy of Sciences is a stable institution, with appropriately prepared scientific staff and good technical facilities, developing high-level, modern dendrological research in the field of introduction and acclimatization, systematics and geography, genetics and environmental protection, fulfilling an important role in the broadly understood theoretical and practical knowledge of woody plants. I consider the further development of the Institute to be purposeful and desirable".

In the mid-1990s, the Institute employed approximately 120 people. The duties of the director were entrusted to Prof. Tadeusz Przybylski (fig. 5), who managed it for three years. The scientific research program, which is to some extent reflected in the very extensive internal structure of the Institute, was very broad at that time. The research was conducted in six departments: Department of (1) Systematics and Geography, (2) Introduction and Acclimatization, (3) Tree Genetics, (4) Tree Physiology, (5) Tree Resistance, and (6) Seed Biology.

Very dynamic development of the Institute has been visible since the turn of the 20th and 21st centuries – expressed, for instance, by a significant increase in the quantity and quality of publication output by employees and the expansion of the scope of research, thanks to the possibility of applying for financial resources for conducting research in domestic and foreign grant agencies (fig. 6). This development coincided with the periods of management of the Institute by our directors: Prof. Tadeusz Przybylski (in the years 1996–1998), Prof. Gabriela Lorenc-Plucińska (1999–2010), and Prof. Jacek Oleksyn (2011–2018).

The Institute of Dendrology of the Polish Academy of Sciences conducts interdisciplinary research encompassing two scientific disciplines: forestry and biological sciences. Their aim is to learn about the biology and ecology of woody plants at all levels of their organization. Since its inception, the Institute's staff have produced many interesting research results, have published in the best scientific journals in the world, have authored numerous monographs, and have actively disseminated the knowledge gained, including by organizing specialized scientific conferences and conducting popularization activities. All these activities were aimed at realizing the Institute's key mission, stemming from the will of its Founder – Count Władysław Zamoyski.

The Institute's employees have always taken care to disseminate the results of their research. We have published numerous books and scientific articles in domestic and foreign journals, including the best ones indexed in the Web of Science Clarivate database. Our research articles have also been noticed by the scientific community. The number of citations to publications created in Kórnik

Fig. 4. Memorial plaque dedicated to Prof. Władysław Bugała (Photo: Kinga Nowak)

is growing at a very high rate. The dominant thematic areas in which our publications are classified include plant sciences, forestry, ecology, environmental sciences, genetics, biodiversity conservation and others. The analysis of our achievements also shows how intensively we cooperate with scientists from Poland and abroad, solving scientific problems together. Our main scientific partners outside Poland are from the United States, Spain, Australia, Germany, Italy, France, the United Kingdom, and China.

We conduct research in teams that include five scientific departments and auxiliary departments. They are: (1) Department of Biogeography and Systematics, (2) Department of Developmental Biology, (3) Department of Ecology, (4) Department of Genetics and Environmental Interactions, and (5) Department of Symbiotic Associations. A great amount of support is given by: the Arboretum and Experimental Forest, the Library, the Laboratory of Mineral Analyses, and the Herbarium.

The collections gathered in the Arboretum were and are an extremely important object of our research. In 2026, the Arboretum will celebrate its 200th anniversary. The Arboretum is the Institute's flagship. So far, its management has been supervised by Antoni Wróblewski, Prof. Władysław Bugała, and Dr. Tomasz Bojarczuk (1942–2023). The Arboretum's collection of woody plants is one of the largest and oldest in Europe and is constantly being expanded by the Institute. It is, however, not only a place of research, but also a thriving center for tourist and knowledge-promoting activities. The social importance of the Arboretum can be seen from the fact that in recent years it has been visited by more than 100,000

Fig. 5. Prof. Tadeusz Przybylski educational spot

people a year, who also take advantage of its educational offerings, i.e. numerous cultural and sporting events. Currently, this mission is being carried out by Dr. Kinga Nowak, with the support of her team.

The Institute also has an Experimental Forest with an area of 222 ha, where many research sites have been established, including numerous provenance experiments, the scientific value of which increases every year. An extremely valuable resource of the Institute is the Herbarium, which contains over 56,000 sheets with specimens of woody plants. Many of these resources have been digitized in recent years and are widely available. The Library provides important support for us. Our collections include over 50,000 books and journals. Many of them are also accessible from your home computer.

The Institute has always placed emphasis on publishing research results. We also published numerous journals of our own. We currently publish the "Dendrobiology" journal, indexed by Web of Science Clarivate, with open, free access to publications (https://www.idpan.poznan.pl/dendrobiology/).

The education of scientific staff was and is the greatest priority for the Institute. We are currently educating young scientists at the Poznań Doctoral School of the Institutes of the Polish Academy of Sciences, which we co-founded. We have the right to confer doctorates and habilitations, as well as to nostrify scientific degrees in two disciplines: forestry and biological sciences.

We attach great importance to disseminating knowledge, not only in the form of publications. We have been organizing conferences for years, which enjoy constant interest, e.g. "Biology and ecology of woody plants" (2013, 2018, 2023) and

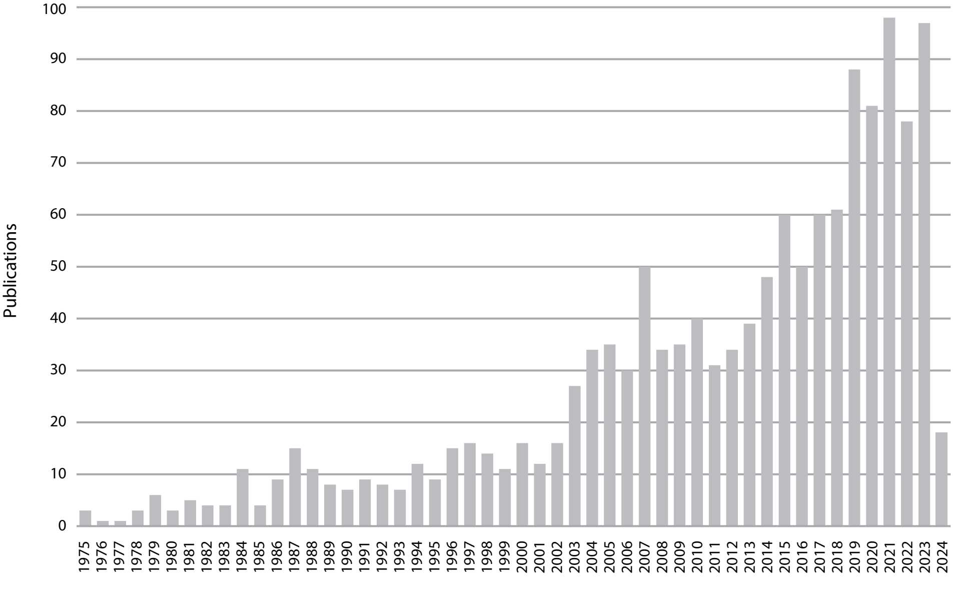

Fig. 6. Number of publications published by the researches of the Institute of Dendrology, Polish Academy of Sciences, in the years 1975–2024 according to Web of Science Clarivate Analytics (as of 17[th] April 2024)

"Trees and forests in a changing environment" (2016, 2021). We also publish numerous popular science articles in biological, forestry and environmental protection magazines. Keeping up with the times, we record numerous films and lectures prepared by our employees to shape awareness of a knowledge-based society (https://www.facebook.com/InstytutDendrologiiPAN; https://www.youtube.com/@InstytutDendrologiiPAN).

Our achievements have been repeatedly and systematically assessed by the Ministry of Science and Higher Education. We can proudly say that the latest thorough analysis of the Institute's achievements (2017–2021) places us among the best scientific institutions in Poland, in both forestry and biological sciences. Moreover, in 2024 we have been recognized by the European Commission and awarded the HR Excellence in Research Logo – the HR Logo is awarded to the institutions that provide the best working conditions for researchers and conduct their recruitment processes transparently and in accordance with the guidelines of the European Charter for Researchers and the Code of Conduct for the Recruitment of Researchers.

More information

Jagodziński A.M. 2018. Badania ekologiczne drzew i lasów w Instytucie Dendrologii PAN. Przegląd Leśniczy 11: 31.

Jagodziński A.M. 2018. Institute of Dendrology, Polish Academy of Sciences, Kórnik. In: Jasińska A.K., Jagodziński A.M., Fazan L., Sękiewicz K., Walas Ł. (eds.). Relict woody plants: linking the past, present and future. The International Scientific Conference. Kórnik, 19th June 2018. Book of Abstracts. Bogucki Wydawnictwo Naukowe, Poznań. pp. 17–24.

Jagodziński A.M. 2019. 110-lecie urodzin twórcy Instytutu Dendrologii PAN w Kórniku. Las Polski 10: 29–30.

Jagodziński A.M. 2019. Instytut Dendrologii PAN – pasja i misja. Kórniczanin 10: 6–7.

Jagodziński A.M. 2022. Instytut Dendrologii PAN. In: Świtoński M., Sobkowska K. (eds.). Oddział PAN w Poznaniu w latach 1972–2022. Agencja Wydawniczo-Poligraficzna GIMPO, Warszawa, pp. 92–94.

Jagodziński A.M. 2022. O prof. Stefanie Białoboku – w 30. rocznicę śmierci. Kórniczanin 14: 11.

Jagodziński A.M. 2023. Instytut Dendrologii PAN – 90. rocznica powstania. Kórniczanin 13: 10–11.

Jagodziński A.M. 2023. Instytut Dendrologii PAN – czerpiąc z historii, tworząc przyszłość. W: Tomaszewski D., Jagodziński A.M. (eds.) 2023. Biologia i ekologia roślin drzewiastych. Konferencja naukowa połączona z obchodami Jubileuszu 90-lecia Instytutu Dendrologii PAN w Kórniku. Kórnik-Poznań, 9–11 października 2023 roku. Materiały konferencyjne. Bogucki Wydawnictwo Naukowe, Poznań, pp. 23–46.

Jagodziński A.M., Biniaś-Szkopek M. 2024. Władysław hr. Zamoyski – nasz Fundator. Kórniczanin 1: 16–17.

Jagodziński A.M., Nowak-Dyjeta K. 2012. Przyrodniczy pomnik historii. Arboretum Kórnickie. Las Polski 12: 18–19.

Jagodziński A.M., Oleksyn J. 2013. Instytut Dendrologii PAN w Kórniku. Kórniczanin 26: 6.

Jagodziński A.M., Ratajczak E., Pers-Kamczyc E. 2019. Prof. dr Stefan Białobok – wybitny dendrolog i zasłużony kórniczanin. Kórniczanin 9: 12–13.

There is a solution to every problem: a contribution to the biography of Władysław Zamoyski (1853–1924)

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The Europe of the 19th century is a Europe without Poland. The world in this century is drowning in constant skirmishes, uprisings, and revolutions. These events lead to a moment when the hitherto existing world totters on its foundations. In the centre of such Europe, Władysław Zamoyski, the first-born son of General Władysław Zamoyski and Jadwiga née Działyńska, was born on 18 November 1853. Throughout his life, he earned the reputation as one of the most interesting and controversial Poles at the turn of the 19th and 20th centuries. An outstanding child of distinguished parents, he came to be dubbed "the strange count", "the iron miser", and even "Don Quixote".

The family lived in Paris, and Władysław was baptised here on the Isle of St Louis. Two months after his birth, the Zamoyski family, active in the fight for

Fig. 1. Władysław Zamoyski (1853–1924), circa 1890, the photograph from the collection of the Polish Academy of Sciences Kórnik Library

Poland's freedom, left for Turkey, where Władysław's father became politically involved in the Crimean War (1853–1856). This is how the boy ended up in Wielkopolska under the care of his grandparents, Celestyna née Zamoyska and Tytus Działyński. He spent the first two years of his life at the Działyński Palace in Poznań and the Kórnik Castle. Władysław grew up surrounded by educated, enlightened, and socially engaged people, full of love for their homeland.

After the Zamoyskis' return from Turkey, little Władysław returned to Paris, and here he soon awaited the birth of his younger brother Witold. The involvement of his entire family in the fight for the country's independence had a significant impact on the children's upbringing. They ensured that the boys – despite being brought up in France – knew the Polish language, as well as the basics of the geography, history, and literature of their own nation. Their parents took care of their spiritual, intellectual, and physical development. As part of the upbringing of the Zamoyski children, special attention was paid to self-discipline, dutifulness, and diligence, as well as elements of the Christian faith and deep patriotism. The contents of many letters, which the members of this family eagerly wrote to each other, indicate that an atmosphere full of love and mutual care prevailed in the Zamoyski and Działyński households. This atmosphere was also supported by regular household members who, living in Władysław's family home, were much closer to the Zamoyski children than servants, and assisted in their upbringing and education. Among these individuals, it is worth mentioning the Englishwoman Anna Birt, as well as two Poles: Leonard Niedźwiecki, and the well-known pedagogue Józef Rustejka.

Education through travel was also an extremely important element in this family. Little Władysław travelled all over Europe with his parents, and as a young boy, he also accompanied them on a trip to the exotic Ottoman Empire, where his politically engaged parents supported the Polish cause. He also toured Italy and visited his grandparents in the Polish lands.

After the first years of his home education, Władysław entered the elite Lycée Charlemagne secondary school in Paris. The family continued to live in Paris, where General Władysław Zamoyski died in 1868. The widow and her three children – two sons and the youngest daughter Maria – remained in France. Here, Władysław attempted embark on a military career and gain admission to the École polytechnique. Unfortunately, his exams did not go well, and despite his attempts, he did not manage to start his studies. In this situation, on the advice of his uncle Jan Działyński and his caring mother, he made an interesting choice. In 1879, he set off as attaché to the French Commission organising the next Universal Exhibition, which took place in Australia this time.

He spent several months on this remarkable continent, assisting in the opening of two major exhibitions – in Sydney and Melbourne. His travel notes show that he was constantly on the move, visiting and exploring all sorts of places which had just been made accessible thanks to modern means of transport. During this trip, which became an around-the-world journey, he visited not only Australia, but also Tasmania, New Caledonia, New Zealand, the Tonga Islands, Samoa, Hawaii, and the United States of America. Władysław's activity was driven by curiosity and the hope that he would be able to use the experience and skills he gained from his travels to benefit his partitioned homeland and its people. He also maintained contacts with the compatriots he encountered during his travels. From this great expedition, which was undoubtedly a turning point in his life, he managed to bring back hundreds of valuable souvenirs, which still constitute one of the most interesting collections of the Kórnik Library of the Polish Academy of Sciences.

Meanwhile, in the territory under the Prussian partition, Władysław's uncle Jan Kanty, the last of the Działyński family, passed away at Kórnik Castle – a place familiar to Władysław. Since he had been without an heir, Jan Kanty had appointed his nephew as his heir apparent, and Władysław received this news while he was still travelling. At that moment, Zamoyski became the landowner and, perhaps more importantly, he took under his wing a valuable collection of manuscripts and precious prints accumulated by his grandfather and uncle, with the support of his parents, which was to become the Kórnik Library; as well as an extraordinary English garden, which already at that time constituted a valuable collection of trees and shrubs imported from various parts of the world – which became the Kórnik Arboretum.

After his return to the lands of the Prussian partition, Władysław began managing the estate. Despite considerable difficulties and burdens associated with this inheritance, Zamoyski managed to improve its financial condition. He was greatly supported in this by the castle administrator Zygmunt Celichowski, who looked after the Zamoyski estates in Wielkopolska until his death. Władysław supported all initiatives beneficial to the Poles. He also took up the fight against Prussian attempts to expel Polish citizens from their land. He actively supported the redemption – often even at a loss to himself – of estates threatened with seizure by the Germans. It was also at his personal initiative that Jadwiga Zamoyska established her School of Women's Home Work in Kórnik. Thanks to its establishment, girls of various backgrounds were able to acquire basic education as well as a trade, thus gaining the desired independence in many cases.

In 1885, the process of forced deportation began in the Prussian partitioned territories, specifically targeting the Polish-speaking population who had not acquired local citizenship. The Zamoyskis were on this list and were thus forced to leave their estates. They moved to the territory of the Austrian partition, to Galicia. Here they continued to support the Poles and joined in all activities aimed at fighting for the Polish cause. One of the biggest undertakings in which the Count became involved was participating in the bidding for the purchase of Zakopane and its surroundings. This decision was related to the indebtedness of his estates but turned out to be one of the best he ever made. On 9 May 1889, Władysław Zamoyski became the owner of the Zakopane estate, and it is only thanks to these events that this area is now within the borders of the Polish state. After acquiring these lands, he initiated active efforts to revitalise and modernise them. In Kuźnice near Zakopane, he eventually established the headquarters of the Women's Home School.

Everyone came to Władysław Zamoyski for support, and he provided it. There is probably no institution in the Zakopane area that does not owe him something. He supported, for example, the Tatra Museum, which is still in operation today, and the Zakopane branch of the Sokol Gymnastic Society, as well as many societies and occasional projects.

The area around Morskie Oko and Czarny Staw is one of the most beautiful and popular tourist destinations in the High Tatras. At the turn of the 20th century, the Fish Creek Valley was the site of a dispute over the course of the state border between Galicia (Polish lands under Austrian annexation) and Hungary. This conflict was exacerbated when the Prussian Prince Christian Hohenlohe-Öhringen became the owner of one of the nearby estates. At the same time, Władysław Zamoyski stood on the other side. The private conflict over the use of these lands eventually gained international publicity and forced the actual course of the disputed border to be determined unequivocally. The matter was brought before Emperor Franz Joseph, who decided to refer the dispute to an international arbitration court. With the financial support of Władysław, detailed historical queries were carried out to certify that the disputed area belonged to the former Poland. Members of the arbitral tribunal conducted a local inspection, during which the Polish population organised a patriotic demonstration. Finally, on 13 September 1902, the tribunal in Graz issued a verdict, awarding the entire disputed area to Galicia. This decision caused widespread enthusiasm among Poles, and today we can state that it is thanks to the Count's struggle that these territories now belong to the Polish state.

The decision to transfer the estate to the nation was taken by the Zamoyskis in Paris in 1912. On the 16th of February 1924, Maria and Władysław Zamoyski signed a donation act, under which they established the Zakłady Kórnickie Foundation, which was to take over the administration of their entire estate. The act on the Foundation came into force on the 30th of July 1925 – thus soon after the passing of the founder, who had died on 3 October 1924.

The year 2024 marks 100 years since the death of Władysław hr. Zamoyski, a great patriot, organizer, founder of the Kórnik Library, and the Kórnik Arboretum. In recognition of his services to the Polish state, the Senate of the Republic of Poland, by a resolution of 7 September 2023, established the year 2024 as the Year of Władysław Zamoyski.

Future of European tree species: how will climate change shape forests?

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Climate is a major determinant of species distributions at the coarsest spatial scales, determining continental patterns of species occurrences. Global environmental changes, especially related to climate warming and decreasing precipitation in the growing season, will significantly alter the climatic ranges of living organisms. With the development of open databases with species distributions and modeling tools, there are more and more studies aiming to assess the climatic range of numerous species. However, conclusions from most studies are burdened by biases in distributional data and uncertainties related to the choice of future climate scenarios and global circulation models. In this talk, I aim to present the workflow of species distribution model development and recent findings on the future of European tree species and forests, highlighting species that will expand their ranges and those that will lose their climatic optimum.

The development of a species distribution model requires two types of data: occurrence and environmental. Occurrence data usually cover presence-only, due to the lack of wide-scale inventories providing reliable absence data. For the detection of environmental drivers of species distributions, presence data within the area of interest should cover at least one edge of species distribution. The biggest dataset with occurrence data is Global Biodiversity Information Facility (GBIF), aggregating published and unpublished datasets, inventories, and citizen science observations from services like iNaturalist. However, coverage of occurrence points in GBIF is uneven, with higher representation in Western than Eastern Europe, as well as with higher representation of dense-populated areas. Before model development, it is highly recommended to check whether GBIF data with distributions of the species of interest cover the whole range of this species. In cases where points sparsely cover a significant part, it is necessary to use additional data sources, e.g., citizen science projects, national forest inventories, or vector range maps.

For environmental data, most researchers use gridded climatic variables from WorldClim2.1 or CHELSA databases. These are the easiest to obtain datasets of current and future climate, usually using 19 bioclimatic variables based on monthly temperature and precipitation data. These variables are not necessarily the best predictors of species occurrence, but due to the availability of future climate projections, they allow for forecasting shifts in climatic optimum. Similarly, due to the lack of global scenarios of soil characteristics changes with increasing human impact (e.g., erosion or nutrient deposition), these variables are either set as constant or omitted because on a larger spatial scale soil conditions are shaped by climate. Predicting future climatic suitability requires a clear statement about the timeline and intensity of climate change in such a timeline. Due to a lack of knowledge regarding how much the climate will change, IPPC provides different scenarios, developed by different teams of researchers. Each team uses different global circulation models (GCMs), which provide different values of bioclimatic variables. For that reason, this source of uncertainty has to be accounted for in studies, and a range of models should be used to minimize uncertainty.

The development of a species distribution model requires a particular algorithm. As many statistical tools allow for binary discrimination (estimation of presence/absence), only a few assume a lack of true absences. For that reason, the most frequently used algorithm is MaxEnt, available either as a stand-alone application or an R package. Other tools are generalized linear models, generalized additive models, and machine learning techniques, especially random forests, boosted regression trees, or neural networks. Regardless of the employed algorithm, the most important part of model development is training it on part of the data (usually 66–80% of observations) and using the remaining part as an independent validation set, to receive model performance statistics not affected by overfitting. After evaluation and assessment of its ecological soundness, the model allows for the assessment of climatic suitability in the area of interest. This value is sometimes incorrectly named 'occurrence probability', but as it is based on climatic data only and pseudoabsences; it is not true probability. Climatic suitability ranges from 0 to 1, and model evaluation allows determining the threshold between points within climatic optimum and outside it. Such determination is based on comparing observed presences and pseudoabsences with model outcomes assuming all values of thresholds and selecting the one balancing the specificity and sensitivity of the model, i.e., the proportions of true negative and true positive cases. After that, it is possible to present climatic suitability not as a continuous variable but as a binary outcome.

Our studies on forest species responses to climate change started in 2016 when we aimed to assess the responses of main forest-forming tree species in Europe to the changing climate (Dyderski et al. 2018). Specifically, we aimed to quantify changes in projected ranges and threat levels for the years 2061–2080 for 12 European forest tree species under three climate change scenarios. We combined tree distribution data from the GBIF, EUFORGEN, and forest inventories, and developed species distribution models using MaxEnt and 19 bioclimatic variables. Models were developed for three climate change scenarios – optimistic (RCP2.6), moderate (RCP4.5), and pessimistic (RCP8.5) – using three GCMs for the period 2061–2080. These three scenarios were provided by the 5th Assessment Report of IPCC. Our study revealed different responses of tree species to projected climate change. The species may be divided into three groups:

"winners" – mostly late-successional species: *Abies alba, Fagus sylvatica, Fraxinus excelsior, Quercus robur,* and *Q. petraea*; "losers" – mostly pioneer species: *Betula pendula, Larix decidua, Picea abies,* and *Pinus sylvestris,* and alien species – *Pseudotsuga menziesii*, *Q. rubra*, and *Robinia pseudoacacia*, which may also be considered as "winners". Assuming limited migration, most of the species studied would face a significant decrease in suitable habitat areas. The threat level was highest for species that currently have the northernmost distribution centers and are coniferous. This study reached a broad audience and increased attention to serious threats to forest-forming tree species in Central Europe, especially Scots pine and Norway Spruce. However, it was not the first study presenting these results (e.g., Sykes et al. 1996) nor the only one (e.g., Thurm et al. 2018). The results of this study are open to the general public by courtesy of Appsilon, a company that created a dedicated application to overview predicted shifts in tree distribution (https://connect.appsilon.com/future-forests/).

Our next step was to assess how much the outcome of the model is dependent on the used global circulation model (Paź-Dyderska et al. 2021). We used *Juglans regia*, a species of great importance for environmental management due to attractive wood and nutritious fruits, but also high invasive potential, as a model species. Using the MaxEnt algorithm, we prepared a species distribution model for the years 2061–2080 using 19 bioclimatic variables. We applied three emission scenarios, expressed by representative concentration pathways (RCPs): RCP2.6, RCP4.5, and RCP8.5, and three GCMs: HadGEM2-ES, IPSL-CM5A-LR, and MPI-SM-LR. Our study predicted a northward shift of the species, with simultaneous distribution loss at the southern edge of the current range, driven by increasing climate seasonality. Temperature Seasonality and Temperature Annual Range were the predictors of the highest importance. General trends are common for the projections presented, but the variability of our projections among the GCMs or RCPs applied (the predicted range will contract from 17.4% to 84.6% of the current distribution area) shows that caution should be maintained while managing *J. regia* populations. Adaptive measures should focus on maintaining genetic resources and assisted migration at the southern range edge, due to range contraction. Simultaneously, at the northern edge of the range, *J. regia* turns into an invasive species, which may need risk assessments and control of unintended spread. This study, together with Goberville et al. (2015) and Thuiller et al. (2019), highlighted the importance of GCM choice in species distribution models.

We also asked how much an increase in data coverage by unpublished observation can change the model outcome, as well as how many of the changes predicted for 2061–80 would be visible in a closer timeline (Puchałka et al. 2021). We used *Robinia pseudoacacia* as a model species, as it is one of the most frequent non-native species in Europe. To fill the gap in current knowledge of *R. pseudoacacia* distribution and improve the reliability of forecasts, we aimed to (1) determine the extent to which the outcome of range modeling will be affected by complementing *R. pseudoacacia* occurrence data with sites from Central, Southeastern, and Eastern Europe, (2) identify and quantify the changes in the availability of climate niches for 2050 and 2070, and discuss their impacts on forest management and nature conservation. The majority of the range changes expected in 2070 will occur as early as 2050. In comparison to previous studies, we demonstrated a greater eastward shift of potential niches of this species and a greater decline of potential niches in Southern Europe. Consequently, future climatic conditions will likely favor the occurrence of *R. pseudoacacia* in Central and Northeastern Europe where this species is still absent or relatively rare. Therefore, controlling the spread of *R. pseudoacacia* will require monitoring sources of invasion in the landscape and reducing the occurrence of this species. The expected effects of climate change will likely be observed 20 years earlier than previously forecast.

The next step after developing the model for *R. pseudoacacia* was to predict the future climatic optimum for other alien tree species in Europe (Puchałka et al. 2023). We compiled species occurrences from biodiversity databases, forest inventories, and literature data. We modeled the availability of potential niches using the MaxEnt method and bioclimatic variables for current conditions, the periods 2041–2060, and 2061–2080. Instead of RCPs from the 5th Assessment Report, we used shared socioeconomic pathways (SSPs) from the 6th Assessment Report (Riahi et al. 2017). We employed four climate scenarios: SSP126 (the most optimistic), SSP245 (intermediate), SSP370, and SSP485 (worst-case scenario). The results confirmed our hypotheses that coniferous species will contract, and deciduous trees will expand their climatic niche. A significant part of the areas where the studied species currently occur will be outside their climatic optimum in the coming decades, and changes in the climatic optimum distribution will be greater in the 2041–2060 period than in 2061–2080. These predicted shifts are relevant for evidence-based management in sites already occupied by the studied alien trees. Our results are also relevant to the development of prevention and early detection measures in areas predicted to become climatically suitable for the studied species.

In conclusion, I summarized the process of species distribution model development and our recent studies applying these models to European trees under a wide range of climate change scenarios. Coniferous forest-forming tree species will lose a significant part of their current climatic optimum, and in some parts of Europe they will potentially be replaced by broadleaved trees. Similar trends are predicted for invasive tree species: the retreat of conifers and the spread of broadleaved species. The discussed shifts will alter the functioning of European forests.

References

Dyderski M.K., Paź S., Freelich L.E., Jagodziński A.M. 2018. How much does climate change threaten European forest tree species distributions? Global Change Biology 24: 1150–1163.

Goberville E., Beaugrand G., Hautekèete N.-C., Piquot Y., Luczak C. 2015. Uncertainties in the projection of species distributions related to general circulation models. Ecology and Evolution 5: 1100–1116.

Paź-Dyderska S., Jagodziński A.M., Dyderski M.K. 2021. Possible changes in spatial distribution of walnut (Juglans regia L.) in Europe under warming climate. Regional Environmental Change 21: 18.

Puchałka R., Dyderski M.K., Vítková M., Sádlo J., Klisz M., Netsvetov M., Prokopuk Y., Matisons R., Mionskowski M., Wojda T., Koprowski M., Jagodziński A.M 2021. Black locust (Robinia pseudoacacia L.) range contraction and expansion in Europe under changing climate. Global Change Biology 27: 1587–1600.

Puchałka R., Paź-Dyderska S., Jagodziński A.M., Sádlo J., Vítková M., Klisz M., Koniakin S., Prokopuk Y., Netsvetov M., Nicolescu V.-N., Zlatanov T., Mionskowski M., Dyderski M.K. 2023. Predicted range shifts of alien tree species in Europe. Agricultural and Forest Meteorology 341: 109650.

Riahi K., van Vuuren D.P., Kriegler E., Edmonds J., O'Neill B.C., Fujimori S., Bauer N., Calvin K., Dellink R., Fricko O., Lutz W., Popp A., Cuaresma J.C., Kc S., Leimbach M., Jiang L., Kram T., Rao S., Emmerling J., Ebi K., Hasegawa T., Havlik P., Humpenöder F., Da Silva L.A., Smith S., Stehfest E., Bosetti V., Eom J., Gernaat D., Masui T., Rogelj J., Strefler J., Drouet L., Krey V., Luderer G., Harmsen M., Takahashi K., Baumstark L., Doelman J.C., Kainuma M., Klimont Z., Marangoni G., Lotze-Campen H., Obersteiner M., Tabeau A., Tavoni M. 2017. The Shared Socioeconomic Pathways and their energy, land use, and greenhouse gas emissions implications: An overview. Global Environmental Change 42: 153–168.

Sykes M.T., Prentice I.C., Cramer W. 1996. A bioclimatic model for the potential distributions of north European tree species under present and future climates. Journal of Biogeography 23: 203–233.

Thuiller W., Guéguen M., Renaud J., Karger D.N., Zimmermann N.E. 2019. Uncertainty in ensembles of global biodiversity scenarios. Nature Communications 10: 1446.

Thurm E.A., Hernandez L., Baltensweiler A., Ayan S., Rasztovits E., Bielak K., Zlatanov T.M., Hladnik D., Balic B., Freudenschuss A., Büchsenmeister R., Falk W. 2018. Alternative tree species under climate warming in managed European forests. Forest Ecology and Management 430: 485–497.

Contribution of quantitative genetics to adapting forests to climate change

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Keywords: adaptation, assisted migration, common garden, genetic variation, provenance, phenotypic plasticity, transplant studies

Tree populations are evolutionarily adapted to their environments. However, due to the ongoing and projected changes in climate and the associated alterations in the intensity/frequency of stresses, they may suffer increasing maladaptation. Provenance or common garden studies are forest experiments where progenies of populations adapted to a variety of environments are planted together in uniform environmental conditions. In such studies, thanks to the proper experimental lay-out and outplanting in several environments, it is possible to separate the effect of genetics and environment on the observed variation in phenotypic traits.

Provenance tests have been crucial in forestry for recognizing the patterns and extent of variation in many important phenotypic traits, the identification of seed sources suitable for use in commercial forestry, and for the delineation of seed/breeding zones in numerous tree species. Initially, they were mostly established for those commercial purposes. However, populations in such experiments are subject to environmental change, which includes changes in climate. Thus, they started to be used for examination of the effect of climate change on population performance. With some assumptions and limitations, provenance studies are ideal for this purpose.

In common garden studies, both the sampled populations and the planting sites could be described in terms of climatic variables. Consequently, the climatic transfers/distances to which populations have been moved (ecodistances; Mátyás 1994) can be defined as a difference between planting sites and population origin for a given climatic parameter. Based on those distances, statistical methods have been developed for studying population response to climate change in common garden studies.

In a given series of provenance tests, several populations of a given species are tested together. In the largest experiments of this type, close to or even over 100 populations and tens of planting sites are represented (Mátyás, Yeatman 1992, Shutyaev, Giertych 1997, Rehfeldt et al. 1999, Sáenz-Romero et al. 2017), but in most cases, due to technical and logistic constraints, those numbers do not exceed a few tens of populations and several planting sites, even in large experiments (Giertych, Oleksyn 1992, von Wühlisch 2004). There are also many smaller-scale series of experiments of this type, which are still useful for this purpose. However, it should be kept in mind that most of the existing provenance tests were not established strictly for examination of climatic response of populations, thus the extremes of a species climatic niche are rarely represented by sampled populations and planting sites. Variation in phenotypic performance of multiple populations to climatic transfer distances at a particular site is examined by the transfer functions approach (Rehfeldt et al. 1999). These functions reflect the local adaptation of maternal populations to their native environment and the effect of transferring them into new conditions (Wang et al. 2010). In contrast, in the response functions approach the performance of a given population across planting sites is examined (Wang et al. 2006). Both of the above approaches are integrated in the analysis of Universal Transfer Functions – UTF (Wang et al. 2010), which take into account the climate of planting sites and population origins.

Species distribution models (ecological niche models) are useful for predicting changes in suitable climatic niches for tree species under projected future climates. According to those predictions, most European tree species will show negative consequences of climate change at the lower latitudinal, elevational or xeric limits of distribution ranges with possible range contractions and/or shifts (Dyderski et al. 2018, Thurm et al. 2018, Buras, Menzel 2019, Chakraborty et al. 2021). However, many such models tend to ignore some mechanisms and phenomena that, in addition to climate, are important for shaping species distributions, such as biotic interactions, dispersal limitations and adaptive genetic variation (Pearson, Dawson 2003, Araujo, Peterson 2012). Thus, there is a growing need for incorporating the knowledge of within-species variation in adaptive traits into models predicting the future of forest tree species.

The slow rates of natural migration for most tree species make it unlikely that they could keep pace with the rapidly occurring changes in climate (Aitken et al. 2008). Thus, assisted migration of species is postulated as a measure that could help future forests to adapt to climate change. However, moving species outside their historic ranges, also called “assisted colonization”, may be environmentally risky and has a strong opposition (McLachlan et al. 2007, Hoegh-Guldberg et al. 2008). In contrast, with regard to tree species, it is more often postulated that individuals or genes be moved within the current range of the species (“assisted gene flow”) to facilitate adaptation of populations to projected changes in climate (Aitken, Bemmels 2015). In some countries, especially in North America, assisted migration measures are currently the active subject of experimentation (O’Neill et al. 2008b, Ukrainetz et al. 2011, Sáenz-Romero et al. 2020).

To help better parameterize species distribution models and to support assisted migration decisions there is a need for information on the extent and patterns of variation in phenotypic traits of adaptive importance and their climatic responsivity for many forest tree species. In fact, the models that integrate such information are starting to appear and provide a more refined picture of species response to projected climate changes (O’Neill et al. 2008a, Benito Garzón et al. 2011, 2019). The information can be gained from existing networks of provenance tests; however, there is a need for new common garden transplant studies that are specially designed for assessing the possibilities and effectiveness of assisted migration.

References

Aitken S.N., Bemmels J.B. 2015. Time to get moving: assisted gene flow of forest trees. Evolutionary Applications 9(1): 271–290.

Aitken S.N., Yeaman S., Holliday J.A., Wang T., Curtis-McLane S. 2008. Adaptation, migration or extirpation: climate change outcomes for tree populations. Evolutionary Applications 1: 95–111.

Araujo M.B., Peterson T. 2012. Uses and misuses of bioclimatic envelope modeling. Ecology 93: 1527–1539.

Benito Garzón M., Alía R., Robson T.M., Zavala M.A. 2011. Intra-specific variability and plasticity influence potential tree species distributions under climate change. Global Ecology and Biogeography 20: 766–778.

Benito Garzón M., Robson T.M., Hampe A. 2019. ΔTraitSDMs: species distribution models that account for local adaptation and phenotypic plasticity. New Phytologist 222: 1757–1765.

Buras A., Menzel A. 2019. Projecting tree species composition changes of European forests for 2061–2090 under RCP 4.5 and RCP 8.5 scenarios. Frontiers in Plant Science 9: 1986.

Chakraborty D., Móricz N., Rasztovits E., Dobor L., Schueler S. 2021. Provisioning forest and conservation science with high-resolution maps of potential distribution of major European tree species under climate change. Annals of Forest Science 78: 26.

Dyderski M.K., Paź S., Frelich L.E., Jagodziński A.M. 2018. How much does climate change threaten European forest tree species distributions? Global Change Biology 24: 1150–1163.

Giertych M., Oleksyn J. 1992. Studies on genetic variation in Scots pine (Pinus sylvestris L.) coordinated by IUFRO. Silvae Genetica 41: 133–143.

Hoegh-Guldberg O., Hughes L., McIntyre S., Lindenmayer D.B., Parmesan C., Possingham H.P., Thomas C.D. 2008. Assisted colonization and rapid climate change. Science 321: 345–346.

Mátyás C. 1994. Modeling climate change effects with provenance test data. Tree Physiology 14: 797–804.

Mátyás C., Yeatman C.W. 1992. Effect of geographical transfer on growth and survival of jack pine (Pinus banksiana Lamb) populations. Silvae Genetica 41: 370–376.

McLachlan J.S., Hellmann J.J., Schwartz M.W. 2007. A framework for debate of assisted migration in an era of climate change. Conservation Biology 21: 297–302.

O'Neill G.A., Hamann A., Wang T. 2008a. Accounting for population variation improves estimates of the impact of climate change on species' growth and distribution. Journal of Applied Ecology 45: 1040–1049.

O'Neill G.A., Ukrainetz N.K., Carlson M.R., Cartwright C.V., Jaquish B.C., King J.N., Krakowski J., Russell J.H., Stoehr M.U., Xie C., Yanchuk A.D. 2008b. Assisted migration to address climate change in British Columbia: recommendations for interim seed transfer standards. Technical Report 048. B.C. Ministry of Forests and Range, Research Branch, Victoria.

Pearson R.G., Dawson T.P. 2003. Predicting the impacts of climate change on the distribution of species: Are bioclimate envelope models useful? Global Ecology and Biogeography 12: 361–371.

Rehfeldt G.E., Ying C.C., Spittlehouse D.L., Hamilton D.A. 1999. Genetic responses to climate in Pinus contorta: Niche breadth, climate change, and reforestation. Ecological Monographs 69: 375–407.

Sáenz-Romero C., Lamy J.-B., Ducousso A., Musch B., Ehrenmann F.F., Delzon S., Cavers S., Chałupka W., Dağdaş S., Hansen J.K., Lee S.J., Liesebach M., Rau H.-M.M., Psomas A., Schneck V., Steiner W., Zimmermann N.E., Kremer A. 2017. Adaptive and plastic responses of Quercus petraea populations to climate across Europe. Global Change Biology 23: 2831–2847.

Sáenz-Romero C., O'Neill G., Aitken S.N., Lindig-Cisneros R. 2020. Assisted migration field tests in Canada and Mexico: lessons, limitations, and challenges. Forests 12: 9.

Shutyaev A.M., Giertych M. 1997. Height growth variation in a comprehensive Eurasian provenance experiment of (Pinus sylvestris L.). Silvae Genetica 46: 332–349.

Thurm E.A., Hernandez L., Baltensweiler A., Ayan S., Rasztovits E., Bielak K., Zlatanov T.M., Hladnik D., Balic B., Freudenschuss A., Büchsenmeister R., Falk W. 2018. Alternative tree species under climate warming in managed European forests. Forest Ecology and Management 430: 485–497.

Ukrainetz N.K., O'Neill G.A., Jaquish B. 2011. Comparison of fixed and focal point seed transfer systems for reforestation and assisted migration: A case study for interior spruce in British Columbia. Canadian Journal of Forest Research 41: 1452–1464.

von Wühlisch G. 2004. Series of international provenance trials of European beech. Improvement and Silviculture of Beech. Proceedings from the 7th International Beech Symposium. IUFRO Research Group 1.10.00. 10–20 May 2004, Tehran, Iran, pp. 135–144.

Wang T., Hamann A., Yanchuk A., O'Neill G.A., Aitken S.N. 2006. Use of response functions in selecting lodgepole pine populations for future climates. Global Change Biology 12: 2404–2416.

Wang T., O'Neill G.A., Aitken S.N. 2010. Integrating environmental and genetic effects to predict responses of tree populations to climate. Ecological Applications 20: 153–163.

Carbon storage estimation in forest ecosystems – research and practice

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Keywords: biomass production, allometric equations, stand density, stand features, BCEF

The forest ecosystem is a complex ecological system dominated by biogeographically specific flora and vegetation, with a particularly high proportion of trees growing in close proximity, which – together with the fauna, the fungi, numerous other microorganisms, the specific climate (including the local climate which the forest contributes to), the soil and the water regime – form a system (network) of interactions, links and interdependencies (Jagodziński 2022).

The importance of forests cannot be underestimated. As one of the most important components of the terrestrial ecosystems, forests occupy ca. 31% of the Earth's land surface (FAO 2022). They provide many ecosystem services which are essential for human well-being (Brockerhoff et al. 2017, Aznar-Sánchez et al. 2018). Forests are critical habitats for biodiversity and they play an essential role in its protection, as a host of many species of animals, plants, fungi and microorganisms; many of them cannot live anywhere else (Jagodziński 2023). They serve as vital shields against extreme weather conditions, e.g. in controlling floods by regulating the water flow in rivers and smaller streams. They regulate water supplies. They prevent erosion by reducing the force of rainfall and by absorbing water. Forests influence regional climates, as the presence of forests alters the climate through modifications in the rates at which heat and water are exchanged. Forests are also a resource of timber and non-timber forest products, e.g. fruits, mushrooms, or medicinal plants. They support cultural and recreational values through the non-material benefits that people derive from nature.

Forests help to stabilize the climate and play a crucial role in mitigating climate change by acting as a carbon sink, absorbing atmospheric CO_2 (Kauppi et al. 2022). They account for more than 60% of the total biomass and over 50% of carbon stocks, and on a global scale, they are a major contributor to carbon uptake and storage. Climate change – a global phenomenon – considered as a change in the average climatic patterns over several decades or longer, is expected to worsen in the next century (Santos et al. 2022). Even if forests alone cannot prevent further warming of the globe *via* carbon capture, they are considered to be an important dynamic carbon pool that reduces the negative effects of human activities on our climate. Carbon is stored in five distinct pools in forest ecosystems, i.e.

in (1) above- and (2) belowground live biomass of trees, shrubs and herbaceous plants, in (3) deadwood, in (4) forest floor litter, and (5) soil.

There are many methods for estimating stand biomass and carbon storage. Since the carbon concentrations in tree biomass components are relatively constant (ca. 50%), most studies focus on stand biomass determination. A stand biomass may be estimated both at the tree and the stand level. The most commonly used method at the tree level is the allometric equation method. Individual tree biomass estimation is based on simply measured tree variables within the stand, e.g. tree diameter at breast height (DBH) and/or tree height (H), and a few sample trees are harvested so that their dry biomass can be determined in the laboratory. The biomass for a plot or a stand level is determined based on allometric equation(s) developed on sample trees, and it is the sum of the biomasses of individual trees. This destructive method is the most precise for biomass estimation, but it is very time- and cost-consuming. However, the main source of error during the estimation of biomass (and then carbon storage) is the choice of the allometric equation. Error in the biomass estimation could be minimized by using original and separate allometric equations for different diameter range classes, and within the range of tree diameters by using an equation specifically developed for determining the biomass of that particular diameter class.

The stand biomass may also be determined based on stand biomass models or biomass expansion and conversion factors (BCEFs). The BCEF is the ratio of the stand biomass (or particular biomass components) to stand volume (kg m^{-3}). Recent studies have shown that stand biomass is related to some stand variables, such as quadratic mean diameter, average height, basal area of the stand, or stand volume. It should be noted that a constant BCEF used for the biomass determination of different stands may be a source of errors or biases, because the BCEF is dependent on the growth conditions of the stand, historical management practices, stand age or stand density. That is why linear or non-linear models have been established between stand volume and stand biomass. However, when more than one biomass component is analyzed for the stand (e.g. stems, branches, needles/leaves, cones, etc.), the stand biomass equation is developed to fit the total biomass and component biomass simultaneously The sum of stand biomass estimations from the several components biomass models and the total biomass models are the same. In such cases, to achieve the additivity of the stand biomass equations, non-linear seemingly unrelated regression and/or seemingly unrelated regression are the most common methods used.

We employed these methods in many research activities to analyze what factors determine tree biomass production and carbon storage; below a few examples were described.

Over the years 2004–2007, we studied the ecological consequences of Scots pine (*Pinus sylvestris* L.) silviculture in different stand densities. The specific objectives of our study were (1) to develop allometric equations for predicting aboveground standing biomass, and (2) to determine how above- and belowground biomass vary with initial stand density. The study was conducted in the Siemianice Experimental Forest (51°14.87'N, 18°6.35'E, 150 m a.s.l.). The stands were established in the spring of 1974, when two-year-old Scots pine seedlings were planted in nine different spacings (stand densities varied from 2500 to 20833 trees/ha^{-1}). From the onset of the experiment no cleanings or thinnings were made, and stand density was reduced only as a result of natural mortality. In 2003, we harvested 8–9 trees that represented the range of DBH in each spacing treatment. In total, we harvested 74 sample trees (aboveground biomass) and divided them into biomass components. All organs were weighed in the field to obtain fresh biomass. Based on the water content in the samples taken from the sample trees, we determined their dry biomass. To assess the aboveground standing biomass we developed a set of allometric equations (stand-specific) and used them to calculate tree biomass per stand area. The belowground biomass of Scots pine stands was calculated based on root excavation from soil pits.

We found that initial stand density had a clear effect on natural tree mortality, mean tree DBH, and total basal area. The total aboveground biomass ranged from 108 to 127 Mg ha^{-1} and was not related to initial stand density. The total belowground biomass ranged from 23 to 54 Mg ha^{-1} and decreased with increasing initial stand density. When the initial stand density increases, the total Scots pine stand biomass decreases. Based on the literature data, we also analyzed other stand features that were influenced by initial stand density (Jagodziński, Oleksyn 2009a–c).

There are many ecological processes that create carbon flux in forest ecosystems. However, when we adopt a global or country perspective we should focus on more general data to determine carbon storage in forest ecosystems (particularly tree stands) and its changes over time in relation to forest management practices. According to IPCC data, the mean carbon content in the aboveground biomass for conifers is 51%, whereas for broadleaved trees it is 48%. In comparison with our detailed data collected over many years, IPCC values are too high for most conifers and too low for broadleaved tree species. However, it is worth mentioning that biomass is more variable than carbon content, thus we should focus on collecting data on biomass.

During the research project "Remote sensing based assessment of woody biomass and carbon storage in forests", conducted in the years 2014–2018 (Biostrateg, The National Centre for Research and Development), we collected data on aboveground biomass for almost 3.5 thousand sample trees from 432 forest stands. We studied the following tree species stands: *Pinus sylvestris, Picea abies, Abies alba*, *Larix decidua*, *Quercus robur*, *Fagus sylvatica*, *Betula pendula*, and *Alnus glutinosa*. Our data range from young trees to 120-year-old stands, and the research sites cover the whole of Poland – and thus different habitat conditions. After measuring all the trees within the tree stand, we selected eight representative trees, which were cut and weighed. We measured the diameters and breast height of more than 100 trees in each stand, and at least 20% of tree heights. Based on DBH distributions, we selected 8 sample trees in each stand which were then harvested.

In the field, we determined the fresh biomass of aboveground biomass and its components, whereas in the laboratory all the samples were oven-dried and the dry biomass was measured. As a result, we determined the oven-dried biomass for all the sample trees and in each sample, we determined the carbon content. Based on these data, we developed site-specific allometric equations for the biomass estimation for each stand, we calculated stand volume and stand biomass, and as a result – the biomass conversion and expansion factors (BCEFs) for each biomass component and total aboveground biomass. Thus, using tree stand measurements we calculated the dry biomass of each tree and we summarized it per area unit.

We used site-specific allometric equations for biomass determination because biomass allocation changes with stand age. If we had used an equation based on old trees rather than young trees, we would have obtained overestimated values, as older trees consist of 80% stem, which in young trees accounts for less than 50%, and as we know from field works, the stem is the heaviest part of the tree.

Just for example, we compared our site-specific biomass estimation data for European larch trees (12 stands, 7–120 years old, 96 sample trees, DBH range: 1.9–57.9 cm) with other published data for this tree species for different biomass components (Jagodziński et al. 2018a). Our data for the aboveground biomass for individual trees are higher than from other studies. So using allometric equations developed in other site conditions may also lead to biomass underestimation. For this species, we provided age-specific and generalized allometric equations, biomass conversion and expansion factors (BCEFs), and biomass models based on forest stand characteristics. The aboveground biomass of European larch stands ranged from 4.46 (7-year-old forest stand) to 445.76 Mg ha^{-1} (106-year-old). We found that stand biomass increased with greater stand age, basal area, mean diameter, height and total stem volume, and decreased with greater density. Our BCEFs of the aboveground biomass and stem biomass were almost constant (mean BCEFs of 0.4688 and 0.3833 Mg m^{-3}, respectively). Moreover, our generalized models at the tree and stand level had a lower bias in predicting the biomass of the forest stands studied than other published models (Jagodziński et al. 2018a).

Within the same research grant, we provided a complete set of tree- and stand-level models for the biomass and carbon content of silver fir (*Abies alba*) (Jagodziński et al. 2019b). We established a set of 12 study plots to cover the whole chronosequence of *A. alba* tree stands from 8 to 115 years old. We measured tree stand structures, and destructively sampled the aboveground biomass of 96 sample trees (DBH range: 0.0–63.9 cm).

Based on the data collected, we provided tree-level models, BCEFs, and biomass models based on forest stand characteristics. We found that aboveground biomass ranged from 0.3 (8-year-old stand) to 293.6 Mg ha^{-1} (82-year-old stand), and stem biomass from 0.1 to 233.8 (8 and 82-year-old stands). Our study shows that the best predictor of biomass at the stand level is stand volume, whereas the worst are tree stand basal area and stand density.

Our models performed better than other published models, allowing for more reliable biomass estimation. We recommend biomass models instead of BCEFs at the stand level, as such models are less biased than BCEF models (Jagodziński et al. 2019b).

Within the research, we also answered the question of whether biomass production and allocation differ between various habitat conditions, i.e. mountain and lowland forests, and indicated how much neglecting this difference biases carbon storage estimations. We studied the chronosequences of 24 *Fagus sylvatica* and 24 *Picea abies* stands, located both in the lowlands and highlands of Poland. We cut and weighed 192 sample trees, and we measured the carbon content in the aboveground biomass components of trees (i.e. wood, bark, branches and leaves/needles).

Based on the data collected, we developed allometric tree- and stand-level models of biomass and carbon mass, and using log-transformed linear models we checked the effect sizes of elevation category on estimate output. Our study revealed small and statistically insignificant differences in biomass allocation patterns, carbon content, and allometric trajectories in trees between lowland and highland stands for both species studied. Moreover, the tree-level allometric models without habitat category had greater accuracy than the models including elevation. As in the previous studies, the aboveground biomass and carbon stock of the species studied were positively correlated with stand volume, age, basal area and height, and negatively with stand density. We also found that the differences between lowland and highland stands were low (Jagodziński et al. 2020).

Our studies have shown that if we use stand merchantable volume per hectare, we may determine the carbon mass stored in the stand aboveground biomass with greater accuracy, however, for younger stands without merchantable volume, other stand parameters must be taken into account (Jagodziński et al. 2014, 2017, 2018b). Our predicted values of carbon storage in the aboveground biomass are very precise for all the species studied. Thus we need local solutions to precisely determine carbon storage in forest stand biomass (e.g. Jagodziński et al. 2019a). The estimations of the aboveground biomass of the stands studied using the values provided by the IPCC guidelines are distinctly higher than those determined by local models, and our BCEFs are lower than those published by the IPCC. Thus to precisely determine local biomass and carbon storage in forest stands we should use regional models for biomass and carbon determination in stands. The variability of BCEFs is geographically based and related to climate conditions.

Our studies allow more general conclusions to be drawn: (1) carbon storage in a forest stand depends on tree stand parameters, (2) forest inventories are a good source of data that may be used for country-level carbon reporting, (3) generalizations from models are limited, thus regional approaches provide more reliable results, and (4) that further investigations are important for calculating the carbon resources in other components of forest ecosystems (not only tree stands).

The management of forest ecosystems may play an important role both in climate change mitigation and adaptation. Sustainable and multifunctional forest management may play a key role in mitigating the negative effects of increasing carbon dioxide concentration in the atmosphere, and thus in slowing global warming. Forest ecosystems are a very important carbon reservoir, and organic carbon may be stored both in living and dead biomass, as well as in soils. However, it is extremely important to increase the accuracy of carbon estimations in forest ecosystems at local scales, as well as regional and global scales.

References

Aznar-Sánchez J., Belmonte-Ureña L.J., López-Serrano M.J., Velasco-Muñoz J.F. 2018. Forest ecosystem services: An analysis of worldwide research. Forests 9: 453.

Brockerhoff E.G., Barbaro L., Castagneyrol B., Forrester D.I., Gardiner B., González-Olabarria J.R., Lyver P.O'B., Meurisse N., Oxbrough A., Taki H., Thompson I.D., van der Plas F., Jactel H. 2017. Forest biodiversity, ecosystem functioning and the provision of ecosystem services. Biodiversity Conservation 26: 3005–3035.

FAO 2022. The State of the World's Forests 2022. Forest pathways for green recovery and building inclusive, resilient and sustainable economies. FAO, Rome. https://doi.org/10.4060/cb9360en

Jagodziński A.M. 2022. Prognozowane problemy z utrzymaniem trwałości lasów wobec postępujących zmian klimatycznych. In: Szabla K. (ed.). Leśnictwo przyszłości. Polskie Towarzystwo Leśne, Stare Jabłonki, pp. 55–75.

Jagodziński A.M. 2023. Różnorodność biologiczna ekosystemów leśnych a zmiany klimatyczne. In: Skrzecz I., Sikora K. (eds.). Wpływ zmian klimatu na środowisko leśne. Instytut Badawczy Leśnictwa, Sękocin Stary, pp. 19–45.

Jagodziński A.M., Dyderski M.K., Gęsikiewicz K., Horodecki P. 2018a. Tree- and stand-level biomass estimation in a Larix decidua Mill. chronosequences. Forests 9: 587.

Jagodziński A.M., Dyderski M.K., Gęsikiewicz K., Horodecki P. 2019a. Effects of stand features on aboveground biomass and biomass conversion and expansion factors based on a Pinus sylvestris L. chronosequence in Western Poland. European Journal of Forest Research 138: 673–683.

Jagodziński A.M., Dyderski M.K., Gęsikiewicz K., Horodecki P. 2019b. Tree and stand level estimations of Abies alba Mill. aboveground biomass. Annals of Forest Science 76: 56.

Jagodziński A.M., Dyderski M.K., Gęsikiewicz K., Horodecki P., Cysewska A., Wierczyńska S., Maciejczyk K. 2018b. How do tree stand parameters affect young Scots pine biomass? – Allometric equations and biomass conversion and expansion factors. Forest Ecology and Management 409: 74–83.

Jagodziński A.M., Dyderski M.K., Horodecki P. 2020. Differences in biomass production and carbon sequestration between highland and lowland stands of Picea abies (L.) H. Karst. and Fagus sylvatica L. Forest Ecology and Management 474: 118329.

Jagodziński A.M., Kałucka I., Horodecki P., Oleksyn J. 2014. Aboveground biomass allocation and accumulation in a chronosequence of young Pinus sylvestris stands growing on a lignite mine spoil heap. Dendrobiology 72: 139–150.

Jagodziński A.M., Oleksyn J. 2009a. Ekologiczne konsekwencje hodowli drzew w różnym zagęszczeniu. I. Wzrost i rozwój drzewostanu [Ecological consequences of silviculture at variable stand densities. I. Stand growth and development]. Sylwan 153(2): 75–85.

Jagodziński A.M., Oleksyn J. 2009b. Ekologiczne konsekwencje hodowli drzew w różnym zagęszczeniu. II. Produkcja i alokacja biomasy, retencja biogenów [Ecological consequences of silviculture at variable stand densities. II. Biomass production and allocation, nutrient retention]. Sylwan 153(3): 147–157.

Jagodziński A.M., Oleksyn J. 2009c. Ekologiczne konsekwencje hodowli drzew w różnym zagęszczeniu. III. Stabilność drzewostanu, fitoklimat, różnorodność biologiczna [Ecological consequences of silviculture at variable stand densities. III. Stand stability, phytoclimate and biodiversity]. Sylwan 153(4): 219–230.

Jagodziński A.M., Zasada M., Bronisz K., Bronisz A., Bijak S. 2017. Biomass conversion and expansion factors for a chronosequence of young naturally regenerated silver birch (Betula pendula Roth) stands growing on post-agricultural sites. Forest Ecology and Management 384: 208–220.

Kauppi P.E., Stål G., Arnesson-Ceder L., Sramek I.H., Hoen H.F., Svensson A., Wernick I.K., Högberg P., Lundmark T., Nordin A. 2022. Managing existing forests can mitigate climate change. Forest Ecology and Management 513: 120186.

Santos F.D., Ferreira P.L., Pedersen J.S.T. 2022. The climate change challenge: A review of the barriers and solutions to deliver a Paris solution. Climate 10(5): 75.

Genomics of population history and adaptive variation in forest trees

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Keywords: population structure, local adaptation, natural selection, SNP genotyping, genetic markers

Many keystone temperate forest tree species have broad distribution ranges and occupy habitats across steep climatic gradients. In response to varying selection pressures across time and heterogeneous environments, they not only maintain a high level of background genetic variation often uniformly distributed at large geographical areas, but they also show considerable levels of phenotypic plasticity and exhibit signatures of local adaptation due to natural selection, despite extensive gene flow between populations. These mechanisms likely evolved to ensure persistence in the face of environmental upheavals, and are of crucial importance in long-lived organisms with limited mobility and a lack of behavioural responses to changing environments.

Increasing pressure on forest ecosystems due to environmental change raises the question of tree conditions and performance, which entails considering their specific life history traits (longevity, long generation time), complex demographic histories, and population structures. Therefore, a better understanding of the link between the geographical distribution of adaptive genetic variation and its molecular signatures, along with the associations between genomes and the environment, is essential. These insights could inform decisions regarding the management of both economically important commercial forests and natural forests of great ecological value that are under increased pressure from environmental changes. However, for many highly outcrossing and wind-pollinated species, such as forest trees, it is mostly unclear how the past population history events influenced the species' genetic structure, and what the genetic architecture of adaptive traits is. Until recently, due to complex genomes, scarce genomic resources, and the usually low resolution of available genetic markers, it was difficult to conduct thorough investigations of past population processes in such non-model plant species as forest trees, and especially conifers.

Study system

Scots pine (*Pinus sylvestris* L.) is a highly outcrossing and wind-pollinated forest tree species that naturally forms random mating populations of high ecological and economic importance in the Northern Hemisphere. The ecological niche of the species is broad, with natural populations covering large parts of Europe and Asia at a distance of more than 14,000 km from western Scotland to eastern Siberia. Due to its great geographical and ecological range, numerous ecotypes of the species have been defined, reflecting its phenotypic and ecophysiological variation. Clear clinal patterns of adaptation to local environmental conditions have been established since postglacial migration, as evident in growth traits, phenology, abiotic stress resistance, and responses to light spectra. Patterns of population differentiation are generally clinal across the range and vary with latitude.

Wide distribution range and current patterns of genetic variation in *P. sylvestris* is the result of several interwoven factors, including demographic changes in distribution and population sizes of different lineages associated with alternating glacial-interglacial periods of Quaternary, past and current levels of inter- and intra-population gene flow, and adaptation to a heterogeneous environment along multiple environmental clines. However, increasing anthropogenic pressures that affect levels of genetic diversity in forest tree species, coupled with unprecedented environmental changes, might influence its adaptive potential, patterns of distribution and mortality rates across its range. Therefore, analysis of the spatial distribution of genetic variation and adaptive potential of Scots pine across its range is crucial to evaluate the climate vulnerability of wild populations and identify those that might exhibit greater resilience to changing environments. This information can guide strategies for enhancing forest management and conservation, ensuring the continued health of culturally and economically significant forests.

Novel genomic resources and applications

So far, population structure assessments using maternally inherited and seed-dispersed mitochondrial (*mt*DNA) markers have been largely limited to variation in a few polymorphic markers. However, due to low spatial resolution of the markers, the population structure of the species has only been achieved at a broad scale. Novel genomic resources have recently been developed for Scots pine and its close relatives with the use of next-generation sequencing approaches (Wachowiak et al. 2015, Donnelly et al. 2017). Among other things, they provide information on the polymorphisms in large fragments of resequenced mitochondrial genomes (Donnelly et al. 2017) and were used for the development of population genetic studies on a set of novel mitochondrial markers (Łabiszak et al. 2019, Zaborowska et al. 2020). Due to the massive size of conifer genomes (over 20 Gbp), whole-genome sequencing for population-level studies is not cost-effective in this group of taxa. Therefore, reduced representation genotyping, such as Axiom single nucleotide polymorphism (SNP) chips, offers a cost-effective and high-throughput alternative for studying genetic variation at population level.

Such custom genotyping arrays (Affymetrix, Thermo Fisher Scientific, US) have recently been developed for Scots pine, including the Axiom PineGAP (Perry et al. 2020) and PiSy50k SNP arrays (Kastally et al. 2021), which comprise thousands of polymorphic loci discovered in transcriptome, exon capture and candidate genes resequencing in Scots pine.

In the presented research, polymorphisms at mitochondrial DNA markers were used to investigate genetic variation within and among Scots pine populations sampled across its broad distribution range. The aim was to search for evidence of the genetic structure of the population in Scots pine and the possible admixture of populations of different origins that might have contributed to the establishment of different ecotypes of the species in Europe and Asia. Then, information from thousands of SNPs markers genotyped across dozens of natural populations of Scots pine was used to better understand the relationship between evolutionary history and demographic factors in this widespread species with a large and complex genome. Such detailed knowledge regarding the neutral genetic structure of the species is necessary when searching for the molecular signatures of natural selection affecting phenotypic variation and local adaptation of ecotypes distributed over a broad geographic range, and it advances the search for genomic regions involved in population divergence. Finally, the genetic analysis of the markers was used to advance our understanding of the genetic basis of phenotypic divergence and adaptation within species, and for the development of predictive models to forecast the likely impacts of environmental changes on the populations.

Signatures of population structure

The mitochondrial markers revealed only a weak population structure in Central and Eastern Europe and suggested postglacial expansion to the middle and northern latitudes from multiple sources. Major mitotype lineages include: the remnants of Scots pine at most-western distribution of the species that colonized the Scottish Highlands; two main lineages (western and central European) that contributed to the contemporary populations in Norway and Sweden; the central-eastern European linage that colonized Baltic countries – Finland, and Russian Karelia; and a separate linage common to most eastern European parts of Russia and western Siberia (fig. 1). We also observed the signatures of a distinct refugium located in the northern parts of the Black Sea basin that contributed to the patterns of genetic variation observed in several populations in the Balkans, Ukraine, and western Russia (Wachowiak et al. 2023).

The findings also show evidence of past admixture events between genetic clusters that were retained despite the potential for effective pollen-mediated gene flow across the species' distribution range, as evident from SNPs markers analysis. The results indicate that Scots pine populations from the main range, including populations from northern and southern margins, share a similar and relatively high level of neutral genetic diversity, but the amount of variation significantly drops within the easternmost population in Russia (fig. 2). Central

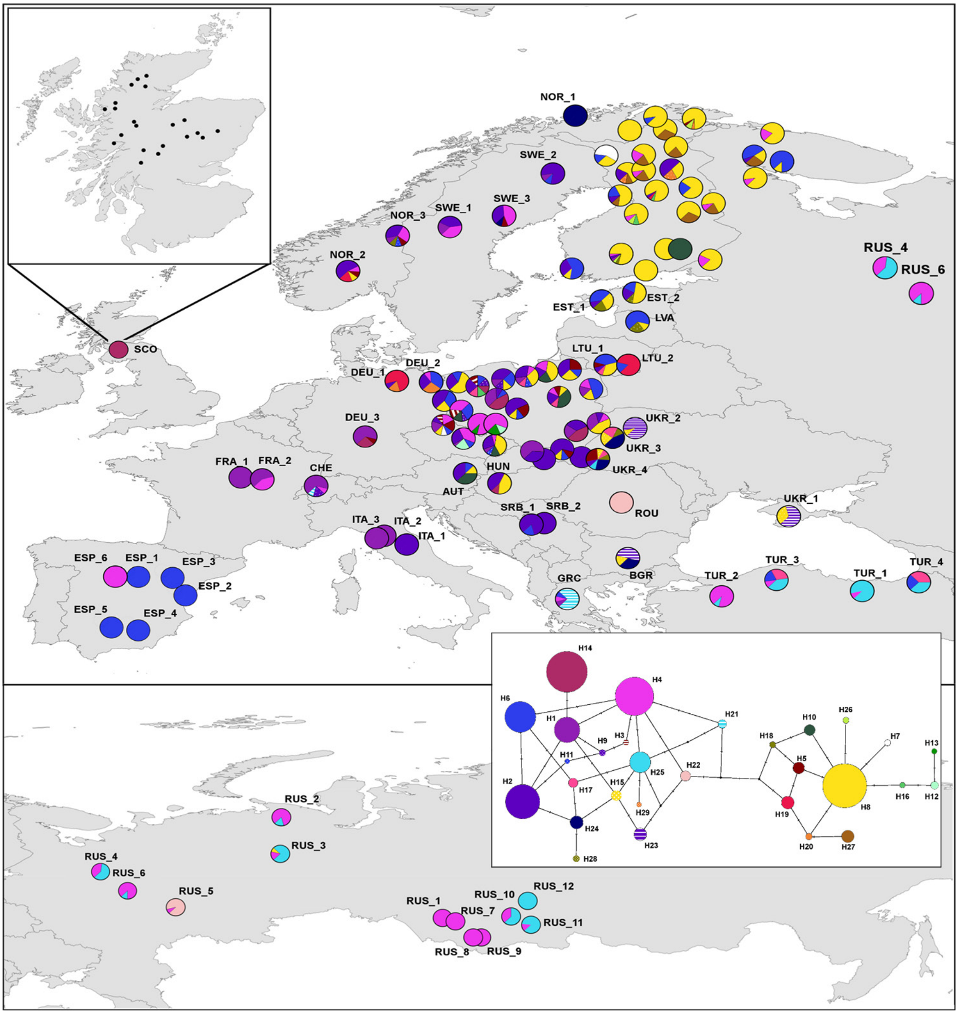

Fig. 1. Network of haplotypes and their distribution in the analyzed populations of Scots pine

Europe was likely the admixture zone between different lineages during the last glacial cycle and possibly in previous glaciation events. Fennoscandia was likely recolonized from two different sources, as evident from the differentiation found between Swedish and Norwegian vs. Finnish populations, with the latter being the most divergent lineage in the main range of Scots pine. The study describes the signatures of distinct genetic lineages persisting across the species distribution range, despite the most recent population range shifts during the last glaciation and possible gene flow following recolonization of Europe (Łabiszak, Wachowiak 2023).

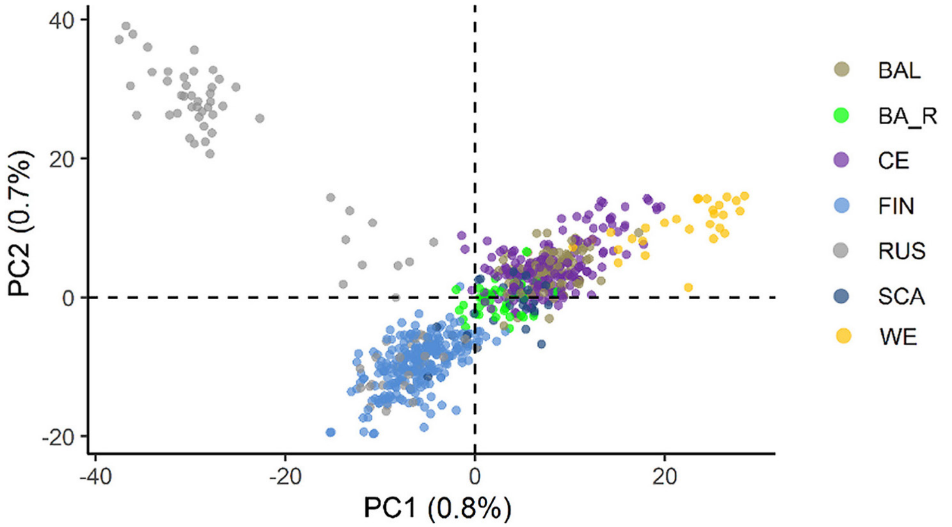

Fig. 2. Principal component analysis (PCA) projections of *P. sylvestris* individuals from the Eurasian distribution range. Total variance explained by each principal component is indicated within parentheses next to the respective principal component header. Different colors reflect the regional groups; however, all populations from Russia (RUS) share the same color, see the description in the main text for details of population grouping

Past demographic events

Phylogenetic analyses and demographic modeling suggest that the observed divergence patterns between genetic lineages likely predate the last glaciation events. Two of the most distinctive groups are represented by trees from the eastern parts of Fennoscandia and Eastern Russia, which have remained separated since the mid-Pleistocene. The patterns of genetic variation also confirm the dual

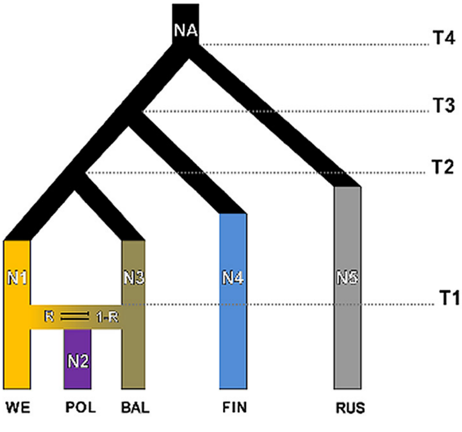

Fig. 3. Schematic representation of the best-fit model inferred by fastsimcoal2 software, showing genetic relationships between five inferred genetic groups of Scots pine in the Eurasian distribution range of the species. We – Western Europe, POL – Poland, BAL – Balkans, FIN – Finland, RUS – Russia. Parameters acronyms: NA: ancestral population effective size; N1:N5: effective population sizes for the five geographical regions analyzed; T1:T4: time for four divergence events in years; R: migration per generation after the divergence (arrows indicate migration direction)

colonization of Fennoscandia and the existence of an admixture zone in Central Europe that was formed during multiple waves of postglacial recolonization (fig. 3). The results suggest that, besides the most recent and well-studied demographic changes during the Holocene period, mid-Pleistocene events influenced the current spatial structure of genetic diversity of tree populations across Europe (Łabiszak, Wachowiak 2023).

Maintenance of genetic identity despite gene flow

Thousands of SNP markers derived from the recently developed Axiom pine genotyping array (Perry et al. 2016) were applied to examine genetic variation across a broad geographical range of Scots pine. The research combined the phylogeographic approach, inference of population structure, and demographic analysis in an approximate Bayesian computation framework to elucidate patterns of distribution and population genetic diversity. The results indicate that the signatures of distinct and ancient lineages can still be detected using high-resolution markers, despite the overall low level of population differentiation across the large geographic distances anticipated for tree species with such effective wind-dispersing mechanisms and mostly continuous distribution range (fig. 4). Notably, the most

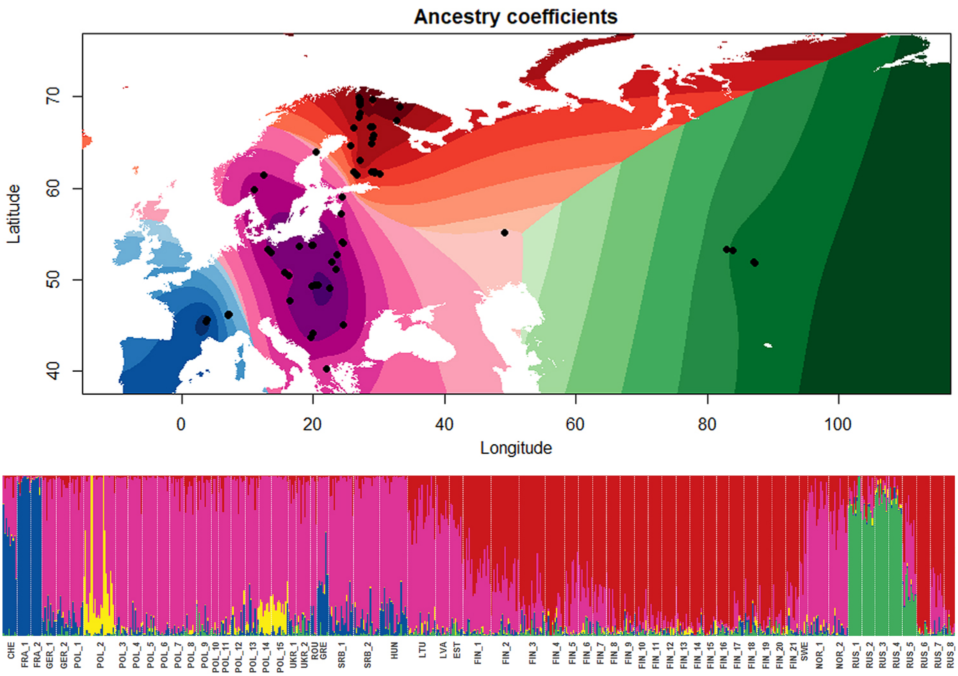

Fig. 4. Bayesian analysis of genetic structure. Plot of the ancestry coefficients of each individual obtained with STRUCTURE based on the admixture model and correlated allele frequencies for K = 5. Different colors of bars correspond to the inferred ancestries

recent range shifts during the last glaciation and possible gene flow following the recolonization of Europe have not erased the signatures of those distinct genetic lineages in Scots pine distribution.

Molecular signatures of selection

Finally, the research demonstrates the molecular signatures of selection in Scots pine and reveals that temperature is the primary factor influencing levels of genetic variation and driving local adaptation in *P. sylvestris*, offering insights for predicting genomic offset in this species. The population genomic approach indicates that local adaptation in Scots pine is primarily influenced by selection acting on numerous loci of small effect. As shown in other plant systems, climate-associated traits were found to be polygenic across many species, with a complex genetic architecture, shaped in response to environmental gradients through multilocus selection. Selection must be high enough to overcome the homogenizing effect of high level gene flow in order to maintain local adaptation. Despite the presumably polygenic character of many traits related to environmental gradients and the usually weak allelic frequency spectra defined with outlier detection approaches, we identified a high correlation of variation at certain loci with temperature that seems to play a key role in local adaptation at the analyzed distribution range of the species. However, the results show that standing genetic variation in many places may be maladapted to rapidly changing environments, and alternative approaches to natural regeneration, might be needed to ensure the resilience of Scots pine populations in the face of climate change (Łabiszak, Wachowiak, in preparation).

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References

Donnelly K., Cottrell J., Ennos R.A., Vendramin G.G., A'Hara S., King S., Perry A., Wachowiak W., Cavers S. 2017. Reconstructing the plant mitochondrial genome for marker discovery: A case study using Pinus. Molecular Ecology Resources 17: 943–954.

Kastally C., Niskanen A.K., Perry A., Kujala ST., Avia K., Cervantes S., Haapanen M., Kesälahti R., Kumpula T.A., Mattila T.M., Ojeda D.I., Tyrmi J.S., Wachowiak W., Cavers S., Kärkkäinen K., Savolainen O., Pyhäjärvi T. 2021. Taming the massive genome of scots pine with PiSy50k, a new genotyping array for conifer research. bioRxiv 2021.2006.2029.450162.

Łabiszak B., Wachowiak W. 2023. Mid-Pleistocene events influenced the current spatial structure of genetic diversity in Scots pine (Pinus sylvestris L.). Journal of Systematics and Evolution (accepted). https://doi.org/10.1111/jse.13013

Łabiszak B., Zaborowska J., Wachowiak W. 2019. Patterns of mtDNA variation reveal complex evolutionary history of relict and endangered peat bog pine (Pinus uliginosa). AoB Plants 11: plz015.

Perry A., Wachowiak W., Downing A., Talbot R., Cavers S. 2020. Development of a single nucleotide polymorphism array for population genomic studies in four European pine species. Molecular Ecology Resources 20: 1697–1705.

Wachowiak W., Trivedi U., Perry A., Cavers S. 2015. Comparative transcriptomics of a complex of four European pine species. BMC Genomics 16: 234.

Wachowiak W., Żukowska W.B., Perry A., Lewandowski A., Cavers S., Łabiszak B. 2023. Phylogeography of Scots pine in Europe and Asia based on mtDNA polymorphisms. Journal of Systematics and Evolution 61: 315–327.

Zaborowska J., Łabiszak B., Wachowiak W. 2020. Population history of European mountain pines Pinus mugo and Pinus uncinata revealed by mitochondrial DNA markers. Journal of Systematics and Evolution 58: 474–486.

Can we conserve exceptional forest tree species *ex situ* in the face of global climate change?

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Keywords: seed storage, cryopreservation, seed longevity, epigenetic, *in vitro* culture, gene banks, pedunculate oak

Classical seed storage, cryopreservation or *in vitro* culture?

Plant species *ex situ* conservation has taken on a new meaning, especially with the unprecedented rate of global climate change. Forest tree species produce seeds that are desiccation-tolerant and desiccation-sensitive. They all quickly lose their viability during storage, even in optimal conditions. Therefore, it is difficult to store them for decades *ex situ* in a gene bank, especially for short-lived seeds of recalcitrant species. To enable *ex situ* germplasm storage of forest tree species, we are focusing on seed biology, their desiccation sensitivity, sensitivity to low temperatures during classical storage at –18°C or/and to cryopreservation at –196°C. *In vitro* techniques are an integral part of the cryopreservation protocols which have been developed. Micropropagation is also used to clone old, historic trees, including monumental oaks, that are hundreds of years old. Experiments were carried out on biotechnological, physiological and molecular levels.

Seeds

The seed biology of forest tree species strongly varies across species and determines their desiccation sensitivity and the selection of the optimum approach to long-term storage. Some very resistant seeds can be desiccated to a very low level of moisture content (*c.* 2%, fresh weight basis) and stored in conventional conditions at a low temperature (e.g. *Alnus glutinosa, Betula pendula, Carpinus betulus, Prunus avium*). Furthermore, there are seeds with a slight sensitivity to desiccation, meaning a critical moisture content at *c.* 5% (e.g. *Fraxinus excelsior, Malus sylvestris, Tilia cordata, Salix* spp., *Populus* spp.). Other seeds, with a clear threshold during drying to *c.* 5–7%, are more sensitive (e.g. *Fagus sylvatica*). There are also

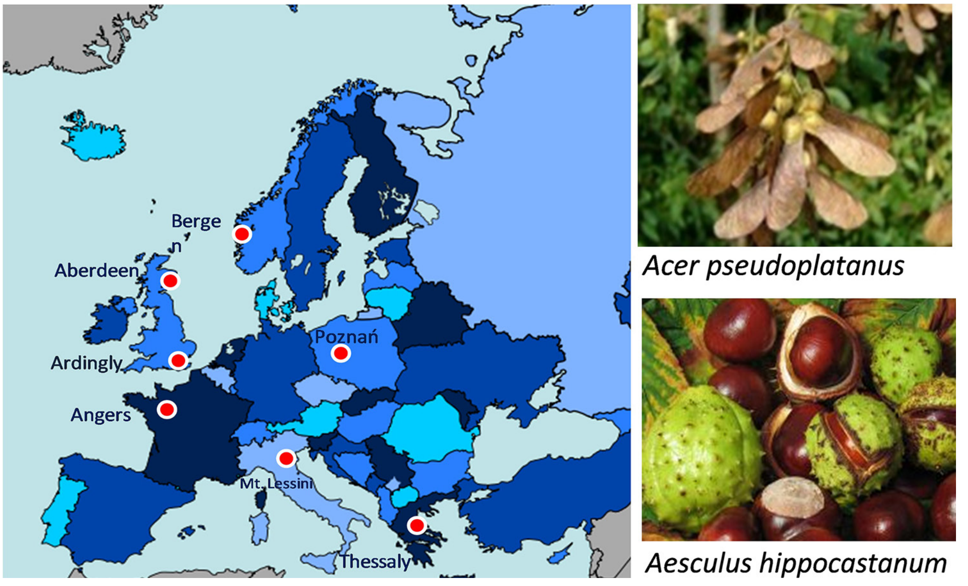


Fig. 1. Sycamore (*Acer pseudoplatanus*) and chestnut (*Aesculus hippocastanum*) seeds were collected in different parts of Europe (red dots; Daws et al. 2004, 2006)

forest tree species producing seeds very sensitive to desiccation, e.g. *Acer pseudoplatanus* (*c.* 24%), *Quercus robur* or *Q. petraea* (*c.* 40%). Environmental conditions (accumulated temperature) may differentiate the biological properties of recalcitrant seeds even within the same species, including their tolerance to desiccation (fig. 1). For example, seeds of *Acer pseudoplatanus* from Norway and Scotland were less tolerant to desiccation and survived desiccation only to 33–39% moisture content (mc), while seeds from the south of Europe – from countries like France and Italy, could be classified as suborthodox and survived desiccation to 15% mc (Daws et al. 2006). Seeds of *Aesculus hippocastanum* which developed in the north of Europe were smaller and more desiccation-sensitive in comparison to seeds from the south of Europe (fig. 1; Daws et al. 2004).

Orthodox and suborthodox seeds tolerate storage *ex situ* in tightly closed containers after desiccation to 5–10% mc, depending on the species, and at a temperature of –18°C (fig. 2).

Orthodox seeds

Our epigenetic studies concerning orthodox seeds of *Pyrus communis* showed some links between seed storage and the global level of 5-methylcytosine (m^5C; fig. 3; Michalak et al. 2013). Comparison of the percentage of DNA methylation revealed a relevant time-dependent increase in 5-methylcytosine. At the same time we did not observe significant differences in germination and seeding emergence of *P. communis* after 0 and 1 year of storage (8.4% seed mc; fig. 3). After 1 year of seed storage, the amount of m^5C in their DNA increased from 3.08 to 5.49% (fig.

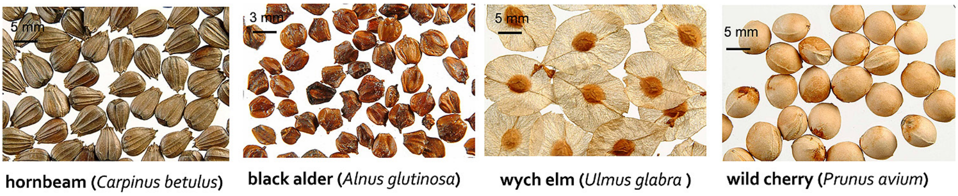


Fig. 2. Examples of forest tree species producing typical, desiccation-tolerant, orthodox seeds

3). The increase of m^{5}C can lead to stopped gene expression and lower production of protective proteins, which can cause a decrease in seed viability. We think that this method could be useful for early determination of seed aging during storage.

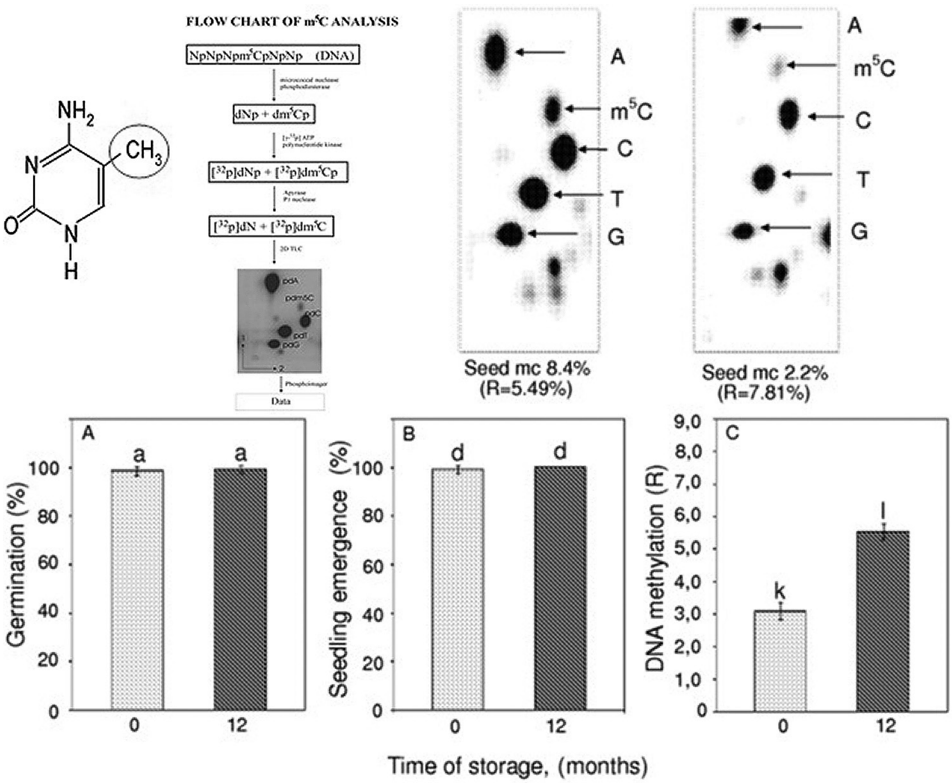
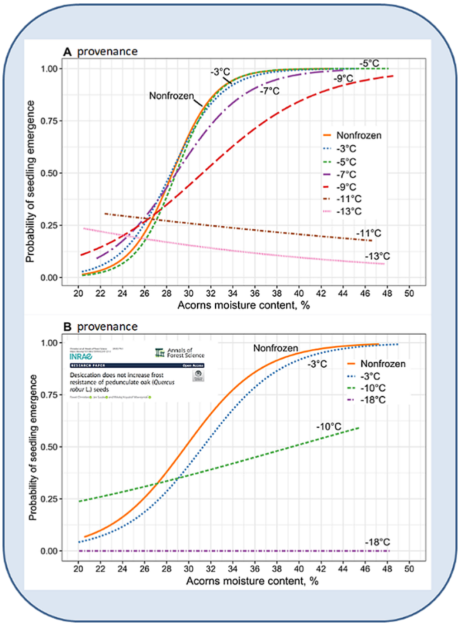


Fig. 3. Assessment of global DNA methylation of *Pyrus communis* seeds with different levels of moisture content (8.4 and 2.2%) or stored (mc 8.4%) for 0 and 1 year. 2-deoxynucleotides derived from DNA hydrolysis (labelled spots) and RNA contamination (unlabelled spots) are clearly separated. A – adenine, m^{5}C – 5-methylcytosine, C – cytosine, T – thymine, G – guanine. A method for determining the level of methylated cytosine according to Barciszewska et al. (2007). Data from Michalak et al. (2013)

Recalcitrant seeds

Recalcitrant seeds produced by oaks (acorns) do not tolerate a temperature of –18°C (Chmielarz et at. 2022), and cannot be stored as whole seeds in gene banks for longer periods. The probability of seedling emergence of non-frozen acorns with moisture content above 38% was 100% (fig. 4). Acorns with a moisture content of 30.7–30.9% and cooled to –3 or –5°C had a seedling emergence probability of 75%. The probability of seedling emergence from acorns desiccated before freezing at –3°, –5°, –7°, or –9°C to a moisture content range of 25.4–26.4% was only 25%. The probability of obtaining seedlings from acorns frozen for two weeks at –18°C was 0% (fig. 4).



pedunculate oak ***(Quercus robur)***

Fig. 4. Probability of *Quercus robur* seedling emergence depending on acorn moisture content and freezing temperature, i.e., –3°, –5°, –7°, –9°, –11° or –13°C (seed lot A) and –3°, –10°, or –18°C (seed lot B), for 2 weeks (Chmielarz et al. 2022)

Cryopreservation

Orthodox seeds

In the case of orthodox seeds, their cryopreservation is used as a backup to seeds stored by traditional methods. Safe seed moisture ranges have been developed for different forest tree species that can tolerate liquid nitrogen temperatures at the species specific seed moisture contents range. For example, for *Fraxinus excelsior* seeds it is 7.2–19.5% (mc of samaras; fig. 5; Chmielarz 2009a).

Seedling emergence for *Fraxinus excelsior* seeds treated with LN at mc 5.2% was significantly lower (3%) in comparison to control seeds. For seeds frozen at

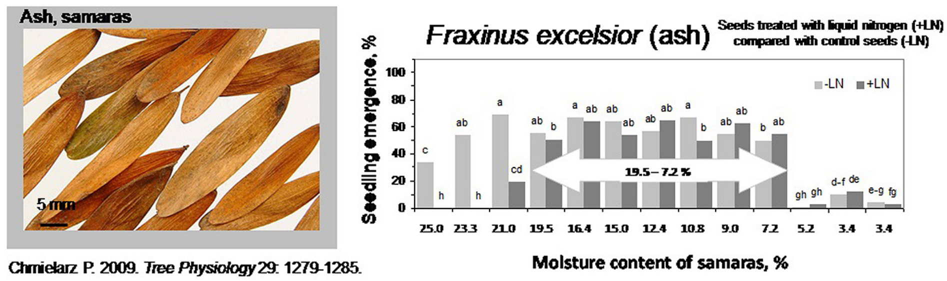


Fig. 5. *Fraxinus excelsior* seedling emergence after drying or moisturizing of seeds (samaras) to 13 levels of moisture content (mc) (3.4–25%, samaras), untreated -LN and treated +LN for 24 h with LN, provenance Poznań; $p < 0.05$, Tukey test (Chmielarz 2009a)

mc 21%, seedling emergence decreased from about 60% to 20% (fig. 5). At an even higher seed mc during cryostorage, no seedlings emerged after thawing was observed (fig. 5; Chmielarz 2009a).

Safe ranges of seed moisture content (%) for seeds (dormant and non-dormant) of tree species of Polish provenances are presented in tab. 1. The lower limit of the mc range is the critical moisture content of severely desiccated seeds (here in green colour). The upper limit of the range is the High Moisture Freezing Limit (HMFL) of seeds store in liquid nitrogen (in purple; tab. 1; Chmielarz 2009ab, Chmielarz 2010a-d).

Tab. 1. Safe ranges of seed moisture contents of orthodox forest tree species (dormant and non-dormant) stored in liquid nitrogen (–196°C)

	Species	Range
Species with dormant seed	common hornbeam (*Carpinus betulus*)	3.2–16.5
	European ash (*Fraxinus excelsior*)	7.2–19.5
	wild cherry (*Prunus avium*)	9.0–16.9
	small-leaved lime (*Tilia cordata*)	5.2–20.1
Species with non-dormat seeds	black alder (*Alnus glutinosa*)	2.7–19.2
	silver birch (*Betula pendula*)	2.0–23.2
	wych elm (*Ulmus globara*)	3.3–17.7

Recalcitrant seeds

In contrast to orthodox seeds, the cryopreservation of recalcitrant seeds is the only method (if it is possible to develop it for individual species) for storing the germplasm of a species *ex situ* in a gene bank. The cryopreservation of plumules isolated from acorns (shoot meristems, 1 mm in size) of *Quercus robur* (Chmielarz et al. 2011) and *Quercus petraea* (Wasileńczyk et al. 2024) allows the genetic resources of these two oak species, growing in Poland, to be conserved in gene banks. After thawing from liquid nitrogen, the regeneration of whole plants from

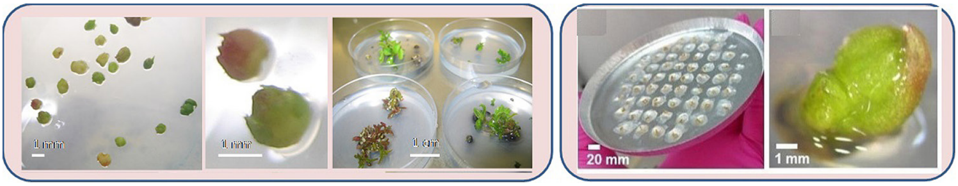


Fig. 6. Plumules of *Q. robur* (Chmielarz et al. 2011) on the left and plumules of *Q. petraea* (Wasileńczyk et al. 2024) regenerated *in vitro* after cryopreservation. For *Q. petraea* plumules were cryoprotected, desiccated and cooled in liquid nitrogen on aluminium cryo-plates covered by calcium alginate gel

plumules is carried out using *in vitro* culture (fig. 6; Chmielarz et al. 2011, Wasileńczyk et al. 2024). For *Q. petraea*, successful cryopreservation (Wasileńczyk et al. 2024) isolated plumules were attached to the cryo-plate by calcium alginate gel, then osmoprotected in solution containing 2.0 M glycerol and 1.0 M sucrose for 40 min at 25°C. Next, the plumules on the cryo-plate were desiccated under a laminar air flow cabinet for 2.0 h at 25°C and plunged directly into the liquid nitrogen, then regenerated after cryostorage using *in vitro* culture (fig. 6).

The results of research concerning classical and cryogenic storage of forest tree germplasme are applied in forest practice in the Kostrzyca Forest Gene Bank, Poland (Zimnoch-Guzowska et al. 2022).

In vitro culture

Micropropagation of oaks

Old oak trees appeared to be recalcitrant to micropropagation. However successful *in vitro* shoot multiplication of ca. 800-year-old *Q. robur* trees growing in Poland were recently described for the first time (Chmielarz et al. 2023). Genotype and tree age had significant effects on the efficiency of epicormic shoot formation during the pot culture (preparation of explants) and shoot multiplication *in vitro*. Old trees display many features valuable for silviculture. For example, the resistance to oak powdery mildew (*Erysiphe alphitoides*) of some specimens. They have a cultural and historical value for society; however, in many regions of the world, a rapid decline in the largest and oldest trees has been observed (Lindenmayer et al. 2012).

There is a need to protect these monumental trees for local communities. Unfortunately, vegetative reproduction, which can be used with many species, is not possible in the case of oaks. In addition, the process of rooting oak shoots is particularly ineffective in the aged trees we studied. The method of grafting an oak sapling can be used, but it does not ensure that a complete copy of the mother tree's genome is obtained, since in grafting the rootstock comes from another plant of the same species. The only method for an oak species that will result in a genetically identical individual to the mother tree is to obtain it using sterile *in vitro* cultures.

To initiate such *in vitro* cultures we used woody shoots of pedunculate oaks trees. The woody shoots were placed in vase culture at high humidity and a temperature of 20°C. Over four weeks, sprouts 2–20 cm long grew from the epicormic (dormant) buds under the cork of the woody shoots (the first stage of tissue rejuvenation). Small fragments of these shoots (explants), about 2 cm long, containing 1–2 buds, were then used to initiate *in vitro* cultures. After sterilisation explants were cultured *in vitro* on Woody Plant Medium (WPM), a mixture of chemical compounds containing essential macro and microelements needed for plant growth – with cyclic transfers to fresh medium of the same composition (the second stage of rejuvenation). The medium was also supplemented with vi-

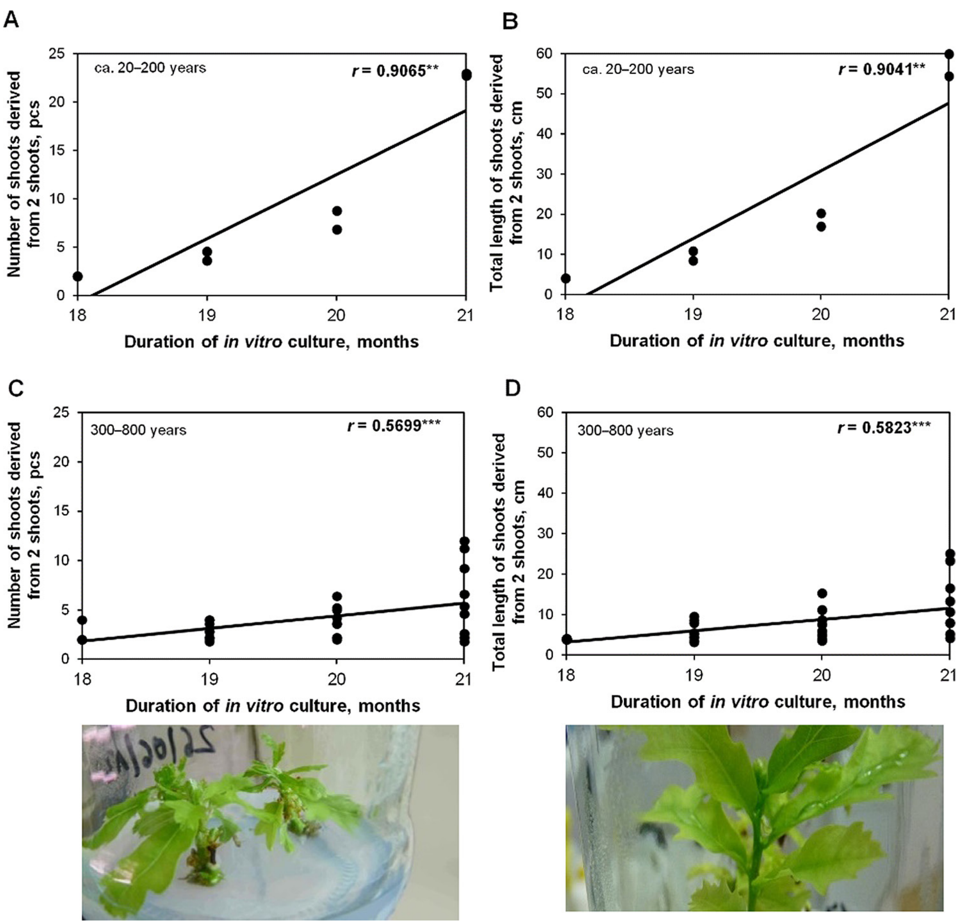


Fig. 7. Pearson correlation coefficient (r) between the number (pcs-pieces) (A,C) or total length of shoots (B,D) and *in vitro* culture duration from 18 to 21 months in oaks aged *c*. 20–200 years (A,B) and monumental oaks (*c*. 300–800 years old) (C,D). Correlation significant at the ** $p \leq 0.01$ or *** $p \leq 0.001$ level. The x-axis represents the months from the start of the culture after sterilization (Chmielarz et al. 2023)

tamins, amino acids, sugar (as an energy source), and growth regulators, mainly 6-benzylaminopurine (BAP; Chmielarz et al. 2023). As a result, genotype and tree age had significant effects on the efficiency of epicormic shoot formation and shoot multiplication *in vitro* (fig. 7). Generally, younger oaks displayed a higher potential of *in vitro* growth (fig. 7; Chmielarz et al. 2023).

In vitro derived small trees – clones of monumental trees – were planted next to their mother trees (being its genetic copy) or in other places in Poland. On 14 April 2019, the first oak clone with an 800-year-old genotype obtained by *in vitro* culture was planted in Rogalin, which is home to Europe's largest cluster of such trees (fig. 8). Clones of monumental oaks are also now growing in the Arboretum of the Institute of Dendrology PAS in Kórnik and in the Arboretum of the Kostrzyca Forest Gene Bank. This study demonstrates that we can protect the genotypes of *c.* 800-year-old *Q. robur* trees using an optimized micropropagation protocol (fig. 8; Martins et al. 2022, 2023, 2024, Chmielarz et al. 2023).

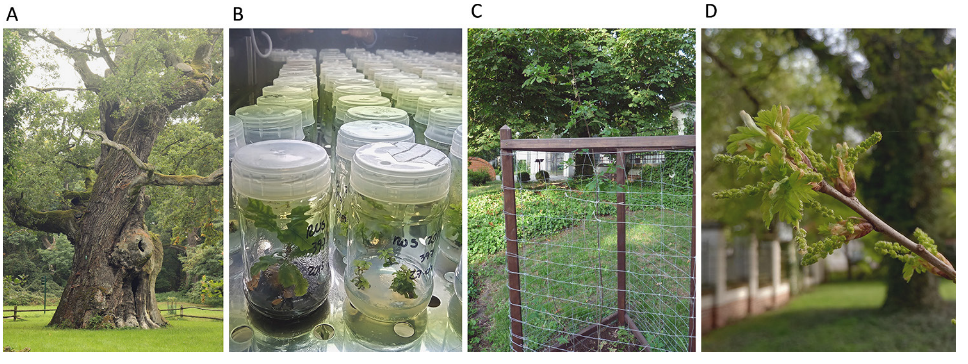


Fig. 8. 800-year-old pedunculate oak (*Quercus robur* L.) from Rogalin (oak Rus), Photo: K. Borkowski (A). *In vitro* multiplication of shoots (the right jars) and shoot rooting on the medium with activated charcoal (left jar) (B). The 4-year-old clone of oak Rus, 2 m high, planted in Rogalin (C). Oak clones "remember" their genetic age and immediately after planting produce flowers and acorns (D). Photo: P. Chmielarz

References

Barciszewska M.Z., Barciszewska A.M., Rattan S.I.S. 2007. TLC-based detection of methylated cytosine: application to aging epigenetics. Biogerontology 8: 673–678.

Chmielarz P. 2002. Sensitivity of Tilia cordata seeds to dehydration and temperature of liquid nitrogen. Dendrobiology 47: 71–77 Supplement.

Chmielarz P. 2009a. Cryopreservation of dormant European ash (Fraxinus excelsior) orthodox seeds. Tree Physiology 29: 1279–1285.

Chmielarz P. 2009b. Cryopreservation of dormant orthodox seeds of forest trees: mazzard cherry (Prunus avium L.). Annals of Forest Science 66: 405.

Chmielarz P. 2010a. Cryopreservation of dormant orthodox seeds of European hornbeam (Carpinus betulus). Seed ScienceTechnology 38: 146–157.

Chmielarz P. 2010b. Cryopreservation of the non-dormant orthodox seeds of Ulmus glabra. Acta Biologica Hungarica 61: 224–233.

Chmielarz P. 2010c. Cryopreservation of orthodox seeds of Alnus glutinosa. CryoLetters 31: 139–146.

Chmielarz P., 2010d. Cryopreservation of conditionally dormant orthodox seeds of Betula pendula. Acta Physiologiae Plantarum 32: 591–596.

Chmielarz P., Kotlarski S., Kalemba E.M., Martins J.P.R., Michalak M. 2023. Successful in vitro shoot multiplication of Quercus robur L. trees aged up to 800 years. Plants 12(12): 2230.

Chmielarz P., Michalak M., Pałucka M., Wasileńczyk U. 2011. Successful cryopreservation of Quercus robur plumules. Plant Cell Reports 30: 1405–1414.

Chmielarz P., Suszka J., Wawrzyniak M.K. 2022. Desiccation does not increase frost resistance of pedunculate oak (Quercus robur L.) seeds. Annals of Forest Science 79: 3.

Daws M.I., Cleland H., Chmielarz P., Gorian F., Leprince O., Mullins C.E., Thanos C.A., Vandvik V., Pritchard H.W. 2006. Variable desiccation tolerance in Acer pseudoplatanus seeds in relation to developmental conditions: a case of phenotypic recalcitrance? Functional Plant Biology 33: 59–66.

Daws M.I., Lydall E., Chmielarz P., Leprince O., Matthews S., Thanos C.A., Pritchard H.W. 2004. Developmental heat sum influences recalcitrant seed traits in Aesculus hippocastanum across Europe. New Phytologist 162: 157–166.

Lindenmayer D.B., Laurance W.F., Franklin J.F. 2012. Global decline in large old trees. Science 338: 1305–1306.

Martins J.P.R., Wawrzyniak M.K., Kalemba E.M., Ley-López J.M., Lira J.M.S., Chmielarz P. 2024. In vitro rooting of Quercus robur, activated charcoal vs. exogenous auxin: a morphophysiological approach. Plant Cell, Tissue and Organ Culture 156: 24.

Martins J.P.R., Wawrzyniak M.K., Kalemba E.M., Ley-López J.M., Mendes M.M., Chmielarz P. 2023. Calcium silicate mitigates the physiological stress induced by 6-benzylaminopurine during the in vitro multiplication of Quercus robur. Industrial Crops & Products 194: 116377.

Martins J.P.R., Wawrzyniak M.K., Ley-López J.M., Kalemba E.M., Mendes M.M., Chmielarz P. 2022. 6-Benzylaminopurine and kinetin modulations during in vitro propagation of Quercus robur (L.): an assessment of anatomical, biochemical, and physiological profiling of shoots. Plant Cell, Tissue and Organ Culture 151(1): 149–164.

Michalak M., Barciszewska M.Z., Barciszewski J., Plitta B.P., Chmielarz P. 2013. Global changes in DNA methylation in seeds and seedlings of Pyrus communis after seed desiccation and storage. PLoS ONE 8(8): e70693.

Plitta B.P., Michalak M., Kotlarski S., Chmielarz P. 2013. Kriogeniczne przechowywanie nasion. Sylwan 157: 723–729.

Wasileńczyk U., Wawrzyniak M.K., Martins J.P.R., Kosek P., Chmielarz P. 2024. Cryopreservation of sessile oak (Quercus petraea (Matt.) Liebl.) plumules using aluminium cryo-plates: influence of cryoprotection and drying. Plant Methods 20: 53.

Zimnoch-Guzowska E., Chmielarz P., Wawrzyniak M.K., Plitta-Michalak B.P., Michalak M., Pałucka M., Wasileńczyk U., Kosek P., Kulus D., Rucińska A., Mikuła A. 2022. Polish cryobanks: research and conservation for plant genetic resources. Acta Societatis Botanicorum Poloniae 91: 9121.

Factors determining seed viability and successful seed germination in a changing world

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Keywords: seed ageing, seed longevity, oxidative stress, redox status, seed germination, seedling establishment

Seed longevity is a global challenge associated with ecosystem biodiversity and the success of plant reproduction and forest restoration. Seed longevity is a polygenic feature that requires the coordination of many biological processes shaping the stability of seeds in the soil and their ex situ storage time. Our research contributes to understanding the problem of seed ageing at the molecular, cellular, and physiological levels, and to the multi-faceted characterization of the basis for the reduced longevity phenotype in seeds. Many mechanisms, including protection, repair, and detoxification, are developed at the stage of seed maturation and directly affect the quality of seeds and the possibility of their storage. Learning about new compounds, discovering new interactions and biochemical processes, make it possible to explain the heterogeneity of seed longevity in different plant/tree species. To meet the need to protect the world's plant genetic resources for the future, many modern seed banks have been established to function as gene banks. During storage, the seeds undergo an ageing process, which results in reduced seed viability and loss of germination ability. The deterioration of the physiological condition of seeds is the most important problem associated with gene banks, and it is also the most difficult to solve. The knowledge gained from our research is useful for optimizing seed banking protocols that ensure seed viability is maintained for as long as possible. The ability of seeds to survive until the moment of germination depends on the synergistic action of endogenous proteins, hormones, and metabolites, as well as on storage conditions – here temperature and humidity are of the greatest importance. Optimal storage conditions guarantee seed viability preservation, thus ensuring high-quality sowing material for the establishment of healthy seedlings. Storage of high-quality seeds in conditions that do not impair their viability is crucial for biodiversity conservation. Detailed development of the technology for handling seeds after harvesting, together with optimal conditions for seed storage and stratification, assure the continuity of seedling production in forest management. Successful seed banking allows the preservation of the genetic resources of trees and shrubs in terms of valuable genotypes.

Reactive oxygen species (ROS) as seed longevity determinants

Literature data in the field of seed biology clearly indicate that ROS overproduction and accumulation contribute to the gradual deterioration of seeds and seed ageing (Jeevan Kumar et al. 2015), whereas lower concentrations of ROS play a signaling role in the regulation of many physiological processes, including seed dormancy and germination (Bailly 2019). In this context, ROS exhibit a dual function. ROS are postulated as the most important factors initiating the seed ageing process because the excess of ROS is inevitably harmful to seeds during their storage. Seed deterioration refers to seeds of all categories, even those resistant to desiccation, because ROS can interact with any macromolecule of biological importance (proteins, lipids, DNA), resulting in oxidative damage to various cellular components and even programmed cell death (Zhang et al. 2021). Incomplete reduction of molecular oxygen (O_2) leads to the formation of superoxide anion ($O_2^{\bullet -}$), which is converted enzymatically to hydrogen peroxide (H_2O_2). There are plenty of cellular antioxidants able to decompose H_2O_2 into water and oxygen. H_2O_2 can be further converted to the hydroxyl radical ($^{\bullet}OH$), for which no enzymatic or non-enzymatic scavenger exists. This reaction presents the cascade by which O_2 is converted into H_2O through reactive intermediates:

$O_2 \rightarrow O_2^{\bullet -} \rightarrow H_2O_2 \rightarrow {}^{\bullet}OH \rightarrow H_2O$

Importantly, $^{\bullet}OH$ might be converted to H_2O only when another hydrogen atom (H^+) is added. Unfortunately, in the oxidative state, there is a constant deficit of H^+, therefore $^{\bullet}OH$ accumulates in ageing tissues in proportion to the advancement of the ageing process. Seeds of the recalcitrant category are metabolically active during storage, therefore the content of $O_2^{\bullet -}$ and H_2O_2 is subject to constant changes due to the activity of superoxide dismutase that decomposes $O_2^{\bullet -}$ to H_2O_2 and, among others, catalase, glutathione, and ascorbate that decompose H_2O_2 into water.

Antioxidant system

The amount of ROS is controlled by the antioxidant system, which consists of enzymes involved in the removal of ROS, such as catalase (CAT), enzymes converting ROS – for example, superoxide dismutase (SOD) or peroxidases, and non-enzymatic ROS scavenging compounds, such as ascorbate and glutathione, which cooperate in the plant-specific ascorbate-glutathione cycle (Jajic et al. 2015). The above molecules are the first-line defense because they directly react with ROS. The second-line defense includes: 1) redoxins able to reduce disulfide groups to –SH groups such as thioredoxins and glutaredoxins, 2) methionine sulfoxide reductases (MSRs) reducing methionine sulfoxides (MetO) to methionine (Lourenço Dos Santos et al. 2018). The reduced form of nicotinamide adenine dinucleotide phosphate (NADPH) is involved in the functioning of all the above antioxidants and repair systems acting on oxidized proteins, clearly indicating that NADPH is always needed for protein reduction reactions.

Seed categories

Seeds are the most important form of plant reproduction and play an important role in the spread of plants. Changes in the hydration of seed tissues accompany important stages in seed development. Seeds dry during ripening (Leprince et al. 2017), remain in a dry state through the dormant phase, and then start germination after imbibition (Bewley et al. 2013). Therefore, it is important to determine the defense mechanisms and their elements that allow the maintenance of seed viability. On the basis of water content limits that ensure the maintenance of plant viability, there are two types of tolerance to water loss (Hoekstra 2005): drought tolerance, when the variable degree of tolerable dehydration is up to approximately 23% in fresh weight (~0.3 g H_2O g^{-1} dry weight (DW)), and resistance to desiccation, i.e., the tolerance of water loss below 0.07 g H_2O g^{-1} DW (Walters 2015). Seeds resistant to desiccation are classified as orthodox, while seeds resistant to drought and sensitive to desiccation are classified as recalcitrant (Roberts 1973). Seeds with in-between characteristics belong to the intermediate category (Ellis et al. 1990). The determination of the physiology of seeds exhibiting intermediate and recalcitrant behavior remains a major challenge in the effort to develop specific rules of seed management that will ensure seed viability. Our studies focus on the physiology of intermediate category seeds of European beech (*Fagus sylvatica* L.) and recalcitrant category seeds of pedunculate oak (*Quercus robur* L.). The recalcitrant phenotype of seeds is investigated also in sycamore (*Acer pseudoplatanus* L.) as compared to genetically closely related orthodox category seeds of Norway maple (*Acer platanoides* L.).

Seeds inevitably deteriorate. Excessive accumulation of ROS causes three-phasic damage to seeds of all categories: (phase 1) slight reduction in vigor, (phase 2) oxidative damage, including lipid peroxidation and electrolyte leakage, and (phase 3) irreversible damage to the genetic material (Ebone et al. 2019). The first phase may be unnoticeable, while the third phase inevitably leads to the death of cells and entire seeds. In this context, detailed monitoring of the symptoms of the second phase is necessary. Providing new measurement methods to estimate seed viability by analyzing the oxidation state through the amount of ROS and mitochondria activity might become extremely useful in seed practice.

European beech (*Fagus sylvatica*)

Typical orthodox seeds acquire tolerance to desiccation during maturation (Dekkers et al. 2015) and maintain this tolerance during storage in a dry state and dormancy alleviation until the root emerges from germinating seeds (Buitink, Leprince 2008). Beech seeds acquire desiccation tolerance during maturation (Kalemba et al. 2008); however, because of storage difficulties, these seeds are classified as intermediate (León-Lobos, Ellis 2002). Our results demonstrated that ROS levels increase as beech seeds age (Pukacka, Ratajczak 2007, Kalemba, Pukacka 2014, Ratajczak et al. 2015), confirming the oxidative theory of seed ageing. ROS localization tests showed the presence of $O_2^{\bullet-}$ and H_2O_2 in the area of the apical meristem of the embryonic axis (Ratajczak et al. 2015). Increased ROS fluorescence signal in the area of the apical meristem was reported in seeds stored 8–11 years. After 13 years of storage, the fluorescence signal dispersed at neighboring areas in the embryonic axis, indicating the intensification of ageing processes.

The signaling role of $O_2^{\bullet -}$ and H_2O_2 has been demonstrated during beech seed germination (Kalemba et al. 2019). Both these ROS were reported to be synthesized in the apical meristem of the embryonic axis elongating into radicle, and the area in which the ROS spread covered up to half of the length of the radicle as it grew. Drought stress in germinating seeds inhibited the synthesis of $O_2^{\bullet -}$ and root growth and caused the accumulation H_2O_2, which intensified oxidative stress.

The cellular origin of ROS in beech seeds

ROS are produced during the whole life of a seed – in the cytosol, plasma membrane and cell wall, and particularly in organelles containing the electron transport chain (ETC) such as mitochondria, chloroplasts, and peroxisomes (glyoxy-somes). Our research indicates that the cellular structure where the initiation of the ageing process in seeds might take place is the mitochondria (Małecka et al. 2021). Mitochondria are important cellular organelles responsible for the synthesis of energy and they are involved in the regulation of the redox state in cell signaling. Mitochondria generate ROS, which in excess have a destructive effect on mitochondria structure and lead to their impairment (Małecka et al. 2021). Our study revealed that in stored beech seeds, changes present in the mitochondrial structure reduce seed viability.

Metabolic processes during germination require a large amount of energy to be synthesized in mitochondria. By measuring the respiratory activity of seeds, and thus their energy status, it is possible to assess their level of viability. Using the Seahorse analyzer, which enables the respiratory activity of seeds to be measured, the respiratory capacity of mitochondria isolated from beech seeds displayed storage time-dependent gradual decrease in non-phosphorylating respiration and phosphorylating respiration. The longer the storage time, the greater the impairment of respiratory function was reported (Fuchs et al. 2023).

Oxidative modifications of proteins in beech seeds

ROS oxidize proteins at specific amino acids by adding a carbonyl group, a process known as carbonylation, which is considered irreversible. We have established that the amount of carbonylated proteins increases with the storage time of beech seeds (Kalemba, Pukacka 2014). The amount of carbonylated proteins measured in whole beech seeds was the lowest in the youngest seeds. Seeds stored for 5–11 years contained two times more carbonylated proteins than young seeds, whereas seeds stored for 13 years contained four times more carbonylated proteins than young seeds, which clearly indicates the progression of oxidative stress. Comparing changes in the level of ROS and carbonylated proteins (Kalemba, Pukacka 2014), it can be concluded that the accumulation of ROS is the cause of oxidative stress, and protein carbonylation is the effect of oxidative stress in stored beech seeds. The high correlation coefficient between the increasing concentration of $O_2^{\bullet -}$ (r = 0.92), H_2O_2 (r = 0.71), and protein carbonyls confirms this hypothesis. The Pearson correlation coefficient between the germination capacity and the level of protein carbonylation for whole beech seeds stored for 2, 5, 8, 11, and 13 years was very high (r = – 0.93). We have indicated there is a strong relationship between the level of protein carbonyls and the germination capacity of aged beech seeds, i.e., the more carbonylated proteins there are, the lower the germination capacity will be.

ROS oxidize methionine (Met) to methionine sulfoxide (MetO), and the activity of methionine sulfoxide reductases (MSR) restores the reduced form of Met (Rey, Tararago 2018). Met sulfoxidation is reversible, and MSRs function as antioxidants. Our studies revealed that the abundances of B1 and B2 types of MSR decreased during long-term storage of beech seeds (Wojciechowska et al. 2021). MsrB2 was linked with the longevity of beech seeds via its subcellular localization and association with the proper utilization of seed storage material during germination.

Cysteine is another amino acid sensitive to oxidation because of the presence of a thiol (-SH) group. The level of thiol groups in proteins decreased during storage of beech seeds (Ratajczak et al. 2019).

Antioxidants in beech seeds

Peroxiredoxins (PRXs) are thiol-proteins involved in thiol-disulfide redox switches in plants and seeds via the removal of H_2O_2. Our studies demonstrated the occurrence of several PRXs in stored beech seeds, including cytosolic 1CysPRX and PRXIIC, mitochondrial PRXIIF, and plastidial PRXIIE, 2CysPRX, and PRXQ. Among them, 2CysPRX was assumed to perform an important function in the regulation of the redox state during storage of beech seeds (Ratajczak et al. 2019).

Catalase, the main cellular enzyme responsible for the removal of H_2O_2, exhibited decreasing activity as beech seeds aged (Małecka et al. 2019), indicating that the antioxidant system is less sufficient in aged seeds. Beech seeds contain the major form of catalase (CAT55) and catalase-domain-containing protein (CAT 55). CAT55 acts predominantly in embryonic axes, whereas CAT37 is the major enzyme responsible for H_2O_2 removal in cotyledons (Kalemba et al. 2021a).

Pedunculate oak (*Quercus robur*)

Our results demonstrated that in metabolically active seeds of the recalcitrant category, measurement of the $^{\bullet}OH$ content seems to be a new, effective method of seed quality control during storage. In studies of stored recalcitrant oak seeds, we showed that among the tested ROS ($O_2^{\bullet -}$, H_2O_2, $^{\bullet}OH$), only the $^{\bullet}OH$ content was correlated with all seed viability parameters (Kalemba et al. 2021b). This means that increasing the $^{\bullet}OH$ level increases the outflow of electrolytes and, consequently, reduces the germination capacity and the efficiency of seedling establishment. Our experiments are designed to investigate how to manipulate storage temperature to reduce ROS production and extend storage time while maintaining the high quality of pedunculate oak seeds. The search for and optimization of new methods of storing oak seeds involve thermotherapy and lower than recommended constant storage temperatures, and combinations of storage temperatures leading to acclimatization to cold. Our results revealed that thermotherapy of pedunculate oak before storage mimics seed priming and adjusts the redox status in seeds. Seed thermotherapy used to eliminate pathogens, mainly *Ciboria batschiana*, did not increase the level of ROS in the initial stages of seed preparation for long-term storage. Moreover, H_2O_2 levels did not increase only in seeds treated with thermotherapy and stored at constant temperatures or with a gradual temperature reduction regimen to –5 and –7°C, unlike the other two types of ROS ($O_2^{\bullet-}$, $^{\bullet}OH$). This suggests that thermotherapy has a beneficial effect on the ability to eliminate excess hydrogen peroxide in pedunculate oak seeds during short-term storage. Metabolomics studies have shown that one of the causes of ROS accumulation and thus deterioration of the quality of pedunculate oak seeds during storage at –7°C is oxidative DNA damage caused by 1,2,4-benzenetriol (Szuba et al. 2022), which indicates the most advanced seed damage phase (Ebone et al. 2019).

References

Bailly C. 2019. The signalling role of ROS in the regulation of seed germination and dormancy. The Biochemical Journal 476(20): 3019–3032.

Bewley J.D., Bradford K., Hilhorst H., Nonogaki H. 2013. Seeds: Physiology of development, germination and dormancy. 3rd edition. Springer, New York, pp. 247–297.

Buitink J., Leprince O. 2008. Intracellular glasses and seed survival in the dry state. Comptes Rendus Biologies 331(10): 788–795.

Dekkers B.J.W., Costa M.C.D., Maia J., Bentsink L., Ligterink W., Hilhorst H.W.M. 2015. Acquisition and loss of desiccation tolerance in seeds: from experimental model to biological relevance. Planta 241(3): 563–577.

Ebone L.A., Caverzan A., Chavarria G. 2019. Physiologic alterations in orthodox seeds due to deterioration processes. Plant Physiology and Biochemistry 145: 34–42.

Ellis R.H., Hong T.D., Roberts E.H. 1990. An intermediate category of seed storage behaviour? I. COFFEE. Journal of Experimental Botany 41(230): 1167–1174.

Fuchs H., Malecka A., Budzinska A., Jarmuszkiewicz W., Ciszewska L., Staszak A.M., Kijowska-Oberc J., Ratajczak E. 2023. High-throughput method for Oxygen Consumption Rate measurement (OCR) in plant mitochondria. BMC Plant Biology 23: 496.

Hoekstra F.A. 2005. Differential longevities in desiccated anhydrobiotic plant systems. Integrative and Comparative Biology 45(5): 725–733.

Jajic I., Sarna T., Strzalka K. 2015. Senescence, stress, and reactive oxygen species. Plants 4(3): 393–411.

Jeevan Kumar S.P., Rajendra Prasad S., Banerjee R., Thammineni C. 2015. Seed birth to death: dual functions of reactive oxygen species in seed physiology. Annals of Botany 116(4): 663–668.

Kalemba E.M., Alipour S., Wojciechowska N. 2021a. NAD(P)H drives the ascorbate-glutathione cycle and abundance of catalase in developing beech seeds differently in embryonic axes and cotyledons. Antioxidants 10(12): 2021.

Kalemba E.M., Bagniewska-Zadworna A., Suszka J., Pukacka S. 2019. Dehydration sensitivity at the early seedling establishment stages of the European beech (Fagus sylvatica L.). Forests 10(10): 900.

Kalemba E.M., Janowiak F., Pukacka S. 2008. Desiccation tolerance acquisition in developing beech (Fagus sylvatica L.) seeds: the contribution of dehydrin-like protein. Trees 23(2): 305.

Kalemba E.M., Pukacka S. 2014. Carbonylated proteins accumulated as vitality decreases during long-term storage of beech (Fagus sylvatica L.) seeds. Trees 28(2): 503–515.

Kalemba E.M., Wawrzyniak M.K., Suszka J., Chmielarz P. 2021b. Thermotherapy and storage temperature manipulations limit the production of reactive oxygen species in stored pedunculate oak acorns. Forests 12(10): 1338.

León-Lobos P., Ellis R.H. 2002. Seed storage behaviour of Fagus sylvatica and Fagus crenata. Seed Science Research 12(1): 31–37.

Leprince O., Pellizzaro A., Berriri S., Buitink J. 2017. Late seed maturation: drying without dying. Journal of Experimental Botany 68(4): 827–841.

Lourenço Dos Santos S., Petropoulos I., Friguet B. 2018. The oxidized protein repair enzymes methionine sulfoxide reductases and their roles in protecting against oxidative stress, in ageing and in regulating protein function. Antioxidants 7(12): 191.

Małecka A., Ciszewska L., Staszak A., Ratajczak E. 2021. Relationship between mitochondrial changes and seed aging as a limitation of viability for the storage of beech seed (Fagus sylvatica L.). PeerJ 9: e10569.

Pukacka S., Ratajczak E. 2007. Age-related biochemical changes during storage of beech (Fagus sylvatica L.) seeds. Seed Science Research 17(1): 45–53.

Ratajczak E., Małecka A., Bagniewska-Zadworna A., Kalemba E.M. 2015. The production, localization and spreading of reactive oxygen species contributes to the low vitality of long-term stored common beech (Fagus sylvatica L.) seeds. Journal of Plant Physiology 174: 147–156.

Ratajczak E., Staszak A.M., Wojciechowska N., Bagniewska-Zadworna A., Dietz K.J. 2019. Regulation of thiol metabolism as a factor that influences the development and storage capacity of beech seeds. Journal of Plant Physiology 239: 61–70.

Rey P., Tarrago L. 2018. Physiological roles of plant methionine sulfoxide reductases in redox homeostasis and signaling. Antioxidants 7(9): 114.

Roberts E.H. 1973. Predicting the storage life of seeds. Proceedings. Seed Science and Technology 1: 499–514.

Szuba A., Kalemba E.M., Wawrzyniak M.K., Suszka J., Chmielarz P. 2022. Deterioration in the quality of recalcitrant Quercus robur seeds during six months of storage at subzero temperatures: ineffective activation of prosurvival mechanisms and evidence of freezing stress from an untargeted metabolomic study. Metabolites 12(8): 756.

Walters C. 2015. Orthodoxy, recalcitrance and in-between: describing variation in seed storage characteristics using threshold responses to water loss. Planta 242(2): 397–406.

Wojciechowska N., Bagniewska-Zadworna A., Minicka J., Michalak K.M., Kalemba E.M. 2021. Localization and dynamics of the methionine sulfoxide reductases MsrB1 and MsrB2 in beech seeds. International Journal of Molecular Sciences 22(1): e402.

Zhang K., Zhang Y., Sun J., Meng J., Tao J. 2021. Deterioration of orthodox seeds during ageing: Influencing factors, physiological alterations and the role of reactive oxygen species. Plant Physiology and Biochemistry 158: 475–485.

Climate legacy in seed and seedling traits of European beech populations

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Trees grow in a wide range of environments and have developed a variety of adaptive mechanisms. One of the most important mechanisms is seed dormancy, which ensures seed survival through the winter and plant germination in spring, when the conditions are most suitable for the development of seedlings in temperate climates (Klupczyńska, Pawłowski 2021). Environmental factors such as temperature, light intensity, and water availability are key factors controlling seed germination and, as a result, affect the reproductive success of plants. Global climate change alters environmental conditions and thus affects the reproduction of plants from seeds. Recent studies concentrate on the questions of whether the adaptation, manifested as dormancy, is plastic, and whether trees have the potential to adapt to environmental changes. In this lecture, recent knowledge concerning the adaptation and survival of trees under conditions of global warming in the context of seed dormancy and germination will be presented.

At the northern edges of tree species ranges in Europe, an increase in temperature is expected to broaden the suitable niche for some species (Chakraborty et al. 2021). However, direct genetic adaptations to the specific local habitats may reduce such expectancy. Environmental changes may cause disturbances in tree regeneration if future climatic conditions do not match the historically imprinted adaptation requirements for seed germination and seedling fitness (Klupczyńska, Pawłowski 2021).

The ecological niche of tree species is typically narrower at earlier life stages than at later ones, and intraspecific genetic variation of early fitness traits may define the response of tree populations to changing environmental conditions (Solé-Medina et al. 2020). To assess the adaptative potential, there is a need to recognize intraspecific genetic variation in fitness-related traits and its environmental determinants (Kurpisz, Pawłowski 2022). Seed dormancy breaking and the timing of germination have important implications for the success of plant establishment in the face of environmental risk factors (Pausas et al. 2022). However, the role of seed dormancy and germination in the success of trees remains underestimated. Thus, improving our understanding of intraspecific genetic variation in early fitness traits, such as seed dormancy, germination, seedling establishment, and seedling shoot and root characteristics, is essential for investigating the potential ability of tree populations to regenerate and persist under climate change.

European beech (*Fagus sylvatica* L.) is a sub-Atlantic tree species and an important component of forest communities in Western and Central Europe. Beech forests have mainly developed by natural regeneration, and ecotypes have developed that are adapted to the local climatic conditions (Leuschner, Ellenberg 2017). However, the plasticity in the response of beech populations to climatic variability has also been discussed, and in particular, the plasticity of seed dormancy and germination have not been sufficiently researched. Seeds ripen and are released in late autumn; during winter, they lose dormancy, and in spring the next year, they germinate when conditions are favorable for seedling establishment.

There is growing concern about the future survival and sustainability of beech ecosystems throughout Europe, as beech may be unable to adapt to future environmental conditions in its current natural range (Stojnić et al. 2018). Populations growing at marginal sites are generally subject to unsuitable conditions and have a lower chance of survival. The future climatic conditions in parts of the distribution centre and at the rear (southern) edge might thus become limiting for natural beech regeneration by seed, as the likelihood of extreme heat and drought events is expected to increase (Muffler et al. 2021). At the leading edge, increasing temperatures may increase germination success and beech regeneration, but more detailed studies are needed to predict the possible effect of climatic warming on beech regeneration.

The aim of the study was to determine intraspecific genetic variability among the north-eastern European beech populations in early fitness traits – seed dormancy and germination in relation to the climate at population origin (Pawłowski et al. in preparation). The early growth of seedlings was also investigated in a nursery common-garden experiment to define how the climate of seed origin affects the fitness of seedlings. It was expected that the results would allow recognition of the climatic factors related to the ability of individual beech populations to cope with and adapt to climate variability. This understanding is crucial for the assessment of the future of beech forests in Europe. It was hypothesized that variation in dormancy and germination of beech seeds is associated with climatic conditions acting on the populations; climatic characteristics of seed provenances affect seedling traits. It was expected that the depth of dormancy of beech populations will increase from north to south within the analysed geographic region in accordance with the temperature gradient. It was also expected that seed germination success and seedling growth will be better in populations from the sites with milder climate.

Germination and seedling growth

The seeds for the research were collected from 26 European beech stands (populations) representing different natural forests and climatic regions in Poland near the north-eastern border of the natural distribution range of beech. Seeds were collected immediately after falling from the trees, dried, and stored at –3°C, and their assessment was carried out (viability and seed mass).

To analyse the depth of dormancy and germination traits of seeds, the seedlots were subjected to a stratification-germination test at 3°C. The germination was characterized by the following parameters: T_0 – time of germination onset, germination capacity, T_E – time at germination capacity, germination speed, and T_{50} – the time necessary for 50% of viable seeds to germinate. T_0 and T_{50} reflect seed dormancy depth, while germination capacity and speed reflect seed quality.

A nursery common-garden experiment was set up in the experimental field of the Institute of Dendrology, Polish Academy of Sciences in Kórnik, Poland. The seeds were sown after stratification in May 2021 in a randomized complete block design with 4 replications of 30 seeds per population. In September 2021, seedlings were harvested and measured for their height growth, root-collar diameter, and above- and belowground biomass. The set of eleven populations was used for a more detailed analysis of seedling morphological traits.

Climatic data for maternal stands were obtained from the ClimateEU v4.63. Correlation analysis was used to test the relationships between climatic variables and seed germination and seedling traits.

Germination and seedling traits vs climatic variables at the population origin

The results showed that significant variation among populations was found for all germination parameters. Variation between the two regions of seed origin was also statistically significant. The group of northern populations started to germinate later, and had a longer T_{50}, but lower speed of germination than the group of southern populations. The analysis of the relationships between seed germination parameters and climatic variables at the seed origin revealed that germination capacity was negatively correlated with temperature and moisture availability parameters, and positively correlated with precipitation parameters.

Significant variation was also found for seedling growth and morphology among populations and between the two regions of origin. Leaf-related traits were characterized by a low coefficient of variation. However, the highest variability was observed in root-related traits. On average, populations from the southern region had seedlings with larger dimensions and greater final biomass than seedlings from the northern region. Seedling shoot length and diameter were negatively correlated with T_0 and positively correlated with germination speed. Shoot length and total seedling mass were positively correlated with germination capacity, and all seedling traits were positively correlated with seed mass. Seed viability showed a negative association with the dry matter content of leaves and stems. Later seed germination time negatively affected leaf and stem traits, but positively affected root traits. Faster germination was associated with lower biomass allocation to leaves.

The relationships between seedling growth traits and climatic data at the seed origin indicated that better growth of seedlings after germination in the nursery was associated with cooler and moister conditions at the site of seed origin. Considering root traits, they showed significant correlations with climate. Temperature at the sites of origin exhibited strong positive effects that increased from the coldest to warmest sites, whereas a negative effect was reflected by a decline at the warmer sites. Populations originating from sites with lower water produced seedlings with shorter and tougher roots than those from sites with more favorable climatic moisture.

The study described how adaptation to climatic conditions of population habitats influences the seed dormancy and germination traits of European beech. It was showed that seed traits differed among populations and regions of seed origins. The northern populations had deeper seed dormancy and required longer stratification for germination, but had greater seed viability and germination speed, and lower germination capacity and seed mass than the group of southern populations. The presented results show that seed dormancy in beech is influenced by the climate of the seed origin site and latitude – surprisingly, however, less by the mean annual temperature or precipitation, but more so by the frost and evaporation.

Seed dormancy and germination traits are related to plant fitness through their effects on the timing and speed of seedling emergence. The presented results showed that climate affects the seed germination traits of beech populations. The strongest negative influences on dormancy depth were found for frost and evaporation. The germination capacity was positively correlated with precipitation and negatively with temperature and moisture deficit at the seed origin site. These results indicate that the dormancy and germination traits of seeds driven by historical environment are limited by water deficit and temperature conditions, where frost decreases dormancy depth, whereas high temperature and low moisture availability decrease germination capacity. It can be concluded that the germination of beech seeds depends on the climatic conditions of the provenance, associated with seasonal variation of temperature and rainfall. The patterns of variation found in these parameters indicate the adaptation of populations to the climate of the seed origin site.

In the present study, evidence was found of local adaptation of seedling growth. Significant variation was found both among populations and between the two regions of seed origin. Populations from the southern region produced seedlings of greater final biomass than the northern ones. Root and shoot growth were negatively correlated with temperature and water deficit, but positively with precipitation. This indicates that better growth of seedlings was associated with cooler and moister conditions. The sensitivity of beech seed germination and fitness to the water supply can be explained by the suborthodox (intermediate) character of these seeds. They are sensitive to long term storage in a desiccated state. To sum up, beech seed germination and seedling development may be exposed to threatening conditions of water deficit and increased temperatures.

Analysis of correlation between seed and seedling traits indicates that seedling shoot growth was negatively correlated with seed dormancy depth and positively with germination speed. Shoot length and total seedling mass were positively correlated with germination capacity, and all seedling traits, except root length, were positively correlated with seed mass. These results suggest that the fitness of beech correlates with shallow seed dormancy, higher germination capacity, and heavier seed mass.

Analysis of morphological seedling traits showed that they differed among provenances with the highest variability observed in root-related traits. The temperature of the origin sites exhibited strong positive effects on root mass, whereas negative effects were reflected by a shorter root length in populations from warmer sites. The production of longer and finer roots was associated with higher moisture availability at the sites of seed origin. These root traits may allow for effective water absorption from the soil, but may also reduce the ability to withstand drought (Zadworny et al. 2021). The results suggest that even those shorter and tougher roots were effective in providing enough water to the aboveground parts.

Climate change and beech regeneration

The presented results imply that increasing temperatures and decreasing precipitation may be factors limiting beech regeneration in the future. Although individual climatic variables describing the origin sites in the study explained only a moderate part of the variation among examined beech populations in seed germination and seedling traits, there appears to be a clear pattern that populations originating from warmer and drier sites (mostly located in the northern region), when compared with those from the opposite end of the climatic gradient, germinated later and with lower success, and produced smaller seedlings with shorter and tougher roots. This trend is worrisome for several reasons. Firstly, it indicates that those populations require a longer stratification period for seed dormancy breakage, which may be disturbed by additional climatic warming, especially in winter. Particularly concerning is that later-emerging seedlings may be subject to drought, especially since their root morphological traits may be less favorable for water absorption from the soil than in populations from the southern group.

The effects of climate change are generally expected to reduce tree growth and survival, predispose forests to disturbance by wildfire, insects, and disease, and ultimately change forest structure and composition at the landscape scale (Chmura et al. 2011). In the light of the results of the present study, warming can disturb the correct progression of seed germination and in consequence beech regeneration. Water deficit and ground frost may be limiting factors for beech regeneration. The plasticity of beech populations might not be sufficient to ensure its regeneration in the future due to extreme heat and drought events at the rear edge. At the cold distribution margin, high plasticity in the early life-history traits may allow for increasing germination success with increasing temperatures; however, warming may negatively affect seedling establishment and survival, and thus may facilitate natural regeneration in the future only to a limited extent. Because of the differentiation in growth and the phenological responses of beech, it can be suggested that caution should be taken when translocating provenances in anticipation of the predicted climate warming, which we can also infer on the basis of the local habitat-based associations of seed and seedling properties.

Conclusion

The results of the study indicated the existence of differentiation in adaptation in seed and seedling ecology among provenances of *Fagus sylvatica* in Poland, at the northeastern range of the species distribution. This adaptation strategy is related to environmental conditions at the provenance level. The temperature and precipitation variables affecting climatic water availability shaped, to some degree, the seed dormancy and germination pattern among examined beech populations, but less so the variation in seedling growth and morphology. Populations originating from warmer and drier sites in our study required a longer period of seed stratification, germinated later and with lower success, and produced smaller seedlings. When combined, these factors would expose them to the risks associated with projected climate change. On the other hand, the root morphology of these populations may indicate adaptation to lower water availability. Understanding how beech populations adjust to new environments is critically important given the major role of northern populations in broadening their distribution range. Varying requirements for seed germination, the differences in the success of seedling emergence, and seedling growth and morphology will contribute to variation in regeneration success within the species.

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References

Chakraborty D., Móricz N., Rasztovits E., Dobor L., Schueler S. 2021. Provisioning forest and conservation science with high-resolution maps of potential distribution of major European tree species under climate change. Annals of Forest Science 78: 26.

Chmura D.J., Anderson P.D., Howe G.T., Harrington C.A., Halofsky J.E., Peterson D.L., Shaw D.C., Brad St.Clair J. 2011. Forest responses to climate change in the northwestern United States: Ecophysiological foundations for adaptive management. Forest Ecology and Management 261: 1121–1142.

Klupczyńska E.A., Pawłowski T.A. 2021. Regulation of seed dormancy and germination mechanisms in a changing environment. International Journal of Molecular Sciences 22: 1357.

Kurpisz B., Pawłowski T.A. 2022. Epigenetic mechanisms of tree responses to climatic changes. International Journal of Molecular Sciences 23: 13412.

Leuschner C., Ellenberg H. 2017. Ecology of Central European forests: vegetation ecology of Central Europe. Vol. 1. Springer Cham.

Muffler L., Schmeddes J., Weigel R., Barbeta A., Beil I., Bolte A., Buhk C., Holm S., Klein G., Klisz M., Löf M., Peñuelas J., Schneider L., Vitasse Y., Kreyling J. 2021. High plasticity in germination and establishment success in the dominant forest tree Fagus sylvatica across Europe. Global Ecology and Biogeography 30: 1583–1596.

Pausas J. G., Lamont B. B., Keeley J. E., Bond W. J. 2022. Bet-hedging and best-bet strategies shape seed dormancy. New Phytologist 236: 1232–1236.

Solé-Medina A., Heer K., Opgenoorth L., Kaldewey P., Danusevicius D., Notivol E., Robledo-Arnuncio J.J., Ramírez-Valiente J.A. 2020. Genetic variation in early fitness traits across European populations of silver birch (Betula pendula). AoB Plants 12(3): plaa019.

Stojnić S., Suchocka M., Benito-Garzón M., Torres-Ruiz J. M., Cochard H., Bolte A., Cocozza C., Cvjetković B., de Luis M., Martinez-Vilalta J., Ræbild A., Tognetti R., Delzon S. 2018. Variation in xylem vulnerability to embolism in European beech from geographically marginal populations. Tree Physiology 38: 173–185.

Zadworny M., Mucha J., Jagodziński A. M., Kościelniak P., Łakomy P., Modrzejewski M., Ufnalski K., Żytkowiak R., Comas L.H., Rodríguez-Calcerrada J. 2021. Seedling regeneration techniques affect root systems and the response of Quercus robur seedlings to water shortages. Forest Ecology and Management 479: 118552.

Ectomycorrhizal fungal communities in forest ecosystems: a journey through different research methods and questions for future research

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The term "symbiosis" originates from the Greek word (Gk) meaning 'living together'. In this context, "mycorrhizal symbiosis" refers to a structural and functional association involving the soil fungi (Gk: *mykes*; Latin: *mycor*) and fine roots (Gk: *rhiza*) of most terrestrial ecosystems. In historical overviews, the discovery of mycorrhiza is often attributed to the German phytopathologist B. Frank. However, it was a Polish professor, F. Kamieński, who first claimed in 1881 that root fungi may be beneficial to plants. Nowadays, it is widely accepted that most vascular plants cannot grow and reproduce successfully without mycorrhiza (plural: mycorrhizae or mycorrhizas) provided by networks of soil fungi. Based on anatomical and morphological features, several types of mycorrhizal symbiosis have been distinguished (ectomycorrhizal, arbuscular, arbutoid, ericoid, orchid, and monotropoid; common mycorrhiza types, such as ectendomycorrhiza, are considered to be structural derivatives of ectomycorrhiza).

Many economically and ecologically important trees in temperate, boreal, and tropical forests are involved in ectomycorrhizal (ECM) symbiosis, which is the main focus of this review. The predominately ECM tree families include Pinaceae, Fagaceae, and Betulaceae. Some primarily ECM woody plants may also harbour arbuscular mycorrhizal fungal symbionts in their roots. Such dual mycorrhizal associations have been frequently reported in the roots of *Salix, Populus, Alnus, Eucalyptus*, and some *Quercus* spp.

The fungal species forming ECM associations cover three divisions (Ascomycota, Basidiomycota, and Zygomycota), > 250 genera, and an estimated 20,000 species. In an ECM symbiosis, fungi provide nutrients (notably nitrogen and phosphorus) and water to plants, acquired from the soil via their mycelium networks. In exchange, plants provide them with photo-assimilated C-compounds (e.g., simple sugars). Mycorrhizal fungi also protect their hosts from toxins and pathogens.

In ECM symbiosis, the fungi penetrate between epidermal and cortical cells (the so-called Hartig net) but also form a mycelial mantle that covers fine root tips and changes their morphology into different "morphotypes" that differ in colour, shape, and branching (fig. 1).

Fig. 1. Different ectomycorrhizal morphotypes identified on forest trees

Determining above- and below-ground ectomycorrhizal fungal community structure

Community ecology is the study of species that co-occur together in time and space. The particular species present, and their relative abundances, determine what is frequently referred to as “community structure”. The ECM fungal (ECMF) community structure on the root system merits special attention because of its functional significance for forest trees and attempts to define the forces in nature affecting that structure. A comprehensive review of different methods for identifying and measuring the diversity of ECM fungi is presented by Janowski and Leski (2023). In the present contribution, I refer to the usage of different methods in the identification of ECMF communities in diverse forest ecosystems as conducted at the Department of Symbiotic Associations of the Institute of Dendrology over the past twenty years.

Sporocarp inventories: Over many years, descriptions of ECM species compositions and their spatial patterns have been almost exclusively based on sporocarp inventories, assuming that sporocarp production reflects the relative abundance and importance of the species on the root system. However, such surveys have to be performed in considerable detail and over long periods (3–9 years), because sporocarp abundance varies greatly in space and time, as a result of a wide range of external factors. Finally, it appeared that in terms of species composition, the sporocarp surveyonly moderately reflects the analysis of ECM tips. We showed it very clearly in some of our papers that, in terms of species composition, the sporocarp survey was not compatible with the analysis of the ECMF tips of Scots pine, and largely supplemented the belowground view (Rudawska et al. 2011, Leski et al. 2019).

Morphotyping: As an alternative to sporocarp inventories, a supplementary approach to studying ECMF composition was developed based on morphological characterization of ECM roots – so-called morphotyping. The macroscopic and microscopic characteristics of ECM roots have become widely used for identification of the ECM fungi. Our first papers based only on morphotyping were done on Scots pine seedlings from forest nurseries (Rudawska et al. 2001). However, the resolution of such morphological groupings was often low and highly dependent on the training and experience of the investigator, and hence an unambiguous species-specific identification of mycorrhizas was not possible. Nevertheless, in the early stages of the research on ECMF community structure, morphotyping was a very important approach. Though the taxonomy of fungi based on morphological characteristics enabled preliminary description and identification of ECMF species, classification of fungi that is based only on their morphological characteristics was not sufficient and, with time, molecular methods provided excellent tools for studying both, above- and below-ground communities at the species level.

Molecular methods: Different molecular techniques can be chosen to identify ECMF communities, depending on the aims of the study, research hypotheses and the fungal material. Molecular methods based on polymerase chain reaction (PCR) are among the most useful approaches for the identification of fungi from ectomycorrhizas, or ECM mycelium (e.g., sporocarps, extrametrical mycelium or rhizomorphs), because they require only minute sample sizes and can be targeted at several different taxonomic levels. If a barcoding DNA sequence obtained from an ECM root matches one recorded from a sporocarp, it is inferred that they belong to the same ECMF taxon. DNA sequencing of barcode genes, such as the ITS (internal transcribed spacer) rDNA, for the identification and discovery of fungal specimens directly from ectomycorrhizas, has become nowadays of routine use and appears to be a relatively fast and inexpensive approach. Following DNA extraction and PCR, various techniques such as RFLP (restriction fragment length polymorphism), cloning and sequencing resolve the identification of individual fungi or develop ECMF community profiles.

In our research, molecular methods appeared to be especially useful and very efficient for determining ECM fungi in forest nurseries where, due to the intensive management, the appearance of sporocarps is infrequent. For the first time, we investigated the species richness and composition of mycorrhizal fungi naturally colonising one- and two-year-old *Pinus sylvestris* L. seedlings from bare-root nurseries in Poland, using RFLP of the ITS of fungal rDNA (Iwański, Rudawska 2006). Shortly thereafter we substituted RFLP analysis with Sanger sequencing of ITS region of DNA, and over the next consecutive years, we explored the mycorrhizal status of the nursery planting stock of tree species that predominate in temperate forests using Sanger sequencing on such tree species as *P. sylvestris* (Iwański et al. 2006, Aučina et al. 2014, Rudawska, Leski 2021), *Picea abies* (Rudawska et al. 2006, Trocha et al. 2006), *Larix decidua* (Leski et al. 2008), *Quercus* spp. (Leski et al. 2010), *Fagus sylvatica* (Pietras et al. 2013) and admixture tree-seedlings like *Betula pendula*, *Tilia cordata* and *Carpinus betulus* (Rudawska et al. 2019). Our results showed the high diversity of ECM fungi colonizing the nursery stock of different tree species (Rudawska, Leski 2009). They contradicted the opinions about the scarcity of the ECMF symbionts inhabiting seedlings reared in the conditions of forest nurseries, as was advertised by large-scale nurseries.

Direct analyses of ECMF communities from roots or soil samples have been greatly accommodated by the development of next-generation sequencing (NGS) technology, which has resulted in greater sequence yields at lower costs, making these technologies affordable to a variety of users. We applied high-throughput Illumina MiSeq sequencing of fungal ITS1 amplicons to determine the ECMF communities in formerly managed forest reserves (established around 50 years ago) and forests under standard forest management. This new technology allowed us to describe the ECMF communities but also species of fungi from other trophic groups like saprotrophs, and pathogens (Kujawska et al. 2021).

With this decline in the cost of massively paralleled sequencing applications, direct metabarcoding analyses become the most efficient method to query the structure of resident communities. Despite the enormous capabilities of the NGS method and the discovery of high fungal diversity in the soil of tested forests, not all fungi found by mycorrhizal root tips analysis and sporocarps surveys (Leski et al. 2019, Rudawska et al. 2022) were recovered in our parallel NGS studies (Kujawska et al. 2021). Therefore, to obtain a comprehensive and complete understanding of the diversity and functioning of soil mycobiota, varied methods should be applied (see the recommendation for future research below).

Research on ectomycorrhizal fungal communities and their environmental drivers

Based on the combined approach, i.e. morphotyping of fine roots into ECM morphotypes, followed by Sanger molecular identification of a selected small number of samples from each morphotype or NGS technology, we have examined the species diversity of ECM communities in a very wide range of environments and on many different tree species. Our research comprised native tree species like *P. sylvestris* (Iwański et al. 2007, Stankeviciene et al. 2008, Rudawska et al. 2011, 2017, 2018, Trocha et al. 2012, Aučina et al. 2019), *P. mugo* (Aučina et al. 2011), *P. abies* (Rudawska 2007), *Abies alba* (Rudawska et al. 2016, Kujawska et al. 2023), *Larix decidua* (Leski, Rudawska 2012), *Betula pendula* (Trocha et al. 2007), or *Quercus* sp. (Trocha et al. 2012), as well as several alien trees like *Carya ovata* (Rudawska et al. 2018, Wilgan et al. 2020), *A. grandis* (Kujawska et al. 2023) and *Q. rubra* (Trocha et al. 2012). We showed that irrespective of the origin (Europe vs North America), congeneric tree species are characterised by a comparable number of ECMF communities, although with different compositions and structures. This finding implies that alien tree species planted outside their natural range may be an interesting reservoir of ectomycorrhizal fungi in a climate change scenario where conserving diverse ECMF assemblages may be of ecological importance. Other areas of research using combined morpho-molecular studies focused on the effect of the genotype of different tree species, like *P. sylvestris* (Leski et al. 2010), *Populus* sp. (Karliński et al. 2013, 2020) and *Abies* sp. (Kujawska et al. 2023), on ECMF communities. We also revealed the effect of host specificity (Trocha et al. 2012), soil type, soil moisture, season, natural nutrient gradients, and the quality and quantity of organic matter (e.g., Aučina et al. 2014, Rudawska et al. 2017, 2022). ECMF communities were also investigated under standard forest management practices as well as protected forests. The results indicated a rather slow process of transformation of ECMF communities in forest reserves originating from previously managed forests (Leski et al. 2019, Kujawska et al. 2021, Rudawska et al. 2022).

Along with the analysis of the ECMF community structure, we were able to extend our research to include the range of biotic and abiotic drivers influencing these communities. The tree chronosequence appeared to be an important factor influencing the ECMF community both in forest nurseries (Rudawska, Leski 2021) and in mature forests (Rudawska et al. 2018). The observed ECMF species richness did not significantly change along the tree chronosequence, but shifts in identified taxa were significant. Though some studies suggest that on a broad scale, soil fungal communities are highly influenced by stochastic processes, ample evidence suggests that on a local or regional scale, the most influential drivers of fungal species composition are climate (e.g., temperature and precipitation), forest stand structure (e.g., canopy cover, tree species, and stand age), and site characteristics (e.g., slope, aspect, altitude, and soil properties). In most of our studies, the most important drivers determining the structure of forest soil ECMF communities related to the biotic features of stands, such as the total number of tree species, the number of ECM tree species, and the number of trees with a DBH > 10 cm, etc. ECMF diversity was also affected by a wide range of soil chemical properties corresponding to pH, C and N (Leski et al. 2019, Rudawska et al. 2022), but also differentiated nitrogen fertilization (Rudawska et al. 2001), toxic metal contamination (Rudawska et al. 2000, 2003, 2011), and other forms of anthropogenic pollution (Rudawska et al. 2011). It is worth underlining that molecular analysis of ectomycorrhizas also allows the identification of several conservation-relevant species not found by sporocarp surveys. Species of conservation value (red-listed and rare species in Poland) have been noted in both forest reserves and managed forests, showing that each management regime contributes a certain number of taxa not found in the other (Leski et al. 2019, Kujawska et al. 2021, 2023, Rudawska et al. 2022).

The most important challenges in ectomycorrhizal research

The knowledge about ECMF communities is of great importance for science, management, conservation, and policy. Despite the considerable number of studies conducted on ECMF community structure, there are still many unanswered questions. Distributions, diversity hotspots, dominance and rarity, and indicators of forest changes are very important areas which remain to be explored in ECMF interactions and their dynamics. The most important challenges are:

1. Development of new methods more accurately describing ECMF communities. There are several methods available to study ECMF diversity, but each has its limitations. Therefore, it is advisable to use a variety of methods, incorporating the results of traditional and the newest molecular methods to obtain reliable results. Long-term observation in the field is also highly advisable.
2. Delivery of accurate mycological data in man-made, native, or disturbed forests to better manage plant and fungi biodiversity.
3. Filling numerous knowledge gaps in the occurrence of ECM fungi with a particular emphasis on little-known geographic areas, ecosystems, host trees and fungal groups to face the challenges the ECMF community poses, and to discover more about their roles in the biosphere.
4. Development of present-day molecular tools along with standard PCR analysis and transcriptome analysis, such as oligoarray-based transcriptome profiling, to understand the molecular mechanisms of ECM influence on host plants, including N and P acquisition and carbohydrate transport.
5. Application of ECM fungi in sylviculture based on the latest knowledge and observing nature should be developed. The inoculation of nursery seedlings with appropriate ECM fungi is known to promote the uptake of nutrients and water, protection against various stresses, increased resistance against some pathogens and enhanced seedling regeneration and performance. Focus should be placed on applications of a multi-species fungal inoculum.

References

Aučina A., Rudawska M., Leski T., Ryliškis D., Pietras M., Riepšas E. 2011. Ectomycorrhizal fungal communities on seedlings and conspecific trees of Pinus mugo grown on the coastal dunes of the Curonian Spit in Lithuania. Mycorrhiza 21: 237–245.

Aučina A., Rudawska M., Leski T., Skridaila A., Pašakinskiene I., Riepšas E. 2014. Forest litter as the mulch improving growth and ectomycorrhizal diversity of bare-root Scots pine (Pinus sylvestris) seedlings. IForest 8: 394–400.

Aučina A., Rudawska M., Wilgan R., Janowski D., Skridaila A., Dapkūnienė S., Leski T. 2019. Functional diversity of ectomycorrhizal fungal communities along a peatland–forest gradient. Pedobiologia 74: 15–23.

Iwański M., Rudawska M. 2007. Ectomycorrhizal colonization of naturally regenerating Pinus sylvestris L. seedlings growing in different micro-habitats in boreal forest. Mycorrhiza 17: 461–467.

Iwański M., Rudawska M., Leski T. 2006. Mycorrhizal associations of nursery grown Scots pine (Pinus sylvestris L.) seedlings in Poland. Annals of Forest Science 63(7): 715–723.

Janowski D., Leski T. 2023. Methods for identifying and measuring the diversity of ectomycorrhizal fungi. Forestry: An International Journal of Forest Research 96(5): 639–652.

Karliński L., Ravnskov S., Rudawska M. 2020. Soil microbial biomass and community composition relates to poplar genotypes and environmental conditions. Forests 11: 262.

Karliński L., Rudawska M., Leski T. 2013. The influence of host genotype and soil conditions on ectomycorrhizal community of poplar clones. European Journal of Soil Biology 58: 51–58.

Kujawska M., Rudawska M., Wilgan R., Banach J., Leski T. 2023. Comparable ectomycorrhizal fungal species richness but low species similarity among native Abies alba and alien Abies grandis from provenance trials in Poland. Forest Ecology and Management 546: 121355.

Kujawska M.B., Rudawska M., Wilgan R., Leski T. 2021. Similarities and differences among soil fungal assemblages in managed forests and formerly managed forest reserves. Forests 12: 353.

Leski T., Aučina A., Skridaila A., Pietras M., Riepšas E., Rudawska M. 2010. Ectomycorrhizal community structure of different genotypes of Scots pine under forest nursery conditions. Mycorrhiza 20: 473–481.

Leski T., Pietras M., Rudawska M. 2010. Ectomycorrhizal fungal communities of pedunculate and sessile oak seedlings from bare-root forest nurseries. Mycorrhiza 20: 179–190.

Leski T., Rudawska M. 2012. Ectomycorrhizal fungal community of naturally regenerated European larch (Larix decidua) seedlings. Symbiosis 56: 45–53.

Leski T., Rudawska M., Aučina A. 2008. The ectomycorrhizal status of European larch (Larix decidua Mill.) seedlings from bareroot forest nurseries. Forest Ecology and Management 256(12): 2136–2144.

Leski T., Rudawska M., Kujawska M., Stasińska, M., Janowski D., Karliński L., Wilgan R. 2019. Both forest reserves and managed forests help maintain ectomycorrhizal fungal diversity. Biological Conservation 238: 108206.

Pietras M., Rudawska M., Leski T., Karliński L. 2013. Diversity of ectomycorrhizal fungus assemblages on nursery-grown European beech seedlings. Annals of Forests Sciences 70: 115–121.

Rudawska M. 2007. Mycorrhiza; The mycorrhizal status of Norway spruce. In: Tjoelker M.G., Boratynski A., Bugala W. (eds.). Biology and ecology of Norway spruce. Springer, Forestry Sciences 78: 182–194.

Rudawska M., Kieliszewska-Rokicka B., Leski T. 2000. Effect of aluminium on Pinus sylvestris seedlings mycorrhizal with aluminium-tolerant and aluminium-sensitive strains of Suillus luteus. Dendrobiology 45: 93–100.

Rudawska M., Kieliszewska-Rokicka B., Leski T., Staszewski T., Kubiesa P. 2003. Mycorrhizal community structure of Scots pine trees influenced by emissions from aluminum smelter. Developments in Environmental Science 3: 329–344.

Rudawska M., Kujawska M., Leski T., Janowski D., Karliński L., Wilgan R. 2019. Ectomycorrhizal community structure of the admixture tree species Betula pendula, Carpinus

betulus, and Tilia cordata grown in bare-root forest nurseries. Forest Ecology and Management 43: 113–125.
Rudawska M., Leski T. 2009. Znaczenie wiedzy o zbiorowiskach grzybów mikoryzowych w szkółkach leśnych dla rozwoju mikoryzacji sterowanej. Sylwan 153(1): 16–26.
Rudawska M., Leski T. 2021. Ectomycorrhizal fungal assemblages of nursery-grown Scots pine are influenced by age of the seedlings. Forests 12(2): 134.
Rudawska M., Leski T., Aučina A., Karliński L., Skridaila A., Ryliškis D. 2017. Forest litter amendment during nursery stage influence field performance and ectomycorrhizal community of Scots pine (Pinus sylvestris L.) seedlings outplanted on four different sites. Forest Ecology and Management 395: 104–114.
Rudawska M., Leski T., Gornowicz R. 2001. Mycorrhizal status of Pinus sylvestris L. nursery stock in Poland as influenced by nitrogen fertilization. Dendrobiology 46: 49–58.
Rudawska M., Leski T., Stasińska M. 2011. Species and functional diversity of ectomycorrhizal fungal communities on Scots pine (Pinus sylvestris L.) trees on three different sites. Annals of Forest Science 68: 5–15.
Rudawska M., Leski T., Stasińska M., Karliński L., Wilgan R., Kujawska M.B. 2022. The contribution of forest reserves and managed forests to the diversity of macrofungi of different trophic groups in European mixed coniferous forest ecosystem. Forest Ecology and Management 518: 120274.
Rudawska M., Leski T., Trocha L.K., Gornowicz R. 2006. Ectomycorrhizal status of Norway spruce seedlings from bare-root forest nurseries. Forest Ecology Management 236: 375–384.
Rudawska M., Leski T., Wilgan R., Karliński L., Kujawska M., Janowski D. 2018. Mycorrhizal associations of the exotic hickory trees, Carya laciniosa and Carya cordiformis, grown in Kórnik Arboretum in Poland. Mycorrhiza 28: 549–560.
Rudawska M., Pietras M., Smutek I., Strzeliński P., Leski T. 2016. Ectomycorrhizal fungal assemblages of Abies alba Mill. outside its native range in Poland. Mycorrhiza 26: 57–65.
Rudawska M., Wilgan R., Janowski D., Iwański M., Leski T. 2018. Shifts in taxonomical and functional structure of ectomycorrhizal fungal community of Scots pine (Pinus sylvestris L.) underpinned by partner tree ageing. Pedobiologia 71: 20–30.
Stankeviciene D., Kasparavicius J., Rudawska M., Iwański M. 2008. Studies of ectomycorrhizal fungi aboze-and belowground in the 50-year-old Pinus sylvestris forest. Baltic Forestry 14(1): 7–15.
Trocha L.K., Kałucka I., Stasińska M., Nowak W., Dabert M., Leski T., Rudawska M., Oleksyn J. 2012. Ectomycorrhizal fungal communities of native and non-native Pinus and Quercus species in a common garden of 35-year-old trees. Mycorrhiza 22: 121–134.
Trocha L.K., Oleksyn J., Turzańska E., Rudawska M., Reich P.B. 2007. Living on the edge: Ecology of an incipient Betula-fungal community growing on brick walls. Trees – Structure and Function 21: 239–247.
Trocha L.K., Rudawska M., Leski T., Dabert M. 2006. Genetic diversity of naturally established ectomycorrhizal fungi on Norway spruce seedlings under nursery conditions. Microbial Ecology 52: 418–425.
Wilgan R., Leski T., Kujawska M., Karliński L., Janowski D., Rudawska M. 2020. Ectomycorrhizal fungi of exotic Carya ovata (Mill.) K. Koch in the context of surrounding native forests on Central European sites. Fungal Ecology 44: 100908.

Biogeography of potentially invasive non-pathogenic fungi in Europe

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Alien or invasive organisms can be plants, animals, fungi, or other organisms that, intentionally or accidentally, were moved away from their natural origin. In a new area, foreign species may change the structure and species composition of native ecosystems by competition for life resources and space. Nowadays, the occurrence of alien and invasive organisms is one of the most important problems of nature conservation (Vitousek et al. 1996). Most of the currently conducted studies focus on the introduction of various plants and animals, mainly those with high economic importance (Desprez-Loustau et al. 2007) and pathogenic organisms. However, little is known about the spread of foreign species of fungi, especially non-pathogenic ones, beyond their natural range.

The history of biological introductions into new areas began almost 500 years ago, after Columbus's first voyage to the Americas. Globally, there is no detailed information on the number of introduced organisms. It is estimated that during the last 500 years, approximately 50,000 species have been transported to North America alone. The estimated cost of biological invasions in the United States has currently reached approximately 137 million dollars per year (Pimentel et al. 2000). In New Zealand more than 25,000 plant species, including 2,500 invasive taxa, have been introduced in the last 100 years. Most of the introductions described so far were caused by intentional human activities linked with transferring goods, crops, or economically important organisms. For example, many economically important trees, such as fast-growing pines or Douglas fir were moved intentionally, because of fast timber production. Similarly, several plant species were introduced into new areas due to their high ornamental value. In contrast to intentionally transferred plants, microorganisms, including fungi, are mostly introduced circumstantially, mainly by international trade or tourism. For example, the transfer of the fungal-like oomycete, *Phytophthora infestans*, which caused potato blight diseases in Ireland in the mid-19th century, resulted in one of the greatest famines in the history of the world. The list of the "One Hundred Most Dangerous Organisms of Alien Origin" includes only two fungal pathogens affecting trees, *Ophiostoma ulmi*, which causes Dutch elm disease, and *Phytophthora cinnamomi* – an oomycete causing the death of many tree species (Lowe et al.

2000). In contrast to pathogens listed above, the occurrence of less economically important alien non-pathogenic fungi falls outside the interests of the scientific community. Due to their cryptic nature, fungi can exist for several years as a mycelium overgrowing the soil substrate without the appearance of sporocarps. Therefore, one of the most significant gaps limiting research on the biogeography of alien non-pathogenic fungi is the low level of knowledge about species distributions on regional and continental scales. It is assumed that the small number of records of foreign ectomycorrhizal fungi is closely related to the small number (and often the lack) of publications from a given country (Vellinga et al. 2009), which is especially evident from European lists of mycobiota. However, symbiotic fungi show the potential to be widely introduced beyond their natural range. Yet, until recently, non-native ectomycorrhizal fungi received scant consideration in invasion ecology. The review presented by Vellinga et al. (2009) revealed more than 190 publications reporting ectomycorrhizal introductions and at least 200 species that have been moved from native ranges to novel habitats. However, only a few thorough studies have been conducted that describe ectomycorrhizal expansion in the Northern Hemisphere, including Europe. *Laccaria fraterna*, an Australian fungus associated with eucalypt trees has been introduced to Europe and now occurs with rockroses, European pines, and oaks (Diez et al. 2005). *Aureoboletus projectellus* is well known as a highly adapted mycorrhizal fungus that spreads successfully in pine forests, forming ectomycorrhizal assemblages with native European tree partners (Wrzosek et al. 2017). In these cases, fungi persist with the introduced hosts and are also able to spread to local tree species. However, this invasion pattern is not common in the invasion biology of fungi. The most frequently recorded outcome of the introduction of symbiotic fungi involves introducing fungi with foreign host plants that do not spread to native trees. These fungi are naturally associated with trees planted for forestry, mainly conifers. This phenomenon, called coinvasion, is undoubtedly the most observed process described in plant-fungus invasion ecology (Dickie et al. 2010).

According to the Delivering Alien Invasive Species Inventories for Europe database (DAISIE; http://www.europe-aliens.org/), there are 54 alien conifer species in Europe. However, only the North American *Pinus strobus* is regarded as an invasive coniferous species in Europe (Carrillo-Gavilan, Vila 2010). The consequence of the introduction of foreign conifers may be the introduction of their ectomycorrhizal fungi. Suilloid fungi, specifically the genera *Suillus* and *Rhizopogon* (Boletales, Basidiomycota), are regarded as a crucial driver of pine (and related Pinaceae trees) invasions at the global scale (Policelli et al. 2019). Therefore, a large body of literature has been previously published describing coinvasions of Pinaceae trees and their associated symbionts, but most of the conducted studies have been performed in the Southern Hemisphere, where several non-native pine species were naturalized and the coinvasion processes have been intensively studied (Vellinga et al. 2009). In the Northern Hemisphere, the role of symbiotic fungi in coinvasion has been less documented. Besides our previous papers dealing with the distribution of Douglas fir (Pietras et al 2018), North American pines (Pietras, Kolanowska 2019) and their associated suilloid fungi, little is known about the non-native trees transferred to Europe in terms of their symbiotic fungi (Policelli et al. 2019).

The third possibility in the colonization of new areas is simply to not be dependent on mutualism. For example, many common plant invaders are non-mycorrhizal. On the other hand, some non-mycorrhizal fungi can be moved into new areas independently, without plant or animal vectors. For example, species having low dependence on symbioses or being non-mycorrhizal invasive organisms with no need to establish mycorrhizal associations might limit their invasion. The relatively few studies of non-pathogenic fungi focused mainly on saprobic fungi. The expansion of Australian *Clathrus archeri* (Parent et al. 2000, Pietras et al. 2016, 2021) and wood-inhabiting *Favolaschia calocera* (Vizzini et al. 2009) are among a few examples where probable pathways of introduction have been described.

Can non-pathogenic fungi be invasive?

The International Union for Conservation of Nature (IUCN) defines invasive species as a species that is established outside its natural past or present area of distribution, the introduction and/or spread of which threatens biological diversity. However, according to the definition proposed by Blackburn et al. (2011) the term "invasive" can be used for organisms with self-sustaining populations at significant distances from the place of original introduction. The crucial difference between these two definitions is the necessity of the particular species to threaten biological diversity. Thus, based on IUCN requirements, any alien, non-pathogenic fungi in Europe do not yet qualify to be termed "invasive", but rather potentially invasive. The presentation focus is on non-pathogenic taxa non-native to Europe, specifically ectomycorrhizal *Aureoboletus projectellus*, suilloid fungi like *Suillus lakei*, *S. placidus*, *Rhizopogon salebrosus*, and *R. verii*, as well as two saprotrophic taxa: *Stropharia rugosoannulata* and *Clathrus archeri*. The main aim of the lecture is to present various methodological approaches, from the classical taxonomy of fungi, and molecular biology methods, to ecological modeling tools that can be used in biogeographical studies of non-pathogenic fungi; and to consider the associated limitations and biases, as well as perspectives and challenges in invasion ecology and biogeography of fungi.

Where is the problem?

In general, in situ detection of non-native fungi occurring in native ecosystems is very difficult. Fungi are usually cryptic organisms and are found mostly using field surveys of conspicuous fruiting bodies, although the mycelia and mycorrhizal root tips of these species may have persisted for many years before fruiting bodies were produced. The second bias of research on biogeography and ecology on non-pathogenic fungi is the poorly documented geographic ranges of many species or even lineages of fungi. The biogeography of fungi, in general, remains largely unknown, consequently very limited attention has been paid to the spreading of non-pathogenic fungi outside their native range. Recently, this knowledge gap has been particularly worrisome with regard to some suilloid (Pietras et al. 2018, Pietras 2019, Pietras, Kolanowska 2019) and boletoid (Wrzosek at al. 2017, Banasiak et al. 2019a, b) fungi; both groups originated from North America and are spreading in Europe. Still, relatively little is known about the plant-fungus and fungus-ecosystem interactions outside the natural ranges of fungi.

The next problem is that fungi encompass several million species, but most of them are undiscovered (Taylor et al. 2014). Moreover, fungi can be described for the first time or known only from an invasive range (Galan, Moreno 1998). Species concepts in fungi are also often unclear, making identification and range mapping more difficult. This explains why non-pathogenic fungi are usually poorly represented in databases of alien organisms, if at all. According to Desprez-Loustau et al. (2007), the main reason for poor fungi representation in databases of alien organisms is the limited baseline data on fungal communities.

Studied macrofungi

Fungal ecology and biogeography is one of the areas most explored by scientists from the Institute of Dendrology PAS. In recent years, three scientific projects dealing with the early detection, biogeography, and impacts of non-pathogenic fungi have been carried out. Moreover, over the last 10 years almost 200 records of non-native macrofungi have been gathered and published in open databases or deposited in fungaria by mycologists from the Institute, significantly increasing the level of knowledge about the biogeography and ecology of non-native fungi worldwide (Pietras et al. 2018, 2021, Banasiak et al. 2019a, b, Pietras, Kolanowska 2019, Pietras 2019). The main interests of the conducted research have focused on macrofungi with distinct morphological features that are easily recognizable in field conditions, making their identification easy even to the naked eye. Moreover, the geographic distribution of fungi presented below is well recognized in native and non-native ranges, and the chosen fungi are widespread in areas where they occur naturally and frequently in areas where have been introduced. All these aspects allow the studied taxa to be classified as model organisms suitable for biogeographical research.

Aureoboletus projectellus is a North American bolete that was reported for the first time on the Baltic Sea shore in 2007 (Motiejunaite et al. 2011, Wrzosek et al. 2017). Since then, the fungus has been rapidly extending its range and has been found to be a common component of ectomycorrhizal assemblages of European pines. Dickie et al. (2016) classified *A. projectellus* as an example of an invasive ectomycorrhizal fungus, but its impacts on native mycobiota and ecosystems has not been studied (Wrzosek et al. 2017), thus this taxon cannot be classified as an invasive according to the IUCN's definition.

Suillus lakei is a commonly recorded ectomycorrhizal partner of Douglas fir in North America. The presence of *S. lakei* has also been documented outside its natural range: in South America, Europe, and New Zealand (Pietras et al. 2018, Vellinga et al. 2009 and references therein), as a result of its introduction with

Douglas fir. In Europe, Douglas fir plantations cover 750,000 ha, but the coinvasion of *S. lakei* and its mycorrhizal partner Douglas fir is limited. Therefore, their impact on native ecosystems and potential invasiveness in this region has not been studied.

Clathrus archeri is a saprotrophic fungus native to Australia and New Zealand, and it has been gradually spreading throughout the world (Perent et al. 2000). In Europe, it was first recorded in 1914, in France, presumably as a result of international trade or military supplies at the beginning of the First World War. Since then basidiocarps of this fungus have been recorded hundreds of times, mostly in Western and Central Europe, and recently the species has also been recorded more than 150 times in Poland, Ukraine, and Lithuania (Pietras et al. 2016). Thus, the conclusion can be made that *C. archeri* is the most widespread exotic non-pathogenic fungus in Europe.

Stropharia rugosoannulata is a North American agaric fungus widespread and fairly common east of the Great Plains, and occasionally reported in Washington and British Columbia. The fungus is commonly known as the wine cap stropharia, "garden giant", burgundy mushroom, or king stropharia. *Stropharia rugosoannulata* was introduced to Europe in the mid-20th Century for cultivation and was similarly moved to Australia and New Zealand. Escape from cultivation occurs in different soil environments, but always on straw mulch, woody debris, or rich, humus soil.

How to find an alien?

Nowadays, the constant development of new technologies and the increasing reliance on social media offer excellent opportunities for comprehensive mycological research (Heilmann-Clausen et al. 2016), and allow us to obtain detailed information from many places on Earth (Andrew et al. 2018). Using open databases such as the UNITE sequence database, MycoPortal, and the Global Biodiversity Information Facility is crucial for achieving comprehensive information on the distribution and occurrence of non-native fungi. An additional source of knowledge can be information associated with the specimens deposited in herbaria. Using standardized biogeographic data obtained from open databases and thanks to the possibility of querying herbarium specimens, we were able to conduct studies on the occurrence of *Suillus lakei* (Pietras et al. 2018), *Clathrus archeri* (Pietras et al. 2021), and *Aureoboletus projectellus* (Banasiak et al. 2019a, 2019b). Citizen science was also used to detect human vectors in the spread of *A. projectellus* in Poland. An additional advantage of using data related to herbarium specimens is the possibility of later use of the collected vouchers in research based on advanced molecular methods, e.g., population genetics.

Molecular tools in fungal biogeography

The development of molecular tools contributed to the development of the biological sciences, including mycology. The genetic approach can be used in taxonomic, ecological, and population studies. The recognition of the ITS marker as a "barcode" used for molecular identification of fungi is also of great importance. Based on this approach, molecular identification of ectomycorrhizae was performed to investigate fungi assembled with North American Douglas fir and early detection of *Suillus lakei* in Europe (Pietras et al. 2018). Similar research was conducted in Eastern white pine plantations (Pietras, Kolanowska 2019), where the presence of the North American fungus *Rhizopogon salebrosus* was confirmed in Pomerania. This finding is the first record of the fungus in Poland (Pietras, Kolanowska 2019). The documentation of both *S. lakei* and *R. salebrosus* in Poland proves that molecular markers are helpful in the early detection of alien symbiotic fungi, even though the fungus does not form visible sporocarps. The ITS-based approach opens the way to develop species specific primers allowing to detect non-native fungi directly from the soil, without time-consuming sporocarps surveys.

Species distribution models

The niche modeling tools, using presence-only data, allow us to estimate the potential range of a fungus based on the analysed climatic or environmental variables. The research carried out showed the potential occurrence of *Suillus lakei* on a regional and global scale (Pietras et al 2019). The performed models showed that the distribution of the climatic niche useful for the fungus, is on the one hand comparable to the range of the tree's potential occurrence, and on the other hand, much wider than the currently known range of the fungus. In Europe, where this taxon is relatively rare and occurs mainly in Central and Southern Europe, the range of the bioclimatic niche is much wider. In a study dealing with the distribution of the bioclimatic niche of *Rhizopogon salebrosus* the species distribution models were used to assess the distribution of the fungus in Europe (Pietras, Kolanowska 2019). The conducted analysis shows that the currently known distribution of the fungus covers a potential natural range. In Europe, the predicted highest occurrences of the fungus were in mountainous regions, including the Alps, the Sudetes and other mountain regions, such as the Cordilleras, Carpathian Mountains, Dinaric Mountains, Pontic Mountains, Taurus Mountains, Caucasus, and Middle Iranian Mountains. Modeling tools can also give information on historical biogeography and the impact of future climate change on the distribution of fungi. The study describing the distribution of *Clathrus archeri* showed historical and future range changes caused by climate change (Pietras et al. 2021). This study revealed that observed climate changes may significantly accelerate the loss of suitable area in the natural range, but on the other hand, the fungus can extend its area beyond its natural range, making it a potentially invasive species.

Perspectives and challenges

Detailed descriptions of the biogeography of non-native macrofungi are still needed. However, the most challenging aspect of their ecology is to investigate the impact of fungal introduction on native ecosystems, especially regarding competition with native mycobiota, the influence on soil carbon cycle, and impacts on other components of native ecosystems. At the Institute of Dendrology PAS we are employing several approaches focused on population genetics, ecosystem functioning and molecular biology of potentially invasive fungi, to better understand the invasion ecology of non-pathogenic fungi and to prove their negative impacts on native ecosystems. Therefore, our everyday work connects different science disciplines (such as fungal ecology, mycology, environmental sciences, and molecular biology) that consider the invasion ecology of fungi.

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References

Andrew C., Diez J., James T.Y., Kauserud H. 2018. Fungarium specimens: a largely untapped source in global change biology and beyond. Philosophical Transactions of the Royal Society B: Biological Sciences 374: 20170392.

Banasiak Ł., Pietras M., Wrzosek M., Okrasińska A., Gorczak M., Kolanowska M., Pawłowska J. 2019a. Aureoboletus projectellus (Fungi, Boletales) – An American bolete rapidly spreading in Europe as a new model species for studying expansion of macrofungi. Fungal Ecology 39: 94–99.

Banasiak Ł., Pietras M., Wrzosek M., Okrasińska A., Gorczak M., Kolanowska M., Pawłowska J. 2019b. Aureoboletus projectellus (Fungi, Boletales) occurrence data, environmental layers and habitat suitability models for North America and Europe. Data in Brief 23: 103779.

Blackburn T.M., Pysek P., Bacher S., Carlton J.T., Duncan R.P., Jarosik V., Wilson J.R., Richardson D.M. 2011. A proposed unified framework for biological invasions. Trends in Ecology & Evolution 26: 333–339.

Carrillo-Gavila M.A., Vila M. 2010. Little evidence of invasion by alien conifers in Europe. Diversity and Distributions 16: 203–213.

Desprez-Loustau M.L., Robin C., Buée M., Courtecuisse R., Garbaye J., Suffert F., Sache I., Rizzo D. 2007. The fungal dimension of biological invasions. Trends in Ecology & Evolution 22: 472–487.

Dickie I.A., Bolstridge N., Cooper J.A., Peltzer D.A. 2010. Coinvasion by Pinus and its mycorrhizal fungi. New Phytologist 187: 475–484.

Dickie I.A., Nunez M.A., Pringle A., Lebel T., Tourtellot S., Johnston P.R. 2016. Towards management of invasive ectomycorrhizal fungi. Biological Invasions 18: 3383–3395.

Díez J. 2005. Invasion biology of Australian ectomycorrhizal fungi introduced with eucalypt plantations into the Iberian Peninsula. Biological Invasions 7: 3–15.

Galan R., Moreno G. 1998. Ruhlandiella berolinensis, an exotic species in Europe. Mycotaxon 68: 265–271.

Heilmann-Clausen H., Maruyama P.H., Bruun H.H., Dimitrov D., Læssøe T., Frøslev D.G., Dalsgaard B. 2016. Citizen science data reveal ecological, historical and evolutionary factors shaping interactions between woody hosts and wood-inhabiting fungi. New Phytologist 212: 1072–1082.

Lowe S., Browne M., Boudjelas S., De Poorter M. 2000. 100 of the World's Worst Invasive Alien Species A selection from the Global Invasive Species Database Published by The Invasive Species Specialist Group (ISSG) a specialist group of the Species Survival Commission (SSC) of the World Conservation Union (IUCN).

Motiejunaite J., Kasparavicius J., Kacergius A. 2011. Boletellus projectellus – an alien mycorrhizal bolete new to Europe. Sydowia 63: 203–213.

Parent G.H., Thoen D., Calonge F.D. 2000. Nouvelles données sur la répartition de Clathrus archeri en particulierdansl'Ouest et le Sud-Ouest de l'Europe. Bulletin de la Société mycologique de France 116: 241–266.

Pietras M. 2019. First record of North American fungus Rhizopogon pseudoroseolus in Australia and prediction of its occurrence based on climatic niche and symbiotic partner preferences. Mycorrhiza 29: 397–401.

Pietras M., Kolanowska M. 2019. Predicted potential occurrence of the North American false truffle Rhizopogon salebrosus in Europe. Fungal Ecology 9: 225–230.

Pietras M., Kolanowska M., Selosse M.-A. 2021. Quo vadis? Historical distribution and impact of climate change on the worldwide distribution of the Australasian fungus Clathrus archeri (Phallales, Basidiomycota). Mycological Progress 20: 299–311.

Pietras M., Litkowiec M., Gołębiewska J. 2018. Current and potential distribution of the ectomycorrhizal fungus Suillus lakei ((Murrill) AH Sm & Thiers) in its invasion range. Mycorrhiza 28: 467–475.

Pietras M., Rudawska M., Iszkuło G., Kujawa A., Leski T. 2016. Distribution and molecular characterization of an alien fungus, Clathrus archeri, in Poland. Polish Journal of Environmental Studies 25: 1197–1204.

Pimentel D., Lach L., Zuniga R., Morrison D. 2000. Environmental and economic costs associated with non-indigenous species in the United States. BioScience 50: 53–65.

Policelli N., Bruns T.D., Vilgalys R., Nunez N.A. 2019. Suilloid fungi as global drivers of pine invasions. New Phytologist 222: 714–725.

Taylor D.L., Hollingsworth T.N., McFarland J.W., Lennon N.J., Nusbaum C., Ruess R.W. 2014. A first comprehensive census of fungi in soil reveals both hyperdiversity and fine-scaleniche partitioning. Ecological Monographs 84: 3–20.

Vellinga E.C., Wolfe B.E., Pringle A. 2009. Global patterns of ectomycorrhizal introductions. New Phytologist 181: 960–973.

Vitousek P.M. 1996. Biological invasions as global environmental changes. American Scientist 84: 468–478.

Vizzini A., Zotti M., Mello A. 2009. Alien fungal species distribution: the study case of Favolaschia calocera. Biological Invasions 11: 417–429.

Wrzosek M., Motiejūnaitė J., Kasparavičius J., Wilk M., Mukins E., Schreiner J., Vishnevskiy M., Gorczak M., Okrasińska A., Istel Ł., Pawłowska J. 2017. The progressive spread of Aureoboletus projectellus (Fungi, Basidiomycota) in Europe. Fungal Ecology 27: 134–136.

What's the point of toothed leaves?

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Leaves play a crucial role in the growth, development, and survival of plants. They are engaged in capturing light, transporting water together with mineral salts and assimilates, facilitating gas exchange, dissipating heat, and providing defence against herbivores and pathogens (Nicotra et al. 2011). The diversity in their morphology and structure is shaped by these functions and the changing environmental conditions (Nicotra et al. 2011, McKee 2017). In particular, through the process of evolution, angiosperms have developed the widest variety of leaf forms. The evolution, growth, and development of these forms are intricately linked to both the species' evolutionary background and the environmental conditions, as well as the developmental stage at which they find themselves (Nicotra et al. 2011).

It is believed that teeth may provide protection against herbivores. For example, insects often refrain from feeding on leaves inhabited by other insects. In this case, the teeth visually resemble insect damage, which deters insects (Ehrlich, Raven 1967, Curtis, Lersten 1974, Givnish 1979, Rivero-Lynch et al. 1996). Teeth may also be the result of hydraulic constraints of the xylem (Givnish 1979, Zwieniecki et al. 2004, Givnish, Kriebel 2017). The leaf is a complex hydraulic system. Delivering water to surfaces located between conducting bundles is relatively difficult, so during leaf development there is a restriction of tissues difficult in such transport, ensuring the most efficient water delivery. On the other hand, teeth may accelerate water removal (Field et al. 2005). Water stomata in teeth prevent water from filling the intercellular spaces in plants, with the function of hydathodes being most significant in young plants. Additionally, the functional consequence of hydathode development on teeth is easier guttation (Royer, Wilf 2006). Accelerating the flow of phloem sap or efficient removal of excess water is related to a large number of hydathodes and stomatal apparatuses (Roth et al. 1995, Feild et al. 2005).

Teeth on leaves could significantly enhance the carbon absorption rate at the start of the vegetation period (Baker-Brosh, Peet 1997, Feild et al. 2005, Royer, Wilf 2006, Peppe et al. 2011). Observations reveal that the teeth on young, developing leaves are notably larger than those on mature leaves (Baker-Brosh, Peet 1997, Feild et al. 2005). The teeth found on young *Ulmus* leaves are characterized by a well-developed spongy mesophyll, suggesting a more vigorous gas exchange compared to the central portion of the leaf (Baker-Brosh, Peet 1997, Nicotra et al. 2011). This observation is supported by Zwieniecki's (2004) research on *Quercus rubra*, which confirms that the developing leaves have a larger surface area.

The large surface area of teeth in developing leaves may indicate a functional relationship between the presence of irregularity on the leaf blade edge and the intensity of photosynthesis in the early stages of vegetation (Baker-Brosh, Peet 1997, Wright et al. 2005). The hypothesis that increased gas exchange at the leaf blade's edge can explain this relationship posits that such an adaptation enhances early-stage photosynthesis. This adaptation could lead to more efficient export and assimilate supply by young and developing leaves, especially when the crown is less dense, allowing more light to reach the lower parts, and when water availability is high in spring (Zohner et al. 2019). As the irregularities on the leaf blade's edge diminish with maturation, it is presumed that light conditions can affect the rate of these changes. Consequently, one might expect to find more pronounced irregularities on the edges of leaves in shaded areas. Yet, to date, no such comparative studies have been undertaken.

Another potential role of irregularity on the leaf blade edge is associated with the improvement of light conditions of lower leaves. Teeth cause light to refract, disperse, and consequently improve its penetration to the lower parts of the crown (Niklas 1989, Nicotra et al. 2011, Rubio de Casas et al. 2011, Maslova et al. 2018).

Leaf morphology is shaped by numerous factors, including adaptation to environmental conditions and their primary function of carbon dioxide assimilation (McDonald et al. 2003, Nicotra et al. 2011). Early in the 20th century, scientists observed a link between climate and leaf blade edge irregularities, noting that species with toothed leaves were more common in cooler climates than in warmer ones (Bailey, Sinnott 1915, 1916). Paleobotany uses this observation to deduce climate conditions from the species composition of fossil floras (Wolfe 1978, 1993, Huff et al. 2003, Greenwood et al. 2004, Nicotra et al. 2011, McKee 2017). Numerous studies have verified the correlation between average temperature and the presence of species with toothed versus entire leaf blades in local floras (Wolfe 1979, Traiser et al. 2005, Royer, Wilf 2006, Royer et al. 2009, Little et al. 2010, Peppe et al. 2011, Yang et al. 2015). However, the specific causes behind the irregularity of the leaf blade edge remain largely unknown (Wolfe 1993, Roth et al. 1995, Baker-Brosh, Peet 1997, Wilf 1997, Field et al. 2005). Further research is needed to explore this dependency within the context of assessing climatic conditions of fossil floras and to better understand this phenomenon. However, the relationship does not appear to be straightforward or universal. For instance, it has been pointed out that in cooler conditions (around up to 5°C) this dependency is often not observed (Peppe et al. 2011). An explanation might be the predominant presence of evergreen shrubs that overwinter under snow in cooler local climates, where species with evergreen leaves tend to have entirely edged blades, dominating over species with toothed leaves (Givnish 1979, Wolfe 1993, Peppe et al. 2011).

Phylogenetic studies also indicate the decisive influence of temperature on the appearance of irregularities on the leaf blade (Little et al. 2010). Teeth are commonly found in 42 orders and 173 families of angiosperms (Rios et al. 2020). Jones et al. (2009) suggest that leaves with teeth have evolved many times independently in unrelated groups of plants, and that this is a labile trait indicating adaptive adaptations and response to environmental conditions (Baker-Brosh, Peet 1997, Iszkuło, Myślicka 2015, McKee 2017). To date, the causes and mechanisms that would confirm that a cooler climate may influence the irregularity of the leaf blade edges, and that among closely related species the relationship between leaf form – climate and leaf habit will be preserved, have not been clearly identified.

The hypothesis of bud packing emerged while seeking answers to questions regarding the causes behind the formation of teeth and irregularities on the leaf blade. The development of leaf primordia and the way they are packed in winter buds may influence the shaping of the leaf blade edge appearing early in spring (Kobayashi et al. 1998, Couturier et al. 2012, Edwards et al. 2016, 2017). The presence of teeth or lobing may affect denser packing of leaves in buds, thereby contributing to their earlier appearance in spring (Lopez et al. 2008). This was confirmed by the observations of Edwards et al. (2016) indicating that in *Viburnum* leaves appearing earlier on the shoot are leaves that develop longer in buds: they are more toothed than those that appear later. On the other hand, Givnish and Kriebel (2017) argue that this hypothesis may rather explain the development of lobing of the leaf blade, not the teeth themselves. Therefore, the cause shaping the irregular edges of the leaf blade remains unknown.

References

Bailey I.W., Sinnott E.W. 1915. A botanical index of cretaceous and tertiary climates. Science 41: 831–834.

Bailey I.W., Sinnott E.W. 1916. The climatic distribution of certain types of angiosperm leaves. American Journal of Botany 3: 24–39.

Baker-Brosh K.F., Peet R.K. 1997. The ecological significance of lobed and toothed leaves in temperate forest trees. Ecology 78: 1250–1255.

Couturier E., Brunel N., Douady S., Nakayama N. 2012. Abaxial growth and steric constraints guide leaf folding and shape in Acer pseudoplatanus. American Journal of Botany 99: 1289–1299.

Curtis J.D., Lersten N.R. 1974. Morphology, seasonal variation, and function of resin glands on buds and leaves of Populus deltoides (Salicaceae). American Journal of Botany 61: 835–845.

Edwards E.J., Chatelet D.S., Spriggs E.L., Johnson E.S., Schlutius C., Donoghue M.J. 2017. Correlation, causation, and the evolution of leaf teeth: A reply to Givnish and Kriebel. American Journal of Botany 104: 509–515.

Edwards E.J., Spriggs E.L., Chatelet D.S., Donoghue M.J. 2016. Unpacking a century-old mystery: Winter buds and the latitudinal gradient in leaf form. American Journal of Botany 103: 975–978.

Ehrlich P.R., Raven P.H. 1967. Butterflies and plants. Scientific American 216: 104–114.

Feild T.S., Sage T.L., Czerniak C., Iles W.J.D. 2005. Hydathodal leaf teeth of Chloranthus japonicus (Chloranthaceae) prevent guttation-induced flooding of the mesophyll. Plant, Cell and Environment 28: 1179–1190.

Givnish T.J. 1979. On the adaptive significance of leaf form. The American Naturalist 120: 375–407.
Givnish T.J., Kriebel L. 2017. Causes of ecological gradients in leaf margin entirety: Evaluating the roles of biomechanics, hydraulics, vein geometry, and bud packing. American Journal of Botany 104: 354–366.
Greenwood D.R., Wilf P., Wing S.L., Christophel D.C. 2004. Paleotemperature estimation using leaf- margin analysis: Is Australia different? Palaios 19: 129–142.
Iszkuło G., Myślicka K. 2015. Is climate warming advantageous for plants with untoothed leaves? Basic and Applied Ecology 16: 386–393.
Jones C.S., Bakker F.T., Schlichting C.D., Nicotra A.B. 2009. Leaf shape evolution in the South African genus Pelargonium L'Her. (Geraniaceae). Evolution 63: 479–497.
Kobayashi H., Kresling B., Vincent J.F.V. 1998. The geometry of unfolding tree leaves. Proceedings of the Royal Society B. Biological Sciences 265: 147–154.
Little S.A., Kembel S.W., Wilf P. 2010. Paleotemperature proxies from leaf fossils reinterpreted in light of evolutionary history. PLoS ONE 5(12): e15161.
Lopez O.R., Farris-Lopez K., Montgomery R.A., Givnish T.J. 2008. Leaf phenology in relation to canopy closure in southern Appalachian trees. American Journal of Botany 95: 1395–1407.
Maslova N.P., Karasev E.V., Xu S.-L., Spicer R.A., Liu X.-Y., Kodrul T.M., Spicer T.E.V., Jin J.-H. 2021. Variations in morphological and epidermal features of shade and sun leaves of two species: Quercus bambusifolia and Q. myrsinifolia. American Journal of Botany 108: 1441–1463.
McDonald P.G., Fonseca C.R., Overton J.M., Westoby M. 2003. Leaf-size divergence along rainfall and soil-nutrient gradients: is the method of size reduction common among clades? Functional Ecology 17: 50–57.
McKee M. 2017. How does temperature impact leaf size and shape in four woody dicot species? Testing the assumptions of leaf physiognomy-climate models. Wesleyan University [thesis].
Nicotra A.H., Leigh A., Boyce K., Jones D., Niklas E., Royer D.L., Tsukaya H. 2011. The evolution and functional significance of leaf shape in the angiosperms. Functional Plant Biology 38: 535–552.
Niklas K.J. 1989. The effect of leaf-lobing on the interception of direct solar radiation. Oecologia 80: 59–64.
Peppe D.J., Royer D.L., Cariglino B., Oliver S.Y., Newman S., Leight E., et al. 2011. Sensitivity of leaf size and shape to climate: Global patterns and paleoclimatic applications. New Phytologist 190: 724–739.
Rios A.B.M., de Oliveira Menino G.C., Dalvi V.C. 2020. Leaf teeth in eudicots: what can anatomy elucidate? Botanical Journal of the Linnean Society 193: 504–522.
Rivero-Lynch A.P., Brown V.K., Lawton J.H. 1996. The impact of leaf shape on the feeding preference of insect herbivores: Experimental and field studies with Capsella and Phyllotreta. Philosophical Transactions of the Royal Society B: Biological Sciences 351: 1671–1677.
Roth A., Mosbrugger V., Belz G., Neugebauer H.J. 1995. Hydrodynamic modelling study of angiosperm leaf venation types. Botanica Acta 108: 121–126.
Royer D.L., Meyerson L.A., Robertson K.M., Adams J.M. 2009. Phenotypic plasticity of leaf shape along a temperature gradient in Acer rubrum. PLoS ONE 4(10): e7653.
Royer D.L., Wilf P. 2006. Why do toothed leaves correlate with cold climates? Gas exchange at leaf margins provides new insights into a classic paleotemperature proxy. International Journal of Plant Sciences 167: 11–18.

Rubio de Casas R., Vargas P., Pérez-Corona E., Manrique E., García-Verdugo C., Balaguer L. 2011. Sun and shade leaves of Olea europaea respond differently to plant size, light availability and genetic variation. Functional Ecology 25: 802–812.

Wilf P. 1997. When are leaves good thermometers? A new case for Leaf Margin Analysis. Paleobiology 23(3): 373–390.

Wolfe J.A. 1978. A paleobotanical interpretation of tertiary climates in the northern hemisphere: data from fossil plants make it possible to reconstruct Tertiary climatic changes, which may be correlated with changes in the inclination of the earth’s rotational axis. American Scientist 66: 694–703.

Wolfe J.A. 1979. Temperature parameters of humid to mesic forests of Eastern Asia and relation to forests of other regions of the Northern Hemisphere and Australasia. Geological Survey Professional Paper 1106: 1–37.

Wolfe J.A. 1993. A method of obtaining climatic parameters from leaf assemblages. Geological Survey Bulletin 2040: 1–75.

Wright I.J., Reich P.B., Cornelissen J.H.C., Falster D.S., Garnier E., Hikosaka K., Lamont B.B., Lee W., Oleksyn J., Osada N., Poorter H., Villar R., Warton D.I., Westoby M. 2005. Assessing the generality of global leaf trait relationships. New Phytologist 166: 485–496.

Yang J., Spicer R.A., Spicer T.E.V., Arens N.C., Jacques F.M.B., Su T., Kennedy E.M., Herman A.B., Steart D.C., Srivastava G., Mehrotra R.C., Valdes P.J., Mehrotra N.C., Zhou Z.-K., Lai J.-S. 2015. Leaf form-climate relationships on the global stage: an ensemble of characters. Global Ecology and Biogeography 24: 11131–1125.

Zohner C.M., Ramm E., Renner S.S. 2019. Causation of toothed leaves in northern floras. Examining the support-supply and bud-packing hypotheses for the increase in toothed leaf margins in northern deciduous floras. American Journal of Botany 106: 1404–1411.

Zwieniecki M.A., Boyce C.K., Holbrook N.M. 2004. Hydraulic limitations imposed by crown placement determine final size and shape of Quercus rubra L. leaves. Plant, Cell and Environment 27: 357–365.

Recent contributions to the biodiversity of the Stołowe Mountains National Park: research and practice

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Stołowe Mountains National Park (SMNP) is one of the most explored national parks in Poland in terms of scientific research conducted by scientists from the Institute of Dendrology PAS. Just in the last few years, we conducted three research projects aimed at the recognition of habitats in the vicinity of main streams, small rivers and springs covering the whole SMNP area (fig. 1). The research took place in 2017, 2021 and 2023, and the results and conclusions served

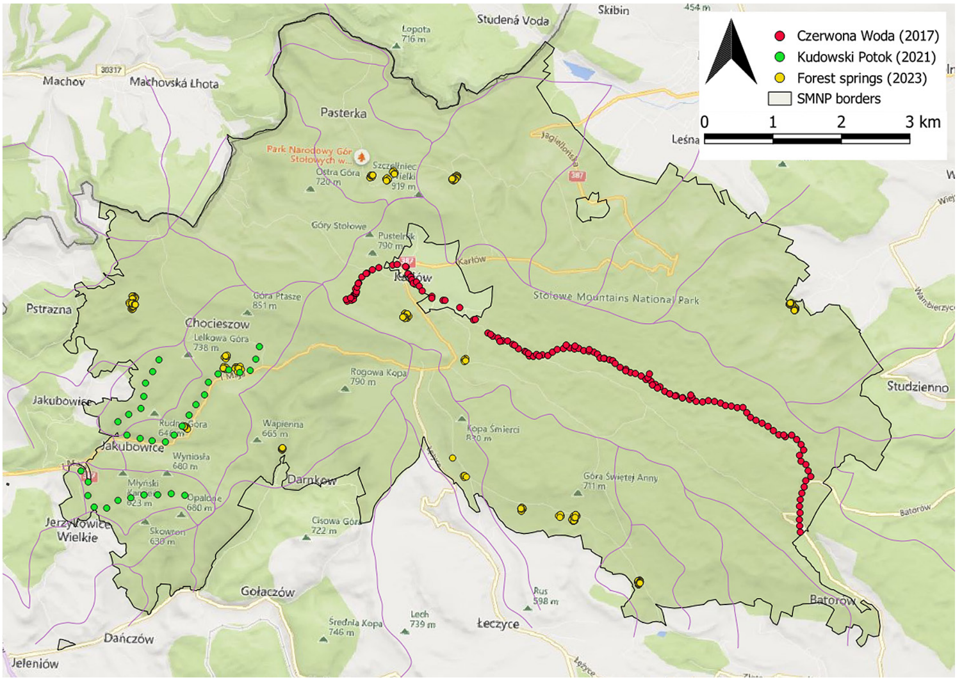

Fig. 1. Locations of the study sites along the Czerwona Woda River and the Kudowski Potok River, and forest springs within the Stołowe Mountains National Park (QGIS Development Team, 2024, QGIS Geographic Information System. Open Source Geospatial Foundation Project)

as guidelines for the renaturalization processes of anthropogenically modified semi-natural habitats. Our intense research in recent years significantly increased the level of biological and ecological knowledge of SMNP ecosystems (e.g. Dyderski et al. 2018, Kamczyc et al. 2018, 2023, Pielech et al. 2018, Wierzcholska et al. 2018, 2024).

"Recognition and assessment of the current state of natural habitats in the vicinity of the Czerwona Woda River"

Our first research project conducted in SMNP in 2017 aimed to identify natural values and their mutual relationships through a qualitative assessment of the structure and functioning of ecological systems located in the vicinity of the Czerwona Woda River (fig. 1).

The research covered the Czerwona Woda River valley along its entire length, from its source to the end of its course in SMNP. The area of the Czerwona Woda River valley was determined using a digital terrain model, and in its widest part it was limited to a width of 100 m. In total, the research covered 91.2 ha. Within a set of sample plots we assessed the state of vegetation, taking into account:

(1) the degree of its naturalness/preservation, (2) the state of herbaceous plant biomass in selected patches of actual vegetation, (3) the diversity of lichenized fungi and macrofungi, with indications of the locations of rare, endangered and protected species, (4) the assemblages of Mesostigmata mite communities as bioindicators of changes occurring in the soil environment, (5) the relative solar radiation intensity (as a proxy for light availability, we used diffuse non-interceptance – DIFN), (6) the state of tree fine root biomass and morphology as a reaction to the variability of habitat conditions, and (7) natural tree regeneration as a reflection of habitat conditions.

The studied area, despite significant transformations in relation to the potential natural vegetation, has high natural value. In total, 196 taxa of vascular plants were found there. On the background of the entire SMNP flora, numbering 1,006 taxa in total (and currently 868 taxa; Świerkosz 2007), this constitutes 22.5%, while the area of the Czerwona Woda River valley (91.2 ha) constitutes 1.4% of the area of SMNP. Such a large part of the plant species pool of a given region, accumulated in a disproportionately small area, is a characteristic feature of riverine ecosystems (Naiman et al. 1993, Dolanc, Hunsaker 2017, González et al. 2017).

The occurrence of 148 bryophyte taxa was also found. The analysis of the fungal biota of the Czerwona Woda River valley showed the occurrence of 148 taxa of macrofungi and 2 species of slime molds. This number is almost three times higher than the number given in the SMNP conservation plan – 56 (Chachulski 2013). The occurrence of 106 lichen taxa (lichenized fungi) was also found. This number constitutes 48.3% of the 211 taxa identified during the elaboration of the SMNP conservation plan (Chachulski 2013). The analysis of samples of Mesostigmata soil mites showed the presence of 57 taxa, which constitutes 60.6% of the SMNP acarofauna (Kamczyc, Skorupski 2014).

The analysis of species richness in the longitudinal river gradient showed the highest species richness between the middle and lower reaches of the river. For the analyzed groups of organisms, statistically significant relationships were found in the cases of all bryophytes ($r^2 = 0.12$), fungi ($r^2 = 0.14$) and Mesostigmata mites ($r^2 = 0.06$). The low values of the determination coefficients result from the large internal diversity of the analyzed samples. For bryophytes in general and epigeic bryophytes, the differences in responses resulted from the more frequent occurrence of deciduous trees in the lower reaches that are important phorophytes for epiphytic species. In the case of fungi, the lower river course is the poorest in terms of species richness, mainly due to the tree stand composition and presence of more fertile habitats there. The analysis of the spatial distribution of species richness showed similar relationships to the analysis of the distribution along the longitudinal gradient of the Czerwona Woda River. The greatest species richness of vascular plants in the middle reaches and lichens in the upper reaches is particularly visible. For Mesostigmata mites, two centers of species richness are located in the middle reaches and the lower river course within SMNP. In the case of epigeic bryophytes, there is a large local variation in species richness, which results from the diversity of micro-relief and light conditions.

Analysis of the relationships between species richness and light availability indicates the lack of clear relationships for most of the studied groups. This may be due to the high species diversity in the Czerwona Woda River valley, which is usually followed by functional diversity. The strongest, but still statistically insignificant relationships were found for lichens, dominated by lichens occurring on wood. For this group of organisms, the light availability is crucial. For bryophytes, a negative but statistically insignificant relationship is visible.

Analysis of the relationships between species richness and soil pH indicates more clear relationships than for light availability. The strongest relationships were recorded for vascular plants, epigeic and epilithic bryophytes, and macrofungi. The only group of plants whose species richness increases with soil reaction are vascular plants. The lack of statistical significance for organisms from habitats other than soil is understandable due to their independence from soil resources.

The species richness of individual groups of organisms was weakly related. The closest relationships were found between bryophytes and lichens ($r = 0.829$), mites and bryophytes ($r = -0.364$), fungi and vascular plants ($r = -0.337$), and bryophytes and vascular plants ($r = -0.262$). Low values of correlation coefficients may also result from the high variability of light and trophic conditions, which mask the existing relationships.

The obtained results indicate large differences in the response of individual taxonomic groups to water availability against the backdrop of trophic gradients and light availability. The main factor that may shape biodiversity locally is the microhabitat variability of water conditions. It results from factors related to the tree stand structure, micro-relief, and the influence of the river's hydrological regime. The tree stand structure determines the openness of the canopy, and therefore the inflow of solar energy, determining the rate of water evaporation (von Arx et al. 2012, Mueller et al. 2016), and additionally, the rate of transpiration is related to the assimilation apparatus area (Cienciala et al. 1992, Kozlowski, Pallardy 1997, Sarkkola et al. 2010). Microrelief affects the local accumulation of water in ground depressions and the possibility of creating peaty basins. These microhabitats, also related to the occurrence of sphagnum facies, increase water retention in the ecosystem (Rydin et al. 2006). On the other hand, these microhabitats, despite the presence of numerous seedlings, are not suitable for further stand development (Macdonald, Yin 1999, Ohlson et al. 2001, Jagodziński et al. 2017). The mosaic nature of the microrelief is also related to the occurrence of dead trees, especially those fallen with the root system (Swanson et al. 2011, Chećko et al. 2015).

An important element in recognizing the natural values of the Czerwona Woda River valley is the distinction between spruce stands of artificial origin, occupying primarily acidic beech forest habitats, and the swamp spruce forests of *Vaccinio uliginosi-Piceetum*, which constitute natural communities. Due to the occurrence of rare and endangered species, as well as natural syngenesis, these swamp spruce forests are highly valuable systems. They are also a natural habitat of the Natura 2000 network no. 91D0-4 called "wet and peat bog mountain spruce forest" (Potocka 2004). Due to the large share of bryophytes in the undergrowth biomass, they constitute an important reservoir of water, but are also at risk due to the existence of former drainage ditches. Despite overgrowing vegetation, drainage ditches may continue to cause water runoff from peat bog catchments for a long time (Ilnicki 2002, Kujawa-Pawlaczyk, Pawlaczyk 2005, Herbichowa et al. 2007, Herbich, Herbichowa 2011, Dyderski et al. 2015, Dyderski, Jagodziński 2016). These systems are particularly vulnerable to further disruption of water conditions, although they are capable of regeneration, as evidenced by the small share of species typical of drained peat bogs, e.g., *Molinia caerulea*.

"Recognition and assessment of the current state of natural habitats in the vicinity of the Kudowski Potok River and its main tributaries"

The second research project was conducted in SMNP in 2021 and aimed to identify natural values and their mutual relationships through a qualitative assessment of the structure and functioning of ecological systems located in the vicinity of the Kudowski Potok River and its tributaries (fig. 1). The Kudowski Potok River is a mountain stream bearing water from the southwestern slopes of SMNP. The total length of the river is 7.3 km, of which 3.2 km lies within SMNP. The total area of the Kudowski Potok River catchment is 1,110.39 ha, while within SMNP it is 657.84 ha. The total length of the river network in the Kudowski Potok River catchment within SMNP is 15.1 km. The Kudowski Potok River is located in the watershed of the North Sea (Elbe basin).

Due to the great importance of the Kudowski Potok River and its tributaries for water retention within SMNP, preservation of plant communities in protected forests and SMNP biodiversity, we carried out a number of studies covering forest natural habitats in the vicinity of the Kudowski Potok River (3.2 km) and its two main tributaries: the tributary from Lelkowa Mount (1.6 km) and the tributary from Krucza Kopa (2.4 km). The total length of the selected watercourses is 7.2 km.

Almost the entire area of the Kudowski Potok River catchment and its tributaries within SMNP is forested, which is of great importance for the natural water retention capacity in this area. The watercourses in question are surrounded mainly by well-preserved forest plant communities.

Research carried out in the valley of the Kudowski Potok River and two of its tributaries showed the extraordinary natural value of this area. In the analyzed area (83.7 ha in total), the actual vegetation consists of nine syntaxa, including one meadow community and eight forest communities. The largest area is occupied by forest secondary communities consisting of spruce (43.4%), acidic beech forests (33.0%), and sycamore forests (14.3%). The most valuable riparian forest communities cover 6.9% of the area. The potential vegetation is dominated by the acidic beech forest association.

The analysis of bryoflora revealed the occurrence of 119 species of bryophytes; 39 of them are under protection and six are rare and endangered species. Many valuable species occur on the rocks, including valuable protected species such as *Anomodon attenuatus, A. viticulosus, Homalia trichomanoides, Leucobryum juniperoideum, Neckera complanata, Porella platyphylla* and *Thamnobryum alopecurum*. More species were found on dead wood than on living trees (64 *vs.* 54), which proves the important role of these microhabitats in shaping the bryoflora of the site. Among epiphytic bryophytes, most species were found on sycamore maple (40), European ash (34) and European beech (29). One alien moss species was found within the field research, i.e. *Orthodontium lineare*.

Analysis of the macrofungi in the Kudowski Potok River valley and its two tributaries revealed 217 taxa of macrofungi, including 19 taxa of ascomycetes and 198 taxa of basidiomycetes, as well as 4 species of slime molds. One of them (*Ganoderma lucidum*) is protected according to Polish law, and 48 others are rare and endangered species, including five considered critically endangered in Poland. Species occurring in less than 10 localities in Poland were also recorded, including two (*Trechispora stevensonii* and *Brevicellicium olivascens*) identified in the second locality in the country. High species richness, obtained after research conducted during one growing season, proves the great potential of the surveyed habitats in maintaining fungal diversity. The species richness and functional diversity of fungi reflects the significant habitat diversity of the study area, especially in terms of the composition of the forest stand and the resulting diversity of hosts, litter and dead wood available to fungi.

The lichen biota of the study area consists of 79 species, including 64 epiphytes. The most important phorophytes for lichens are European ash (40 species), Norway spruce (29) and sycamore maple (23). There were 26 species on dead wood and 19 on rocks. 14 of the identified species are on the red list of endangered lichens in Poland, one is under strict protection – *Peltigera praetextata*, and one is under partial protection – *Hypogymnia tubulosa*.

Acarological research revealed 5,349 Mesostigmata mites classified into 93 species, including 20 previously unrecorded in the SMNP area. The mites detected represented 18 families, and the most numerous were Parasitidae (974 individuals). The presence of so many species of Mesostigmata mites in such a small area proves the exceptionally high natural value of this part of SMNP and its high habitat potential. The demonstration of approximately 15% of the share of mites from the Parasitidae family is an expression of a much weaker influence of spruce stands on the developing mite communities, especially in comparison to the results obtained in the Czerwona Woda River catchment, where this share reached as high as 30%. The high share of important indicator species of mites from the Uropodina suborder (of the genera *Trachytes, Cilliba, Olodiscus* or *Urodiaspis*), as well as mites from the Macrochelidae or Pachylaelapidae families, indicates a high species diversity of mites from this order, but also a relatively small transformation of the soil environment in this part of SMNP.

The tree stands in the Kudowski Potok River valley and its two tributaries were characterized by high diversity in terms of species composition. They passed continuously from spruce forests through beech forests to slope and riparian forests with sycamore maple and ash. Their biometrical structure mostly indicated the continuity of the natural regeneration process and the regeneration of secondary forest communities with spruce towards beech forests. The forest stands accumulated biomass ranging from 141.6 to 569.4 Mg ha^{-1} (average 389.3±17.1 Mg ha^{-1}). In the regeneration layer, ash was the most abundant, but was represented mainly by seedlings which did not advance to the upper forest layers (above 50 cm in height). Beech and sycamore natural regeneration occurred in large numbers in all layers; the young spruce generation was limited only to some of the stands where spruces appeared in the main canopy. The observed mortality of spruce and ash contributes to the increased share of dead trees and creates diverse light conditions within stands. European beech is the species that most effectively recruits to the higher forest layers. The regeneration of Norway spruce is characterized by low dynamics. The natural regeneration densities of the studied species were determined by light availability, ectohumus thickness, stand species composition and soil pH.

The analysis of environmental factors within the gradient of tree species composition showed the direct and indirect impacts of dominant tree species on the formation of biomass pools in SMNP forest ecosystems. The ectohumus thickness ranged from 0.5 cm in slope and riparian forests to 11.5 cm in spruce forests, the leaf area index (LAI) ranged from 2.71 to 7.07 m^2 m^{-2}, the availability of light expressed as DIFN ranged from 0.5% in mixed stands up to 15.0% in spruce stands. The biomass of bryophytes ranged from 0.0 to 497.5 g m^{-2}, and the biomass of vascular plants ranged from 0.1 to 249.2 g m^{-2}. The litter mass ranged from 113.5 g m^{-2} in black alder stands to 10,779.3 g m^{-2} in spruce stands, and the biomass of fine roots ranged from 34.6 to 817.3 g m^{-2}. The undergrowth biomass was characterized by the greatest variability while the soil pH by the smallest. Using structural equation modelling analysis, we showed that the tree stand species composition directly and indirectly determines the biomass of undergrowth and fine roots, as well as the litter mass. These impacts are related to the habitat-forming role of trees, which modify soil chemistry and light transfer to the forest floor. Our results allow us to predict how labile carbon pools in the ecosystem may change with forest stand composition, and the resulting changes in light availability. These two factors are key tools for forest management, hence their modification determines the functioning of the forest ecosystem and the allocation of carbon in its various pools. Reducing the share of Norway spruce reduces the availability of light, soil acidification and the pools of undergrowth and litter biomass.

"Recognition and assessment of the current state of natural habitats in the vicinity of forest springs in the Stołowe Mountains National Park"

Forested springs differ significantly from riverside forests, in which the main factor shaping the vegetation is the hydrological regime of the stream and the characteristics of the entire catchment. In the case of forest spring areas, local factors such as sunlight, exposure, ground and soil type or altitude are more important (Pielech 2015). Springs are relatively rich terrestrial-aquatic habitats that perform numerous ecosystem functions that are disproportionate to the area they occupy. Their physiognomy and character are of key importance for the preservation and protection of mountain ecosystems. Local hydrological conditions are an important factor influencing the biodiversity of spring areas. Minerals from deeper soil layers that are inaccessible to plants reach the surface along with the water flowing out. Spring areas may therefore be more fertile than adjacent areas (Dahm et al. 1998, Bufková, Prach 2006). The chemical composition of water flowing from springs depends primarily on the type of parent rock (Chełmicki, Siwek 2001). Therefore, even small forest marshes, fed by groundwater outflows, may differ significantly from the surroundings. Forest spring areas are also local biodiversity refuges.

Research on the diversity of riparian forests in the Sudetes has shown that two types of riparian forests develop in forested spring areas, i.e., spring ash riparian forest and alder-spruce spring riparian forest (Pielech 2015). Both groups were listed in SMNP. However, communities from the *Montio-Cardaminetea* class that developed in SMNP within forest communities as small patches, mainly formed by the *Caricetum remotae* Kästner 1941 association, are poorly recognized (Kącki et al. 2018). The analysis of literature sources indicates that the assessment of the current state of natural spring habitats occurring in the SMNP area, together with their species richness, has not yet been carried out in a way that would enable understanding the mechanisms determining the functioning of biological diversity in this area. Although the importance of springs as a natural resource of SMNP cannot be overestimated, the recognition and assessment of the condition of such natural habitats has not been carried out in a comprehensive manner so far, and the existing data are fragmentary and constitute only individual observations (Kącki et al. 2018).

The aim of the research was to identify natural values and their mutual relationships through a qualitative assessment of the structure and functioning of ecological systems located within the springs and surrounding forest communities (fig. 1). Due to the observed deterioration of water conditions in the SMNP, recognizing the natural values of forest springs is of great importance for actions taken to increase water retention in the SMNP forest ecosystems.

The geobotanical research carried out on springs in SMNP clearly shows a high vascular plant species richness in the observed areas. Local hydrological conditions are an important factor influencing this diversity. Relatively high light availability and strong hydration are also important factors. All these circumstances create favorable living conditions for a significant number of hygro- and eutrophic as well as photophilous species. Forested springs are often the only refuge for many taxa in areas dominated by spruce monocultures and acidic mountain beech forests, where the undergrowth has a significantly poorer species composition. Identifying and protecting these valuable ecosystems is crucial for maintaining local biodiversity.

As a result of the research, 161 species of vascular plants were identified, including 5 species under protection (*Blechnum spicant, Carex pendula, Dactylorhiza fuchsii, Daphne mezereum* and *Primula elatior*). Based on the phytosociological surveys, the examined patches were classified into three plant communities. Two of them are riparian forests belonging to the *Alnion incanae* association – spring ash riparian forest *Carici remotae-Fraxinetum* Koch 1926 and spring alder-spruce riparian forest *Piceo-Alnetum* Mráz 1959. The third one is a spring forest from the *Piceion abietis* association. It is the wood horsetail and spruce association *Equiseto sylvatici-Piceetum abietis* Šmarda 1950.

Analysis of the bryophyte flora of the study area within all three types of plant communities showed the occurrence of a total of 95 species (22 species of liverworts and 73 species of mosses). All of them were recorded in areas directly related to the water outflow – direct spring areas. In the adjacent areas – in the ecotone – 74 species of bryophytes were recorded, and in the surrounding forest communities – 67 species. In total, the study area revealed the presence of 25 species of bryophytes under protection and 5 species with threat categories on a national scale. The highest species richness was recorded in the alder-spruce riparian community, where a total number of 87 species were recorded, including 23 species under protection. In the ash riparian communities, a total of 79 species of bryophytes were recorded, including 16 protected species, and in the wood horsetail and spruce association, a total of 72 species were recorded, including 16 protected species.

The analysis of the mycobiota showed the occurrence of 291 taxa of macrofungi, including 13 classified of Ascomycota and 278 of Basidiomycota phyla. Metagenomic analysis of soil fungal assemblages revealed 859 taxonomic units identified at the genus level. Such a high species richness of soil mycobiome, obtained in research conducted during only one growing season, proves the high potential of the studied ecosystems for maintaining the diversity of both macrofungal communities and soil mycobiome of SMNP. Four of the recorded taxa have not been previously recorded in Poland, i.e., *Amanita coryli, A. brunneofuliginea* f. *ochraceomaculata, Cyanosporus simulans,* and *Inosperma vinaceum*. Two species under partial protection were found in the study area, i.e., *Suillus cavipes* and *Inonotus obliquus*. Many of the species found that are not protected are on Polish red lists (Chmiel 2006, Mirek et al. 2006), with the following status assigned: EX – 1 species, E – 7 species, V – 10 species, R – 26 species, I – 1 species. Additionally, the next 70 species are listed in the Register of Protected and Endangered Fungi (Gierczyk, Kujawa 2023) and six rare species not included in the registers were also recorded. A total of 127 species were marked as valuable species.

A total of 89 species of lichens were found in the spring areas and their close vicinity. The greatest number of species was recorded in the spring zone (81 species), and the least in the ecotone zone (53 species). Among the lichens found, 12 species have different threat categories, including two species that are protected. The lichen biota within the studied area is dominated by epiphytes – 72 (including 40 species exclusively on this type of substrate), the second group are epixilites – 40 species (10 exclusive), the third group are epilithes – 21 species (8 exclusive), and the fourth epigeites – only 3 species. Among the three types of plant communities, the highest number of lichen species was found in ash riparian forest *Carici remotae-Fraxinetum* (70 species) and the alder-ash riparian forest *Piceo-Alnetum* (71 species), and the least in wood horsetail and spruce communities (40 species). The forest springs, due to their particular microclimate associated with high air humidity, are an important spot for maintaining a high diversity of lichen species. This diversity is shaped not only by the humidity of the habitats themselves, but also by the presence of heterogeneous substrates that are inhabited by lichens, including diversity of tree species and the presence of wood in various degrees of decay.

The analysis of Mesostigmata mite communities showed that the number of mites was higher in the forests that grow in the direct vicinity of springs compared to the ecotone zone, by approximately 35%. A total of 84 species of Mesostigmata mites were detected, including nearly 50 species exclusively in the spring areas, which proves the exceptionally high natural value of this part of SMNP. The presence of approximately 40% of mites from the Parasitidae family in the spring areas itself is an expression of the great influence of Norway spruce stands growing nearby on the formation of mite communities. The obtained results of the number of mites and their density in individual plots indicate the heterogeneity of this environment, which seems to be a natural feature of spring sites. Previous research indicated the role of watercourses as corridors for the spread of mites, but also as creating conditions for the development of various species of these free-living predators. It turns out that the springs are also an ecological corridor of great importance for the migration of mites up the stream, which is of great importance for renaturalization of the anthropogenic spruce forests covering a large area of SMNP.

Analysis of the variability of environmental parameters showed that they differ depending on the type of the analyzed environment within the designated research areas. Within the research plots, 17 tree and shrub species were found in the forest stands, and 27 in the regeneration layer. A total of 30 species of woody plants were found; 14 of them were recorded in both the stand and regeneration layers. Three of all species occurred only in the forest stand layer, and 13 only in the regeneration layer. Natural regeneration of woody plant species in the studied areas was high, on average 261,430 individuals ha^{-1} (±5919) of seedlings, 21,203 individuals ha^{-1} (±422) of older saplings not exceeding 0.5 m in height, and 1,335 individuals ha^{-1} (±61) of older saplings with a height in the range of 0.5–1.3 m. However, the analysis of natural regeneration densities indicates the lack of recruitment of most of the recorded species into higher forest layers. Despite very abundant sowing, tree seedlings die and only a small proportion of them survive the first year. High variability of soil pH was also demonstrated (3.63–6.87). Such a large range of variability of this parameter indicates a relatively homogeneous substrate, modified by the forest stand rather than by the variability of soil types.

The intensity of sunlight reaching the forest floor was also related primarily to the variability of the structure of stands in different zones within and in the vicinity of forest springs.

Summary

The conducted inventory showed a very high species richness of the stream valleys and forest springs in relation to the current state of knowledge about the biological diversity of SMNP. The presence of plant communities that have not been identified in SMNP before has also been demonstrated. Previous studies were carried out on a much larger spatial scale, which omitted small patches of vegetation accompanying the Czerwona Woda River, the Kudowski Potok River and its tributaries, and forest springs. This indicates the need for a broader natural inventory of the SMNP area, especially in terms of mycology, lichenology and bryology.

The obtained results constitute a documentation source of the conditions of the Czerwona Woda River and the Kudowski Potok River valleys, and of forest springs, before the decision to conduct renaturalization procedures. Thanks to the presented characteristics of plant communities, tree stand conditions and biological diversity of the analyzed groups of organisms, it will be possible to assess the effects of the actions taken. Moreover, the collected data constitute the basis for further research that can fill the gap in the current state of knowledge about the dynamics of transformed mountain forests.

The most valuable type of forest communities are riparian forests, the preservation of which should be a priority in the protection of the study area. For bryophytes, lichens and fungi, protection can be achieved primarily by protecting their habitats; maintaining their sites and their surroundings in an unchanged condition is crucial for their survival and undisturbed existence. Of particular importance here is the preservation of intact tree stands and dead tree resources, as well as ensuring that water conditions do not deteriorate. The gradual restoration of the original water conditions could, over time, contribute to an additional macrofungi diversity increment and an increase in the number of biologically valuable species. In slope forests, but also in spruce forests, rocks are an important reservoir of biodiversity. Most of the bryophyte species found were epigeites, and there were also many lichens. The presence of large rocks is also crucial for varying light conditions under the tree stand canopy – they inhibit tree growth and create canopy gaps.

Forest springs, as ecosystems directly related to water flow, are currently subject to transformations related to climate change, anthropopressure related to the disruption of the water regime and water chemistry (including deforestation, tree dieback, eutrophication; Hájek et al. 2002). Among the three distinguished types of areas (spring areas, ecotone, adjacent forest) in all three phytosociological associations – spring sites are characterized by the highest species diversity, as well as the highest share of protected and endangered species, in relation to ecotone and forest sites, and therefore deserve special attention. The most important task is to maintain the current level of diversity of microhabitats that create specific conditions for the occurrence of very rare species. A large variety of trees, and above all the presence of deciduous trees (e.g., alder, ash, willow, sycamore) forming the spring ash riparian forest and alder-spruce spring riparian forest, ensures a variety of microhabitats for lichens and epiphytic bryophytes. This is affected by the different nature of the bark of these trees, such as physical and chemical properties, structure (thickness and width of cracks) and water capacity. The diversity of microhabitats is also complemented by the presence of dead wood in the form of logs, stumps, dead standing trunks without bark, as well as exposed whole root systems. Maintaining this diversity of microhabitats will primarily ensure the maintenance of appropriate and constant humidity, as well as the persistence of the studied flora, funga and fauna in their sites, and their future spread and increase in population ranges.

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References

Bufková I., Prach K. 2006. Linking vegetation pattern to hydrology and hydrochemistry in a montane river floodplain, the Šumava National Park, Central Europe. Wetlands Ecology and Management 14: 317–327.

Chachulski Ł. 2013. Plan ochrony Parku Narodowego Gór Stołowych. Operat ochrony grzybów i porostów. Instytut Ochrony Środowiska – Państwowy Instytut Badawczy, Warszawa.

Chećko E., Jaroszewicz B., Olejniczak K., Kwiatkowska-Falińska A.J. 2015. The importance of coarse woody debris for vascular plants in temperate mixed deciduous forests. Canadian Journal of Forest Research 45: 1154–1163.

Chełmicki W., Siwek J. 2001. Natural and anthropogenic factors controlling spring water quality in the southern part of the Małopolska Upland (southern Poland). In: Gehrels H., Peters N.E., Hoehn E., Jensen K., Leibundgut C., Griffioen J., Webb B., Zaadnoordijk W.J. (eds.) Impact of human activity on groundwater dynamics. International Association of Hydrological Sciences, Wallingford, pp. 317–322.

Chmiel M.A. 2006. Krytyczna lista wielkoowocnikowych grzybów workowych Polski. W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków.

Cienciala E., Lindroth A., Čermák J., Hällgren J.-E., Kučera J. 1992. Assessment of transpiration estimates for Picea abies trees during a growing season. Trees 6: 121–127.

Dahm C.N., Grimm N.B., Marmonier P., Vallet H.M., Vervier P. 1998. Nutrient dynamics at the interface between surface waters and groundwaters. Freshwater Biology 40: 427–451.

Dolanc C.R., Hunsaker C.T. 2017. The transition from riparian to upland forest plant communities on headwater streams in the southern Sierra Nevada, California, United States. The Journal of the Torrey Botanical Society 144: 280–295.

Dyderski M.K., Gazda A., Hachułka M., Horodecki P., Kałucka I.L., Kamczyc J., Malicki M., Pielech R., Smoczyk M., Skorupski M., Wierzcholska S., Jagodziński A.M. 2018. Impacts of soil conditions and light availability on natural regeneration of Norway spruce Picea abies (L.) H. Karst. in low-elevation mountain forests. Annals of Forest Science 75: 91.

Dyderski M.K., Gdula A.K., Jagodziński A.M. 2015. Encroachment of woody species on a drained transitional peat bog in 'Mszar Bogdaniec' nature reserve (Western Poland). Folia Forestalia Polonica, Seria A – Forestry 57: 160–172.

Dyderski M.K., Jagodziński A.M. 2016. Zmiany roślinności rezerwatu przyrody „Mszar Bogdaniec". Leśne Prace Badawcze 77: 104–116.

Gierczyk B., Kujawa A. 2023. Rejestr gatunków grzybów chronionych i zagrożonych w Polsce. Część XI. Wykaz gatunków przyjętych do rejestru w roku 2015. Przegląd Przyrodniczy 34: 3–72.

González E., Felipe-Lucia M.R., Bourgeois B., Boz B., Nilsson C., Palmer G., Sher A.A. 2017. Integrative conservation of riparian zones. Biological Conservation 211: 20–29.

Hájek M., Hekera P., Hájková P. 2002 Spring fen vegetation and water chemistry in the western Carpathian flysch zone. Folia Geobotanica 37(3): 364–364.

Herbich J., Herbichowa M. 2011. Tendencje dynamiczne zbiorowisk roślinnych i prognozy zmian. In: Herbichowa M., Herbich J. (eds.) Przyroda rezerwatów Łebskie Bagno i Czarne Bagno. Fundacja Rozwoju Uniwersytetu Gdańskiego, Gdańsk, pp. 210–215.

Herbichowa M., Pawlaczyk P., Stańko R. 2007. Conservation of Baltic raised bogs in Pomerania, Poland. Experience and Results of the LIFE04NAT/PL/000208 PLBALTBOGS Project. Wydawnictwo Klubu Przyrodników, Świebodzin.

Ilnicki P. 2002. Torfowiska i torf. Wydawnictwo Akademii Rolniczej im. Augusta Cieszkowskiego, Poznań.

Jagodziński A.M., Horodecki P., Rawlik K., Dyderski M.K. 2017. Do understorey or overstorey traits drive tree encroachment on a drained raised bog? Plant Biology 19: 571–583.

Kącki Z., Szymura M., Świerkosz K., Swacha G., Pender K. 2018. Roślinność Parku Narodowego Gór Stołowych. In: Kabała C. (ed.) Góry Stołowe – przyroda i ludzie. Park Narodowy Gór Stołowych, Kudowa-Zdrój, pp. 311–328.

Kamczyc J., Skorupski M. 2014. Mites (Acari, Mesostigmata) from rock cracks and crevices in rock labirynths in the Stołowe Mountains National Park (SW Poland). Biological Letters 51: 55–62.

Kamczyc J., Skorupski M., Dyderski M.K., Gazda A., Hachułka M., Horodecki P., Kałucka I., Malicki M., Pielech R., Smoczyk M., Wierzcholska S., Jagodziński A.M. 2018. Response of soil mites (Acari, Mesostigmata) to long-term Norway spruce plantation along a mountain stream. Experimental and Applied Acarology 76: 269–286.

Kamczyc J., Skorupski M., Dyderski M.K., Horodecki P., Rawlik M., Jagodziński A.M. 2023. Diversity of soil mites (Acari: Mesostigmata) in streamside mountain forests. Land Degradation & Development 34(13): 4046–4056.

Kozlowski T.T., Pallardy S.G. 1997. Physiology of woody plants. Second Edition. Academic Press, San Diego.

Kujawa-Pawlaczyk J., Pawlaczyk P. 2005. Ochrona mokradeł. In: Gwiazdowicz D.J. (ed.) Ochrona przyrody w lasach. II. Ochrona szaty roślinnej. Wydawnictwo Ornatus, Poznańj, pp. 105–119.

Macdonald S.E., Yin F. 1999. Factors influencing size inequality in peatland black spruce and tamarack: evidence from post-drainage release growth. Journal of Ecology 87: 404–412.
Mirek Z., Zarzycki K., Wojewoda W., Szeląg Z. (eds.) 2006. Red list of plants and fungi in Poland. W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków.
Mueller K.E., Eisenhauer N., Reich P.B., Hobbie S.E., Chadwick O.A., Chorover J., Dobies T., Hale C.M., Jagodziński A.M., Kałucka I., Kasprowicz M., Kieliszewska-Rokicka B., Modrzyński J., Rożen A., Skorupski M., Sobczyk Ł., Stasińska M., Trocha L.K., Weiner J., Wierzbicka A., Oleksyn J. 2016. Light, earthworms, and soil resources as predictors of diversity of 10 soil invertebrate groups across monocultures of 14 tree species. Soil Biology and Biochemistry 92: 184–198.
Naiman R.J., Decamps H., Pollock M. 1993. The role of riparian corridors in maintaining regional biodiversity. Ecological Applications 3: 209–212.
Ohlson M., Okland R.H., Nordbakken J.-F., Dahlberg B. 2001. Fatal interactions between Scots pine and Sphagnum mosses in bog ecosystems. Oikos 94: 425–432.
Pielech R. 2015. Formalised classification and environmental controls of riparian forest communities in the Sudetes (SW Poland). Tuexenia 35: 155–176.
Pielech R., Malicki M., Smoczyk M., Jagodziński A.M., Dyderski M.K., Horodecki P., Wierzcholska S., Skorupski M., Kamczyc J., Kałucka I., Hachułka M., Gazda A. 2018. Zbiorowiska roślinne doliny Czerwonej Wody w Parku Narodowym Gór Stołowych [Plant communities of the Czerwona Woda River Valley (Stołowe Mountains National Park)]. Leśne Prace Badawcze 79(2): 181–197.
Potocka J. 2004. Podmokła i torfowiskowa świerczyna górska. In: Herbich J. (ed.) Lasy i bory. Poradniki ochrony siedlisk i gatunków Natura 2000 – podręcznik metodyczny. Tom 5. Ministerstwo Środowiska, Warszawaj, pp. 189–193.
Rydin H., Gunnarsson U., Sundberg S. 2006. The role of Sphagnum in peatland development and persistence. In: Wieder R.K., Vitt D.H. (eds.) Boreal peatland ecosystems. Springer, Berlin Heidelberg, pp. 47–65.
Sarkkola S., Hökkä H., Koivusalo H., Nieminen M., Ahti E., Päivänen J., Laine J. 2010. Role of tree stand evapotranspiration in maintaining satisfactory drainage conditions in drained peatlands. Canadian Journal of Forest Research 40: 1485–1496.
Swanson M.E., Franklin J.F., Beschta R.L., Crisafulli C.M., DellaSala D.A., Hutto R.L., Lindenmayer D.B., Swanson F.J. 2011. The forgotten stage of forest succession: early-successional ecosystems on forest sites. Frontiers in Ecology and the Environment 9: 117–125.
Świerkosz K. 2007. General characteristics of the vascular flora and geobotanical division of the Góry Stołowe Mts, Sudety Mts. (Poland). In: Härtel T., Cílek V., Herben T., Jackson A., Williams R. (eds.) Sandstone landscapes. Academia, Prahaj, pp. 194–200.
von Arx G., Dobbertin M., Rebetez M. 2012. Spatio-temporal effects of forest canopy on understory microclimate in a long-term experiment in Switzerland. Agricultural and Forest Meteorology 166–167: 144–155.
Wierzcholska S., Dyderski M.K., Pielech R., Gazda A., Smoczyk M., Malicki M., Horodecki P., Kamczyc J., Skorupski M., Hachułka M., Kałucka I., Jagodziński A.M. 2018. Natural forest remnants as refugia for bryophyte diversity in a transformed mountain river valley landscape. Science of the Total Environment 640–641: 954–964.
Wierzcholska S., Łubek A., Dyderski M.K., Horodecki P., Rawlik M., Jagodziński A.M. 2024. Light availability and phorophyte identity drive epiphyte species richness and composition in mountain temperate forests. Ecological Informatics 80: 102475.

Abstracts

Protected forest patches conserve both carbon stock and tree stand diversity

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Keywords: aboveground biomass, conservation targets, tree species alpha diversity, stepping stones

The conservation of forest patches aims to achieve sustainable forest management, countering biodiversity loss and addressing the challenges of climate change through the preservation of forest carbon pools. Nevertheless, the complexity of forest ecosystems requires setting priorities on specific conservation targets to ensure the preservation of key forest aspects, such as natural processes, carbon stock retention, and biodiversity conservation. Some structural and environmental characteristics of forests are considered to be appropriate target indicators to cover a wide range of conservation objectives at once, e.g. the total amount of deadwood (Stokland 2012), the occurrence of habitat trees (Bütler et al. 2013), and rare ecosystems/forests containing rare species (Karrer et al. 2022). To address the relationships between tree alpha diversity and stand structural characteristics, we investigated differences among those aforementioned conservation targets and forest types in 247 study plots (0.03 ha) in Austria. The plots represented five broad categories of habitats: coniferous, acidophilous broadleaved, hydrogenic, broadleaved, and mixed forests. We revealed that there are neither trade-offs nor synergies between tree alpha diversity and carbon stock in the protected forests of Austria. Tree species alpha diversity and carbon stock are not contradictory dimensions, but strongly depend on forest types. Although rare ecosystems have lower aboveground biomass, their preservation is indispensable, as these habitats allow less competitive species with specialized habitat preferences to persist. We found the highest biomass in forests that were designated to two conservation targets: habitat trees and deadwood, and in three habitat types: hydrogenic, mixed, and broadleaved forests. To effectively address efforts aimed at both mitigating climate change and promoting conservation to combat biodiversity loss, it is advisable to treat these as distinct and separate objectives. Our study offers guidelines for the selection of appropriate forests for the conservation of both tree species diversity and carbon stock retention.

References

Bütler R., Lachat T., Larrieu L., Paillet Y. 2013. Habitat trees: Key elements for forest biodiversity. In: Kraus D., Krumm F. (eds.) Integrative approaches as an opportunity for the conservation of forest biodiversity. European Forest Institute, Joensuu, 86–95.

Karrer G., Bassler-Binder G., Willner W. 2022. Assessment of drought-tolerant provenances of Austria's indigenous tree species. Sustainability 14: 2861.

Stokland J.N., Siitonen J., Jonsson B.G. 2012. Biodiversity in dead wood. Cambridge University Press, Cambridge.

Can functional groups help explain vegetation development in post-mining sites?

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Keywords: species richness, succession, traits, vegetation

Species richness through time is an important measure often used to assess the success of post-mining sites. We expand this approach by considering functional groups in terms of life-form and pollination strategy as important components of ecosystem function and establishment. The questions posed are: (1) Which species are the most successful in post-mining areas, in terms of life-form and pollination strategy? (2) What is the relationship between the species richness of these functional groups and the time that has passed since spontaneous vegetation establishment? We recorded vascular plant species across 400 plots on post-coal mine heaps in Upper Silesia, Poland. We calculated the species richness of functional groups and estimated with the time since spontaneous vegetation establishment using mixed-effect models.

Among life forms, hemicryptophyte (177 species) exhibited the greatest success, within which *Calamagrostis epigejos*, *Daucus carota*, and *Solidago gigantea* are the most frequent in plots. For pollination strategy, insect-pollinated plants are the most successful, with the most frequent species within studied plots being *Daucus carota*, *Solidago gigantea*, and *S. canadensis*. The species richness of the life-form (chamaephyte, hemicryptophyte, and therophyte) decreases with age, while phanerophyte exhibits an increased trend. The richness of the pollination strategy (insect, self, and wind) decreases with age. Our findings suggest that species colonization during succession in post-mining areas is controlled by functional groups. Also, our results highlighted that the use of life-form and pollination strategies can help improve the description and prediction of spontaneous vegetation development on post-mining sites.

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Assessment of conservation plantings for an endangered species (*Cupressus dupreziana* A.Camus)

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Keywords: Cupressus dupreziana, assessment, conservatory plantations, in-vitro culture

The Tassili cypress, *Cupressus dupreziana,* the unique conifer of the Sahara, is threatened with extinction in its natural range, with fewer than 250 living individuals (Abdoun, Beddiaf 2002). It is described as the only case of apomixis in conifers. This paternal apomixis is considered evidence of the species' senility (Pichot et al. 2001). The IUCN classifies it as critically endangered.

Scientific experts and public institutions have worked since the 1940s to conserve it ex situ. Its introduction to the Mediterranean region was done by seeds. Other plantations are carried out in situ. In vitro cultures developed in Prague in the Czech Republic produced numerous plants (Lábusová et al. 2020) which were brought back to Tassili n'Ajjer in a 3rd mission called "Tarout expedition".

The work we present here is our master's thesis project (defended in July 2023). It consists of an evaluation of conservatory plantations in situ and ex situ (in Algeria). The results reveal that the plantations show variable developments depending on the sites and climatic conditions. Successful cases are observed in Ahaggar (hyper-arid climate) and the Saharan Atlas (semi-arid), while failure cases occur in strictly arid climates sensu stricto.

Plantations managed by INRF (National Institute of Forest Research) show overcrowding and also promising growth cases in an urban forest in Algiers. The evaluation of the "Tarout expedition" plantations reveals variable survival rates and growth traits, probably influenced by environmental factors. Monitoring the growth of the individuals from the 2020 "Tarout expedition" reveals differences in growth, responses and interactions that might be explained by particular edaphic conditions. This study provides information on in vitro plants' reactions and suggests future research directions to conserve this species, given its ecological importance in the context of climate change.

References

Abdoun F., Beddiaf M. 2002. Cupressus dupreziana A. Camus: répartition, dépérissement et régénération au Tassili n'Ajjer, Sahara central. Comptes Rendus Biologies 325: 617–627.

Lábusová J., Konrádová H., Lipavská H. 2020. The endangered Saharan cypress (Cupressus dupreziana): do not let it get into Charon's boat. Planta 251: 63.

Pichot C., El Maâtaoui M., Raddi S., Raddi P. 2001. Surrogate mother for endangered Cupressus. Nature 412: 39.

How can functional traits of herbaceous plants provide information about the adaptive capabilities of plants and environmental changes?

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Keywords: functional traits, adaptation, climate change, ecosystems, intraspecific variability

Functional traits include the morphological, biochemical, physiological, structural and phenological features or behavioural characteristics of organisms that influence their adaptation to environmental conditions. They provide answers to questions about the body's response to biotic and abiotic factors. At the level of plant communities, they have a huge impact on competition, succession and the density of individuals, and in the case of the largest scale – the ecosystem – they significantly influence the circulation of nutrients and biomass production. Functional traits are therefore one of the most promising tools for predicting changes in ecosystems. To fully use them, however, it is necessary to take into account the variability of the examined features at the intraspecific level, because the analysis of this variability allows obtaining information on the adaptation of populations and entire communities.

Databases on plant characteristics are constantly expanding, but there is a lack of data on plants, especially from Poland. This makes it impossible to determine the range of variability of features, which is important for forest ecosystems. This data is valuable because it affects the circulation of substances in ecosystems and the rate of decomposition of plant matter.

Research on the functional characteristics of plants is extremely important in the context of the possibility of using them in broader research on climate change. The results could answer questions about how plants respond to ongoing climate change and identify potentially invasive plants. The range of intraspecific variability of traits indicates the species' ability to adapt to changes, and also allows the identification of the most endangered species with a narrow tolerance range. A good understanding of the mechanisms affecting entire ecosystems will enable taking appropriate preventive actions and also mitigating the effects of climate change.

To achieve this, it is necessary to collect as much data as possible from as large an area as possible, to incorporate them into existing databases, and to systematize them at the regional level. This will facilitate not only the acquisition of valuable data, but also their analysis leading to the explanation of both the sources and consequences of intraspecific variability of functional traits.

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Novel pheromone traps for monitoring *Thaumetopoea pityocampa*

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Keywords: pine processionary moth, monitoring, trap devices, Pinus spp.

Thaumetopoea pityocampa (Denis & Schiffermüller) (Lepidoptera: Thaumetopoeidae) is a serious pest of forests, recreational areas, and urban ecosystems in the Mediterranean region. In recent years, it has spread to Central Europe due to climate change. The larvae feed on species of *Pinus*, causing severe defoliation that weakens or kills trees, especially when followed by attacks from secondary colonizers such as fungi and wood borers. This pest triggers serious allergic reactions, including eye and skin irritation, as well as respiratory disorders in humans and animals due to the release of urticating hairs from the larvae. This fact should not be overlooked after pupation, as the hairs remain active in the environment. Management of *T. pityocampa* with insecticides in various ecosystems is not effective because not all parts of the tree can be treated. Additionally, chemical applications in forests are not recommended for widespread use due to their potential to harm other species and their application in urban and recreational areas is not accepted by residents. Therefore, we propose monitoring male adults using novel pheromone traps.

The experiments were carried out in Attica, Greece. Five prototype traps were compared with a commercial trap during the flight period of *T. pityocampa* (i.e., between August and October). Four normal pentagons were created with a distance of 80 m between traps (in the same block). Captured males were recorded and removed from the traps twice per week. Additionally, traps were rotated clockwise within each block to minimize the effect of individual trap locations. A prototype trap was found to be superior to the other traps tested, outperforming the commercial trap in capturing *T. pityocampa* males throughout their flight activity. This trap should be further considered for large-scale mass trapping of *T. pityocampa* male adults.

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Impact of elevated air humidity on the endophytic fungal communities in foliage of silver birch and Norway spruce

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Keywords: silver birch, Norway spruce, fungi, endophytes, climate change

Silver birch (*Betula pendula* Roth.) and Norway spruce (*Picea abies* (L.) H.Karst.) are some of the most widespread and ecologically important tree species in Northern and Eastern Europe. Their tissues harbour a considerable range of organisms, including numerous fungi. It is predicted that the amount of precipitation will increase in the near future in northern temperate and boreal forest bioregions. More frequent precipitation will be accompanied by a rise in air humidity, especially within forest canopies. The effect of this increase on fungi living in the tissues of forest trees has not been predicted yet. To investigate how elevated air humidity affects fungal communities growing in tree foliage, we sequenced fungal DNA extracted from birch leaves and spruce needles growing in the Free Air Humidity Manipulation experimental site built for simulating the effects of an increase in precipitation on forest ecosystems, located in South-East Estonia.

Silver birch leaves and Norway spruce needles were repeatedly sampled in the middle and at the end of the growing season in three years (2020–2022). Fungal DNA found in samples was sequenced with high-throughput sequencing on the PacBio platform. We found that fungal communities of foliage in elevated air humidity conditions had a different composition than those in atmospheric humidity conditions. We observed no such effect in root fungal communities. The dominant species in leaf communities, such as various *Vishniacozyma* species, *Aureobasidium pullulans* and *Venturia ditricha*, were usually not influenced by elevated air humidity. However, the presence of members of Taphrinomycetes, the class which includes the birch pathogen *Taphrina betulina*, severely decreased in birch leaves due to air humidification treatment. We also aim to evaluate the influence of air humidification on other pathogenic fungi.

The impact of *Prunus serotina* Ehrh. on temperate forest natural regeneration

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Keywords: invasion ecology, biodiversity, forest management, forest functioning

Forest regeneration is a crucial element of forest stability and is particularly crucial in natural forests, where processes are driven by nature. In forest practice in managed forests, in addition to artificial regeneration, e.g. planting young trees or sowing seeds, natural forest regeneration comprises an important way of developing the next generation of stands. In addition to seed availability, natural regeneration is influenced by competition between trees, including invasive tree species.

Prunus serotina is an invasive species widespread in many European countries and is eagerly researched in terms of its impact on biodiversity. Some studies confirm a negative effect on forest regeneration, but none of them focused on quantitative scaling. The negative effect of invasive tree species is related to the transformation of ecosystems.

In our study, we aimed to assess the impact of *P. serotina* on natural regeneration along the quantitative gradient of invasion, determined by aboveground biomass. We established 96 circular plots (0.05 ha, 64 with *P. serotina* and 32 control plots), both on fertile habitats with oaks and poor habitats with pines, in different stand ages. Within each plot, we measured tree diameter at breast height to assess aboveground biomass and we counted the natural regeneration of each species within four 28.27 m^2 subplots per plot. We used generalized linear mixed-effects models including stand age, invasive species biomass, and habitat type as fixed effects, and the year of inventory as a random effect. We hypothesize that *P. serotina* will limit native species regeneration, but particular species will react differently to the presence of *P. serotina*. We also expect that *P. serotina* will facilitate the invasion of other species. Additionally, we anticipate that the negative consequences of *P. serotina* transformation will be stronger in nutrient-poor sites with pines.

Our studies are important in the context of invasive tree species management in protected and managed forests. Since natural regeneration has many advantages, such as reducing the costs of forest regeneration and better adaptation to local environmental conditions, it is worth examining the factors limiting it, including competition from invasive species.

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The influence of changes in temperature on the reactions of forest herbaceous plants on the example of the yellow archangel (*Galeobdolon luteum*)

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Keywords: leaf functional traits, leaf biochemical profile, changes in environmental conditions, infrared spectroscopy

Both long-term and short-term temperature disturbances affect vegetation in many ways, often leading to irreversible changes at various levels of biological organization, from molecules to organized tissues. In our work, attention was paid to the response of forest herbaceous plants. For this purpose, the yellow archangel (*Galeobdolon luteum*) was used. About 300 individuals growing in a breeding room under climatic conditions reflecting those in forests of Central Europe, i.e., 21°C during the day and 13°C at night, with a photoperiod of 16 hours of daylight and 8 hours of darkness, were used in the experiment, which served as the control. The next step involved increasing the temperature for both day and night by 8°C. Leaf samples were collected for the first three days and after a week: at noon and in the middle of the night.

The response at the functional and biochemical levels to the increase in air temperature was analysed. The following parameters were assessed: chlorophyll fluorescence (Fv/Fm), performance index (PI), hydration status (RWC), specific leaf surface area (SLA), and dry matter content (LDMC). The biochemical profiles of leaves obtained using infrared spectroscopy (ATR-FTIR) were also compared. Raising the temperature by 8°C reduced the average water content in yellow archangel leaves by 3% both during the day and at night. The dry matter content in leaves increased by 11.5% during the day and 1.5% at night, and the SLA value was lower by 4.5% during the day and 2.5% at night. The Fv/Fm and PI parameters also changed. Fv/Fm increased on average by 3%, and PI by 43%. Principal component analysis (PCA) of the spectral dataset showed clear differences in the absorbance values obtained for the peaks corresponding to the functional groups assigned to carbohydrates (1021 cm^{-1}), pectic substances (1240 cm^{-1}), lipids (1317 cm^{-1}), cellulose and hemicellulose (1360 cm^{-1}), amide II (1558 cm^{-1}), amide I (1617 cm^{-1}), pectin and lignin (1735 cm^{-1}), and lipids (2852 cm^{-1}), with increasing temperature. The results suggest that the increased temperature is responsible for the increase in plant biomass. Despite the decrease in SLA value, this is possible through an increase in the efficiency of photosynthesis. ATR-FT-IR results confirm changes in the metabolism of plants adapting to higher temperatures.

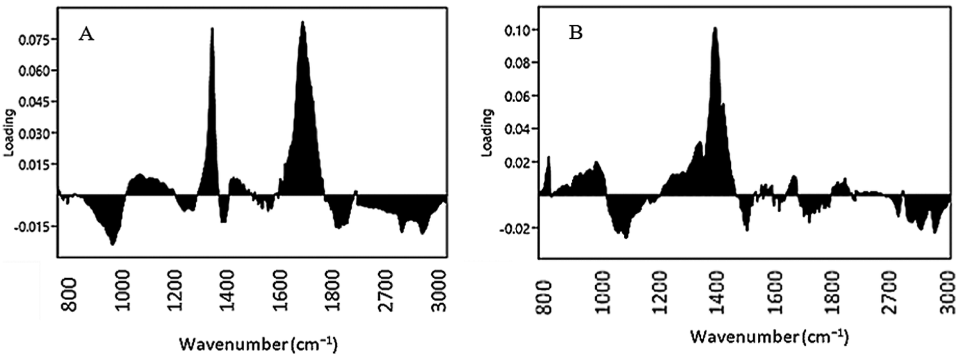

Fig. 1. PCA loadings plots produced (A) by PC 1 with peaks at 1360, 1617 and 2852 cm^{-1}, (B) by PC 2 with peaks at 1021, 1240, 1317, 1558 and 1735 cm^{-1}

Analysis of import demand function of Iranian paper and paperboard using bond test

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Keywords: paper, import demand, long run elasticity, short run elasticity, bond test

Wood pulp and paper are wood products that have undergone many changes in the regional and global market share, in terms of production capacity and consumption patterns, over the past few decades. Due to limited domestic resources and the increasing trend of consumption of wood and its products in Iran, imports have been considered as one of the most important ways to meet the domestic demand. The main aim of this study was to conduct scientific analysis of the paper and paperboard market conditions over the period between 1986 and 2018 using the bond test method. The quantitative impact of the studied variables on the amount of paper and paperboard imports was calculated using

the estimated short- and long-term elasticity. The results showed that among the studied variables, the GDP (without taking into account oil exportations) had an elasticity of 2.16% in the short run and 1.26% in the long run, and the domestic wood production from northern forests with an elasticity of –0.69 and –0.57 in short and long term, respectively, had the greatest impact on paper imports. However, economic sanctions considered as a new variable in this study resulted in a decrease in paper and paperboard imports with coefficients of –0.50 and –0.41 imports, in the short and long run respectively.

Optimizing the assessment of habitat trees in forest landscape management planning

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Keywords: FLMP, forest biodiversity, HT, TreM

Sustainable forest management (SFM) requires a landscape approach, such as forest landscape management planning (FLMP) supported by multi-criteria decision analysis (MCDA). Nevertheless, FLMP may not fully address SFM's influence on forest functions (FFs) without stand-level indicators. For example, habitat trees (HT) are key elements for forest biodiversity conservation, particularly when combined with other indicators to assess SFM's effect on biodiversity. However, existing methods to evaluate HTs (e.g. Asbeck et al. 2021) involve a large number of TreM types, additional data collection efforts and costs, and unclear management outcomes.

We employed a stratified systematic sampling across 74 plots, evenly distributed between beech (*Fagus sylvatica*) and Turkey oak (*Quercus cerris*) high stands within the Natural Reserve of Vico Lake (42.3°N, 12.17°E). For each plot, we assessed HTs and the associated Ecological Value of the Stand (EVS) using the RADAR table (Perrella, Puddu, 2015), alongside forest and topographic measures. Statistical analyses, including GLMs and PLS regression, explored the effect of forest and topographic measures on TreMs abundance, richness, and density. ANOVA and correlation analyses evaluated the stands' discretization power and independence from standard forest measures of EVS.

Regarding tree characteristics, DBH emerged as the primary predictor of TreM abundance and richness, while among the stand characteristics, quadratic mean diameter (QMD), tree density, slope, and aspect explained most of the differences in TreM abundance and richness (fig. 1). EVS exhibited little to no correlation with classical forest measures.

The preliminary results affirm the RADAR table method's validity in optimizing survey procedures, resulting in significant time and cost savings. This will facilitate operational forest biodiversity conservation and its integration into FLMP.

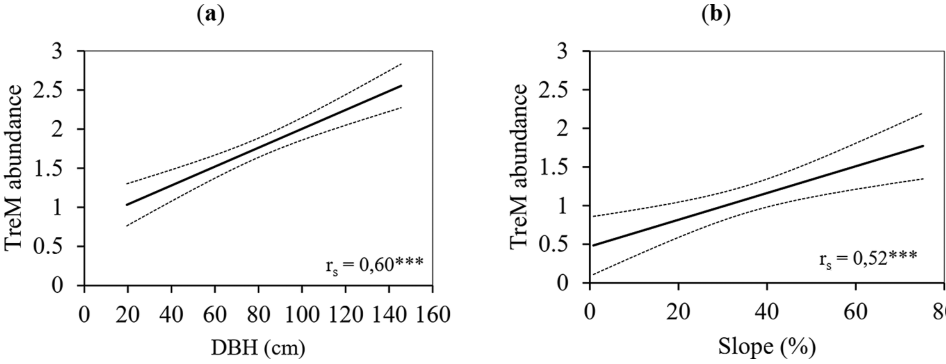

Fig. 1. Plots of the significant predictors, DBH (a) and slope (b), from GLMs of TreM abundance ($p < 0.0001$)

References

Asbeck T., Großmann J., Paillet Y., Winiger N., Bauhus J. 2021. The use of tree-related microhabitats as forest biodiversity indicators and to guide integrated forest management. Current Forestry Reports 7: 59–68.

Perrella P., Puddu G. 2015. Uno strumento innovativo per l'individuazione e la gestione degli alberi habitat: la tabella R.A.DA.R. Gazzetta Ambiente 21(1): 107–128.

Robinia pseudoacacia in urban forestry – management implications based on a literature survey and a case study in Warsaw

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Keywords: Robinia pseudoacacia, urban forestry, biodiversity, alien species, environmental impact

Black locust (*Robinia pseudoacacia*) is a widespread and widely used alien species in Europe, locally considered to be invasive (Klisz et al. 2021). For example, it is cultivated in plantations, planted for decorative purposes, and used in the reclamation of degraded areas, but is also spreading uncontrollably in forests and woodlands.

The aim of this study was to consider how *Robinia pseudoacacia* fits into the needs of urban forestry and what environmental risks are associated with its presence. For this purpose, 80 articles were reviewed. Based on them, black locust stands in urban forests were characterised in three dimensions: social, economic and environmental. This analysis distinguished the trend of changes in the sites in which this species occurs in Europe, such as homogenization, dominance in the stand, and change in biodiversity and species composition. To assess the impact of *R. pseudoacacia* on forest composition and structure in Warsaw, 24 sample plots in black locust-dominated forest patches, and 14 plots in mixed and 24 plots in Scots pine-dominated forest patches, were examined in two study areas: the forests "Na kole" and "Lindego". The results show that, when compared to *Pinus sylvestris, R. pseudoacacia* neither negatively affects the species richness of the undergrowth and ground vegetation nor favors the occurrence of other alien species. Studies indicate that the light-demanding black locust regenerates poorly in urban stands, which is probably due to high level of shade on the forest floor. On the other hand, species such as *Quercus rubra*, *Acer platanoides* and *Acer pseudoplatanus* regenerate extensively under its canopy, creating a future potential change in species composition. Relating the situation of Warsaw's forests to generally observed trends in Europe indicates a need to observe the black locust in the future, especially considering climatic changes, such as more frequent droughts and rising temperatures, which favor the invasiveness of this species (Klisz et al. 2021).

References

Klisz M., Puchałka R., Netsvetov M., Prokopuk Y., Vítková M., Sádlo J., Matisons R., Mionskowski M., Chakraborty D., Olszewski P., Wojda T., Koprowski M. 2021. Variability in climate-growth reaction of Robinia pseudoacacia in Eastern Europe indicates potential for acclimatisation to future climate. Forest Ecology and Management 492: 119194.

Redox dynamics in seeds of woody species: unravelling adaptation strategies for different seed categories

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Keywords: seed physiology, Trx-h1 regulation, metabolic adaptation, seed viability

Seeds of woody plant species, such as Norway maple (*Acer platanoides* L.), sycamore (*A. pseudoplatanus* L.), and common beech (*Fagus sylvatica* L.) exhibit unique physiological traits and responses to environmental stress. The seeds of these species represent three categories, i.e., orthodox, recalcitrant, and intermediate, respectively.

Thioredoxins (Trxs) play a central role in the redox regulation of cells, interacting with other redox-active proteins such as peroxiredoxins (Prxs), and contributing to plant growth, development, and responses to biotic and abiotic stresses. However, the redox system controlled by Trxs in tree seeds remains largely unexplored. Furthermore, there is limited understanding of potential variations in this system between seeds categorized as recalcitrant and orthodox, which could provide insights into adaptive strategies.

In this study, we identified numerous Trx-h1 target proteins, with distinct profiles in each seed type. Recalcitrant seeds presented a diverse range of metabolic, stress response, and signalling-related proteins, suggesting specialized adaptation mechanisms against storage-related stresses. Conversely, the orthodox seeds presented a conserved protein profile, reflecting robust molecular processes conducive to germination and seedling growth. The intermediate seeds exhibit a proteome directed towards genetic information processing and DNA repair. This suggests that intermediate seeds have adopted a strategy for the efficient repair of genetic material exposed to oxidative stress.

These findings underscore the crucial role of redox regulation in seed biology and adaptation to environmental challenges, providing insights into seed viability and storage longevity mechanisms. This study increases the understanding of seed biology and lays the groundwork for improving seed quality and resilience in woody plant species.

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Cell cycle and lipid rearrangements to optimise cryopreservation of *Quercus robur*

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Keywords: cell cycle, cryopreservation, lipidomics, recalcitrant seeds, redox-active molecules

We need to understand the relationship between the limits of desiccation survival and the cell cycle to identify extremes of life persistence at low water contents and safeguard recalcitrant-seeded species. Globally, one third of trees produce recalcitrant seeds that cannot survive desiccation and freezing, thus precluding conventional seed banking (Wyse, Dickie 2017). Cryopreservation is the only option for long-term storage of such species, and severe viability loss occurs early, during partial desiccation before freezing. Desiccation imposes oxidative stress, and checkpoints at the G_1 and G_2 phases of the cell cycle are redox-sensitive (de Simone et al. 2017). Thus, we aim to monitor changes in the phase of the cell cycle during fast desiccation and assess links with viability. We also investigate the influence of exposure to pro- and antioxidants on the cell cycle alongside lipid membrane compositional rearrangements to enhance survival of desiccation, and hence enable potential modulation of seed storage behaviour.

This study is targeting *Quercus robur* (English oak), a threatened keystone tree species, whose survival of desiccation critically hinders explant growth performance following cryopreservation. *In vitro* radicles regenerated ~4-fold more frequently than shoots , compatible with different desiccation responses of the two tissues. Exposing embryonic axes for 1 h to 0.1 M hydrogen peroxide, ascorbate, and glutathione increased by ~3-fold the successful *in vitro* regeneration of both radicle and shoot. We analysed embryonic root and shoot separately, and survival curves during flash-drying guided the choice of desiccation intervals to 1) quantify the percentages of cells in G_1 and G_2 by flow cytometry, and 2) characterise the biochemical composition and oxidation state of fatty acids, phospholipids, and sterols via lipidomics. Pilot results will be discussed in relation to mechanisms that may aid in rationalising cryopreservation and maximising its success on plant *ex situ* conservation.

References

de Simone A., Hubbard R., de la Torre N.V., Velappan Y., Wilson M., Considine M.J. Soppe W.J.J., Foyer C.H. 2017. Redox changes during the cell cycle in the embryonic root meristem of Arabidopsis thaliana. Antioxidants and Redox Signaling 27: 1505–1519.

Wyse S.V., Dickie J.B. 2017. Predicting the global incidence of seed desiccation sensitivity. Journal of Ecology 105: 1082–1093.

The content of trace elements in the soil, bark and wood of *Pinus sylvestris* L. from an area polluted by cement dust deposition

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Keywords: Scots pine, wood, bark, trace elements

Industrial pollution affects the physiology of plants and entire ecosystems. Even though the emission of pollutants has stopped, their effects on the environment remain for many years. We can read it, for example, from the tissue of trees formed under modified environmental conditions.

The cement plant in the city of Kunda in north Estonia emitted a considerable amount of dust from 1870 until 1996 (Ots 2002). During the most intense production period, the dust load in the vicinity of the factory reached up to 2700 g/ m^2 (Ots 2000). This affected the air pollution and, consequently – through the precipitation and deposition – the soil pH, causing soil alkalization, since the pH of dust was strongly alkaline (Mandre 2002). Cement dust can also contain trace elements and heavy metals (Rawat, Katiyar 2015).

From the forest in the vicinity of the cement plant, 21 Scots pines of ca 80-year-old medium in stand were selected. Our research has shown that pine is vulnerable to this type of pollution, as it caused a decrease in needle length (Ots 2000, 2002). Two cores, each at breast height (1.3 m), were taken from each tree, using a Pressler increment borer. Bark was separated from wood, and the outermost annual rings were separated from those closer to the pith. Analogous samples were collected from the forests of Lahemaa National Park, located to the west. The content of selected trace elements was examined in soil, bark and xylem from the outermost and inner parts of the trunk.

Although the trace element content in soil reflected the composition of the cement dust quite well, the dependences in bark and wood were more ambiguous. The results showed significant enrichment of Ca, Cd, Cr, Cu, Fe, Mg, Mn, Ni, and Zn in soil from the cement plant area. In xylem tissue, the most relevant differences were observed for Ca, K and Mg, which were the main components of cement dust. The bioconcentration factors were generally higher for trees from the unpolluted site in Lahemaa Park.

References

Mandre M. 2002. Relationships between lignin and nutrients in Picea abies L. under alkaline air pollution. Water, Air, and Soil Pollution 133(1–4): 363–379.

Ots K. 2000. Morphometric parameters of conifer needles and shoots in the areas near the Kunda cement plant. Metsanduslikud Uurimused 33: 158–176.

Ots K. 2002. Impact of air pollution on the growth of conifers in the industrial region of Northeast Estonia. Estonian Agricultural University, Tartu. [DSc Thesis]

Rawat V., Katiyar R. 2015. A review: On the effects of cement dust on vegetation. International Journal of Scientific and Innovative Research Studies 3(4): 39–45.

Unraveling belowground interactions in okoumé (*Aucoumea klaineana* Pierre) stands: the roles of root anastomoses and mycorrhizal associations

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Keywords: tropical forest ecology, root graft, mycobiota, sustainable management, dendrochronology, metabarcoding

Okoumé (*Aucoumea klaineana* Pierre) is the most harvested timber species in Central Africa. This pioneering light-demanding species favors open environments to form monospecific stands. During forest management activities, thinning operations in these stands were carried out. Through subsequent operations, the stumps of felled trees were discovered to be alive. Their survival was found to be assisted by the graft (anastomose) of their root steles with close conspecifics reaching the canopy. This enabled hydromineral and carbon exchanges between individuals and the extension of the root system of canopy trees.

Despite the widespread occurrence of root anastomose among woody species, the physiological mechanisms involved and their importance are poorly understood. For the first time in the tropics, we investigated it in young interconnected natural stands. We clearcut and excavated three stands using a fire hose to expose roots. Cross-sections of stems, roots, and root grafts were collected to observe growth ring patterns. The impact of root grafts on suppressed tree survival and tree growth through dendrochronology was then analyzed. Preliminary findings show the presence of shared growth rings within root grafts, proving exchanges between individuals.

Knowing that mycorrhizal associations are key components of tropical forests, the belowground interactions among *A. klaineana* trees are unlikely to be exclusively the result of root anastomoses. For that reason, we also investigated the mycorrhizal status of the species to gain an understanding of how these interactions influence their growth, health, and ecological roles. By collecting fine roots, we aimed to elucidate the links between fungal communities (through specific richness and functional diversity) and growth dynamics across populations of varying ages and structures through metabarcoding methods. Among 4,818 OTU, preliminary results show a low pathotroph (4%) but high saprotroph load (36%) compared to symbiotrophic fungi (26%, of which 80% were arbuscular mycorrhizal fungi, AMF), 34% uncertain.

Considering two belowground flows, we shed light on the relative importance of AMF in fungal assemblages in *A. klaineana* populations and were able to reveal the impact of root anastomoses on variations in tree growth. This contributes to filling knowledge gaps regarding *A. klaineana* ecology and interactions at the stand scale to improve management strategies by finding a balance between productivity and sustainable resource use.

Biodiversity indicators for developing forest management and eco-restoration strategies at stand-level in the Indian Western Himalayas

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Keywords: native species, ecological restoration, biodiversity, forests, altitude, threats, Himalayas

Field-based investigations assessing forest stand biodiversity indicators play a crucial role in informing targeted conservation policies and developing eco-restoration strategies effectively. We developed compositional, structural and functional indicators of biodiversity for 12 major forest types along an elevation gradient (350–3450 m a.s.l.) in the Western Himalayas. Compositional indicators results indicated an elevation-associated increase in species richness across all forest types attaining a maximum Shannon diversity value of 3.23 at an elevation of 2820 m a.s.l. TWINSPAN classification yielded three distinct plant communities from the studied forest types based on indicator species analysis. SIMPER analysis highlighted 76 species contributing to 90% of dissimilarity, with 24 indicator species identified across forest types, distributed in tree, shrub, and ground flora layers. Functional indicators showed that environmental (elevation, slope) and anthropogenic variable (grazing) significantly influenced plant composition and community assemblage across forest types ($p < 0.01$), while soil erosion, stem cutting, and fire played lesser roles. A species dissimilarity matrix allows a division of forest types into three groups according to altitude, however, phylogenetic dissimilarity indicates taxonomical similarities between ecosystems from high and low altitudes. We identified key indicator species that perform better under current conditions in each elevation belt as well as site-specific forest types, thus these species can be encouraged for eco-restoration. The investigation provides baseline data that will assist in formulating plans for ecological restoration specific to the prevailing ecological processes in the Himalayan region.

Variation of cell wall chemical composition in the leaves of silver birch (*Betula pendula*) growing in different habitats in the urban-industrial landscape

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Keywords: pectic polysaccharides, immunohistochemistry, Betulaceae, novel ecosystems

The cell wall plays a key role in ensuring the structural and functional integrity of plant cells. It responds both to impulses from inside the cell and factors from the external environment.

This study is aimed at comparing the chemical composition of the cell wall of *Betula pendula* leaves (including pectins) derived from individuals growing in post-mining habitats (coal heap and quarry) and a control site (mixed forest).

Using an immunohistochemical method, labelling of cell wall components was carried out with monoclonal antibodies.

Different habitats of urban post-industrial landscapes consist of unique, specific habitats of the novel ecosystem type that are colonized by some plant species. Among the pioneer species of the open mineral habitats there is also the forest-forming species, *Betula pendula* – a species with a wide range of adaptability to habitat conditions.

The plant material (leaves) was collected randomly. The differences in the biotic and abiotic conditions of the studied sites were estimated. The laboratory work stage included the collection of the material (from the leaf blade), its preparation for immunolabelling, successive fixation, dehydration, and embedding in wax the samples, following the immunohistochemical method by Milewska-Hendel et al. (2017). Cross-sections were made using a microtome. Immunohistochemical reactions were then performed for the presence of pectic epitopes. Fluorescence microscopy was used to visualize and analyze the wall chemistry.

The study revealed differences in the localization of homogalacturonan- and rhamnogalacturonan-rich pectin epitopes in the leaf cell walls of plants from disturbed habitats. Leaves from individuals growing in post-industrial habitats were characterized by the absence of the arabinan epitope compared to controls. Low-esterified HGs were more abundant in leaves from plants growing in disturbed habitats compared to the control site.

Immunohistochemical research makes it possible to study the relationship between plant cell wall composition and its changes depending on the level of environmental disturbance. In the future, these studies can be extended using a wide range of monoclonal antibodies that detect individual pectic polysaccharides or glycoproteins, considering different plant species in a gradient of novel ecosystems.

References

Milewska-Hendel A., Baczewska A.H., Sala K., Dmuchowski W., Brągoszewska P., Gozdowski D., Jozwiak A., Chojnacki T., Swiezewska E., Kurczynska E. 2017. Quantitative and qualitative characteristics of cell wall components and prenyl lipids in the leaves of Tilia x euchlora trees growing under salt stress. PLoS ONE 12: e0172682.

Performances of native tree species in plantations: a synthesis for the Guineo-Congolian region

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Keywords: planting, enrichments, traits, growth rates, survival

In the rainforests of the Guineo-Congolian region, several native tree species have been tested in plantations with different silvicultural methods and objectives. The results of these experiments remained scattered, hampering our ability to identify the key drivers of variability in the survival and growth of planted tree species.

Here, we synthesize the literature by conducting a systematic review. From 45 selected studies, we compiled a database on 89 native tree species planted in different forest types (evergreen, semideciduous and transition). The data included plantation age, survival, height and diameter growth. For each planted species, we gathered information on the planting method (understorey, line planting, gap, degraded area, regrowth and clear-cut), and species functional traits (species guild, dispersal mode, wood density and leaf phenology). We modelled tree survival, height and diameter growth using linear mixed-effect models.

We found that tree survival depended mainly on plantation age, and that the mortality rate was the highest during the seven first years. In contrast, tree survival did not significantly depend on the planting method or species traits. In the studied plantations, height and diameter growth significantly depended on the planting method and species guild. Diameter growth was negatively correlated with wood density. Pioneer, non-pioneer light-demanding and shade-bearer species grew faster in diameter when planted in degraded areas and clear-cuts. Pioneer species grew the fastest in gaps, where they also grew faster than the other species. Although we did not find an effect of forest type on tree survival and growth, the variability between sites was substantial.

This study provided empirical evidence that planting methods need to be adapted to the species guild.

Extension of skid trail network on salvage logging operations: an overview on the north-east Italian Alps

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Keywords: full-mechanized harvesting, cut-to-length wood system, windthrows, Alps

Climate change is contributing to an increase in the frequency and intensity of forest disturbances worldwide. Different disturbance regimes leave distinct types of biological and/or structural legacies, calling for different management strategies. In this context, salvage logging aims to recover economic value and mitigate the consequences of negative abiotic or biotic disasters, such as wildfires, windstorms, and insect outbreaks. This approach has become the most widespread management response to natural disturbances, and it is often subject to rapid decision-making by public authorities. However, at the same time, salvage logging raises ecological issues, including impacts on forest regeneration and biodiversity, as well as soil conservation, especially in mountain areas such as the Alps.

The study aims to estimate the extension, density, and spacing of logging trails in a large windthrow area under a full-mechanized harvesting system, and specifically with the use of harvester and forwarder forest machines. The contribution will provide a general outline of the relationship between terrain and logging trail networks, particularly under intense extraction pressure. The analysis focused on different forestry sites in the eastern Trentino region, and within "Altopiano dei Sette Comuni" in the Veneto region; both areas are facing a significant challenge due to the Vaia storm (2018). More than 700 km of logging trails were identified and analysed according to the forest stand characteristics, terrain morphology, harvesting and transportation issues.

The results highlight high pressure concerning trafficability on forest soils and high variability in terms of logging trail extension and density between different sites. A detailed understanding of the effects of salvage logging is needed to identify possible impacts, conservation targets and ultimately provide management recommendations to preserve post-disturbance bio-heritage.

Variations of ion concentrations in stemflow and throughfall in a beech forest in Germany

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Keywords: stemflow, throughfall, nutrient cycling, beech forest

Stemflow and throughfall constitute essential components of nutrient transport within forest ecosystems, and are subject to diverse influences including tree characteristics, climatic variations, and environmental factors.

This study investigates the impacts of annual and seasonal precipitation, and tree attributes on the temporal and spatial dynamics of stemflow and throughfall in a beech (*Fagus sylvatica* L.) forest located near Ebergötzen, central Germany, from 2017 to 2022. Monitoring of volumes at 15-minute intervals combined with bi-weekly water sampling facilitated a comprehensive assessment of stemflow, throughfall, and gross precipitation volumes and chemical constituents.

The findings unveil significant enrichments in stemflow and throughfall relative to gross precipitation, suggestive of active canopy leaching and the removal of dry deposition. While no universal increase in ion concentrations was observed for larger trees, the highest levels were observed during autumn and summer, intermediate in spring, and lowest during winter.

The studied beech forest exhibited 7% and 56% of the incoming rainfall allocated to stemflow and throughfall, respectively. Furthermore, we observed a positive correlation between annual precipitation and the proportion of rainfall allocated into stemflow and throughfall. Conversely, an inverse relationship was noted between ion concentrations and annual stemflow/throughfall volumes, implying a dilution effect whereby higher rainfall years exhibited lower ion concentrations and vice versa.

These insightful observations into the complexities of stemflow and throughfall dynamics are of the utmost importance for understanding nutrient fluxes and water quality within forest ecosystems. This knowledge serves as a foundation for informed management practices to safeguarding the ecological integrity and sustainability of forest environments.

Proline as a component of metabolic pathways network in plant cells – available knowledge and potential applications

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Keywords: proline metabolism, drought stress, tree seedlings, oxidative changes

The accumulation of proline is a biochemical response of plants to water deficiency. Stress associated with the consequences of climate change, including drought stress, induces excessive accumulation of reactive oxygen species (ROS) in plant cells. Seeds and tree seedlings are particularly vulnerable to oxidative damage to cellular structures caused by ROS.

Proline biosynthesis is enhanced to eliminate ROS and prevent disruptions caused by them. Proline stabilizes the structure of cell membranes and is an important component of many proteins, thus improving their integrity. This amino acid exhibits osmoregulatory properties, limiting water loss by cells. The activation of defense mechanisms against drought also reflects changes in the activity level of enzymes associated with proline catabolism and biosynthesis. The level of proline changes with temperature and humidity conditions, especially in desiccation-sensitive seeds (recalcitrant category). It also depends on the duration of drought and the mass of seeds from which seedlings have developed. Moreover, seedlings of deciduous species and those producing recalcitrant seeds accumulate proline more intensively than coniferous species and those with desiccation-tolerant seeds (orthodox category).

Proline metabolism is regulated not only by various environmental factors but also by numerous physiological processes occurring within cells. This process is associated with electron transfer between mitochondria and chloroplasts, hence proline affects the redox status of these organelles. The constant increase in the content of protein thiols during the development of recalcitrant seed axes indicates that they are unable to stop their metabolism during long-term storage, which can lead to loss of viability during prolonged storage. On the other hand, the content of protein thiols decreases in orthodox seeds after the acquisition of desiccation tolerance, which is a stage in seed maturation. Proline participates in the ascorbate-glutathione cycle, contributes to ROS protection, and increases the activity of antioxidant enzymes e.g. ascorbate peroxidase and the tolerance to high-temperature stress with the participation of nitric oxide (fig. 1). This makes proline a promising biochemical marker that could be used in selecting high-quality forest reproductive material.

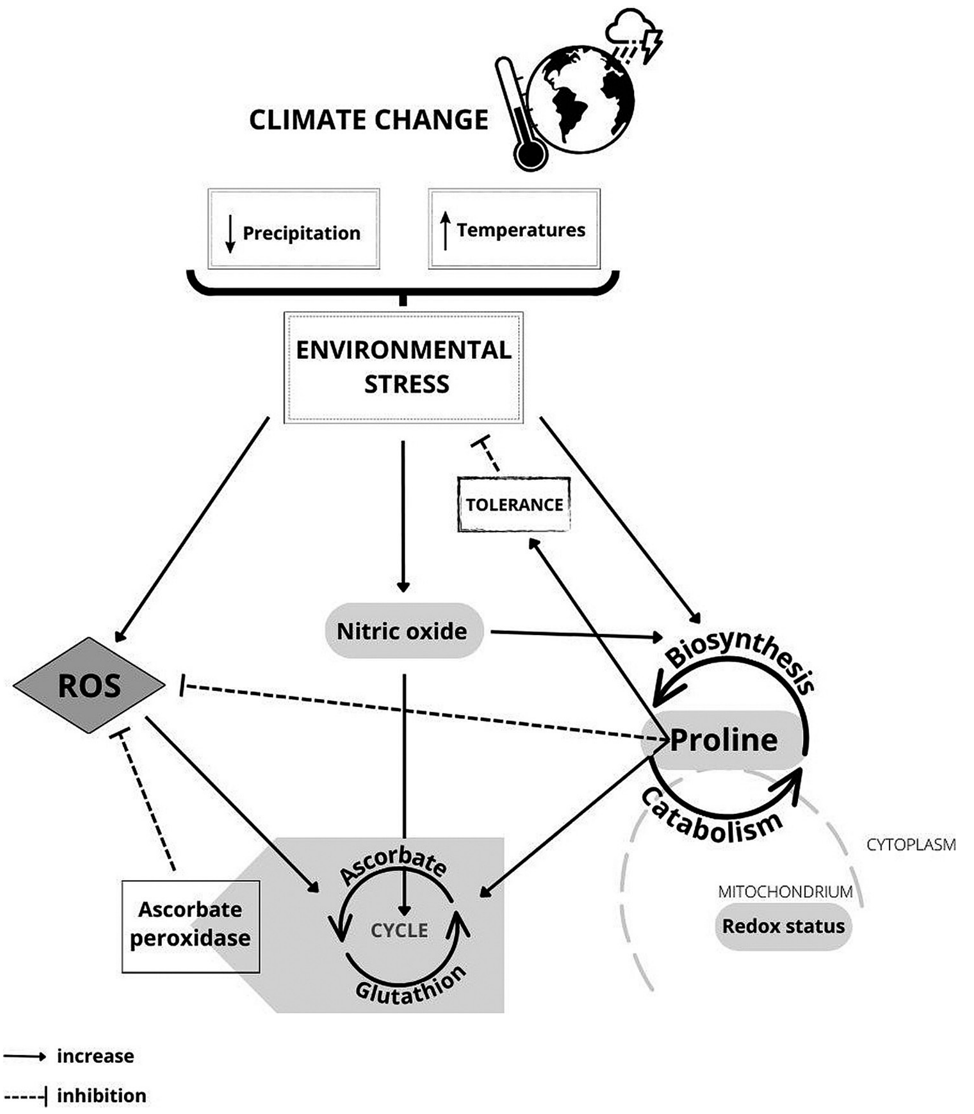

Fig. 1 The relationships between environmental factors and physiological processes within cells, including proline metabolism and elements of antioxidant system

Drivers of intraspecific variation in fecundity in rowan (*Sorbus aucuparia*)

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Keywords: tree fecundity, seed production, mast seeding, reproduction, Sorbus aucuparia

Understanding the variation in fecundity, i.e., the reproductive capacity of individual trees, is crucial for predicting population dynamics and ecosystem functioning. However, estimating tree fecundity is challenging, due to the large variation in seed production observed between trees and across years that necessitates logistically challenging long-term monitoring. This study aimed to investigate the factors influencing fecundity in rowan trees (*Sorbus aucuparia*) using a 22-year fruit-count dataset in 167 individual trees.

We examined the relationship between fecundity and intrinsic (DBH, height, and leaf nutrient concentration) and extrinsic factors (soil nutrients, light availability, and neighborhood crowding). Our findings revealed that diameter at breast height (DBH) better explained variation in fecundity than height.

After accounting for tree size using DBH, light availability had the strongest, positive effect on fecundity and was the major limiting factor for rowan's fecundity. At the same time, a neighborhood crowding index with conspecifics also showed a significant, but negative correlation with fecundity, suggesting competition for pollinators among rowan trees.

Our findings contribute to a better understanding of seed production ecology and can inform management and conservation efforts that aim for increased fruit supply, either for plant populations or fruit consumers.

Soil mycobiome of *Ulmus laevis* grown in diverse habitats

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Keywords: arbuscular mycorrhiza colonization, European white elm, soil fungal community

Elms are a crucial group of deciduous trees grown near rivers and floodplains. Among the elm tree species that predominantly occur in the temperate regions of the northern hemisphere, Europe hosts three distinct species, each widely distributed across most countries: European white elm (*Ulmus laevis* Pall.), wych elm (*Ulmus glabra* Huds.) and field elm (*Ulmus minor* Mill.). Elms constitute an admixture tree species in riparian and mixed broadleaved forests. Due to their ability to thrive in challenging conditions, they are also found as ornamental and roadside trees in rural and urban landscapes. Dutch Elm Disease, caused by the highly aggressive fungal pathogen, *Ophiostoma novo-ulmi*, led to significant losses in many parts of Europe, Asia, and North America. Since its emergence in Europe in 1910, extensive scientific research has focused mainly on disease-related aspects of elm biology, while neglecting others.

The mycobiome, particularly mycorrhizal fungi, plays a vital role in soil ecology. Mycorrhizal fungi, along with other soil fungi, colonize plant roots, enhancing plant nutrition and health, and are thus crucial for trees affected by global diseases. Understanding the factors influencing the mycobiome is essential due to the multifaceted involvement of fungi in plant health, nutrition, and productivity under diverse environmental conditions. Comprehensive knowledge of mycobiome assembly accompanying elms is essential for their sustainable management. *Ulmus laevis*, one of Poland's native elm species, has been chosen for the research as it is widely distributed across the country and has shown resilience to DED.

To establish the structure of soil fungal communities of *U. laevis*, eighteen locations that represent various habitats, including forest (riparian and oak-hornbeam forests) and non-forest habitats (such as forest nurseries, urban, rural, and post-industrial sites) were selected. Throughout the spring and autumn 2021, the roots of the same elm trees at each location were sampled. Our findings reveal significant differences in both mean species richness and the Shannon diversity coefficient across different habitat types. Oak-hornbeam forests exhibited the highest mean species richness, whereas forest nurseries showed the lowest. Arbuscular mycorrhiza colonization was notably higher in forest habitats compared to non-forest ones, and this was particularly evident in urban and post-industrial sites where non-mycorrhizal roots comprised over 50% of the total. Metagenomic analysis also confirmed the prevalence of symbiotic fungi in forest rather than in non-forest habitats. We observed that habitat type, rather than season, had a significant impact on the soil mycobiome structure of *U. laevis*. Interestingly, the level of arbuscular fungal colonization remained unaffected by habitat type.

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Comprehensive genomic analysis of breeding Scots pine (*Pinus sylvestris*) populations in Poland

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Keywords: Scots pine, genetic variability, forest management

Scots pine is the main forest-forming species in Poland, and it has great economic, ecological and social importance. As a dominant tree in forested areas, it plays an important role in forest ecosystems not only as a carbon reservoir but also through interactions with soil and fungi. It is an undemanding tree that grows well in nutrient-poor sandy soils and occupies an exceptionally wide environmental niche with a wide range of distribution in Eurasia. Such environmental plasticity is associated with great phenotypic diversity, known as ecotypic variation, which in the case of Scots pine has been observed for several decades and is reflected in earlier provenance trials.

In this study, we used a set of thousands of polymorphic markers in the Scots pine genome, along with maternally inherited mitochondrial DNA polymorphism, in more than 800 individuals representing the most valuable breeding populations in Poland to provide information on genetic relationships between species and species population history. Generally, our results indicate that the populations share most of the mtDNA haplotypes and show very low differences between the populations (F_{ST} in the nDNA SNP = 0.0017). The high genetic similarity between the populations suggests their common history resulting from postglacial recolonisation and admixture of populations of different origins. The results may be useful in the discussion on existing breeding and forest management strategies used with Scots pine to counteract increasing pressure and the negative consequences of environmental changes.

Disturbance to forest soil related to small-scale forwarding in Mediterranean beech high forests

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Keywords: sustainable forest management, sustainable forest operations, forestry-fitted farm tractor, Apennine, short wood system

Thinning interventions in Italian beech forests are usually carried out by local forest enterprises relying on a low or medium level of mechanization (small-scale forestry). The short wood system is usually applied, with extraction operation performed by forestry-fitted farm tractors equipped with forwarding bins. Despite its wide application, no information is available in the literature about the possible impacts to the forest soil when applying this extraction method. To fulfil this knowledge gap, we developed the first assessment of soil physicochemical (bulk density, penetration resistance, shear resistance, organic matter content) and biological (soil microarthropods biodiversity evaluated with the QBS index) properties for this kind of intervention. We selected three case study areas in Central Italy, and applied an experimental design to evaluate separately the impacts related to the passage of the machine and that of the silvicultural treatment. We used linear mixed effect models to assess the relationship between changes in soil physicochemical and biological features. We found the effect of the silvicultural treatment to be negligible, but there was a strong alteration of the studied variables in the soil affected by the passage of the machine. Soil penetration and shear resistance doubled in the forwarding trails (0.25 MPa and 4.02 t m^{-2}, respectively) in comparison to the other two experimental treatments (control area and soil not affected by the machine passage; about 0.12 MPa and 2.10 t m^{-2}, respectively). Soil organic matter and soil biological quality (QBS index) were reduced by 25% in the forwarding trails (about 30% and 92 respectively) in comparison to the soil not affected by the machine passage (about 39% and about 130, respectively). Such strong disturbance is related to the high number of machine passages needed to extract the woody material with forwarding bins applying the short wood system. We found a significant relationship between soil compaction and soil organic matter removal and microarthropod biodiversity, with the latter which resulted significantly lower in more compacted and organic matter-poor soils. We therefore demonstrated that this small-scale extraction system can have a negative effect on soil features in the short term. We therefore recommend the application of best management practices, for instance placing brush mats and logging residues, on the forwarding trails to limit the soil distur-

bance. We further recommend a wider application of aerial harvesting systems, which generally have lower impacts to the soil.

The study was financed by the project AIMSUSFOR "Extending assessment of the environmental impacts to the forest ecosystem due to forest management: a comprehensive approach to enhance sustainable forestry in the context of climate change", funded under the PASIFIC Call 1 announced by the Polish Academy of Sciences. This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 847639 and from the Polish Ministry of Education and Science. The study was further financed by the statutory research of the Institute of Dendrology PAS.

Adaptation to warming: genetic variability and potential range shifts in European beech (*Fagus sylvatica*) across a temperature gradient in Poland

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Keywords: genetic variability, adaptation, climate warming

With the advance of global warming, many European beech habitats are becoming increasingly inhospitable due to shifts in climate. Rising average temperatures, prolonged droughts, and extreme weather events have heightened the vulnerability of beech trees to pathogens, leading to a decline in their populations across various regions. This study aimed to explore the genetic diversity among young and mature beech trees in natural environments across a temperature gradient. By doing so, we sought to uncover any genetic adaptations linked to environmental factors and forecast potential shifts in the species distribution.

Our approach mirrored the methodology of Capblancq et al. (2020), who examined beech trees in southeastern France. Using the annual mean temperature data (Bio01) from CHELSA (chelsa-climate.org), we identified nine distinct temperature zones across Poland. With assistance from the Forest Data Bank (bdl.lasy.gov.pl), we pinpointed forests where beech trees were prevalent in these zones. We collected leaf samples from both mature trees (exceeding 29 cm at breast height) and saplings (up to 120 cm tall) within these areas. Additionally, we included four herbarium specimens (WRSL) from locations near our study sites in our analysis. We extracted DNA from collected samples and sequenced it using the DArT-seq technique (Diversity Arrays Technology Pty Ltd, Australia). Single nucleotide polymorphisms (SNPs) underwent examination through principal component analysis (PCA), a clustering algorithm (sNMF), and redundancy analysis (RDA) performed in R.

We excluded eight specimens from our study due to their high genetic similarity and ambiguous placement on the PCA plot. The PCA revealed modest variability, with the first three principal components accounting for minimal explained variation (PC1 = 4.1%, PC2 = 2.5%, and PC3 = 1.7%). Notably, two populations (BWP and GBU) were distinct from the rest in our analyses. The cluster analysis identified two main groups, with one aligning with the distinct GBU population. Most individuals were classified in the second group, indicating a generally close genetic relationship among the studied plants.

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References

Capblancq T., Morin X., Gueguen M., Renaud J., Lobreaux S., Bazin E. 2020. Climate-associated genetic variation in Fagus sylvatica and potential responses to climate change in the French Alps. Journal of Evolutionary Biology 33: 783–796.

Insights into annatto's adaptive strategies to drought stress: implications of juvenility

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Keywords: miR156, SPL proteins, gas exchange, water use efficiency

Global temperatures will rise by up to 4°C in the coming years. The increased temperatures will increase drought periods in different Brazilian regions, mainly where *Bixa orellana* L. (annatto) is usually cultivated. This Amazonian woody species is the primary source of bixin dye, used worldwide. We showed that juvenilized annatto plants exhibit elevated ABA levels. Hence juveniles may be more susceptible to drought, which threatens annatto populations and bixin production. Accordingly, we exposed annatto miR156ox (overexpressing miR156) plants, as well STTM156 counterparts (exhibiting reduced activity of miR156) to a water deficit episode (up to 30% of field capacity over 20 days). We measured physiological parameters before, during, and after the drought period. STTM156 plants exhibited higher CO_2 assimilation (*A*) than miR156ox plants under control conditions. However, when subjected to drought, *A* was nearly halved in miR156ox lineages. Moreover, under drought, both transgenic lines displayed different stomatal conductance (*gs*) when compared with Non-transformed (Nt) plants. Remarkably, STTM156 plants displayed higher *gs* values than those in Nt and miR156ox plants, 61 and 300%, respectively. In addition, miR156ox decreased the *gs* under drought whereas STTM156 maintained *gs* following drought which might be circumstantially explained by the higher CO_2 demand. Upon recovery, Nt and STTM156 were capable of re-establishing the *gs* values compared to control conditions. This can be attributed to the required CO_2 for the regrowth of structures lost during drought, such as leaves. The pre-dawn water potential (Ψpd) in Nt miR156ox, and STTM156 followed the expect pattern of *gs*. Furthermore, STTM156 showed a higher water use efficiency (WUE) than miR156ox under control conditions. However, during drought, miR156ox displayed improved WUE compared to both well-watered conditions and Nt plants, a fact attributed to transpiration regulation. Upon recovery, all plants exhibited similar WUE, albeit approximately 50% lower than those under well-watered conditions due to increased transpiration. In summary, our study provides valuable insights into the annatto's physiological responses to drought and how precarious bixin production is in the face of climate change.

The effects of *Pinus sylvestris'* geographical origin on the community and co-occurrence of fungal and bacterial endophytes in a common garden experiment

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Keywords: Scot pine, root microbial communities, tree origin, common-garden, season

Below-ground microorganisms, particularly endophytes, are crucial for plant establishment and functioning through nutrient acquisition, as well as for enhancing resistance to abiotic and biotic stresses. The impact of tree origin within a species on the composition and interaction networks of root endophytic fungi and bacteria has been less explored than plant phylogeny and biological distance. This study investigates the effect of geographic origin on the fungal and bacte-

rial microbiomes of *Pinus sylvestris* root endophytes. Roots from plants grown in a common garden, originating from six locations, were harvested in two seasons. The fungal and bacterial microbiome was analyzed using Illumina MiSeq sequencing, targeting the ITS and 16S rRNA gene regions, respectively. The richness of Operational Taxonomic Units (OTUs) of endophytic fungi and bacteria showed no significant relation to tree origin or season. However, the Shannon diversity index for endophytic fungi was season-related. The composition of endophytic fungal and bacterial communities was affected by both tree origin and season, correlating with host root biochemical parameters such as starch, total non-structural carbohydrates, carbon, nitrogen, and climatic factors like mean annual precipitation and temperature. Moreover, the abundance of specific endophytic fungi and bacteria varied across different *P. sylvestris* origins, depending on the season. The complexity of the co-occurrence networks of fungal and bacterial endophytes within *P. sylvestris* also differed by geographical origin and season. This study highlights the significant role of the biochemical and climatic factors associated with tree origin in shaping interactions with endophytic communities, potentially affecting plant health and adaptability across diverse environments. These insights expand our understanding of the influence of host plant origin, including seed origin climatic conditions (e.g., MAT, MAP), on the assembly of root endophytic communities.

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Study of the chemistry of leaf litter decomposition of selected deciduous trees in environmental conditions

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Keywords: VOCs, leaf litter, GC-MS

For many years, a constant increase in atmospheric pollution caused by photo-oxidants such as ozone, organic and inorganic peroxides, has been observed at the global and regional level. It has been proven that they are produced in chemical processes involving volatile organic compounds (VOCs) and nitrogen oxides (NO_x). Our globe's vegetation is the main source of VOC emissions into the atmosphere. Global phytogenic emissions of organic substances are estimated at 450 million tons per year. The release of these compounds exceeds all anthropogenic sources by 10 times.

Organic matter accumulated on the forest floor in the form of the fallen and decaying leaves of deciduous trees remains unexplored as significant natural sources of reactive VOCs. The aim of the study is to investigate the chemistry of the decomposition of deciduous forest litter from the most common trees in Poland: oak, birch, alder, aspen and hornbeam under environmental conditions. The work undertaken detailed studies of both organic compounds released into the atmosphere from fallen leaves (VOCs) and non-volatile, soluble organic components that constitute substrates for various types of organisms – destructors, as well as those washed out by precipitation into forest soils. The study also examined the chemical composition of volatile substances and extractable organic compounds originating from fallen inflorescences of forest-forming tree species. Due to the complex chemical composition of the tested material, various analysis techniques were used in this study, such as headspace analysis with solid phase microextraction (HS-SPME/GC-MS) and three-stage solvent extraction. To examine the molecular composition of organic compounds leached from leaf litter (birch, hornbeam, poplar, alder) by rainfall, the technique of gas chromatography coupled with mass spectrometry was used. The parameters of chemical oxygen demand (COD) and biochemical oxygen demand (BOD_5) were used as a quantitative indicator of the amount of dissolved organic carbon (DOC) washed from leaf litter by rainwater. To study the composition of VOCs released by the living flowers of woody plants, the HS-SPME/GC-MS technique and polar solvent extraction were used.

The influence of environmental variables on the species composition and richness of oak forest stands in large-scale Scots pine monocultures

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Keywords: old stands, middle-aged stands, habitat islands, vascular plants

The natural vegetation of Europe consists predominantly of deciduous forests, which are considered to be hotspots of biodiversity. However, with the development of civilization, most fertile habitats have been deforested, leading to the fragmentation and reduction of the deciduous forest area. Nowadays, Scots pine monocultures dominate lowland landscapes. Therefore, even small patches of deciduous forests in pine monocultures should significantly increase biodiversity. Stands of native oak species, including pedunculate and sessile oaks, are particularly important, as they form different forest types, such as acidophilous and thermophilous oak forests, oak-hornbeam forests, and riparian forests.

The aim of the study was to assess the composition and species richness of vascular plants in oak forest stands that form habitat islands within pine monocultures.

Data were collected from 100 circular plots (300 m^2) in the Bory Stobrawskie Forest in southwestern Poland. These plots were set in oak stands of two age groups: 41–80 years and 100–180 years, by taking into account habitat fertility and moisture. Variation in species composition was assessed using DCA ordination analysis. Indicator species were selected for each age, fertility and moisture group of the studied oak stands, based on the fidelity (phi coefficient) of species. The groups of oak stands were also compared in terms of species richness, the number of forest species, and ecological indicators calculated with species habitat preferences. Additionally, the variation of species richness, the number of forest species and ecological indicators were assessed, based on the values of correlation coefficients with, among other things, the age of oak stands or the percentage of tree species in a tree layer.

Oak stands, regardless of their age, are an important element for the species diversity in woodlands dominated by pine monocultures. They exhibit diverse species composition. Their species richness depends on the productivity and moisture of the habitat, the species composition of the tree stand layer, and the spatial location of the patches.

Oak forest stands, despite being dispersed and occupying a small area in the studied woodland, are important for biodiversity and enhance non-productive forest functions. They provide favorable conditions for forest species that may not thrive in the surrounding pine monocultures.

The impact of forest management and climate change on bryophyte diversity of the close-to-natural forest

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Keywords: bryophyte, forest management, climate change, natural forest

Bryophytes are one of the most significant parts of the forest. Their ecological characteristics, tied to specific habitats and substrates – e.g. the bark of living trees, and decaying wood, along with their reliance on air humidity – enable them to exhibit immediate responses to environmental alterations. Consequently, they serve as valuable indicators of changes in the forest environment. The presence of various ecological groups of bryophytes, their abundance and species diversity, contributes to the heterogeneity of a forest (by creating microhabitats for dependent organisms), defining its natural biodiversity, stand structure, and ecological continuity. Therefore, bryophytes can serve in forest ecosystems to assess both the degree of naturalness and the impact of human activities.

Gaining a comprehensive understanding of the factors and processes that govern the interactions between managed and natural forests and bryophyte diversity are essential for the conservation of biodiversity. However, there is a limited number of studies elucidating the impact of microhabitat diversity on bryophyte species composition along gradients influenced by extensive human-induced alterations. This knowledge is necessary to improve the proper conservation of lowland forests and their biodiversity.

Studying the impact of forest management activities in close-to-natural forests on changes in the abundance of bryophytes and their diversity opens new perspectives for forest management, where the effects of climate change necessitate dynamic planning of nature conservation measures and sustainable forest resource management.

The aim of our study is to assess the changes in the diversity of forest bryophytes caused by forest management and climate change in close-to-natural forest, and the relative importance of these two drivers of change. We hypothesise that the comparison of natural and managed forest ecosystems will reveal that:

1) the taxonomic diversity of bryophytes depends on the availability of substrata and microhabitats for epiphytic organisms,
2) the functional diversity of bryophytes is higher in natural than managed forests,
3) and the effect of climate change will be negligible compared to changes in biodiversity caused by forest management.

Temporal modulation of the ecophysiology of *Schinus terebinthifolia* and *Dalbergia ecastophyllum* used in the revegetation of a fragment of the Atlantic Forest

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Keywords: green manure, plant growth, photosynthesis

The Atlantic Forest is a rich biome that comprises the coastal region of Brazil. The objective of this work was to investigate the effects of different soil treatments associated with the presence and absence of a type of green manure on the ecophysiology of two species native to the Atlantic Forest: *Dalbergia ecastophyllum* (L.) Taub. and *Schinus terebinthifolia* Raddi.

This study aimed to quantify photosynthetic pigments, Chl *a* fluorescence transient, and growth. The data were subjected to an analysis of variance (ANOVA), and the means were compared by the Tukey test at 5% significance in the statistical program R. A principal component analysis (PCA) was also performed.

The content of photosynthetic pigments was influenced by the treatments with green manure (+GM). At 12 months, *Schinus terebinthifolia* showed an increase in approximately 62.5% (T1), 21.9% (T2), and 82.4% (T3) for the carotenoid content. In turn, at 12 months, *D. ecastophyllum* showed an increase in approximately 8.1% (T1), 6.5% (T2), and 19.0% (T3). Carotenoid pigments play an important role in the absorption of light by the antenna system. They also protect the photosynthetic apparatus against damage from excessive light by mitigating harmful side reactions caused by reactive oxygen species (ROS).

Soil and green manure treatments influenced the JIP test parameters. The photosynthesis process (SFI_{abs}, and PI_{abs}) increased in +GM in all treatments for *S. terebinthifolia* and *D. ecastophyllum* at 12 months in SUBH treatment. In our study, the PI_{abs} and SFI_{abs} parameters demonstrated the mitigating effect of +GM.

The plant height (4.3% and 13.4%) and stem diameter (39.3% and 24.5%) were higher just in SUBH treatment at 6 and 12 months, respectively in *S. terebinthifolia*. The same was observed in *D. ecastophyllum*. Thus, the SUBH technique (T3) that promotes the highest degree of soil decompaction associated with +GM demonstrates its potential use in projects focused on the revegetation of degraded areas.

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Geocaching as a tool for the popularization of forest sciences

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Keywords: geocaching, popularization, environmental education, GPS (Global Positioning System)

Forests are important both for the environment and for humans. A substantial number of people acknowledge the importance of forests and relax in them, but they lack knowledge of the species living there or the functioning of forest ecosystems.

Geocaching is a GPS game which can be used to teach non-professionals about various places, including forests and their elements (Referowska-Chodak 2020, Zecha 2012). Geocaching does not require in-person meetings and thus can be used for learning asynchronously, with no need to gather a group of people at the same place and time. Geocaches – various kinds of boxes ("treasures") with different content – are hidden at given coordinates in the proximity of the locations of interest, which may be of various types: historical objects, old characteristic trees, viewpoints, lakes, or whatever the geocache's author wants to present. On the webpage, every geocache has a description of the place. In some cases, the user has to answer some questions in order to find the geocache, which further motivates them to gain knowledge.

Here I present examples of geocaches which are published on the webpages opencaching.pl and geocaching.com that focus on forest organisms or their relationships. I also discuss the roles of geocaching in the protection of the environment, e.g., meetings of geocachers who collect litter from forests, or geocaching in protected areas. Finally, I invite you to try geocaching in exemplary areas surrounding the conference location.

References

Referowska-Chodak E. 2020. Geocaching in education – a review of international experiences. Part 2. Recipient, location and subject matter of education. Forest Research Papers 81(2): 81–90.

Zecha S. 2012. Geocaching, a tool to support environmental education!? – An explorative study. Educational Research eJournal 1(2): 177–188.

How do phorophytes shape the diversity of dependent epiphytic flora in Central European lowland forests?

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Keywords: bryophyte, commercial forest, phorophyte-epiphyte

Conserving biodiversity is a primary objective of sustainable forest management. Bryophytes, a key part of our flora, offer insights into forest health due to their taxonomic diversity. Accordingly, leveraging this information can prove instrumental in conservation endeavors.

Epiphytic bryophytes, growing on the bark of living trees, influence forest diversity; indirectly, as pioneer organisms, they create microhabitats for other groups of organisms. As phorophytes, trees provide many microhabitats for epiphytic bryophytes, and their physical and chemical characteristics, such as the pH of bark, water holding capacity of the tree bark, and bark sculpture (sculptura), shape epiphyte diversity. Previous studies on epiphytes have provided information on the best-preserved forests in the Central European lowlands, focusing on the Carpathian forests or the protected areas of the Białowieża Forest. The aim of our study is to estimate the species richness and ecological diversity of epiphytic bryophytes in a managed oak-hornbeam forest (*Tilio-Carpinetum*). We hypothesize that: (1) tree type (phorophyte) shapes bryophyte species richness; (2) physical host tree traits such as thickness classes (breast height) influence the diversity and abundance of epiphytic bryophytes, and (3) the pH of bark and water capacity shape guilds of epiphytic bryophytes.

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Needle structure and photochemical efficiency along a needle age gradient in four species of evergreen conifers

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Keywords: chlorophyll a fluorescence, chlorophyll, photosynthesis

The structure and photosynthetic properties of *Abies alba*, *Taxus baccata*, and *Picea abies* needles change with needle age along the horizontal light gradient within the crown. The results of the present study have provided evidence of the synergistic effect of the light level and needle aging on the values of leaf mass-to-area ratio (LMA), photochemical parameters, and the photosynthetic pigments content in needles. Generally, LMA tends to increase with needle age. There is a predictable and continuous decrease in the maximum quantum yield of Photosystem II efficiency (F_v/F_m) as needles age. High F_v/F_m in old needle age classes of the tree species was the result of self-shading, while the decrease in F_v/F_m of current needles of the tree species may result from a photoprotective mechanism. It is evident from this study that the apparent maximal electron transfer rate (ETR_{max}), strongly correlated with the overall photosynthetic capacity, decreases with needle age. It was also discovered that non-photochemical quenching of chlorophyll *a* fluorescence (NPQ) is higher in older needles than in young needles at the same level of fluorescence induction light. There are distinctions in the total chlorophyll levels across the three tree species. The values of photochemical parameters and total chlorophyll content for all the species are in tandem with the structural, physiological and environmental changes associated with increasing needle age.

Aliens are among us… *Colletotrichum acericola* as a threat to native maple species in Poland

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Keywords: invasive species, ash-leaf maple, spillover

Plant invasions are a global phenomenon, with human activity cited as the main cause (Giorgia, Hulme 2023). Alien species can lead to the displacement of native ones, which significantly affects the performance of entire ecosystems (Najberek, Solarz 2016).

In Poland, the ash-leaf maple, *Acer negundo*, was brought to botanical gardens in the early 19th century (Patejuk et al. 2023). In the following decades, the spread of this species outside the gardens was observed, which was the first stage of invasion (Tokarska-Guzik 2005). Alien species can be reservoirs of pathogens that can affect native species, a phenomenon known as spillover (Power, Mitchell 2004).

The aim of the study is to answer the question: does *Colletotrichum acericola*, isolated from ash-leaf maple, pose a phytosanitary threat to the native species *Acer platanoides* and *A. pseudoplatanus*? In order to answer this question, several infection tests, using strains of *C. acericola*, were conducted on both the leaves and shoots of three chosen species of maple trees that are found in Poland, including the invasive *A. negundo*.

Monitoring alien species, which have the potential to become invasive, is a challenging process. Even more daunting is the task of eliminating invasive species from an area (Tokarska-Guzik 2012). Therefore, it is crucial to prevent their invasion into ecosystems, where they can pose significant threats to native organisms and biodiversity.

References

Gioria M., Hulme P.E. 2023. Why are invasive plants so successful? Annual Review of Plant Biology 74: 635–670.

Najberek K., Solarz W. 2016. Gatunki obce. Przyczyny inwazyjnych zachowań i sposoby zwalczania. Kosmos 65: 81–91.

Patejuk K., Czachura P., Baturo-Cieśniewska A., Owczarek-Kościelniak M., Pusz W., Najberek K., Piątek M. 2023. Colletotrichum acericola sp. nov. from seeds of the invasive alien tree species Acer negundo in Poland. Plant Pathology 72(9): 1716–1725.

Power A.G., Mitchell C.E. 2004. Pathogen spillover in disease epidemics. The American Naturalist 164: 79–89.

Tokarska-Guzik B. 2005. The establishment and spread of alien plant species (kenophytes) in the flora of Poland. Wydawnictwo Uniwersytetu Śląskiego, Katowice.

Tokarska-Guzik B., Dajdok Z., Zając M., Zając A., Urbisz A., Danielewicz W., Hołdyński C. 2012. Rośliny obcego pochodzenia w Polsce ze szczególnym uwzględnieniem gatunków inwazyjnych. Generalna Dyrekcja Ochrony Środowiska, Warszawa.

Delayed response of bryophytes to wind disturbance and salvage logging in hemiboreal mixed forests

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Keywords: wind disturbance, post-windthrow succession, storm legacy, windthrow

Climate change accompanied by altered natural disturbance regimes threatens the resilience of boreal and hemiboreal forests. Bryophytes, an important part of plant biomass and diversity, fulfill numerous important functions in forests, thus markedly contributing to the resilience of these ecosystems.

We studied the abundance, richness, diversity and community composition of bryophyte species 20–21 years after wind disturbance in moderately damaged, heavily damaged, and heavily damaged and subsequently salvage-logged stands. Overmature forest stands with similar tree species composition were included in the study as a reference group. The community characteristics were linked with treatment and accompanying environmental variables.

Altogether, 108 bryophyte taxa (81 mosses and 27 liverworts) were identified in the study plots. We found that bryophytes responded to windthrow severity level and salvage logging in terms of species richness, diversity, and composition; the diversity of microhabitats was the most important environmental variable explaining the variation in diversity metrics. Besides exhibiting greater overall species richness, uncleared wind-disturbed plots contained more bryophyte species with high conservational value as well, compared to salvage-logged and control plots.

The largest number of bryophyte taxa was associated with deadwood substrates. Ordination analysis further emphasized the importance of deadwood substrate for liverworts. In order to fulfill the objective of biodiversity conservation, wind-disturbed forests should be left to recover naturally, to ensure a favorable status of all naturally occurring species and to improve the status of endangered and rare species. Retaining a proportion of surviving and/or wind-damaged trees while salvaging might also alleviate the unfavorable environmental conditions for bryophyte species while "lifeboating" some species through the following successional stages.

Noise propagation from chainsaw cross-cutting in forest environments

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Keywords: impact, disturbance, sensitive area, Sustainable Forest Operations

Forests represent an environment in sensitive ecological equilibrium, where activities related to wood harvesting always generate environmental disturbance. Forest ambient background sound level is generally low, and in the past the human presence in the forest during wood harvesting activities caused slight sounds, such as footsteps, creaks, verbal communications, and axe chops. However, with the advent of technology, in particular with the introduction of engines, the noises have become increasingly conspicuous. Forestry operations often involve the use of equipment and machinery that can produce noise and represent a potential permanent or temporary disturbance to wildlife. For this reason, some studies focused attention on the ecological effects of anthropogenic noise in nature, demonstrating negative impacts on the life of many animal species, which are evident from permanent changes in their behaviour and spatial distribution. Although the effects of noise on the natural environment are now widely known, the propagation of noise in natural and semi-natural areas due to forestry operations has not yet been sufficiently explored in the literature.

The work aimed to carry out a first experimental approach to assess the range and intensity of the noise generated by chainsaws with different powers (distinguished in terms of low, medium and high-power chainsaws) by applying an appropriately set up measurement system in order to recreate a realistic condition. The study was conducted in a sensitive forest area of the Sila National Park (southern Italy), where a pilot simulation of cross-cutting was tested on beech and Corsican pine logs. In particular, this research aims to evaluate the human impact on environmentally sensitive areas, in order to gain insight into the phenomena of acoustic disturbance, and to mitigate the negative ecological impact of forest operations. The simulation consisted of wood cutting discs of the same thickness from logs with the same diameter using different chainsaws. The assessment of noise propagation during cross-cutting was carried out using 26 sound dataloggers, which recorded the sound levels in a geometric pattern planned in the test area. Multiple point noise sources and several frequency bands were examined and evaluated statistically to determine factors which have the greatest impact on the dynamics of noise propagation in a forest environment involving cross-cutting operations by motor-manual felling. The case study highlighted how chainsaws are one of the most critical sources in terms of noise level production in a sensitive environment like the forest.

How much do the floral, fruit, and leaf and stem traits of woody plants vary within a species?

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Keywords: floral and fruit traits, plant functional traits, organs' variability, trees and shrubs

While plant functional traits offer numerous advantages in modern ecology, efforts to address their limitations are crucial. These limitations encompass the insufficient recognition of intraspecific trait variability and the uneven coverage of organs, often leading to the neglect of floral and fruit traits (hereafter, reproductive traits). Reproductive traits are frequently sidelined in favor of more commonly used traits, such as specific leaf area (SLA), seed mass, or plant height. However, reproductive traits, often deemed highly conservative, may exhibit low levels of variability, allowing for less biased interspecific comparisons and thus posing great potential for further development of functional ecology.

This study aimed to compare the intraspecific variability of traits across four plant organs: leaves and stems (well-recognized), and flowers and fruits (less recognized). Data were collected for twenty species, each represented by five populations in Greater Poland, separated by at least 5 km. Each population comprised six individuals, from which leaves, stems, flowers, and fruits were collected throughout the vegetation season. Despite occasional incomplete sets of organs due to limited availability, our focus on intraspecific variability and the use of population mean values for analyses mitigated this limitation. We analyzed various functional traits, including dry mass, dimensions, nitrogen (N), and carbon (C) content, and C:N ratio for flowers and fruits; SLA, C and N content, and C:N ratio for leaves; and specific stem density, C and N content, and C:N ratio for stems.

We formulated two hypotheses: (1) reproductive traits would exhibit less intraspecific variability compared to performance-related traits like specific leaf area and specific stem density, and (2) reproductive traits would show stronger phylogenetic associations than the most frequently used functional traits. The results of this research will contribute to our understanding of the intraspecific variability of plant reproductive traits and the assessment of their potential for use in functional ecology research.

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Less productive, but successful. A case of *Cornus sericea* invasion in Lithuania

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Keywords: woody plant, alien, shrub, dogwood

Alien woody species constitute about 12% of the alien flora in Lithuania (of about 700 alien plant species, 83 are woody). Some of these species pose a serious threat to native plants and habitats by altering the characteristics of certain ecosystem elements. The genus *Cornus* is represented by seven taxa in Lithuania, of which only one taxon (*C. sanguinea* subsp. *sanguinea*) is native. *Cornus sericea* has been spreading throughout Lithuania for several decades, yet the invasion and impact of this species have long been overlooked. In neighbouring countries, *C. sericea* spreads in riparian habitats, wetlands, forests, mesic and wet grasslands, and anthropogenic habitats. In Lithuania, the species is found in various habitats, but its invasion of alluvial forests is particularly advanced.

We used phytosociological data from 30 sites of *C. sericea*, mainly from alluvial forests. We compared the ecological traits of four *Cornus* taxa (*C. alba*, *C. sanguinea* subsp. *sanguinea*, *C. sanguinea* subsp. *australis* and *C. sericea*) such as reproductive output, soil seed bank, seedling emergence and annual ring increment. The viability of intact seeds collected from the soil seed bank was tested by a standard staining method (TTC test).

According to the results of our research, *C. sericea* had the lowest number of fruits per infructescence and the lowest number of seeds in the soil seed bank. In addition, the percentage of viable seeds of *C. sericea* ranged from 11.8% to 100% at the individual study sites. The mean seed density of the species in the soil was generally low. Furthermore, these results correlated positively with the low numbers of emerged seedlings. The growth of the annual xylem rings of *C. sericea* was the most pronounced and this species was the fastest-growing of all the *Cornus* taxa studied. Even though this species was less productive, it is one of the most common alien shrubs in Lithuania. We found a negative moderate correlation between the cover of *C. sericea* and the number of herbaceous species per relevè ($r = -0.52$, $p < 0.01$, $N = 30$). Furthermore, there is an increasing propagule pressure and intensive vegetative spread of already established populations. According to the results of the study, *C. sericea* exhibits clear signs of invasiveness. We assume that this species has no competitors in its alien range, including alluvial forest habitats.

Land suitability maps based on the GIS-MCDA-AHP approach for selecting the most prudent forestry ectomycorrhization

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Keywords: GIS, MCDA, AHP, mycoforestry

The strategy for selecting the optimal ectomycorrhization system for a specific case involves the application of the GIS-MCDA-AHP approach. This method enables a meticulous selection of hypogeous mycoforestry operations by generating one or more suitability maps.

The aforementioned multi-criteria analysis and evaluation procedure use various bio-geochemical variables such as slope, altitude, rooting capacity, mechanized access, soil bearing capacity, climate precipitation, land use, management factors, soil drainage, soil depth, soil temperature, soil moisture, soil tolerance to natural vegetation disturbance, disturbance to water flow activities, soil micro and mesofauna, soil erosion disturbance, wildlife impact, soil composition (clay-loam sand), pH, calcium and organic carbon levels, available soil micronutrients, and specific canopy composition. These factors are used to assess potential alternatives for planting ectomycorrhizal forest plants within the operational context.

We applied Multi-Criteria Decision Analysis (MCDA) procedures, an Analytic Hierarchy Process (AHP), and rasterized and georeferenced each criterion in a GIS environment. We also reclassified rasters based on planting suitability classes for each forest system scenario.

Additionally, an extra informational layer was overlaid for each condition of ectomycorrhizal forest planting application, calculating the overall applicability adequacy of each forest ectomycorrhization use system for every pixel in the examined territory. These were categorized according to FAO suitability standards, achieved by summing the suitability value for each criterion, weighted by its relative importance determined by AHP.

This process yields a synthesized land suitability map, facilitating a quantitative evaluation of the appropriateness of planting operations for each pixel in the area of interest. It objectively defines the best mycoforestry operation strategy by extracting the mean suitability value using the zonal statistics raster command in GIS software.

Ultimately, this method offers mycoforesters greater precision in digital mycoforestry management, enabling them to design forest usage plans (both woody and non-woody) for different areas with meticulous and objective operation selection criteria.

Adapting artificial forest regeneration to environmental changes – perspectives from Slovenia

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Keywords: artificial regeneration, climate change, forest genetic resources, forest reproductive material, assisted migration, close to nature forest management

Forestry in Slovenia is based on the principles of sustainability, multifunctionality and close-to-nature forest management (CTNFM). CTNFM can be characterized as a small-scale, uneven-aged forestry system, which follows and uses natural processes with the aim of continuous sustainable provision of all forest ecosystem services. In Slovenia, the prevalent type of regeneration is natural, employing forest reproductive material (FRM) from local seed trees (over 90% of all forest regeneration).

To successfully adapt CTNFM to the effects of climate change, it needs to be enhanced with an active approach to forest regeneration with locally adapted FRM with high levels of species and genetic diversity. This is especially crucial for:

- the long-term conservation and improvement of the status of biological and genetic diversity in forests (by planting and sowing a wide spectrum of locally adapted forest tree and shrub species and provenances),
- facilitating carbon sequestration in forests (timely and successful regeneration of forest stands with high-quality stand trees and seedlings contributes to climate change mitigation and adaptation), and
- ensuring stability, resilience, forest health, and the sustainability of all forest ecosystem services.

The use of artificial regeneration in CTNFM can serve as an “insurance policy” which minimizes risk by facilitating the presence of diverse and climate-adapted FRM in future forests. To achieve this, we are working on several different strategies, including:

- the establishment and monitoring of large- and small-scale provenance trials, including audience beyond forest science (citizen science-based provenance trials);
- creating assisted gene flow (migration) models for the potential transfer of provenances of forest tree species potentially adapted to future climate;
- experimenting with new technologies and techniques to increase the survival of seedlings in extreme conditions (extreme drought, late frost, browsing by animals);
- developing strategies and protocols for monitoring artificial regeneration in all Slovenian forests;

- exploring stakeholder cooperation, policy and business models for the sustainable provision of FRM;
- establishing a new Centre for Forest Reproductive Material and Forest Health at the Slovenian Forestry Institute.

How do herbaceous forest understory plant species reflect global patterns? A study of the relationships between decomposition and plant functional traits

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Keywords: herbaceous layer, decay rate, biomass allocation, carbon cycling

Decomposition, next to photosynthesis, is a crucial ecological process shaping ecosystem properties and influencing all living organisms. Studies of this process in forest ecosystems usually omit the herbaceous layer, focusing on the tree layer. Many previous studies have demonstrated that plant traits influence decomposition rates; however, it is still not clear whether patterns found on a global scale are reproducible at local scales, specifically in different growth forms. The main aim of our study was to assess the impacts of habitat type and plant functional traits on the decomposition process of forest herbaceous species.

We conducted our study in three forest types along a wide gradient of soil hydrologic and nutrient conditions and examined the decomposition of 11 vascular forest understory plant species. We used litter bag methods in our studies, collecting litter bags monthly during the first year of the experiment and at three-month intervals afterward. Additionally, we measured 12 functional traits of herb plant species on our sample plots.

We found statistically significant effects of herb species and the type of forest on litter decomposition. After one year of the experiment, biomass losses were highly diverse, ranging from 26% for *Calamagrostis arundinacea* to more than 99% for *Corydalis cava*. The species studied may be arranged from the fastest to the slowest decomposition rate determined after one year of the study: *C. cava*, *Ficaria verna*, *Anemone nemorosa*, *Oxalis acetosella*, *Mercurialis perennis*, *Maianthemum bifolium*, *Urtica dioica*, *Impatiens parviflora*, *Alliaria petiolata*, *Deschampsia flexuosa*, and *Calamagrostis arundinacea*. Correlation analyses revealed negative relationships between biomass losses after one year of decomposition and stem mass fraction, leaf dry matter content, and the height of plant species. A positive relationship was revealed between biomass losses after one year of decomposition and specific leaf area.

In the studied species set, the litter decomposition rate was generally higher in smaller plants with a lower allocation of biomass to stems. The results are important for a better understanding of the habitat-shaping role of herbaceous species during the decomposition process. This knowledge can aid in the management and protection of forest ecosystems, particularly in the context of changing biodiversity. Moreover, they can help improve the assessment of the forest ecosystem's carbon cycle.

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Natural regeneration potential of black poplar along the Vistula River valley in Poland

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Keywords: black poplar, natural regeneration, genetic variability

Black poplar is the main component of riparian forests, and is an extremely important species from both ecological and economic points of view. Over the last decades, there has been a drastic decline in the number of individuals of this species, which is caused by human activity and the lack of areas suitable for the growth and development of young seedlings. An additional threat is the fact that black poplar may cross with fast-growing hybrid poplar varieties, which may lead to the contamination of its gene pool.

We studied four populations located along different sections of the Vistula River comprising a total of 623 black poplar trees. Each population consisted of a group of mature trees and a group of presumed natural regeneration. The aims of the research were to: (1) determine species purity and clonality, (2) assess the genetic variability, (3) compare the gene pools of populations, and (4) compare the gene pools of potential natural regeneration with the gene pool of mature trees growing in the same location. We also wanted to check whether the selected reproductive strategy differs depending on the location and degree of land transformation, as black poplar may also reproduce vegetatively. The analyses were carried out based on the polymorphism of 14 nuclear microsatellite markers and using species-specific nuclear and chloroplast DNA markers.

We identified nine hybrid individuals in the group of mature trees from the middle section of the river and one in the group of mature trees located in the lower section of the river. The highest percentage of clones was observed in the population from the lower section of the Vistula, where clones constituted 94.92% of the population, and the lowest percentage of clones (3.46%) was found in the population from the middle section of the river. The parameters of genetic variation were generally comparable between groups and the level of genetic variability remains high. The genetic differentiation among the studied groups of presumed natural regeneration was almost twice as high as among the groups of mature trees. The results of the principal coordinates analysis and genetic clustering showed that the population located closest to the influx of the river was the most different from the others. At the level of individual trees, we observed that only in the middle section of the river did the group of presumed natural regeneration reflect the gene pool of the mature trees group. For the remaining populations, the groups of presumed natural regeneration are less similar to the groups of mature trees. Black poplar populations in Poland are usually old and occur mainly in small groups. Moreover, many rivers have been significantly transformed, resulting in a lack of suitable areas for natural regeneration. Taking into account the above and also the results of our previous research, it is necessary to take actions aimed at protecting the gene pool of black poplar in Poland.

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Revolutionizing nursery and garden practices: peat-free substrates and novel fertilization, strategies for sustainable seedling production

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Keywords: peatland conservation, peat-free substrates, sustainable cultivation, liquid fertilizer

In recent times, there has been a growing recognition of the critical importance of peatland conservation, particularly in the context of environmental preservation. This heightened awareness has sparked a quest for alternative organic substrate mediums to replace peat, emphasizing materials that are easily accessible, cost-effective, sustainable, and environmentally friendly. Within the Faculty of Forestry at the University of Agriculture in Kraków, Poland, a dedicated team of researchers has successfully formulated a pioneering organic substrate. This substrate was derived from readily available, highly affordable, and entirely cost-free materials, ensuring sustainability and environmental compatibility. Additionally, this research effort has also yielded a novel recipe for liquid fertilizer.

The developed substrates were adapted to the nutritional requirements of the forest tree seedlings (*Quercus robur* and *Fagus sylvatica*), and their suitability under monitoring. The result provides valuable insights into the complex relationships between fertilization methods and the variance in the chemical composition of oak and beech seedlings after production in the nursery. The substrate mediums developed in this study have demonstrated qualities that are on par with peat-based substrates, as evidenced by robust root system development. The allocation patterns highlighted the variability of nutrient distribution within the seedling, with more nutrients allocated in the root system. Interestingly, the novel treatment recorded the highest mean root values. Based on the results of the study, the peat-free substrates combined with novel fertilization strategies could develop the forestry industry by enhancing the growth and nutrients of tree seedlings without relying on traditional peat-based substrates.

Surface deformation monitoring in forest road networks: a high precision UAV real-time kinematic approach

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Keywords: forest management, terrain analysis, erosion model and earthworks, timber extraction, photogrammetry

The significant increase in hiking, logging, and transportation negatively impacts the natural balance by leading to soil degradation along trails and forest roads. The objective of this project was to develop a Deformation Classification Model for a multi-use road surface, predict sediment deposition, and generate a flood risk map in a predominantly forested region. The eBee X mapping UAV, equipped with the senseFly S.O.D.A. 3D camera and Real-Time Kinematic (RTK) technology, conducted a flight over a 149-hectare study area in Northern Greece at a height of 120 meters, obtaining a spatial resolution of 2.6 cm. The distinctive structure of fixed-wing equipment eliminates the necessity for ground control points in terrain deformation research, a practice that was prevalent in previous studies predominantly utilizing polycopters.

Two distinct classifications were performed using the Digital Surface Model (DSM) and Digital Elevation Model (DEM) for analysis. The Geolocation Errors and Statistics for Bundle Block Adjustment showed high accuracy in the model, with mean values for the X, Y, and Z directions of 0.000023 m, –0.000044 m, and 0.000177 m, respectively. The standard deviation of the error in each di-

rection was 0.022535 m, 0.019567 m, and 0.020261 m. The Root Mean Square (RMS) error values are 0.022535 m, 0.019567 m, and 0.020262 m. There were 20 and 30 altitude categories established with a spatial resolution of 4 cm, each having specified value ranges. Each altitude category was measured in square meters (m^2) and in cubic meters (m^3) for volume. Developing a Deformation Classification Model for the surface of a multi-purpose forest trail, as well as calculating earthworks and generating a flood risks map, is an effective method to support forest managers in creating and maintaining the network of hiking trails and forest roads.

Monitoring and management of *Lymantria dispar* life stages in southern Greece

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Keywords: gypsy moth, trap devices, mating disruption

Lymantria dispar (L.) (Lepidoptera: Erebidae) is a serious defoliator in forests worldwide. It is one of the main pests of oak trees but it can feed on > 400 plant species. Due to the negative impact of chemical insecticides on the environment, alternative approaches to the management of this pest should be adopted. Thus, in 2023 efforts were made to control and monitor *L. dispar* by using trunk, pheromone, and electronic traps. The experiments were conducted in Ilia, Greece. The project included the comparison of three different trunk traps for their effectiveness in capturing *L. dispar* larvae. Additionally, six different prototype pheromone traps were compared with a commercial pheromone trap for the monitoring of *L. dispar* adult males. Adult males were also monitored with electronic traps.

Furthermore, the mating disruption method was applied and compared with an untreated area.

The results revealed that the prototype trunk trap was more effective in capturing *L. dispar* larvae than the two commercial trunk traps. With regard to the pheromone traps, 3 out of 6 prototype traps performed comparably to the commercial trap. Electronic traps recorded data and provided information on the environmental conditions associated with male presence and flight. Captures of males in the mating disruption area decreased in comparison to the untreated area. The overall results indicate the high potential of the alternative methods to monitor and manage the different developmental stages of *L. dispar*.

The LIFE eGymer project is co-funded by the LIFE Programme of the European Union under contract number LIFE20 ENV/GR/000801 and with the contribution of the Green Fund.

The effect of various tree species and time on the process of soil formation on post-mining sites: a common garden experiment with 22 tree species and a chronosequence from Sokolov, Czech Republic

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Keywords: reclamation, pedogenesis, vegetation, soil properties, time

The research presented here evaluated the effect of different tree species, and the effect of time on the initial soil formation processes on post-mining sites. Study locations included four reclaimed spoil heaps in the Sokolov brown coal mining basin in the Czech Republic. A comprehensive analysis of the physical and chemical properties of the soil formed under 23 woody species monocultures of the same age (both broadleaved and coniferous) was performed. The results show that broadleaved species such as *Alnus glutinosa*, *Fagus sylvatica*, *Tilia cordata*, *Acer platanoides*, *A. pseudoplatanus* and *Pyrus communis* exhibited more favorable soil properties and soil formation potential than the other broadleaves and conifers tested (Spasić et al. 2024b). However, the growth potential and resistance to climatic extremes shown by some conifers, both native (*Pinus sylvestris*, *Larix decidua*) and introduced (*Pseudotsuga menziesii*, *Pinus nigra*, *Picea omorika*), demonstrated that these species can be suitable for afforestation of post-mining sites in this region (Vacek et al. 2021, Spasić et al. 2024b). The indisputable effect of time on the initial soil forming processes was assessed through a chronosequence of 3 differently aged forest reclamation sites (Spasić et al. 2024a), as well as through observation of chemical properties on the Lítov spoil heap after 20 years.

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References

Spasić M., Vacek O., Vejvodová K., Borůvka L., Tejnecký V., Drábek O. 2024a. Profile development and soil properties of three forest reclamations of different Ages in Sokolov Mining Basin, Czech Republic. Forests 15(4): 650.

Spasić M., Vacek O., Vejvodová K., Tejnecký V., Vokurková P., Křižová P., Polák F., Vašát R., Borůvka L., Drábek O. 2024b. Which trees form the best soil? Reclaimed mine soil properties under 22 tree species: 50 years later—assessment of physical and chemical properties. European Journal of Forest Research 143: 561–579.

Vacek Z., Cukor J., Vacek S., Linda R., Prokůpková A., Podrázský V., Gallo J., Vacek O., Šimůnek V., Drábek O., Hájek V., Spasić M., Brichta J. 2021. Production potential, biodiversity and soil properties of forest reclamations: Opportunities or risk of introduced coniferous tree species under climate change? European Journal of Forest Research 140: 1243–1266.

Comparison of leaf chemical characteristics between invasive monospecific, native monospecific and native multispecific stands

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Keywords: influence, woody species, invasive, native, multi-species

Trees and shrubs are considered ecosystem engineers. Through the production of leaf litter, they can alter soil conditions, and these changes may determine the plant performance (growth rate, size and fitness) and the species' ability to be expansive.

The IMPAWOS project aims at examining the impact of invasive alien woody species (IAWS) and native dominant (potentially expansive) woody species (NEWS) on the ecosystems of the temperate zone of Central Europe (Czech Republic, Poland and Slovakia). The project focuses on trees and shrubs that can create monospecific stands, i.e., invasive *Acer negundo*, *Prunus serotine* and *Spiraea tomentosa*, and native *Acer platanoides*, *Crataegus monogyna* and *Swida sanguinea*. The control for the IAWS and NEWS monospecific stands are co-occurring native multi-species stands.

In the autumn of the first year of the project, senescent leaves from trees and shrubs were collected and analyzed for several chemical (contents of C, N, Ca, Mg, K, total phenolics and condensed tannins) and microbial properties (abundance of microbial populations using the PLFA method).

We hypothesized that species containing more phenolics in their leaves would be less colonized by microbes, and thus harder to decompose (which will be tested in further stages of the project using the litter bag method). We expected that there would be more such species in the IAWS than in the NEWS group. We also hypothesized that multi-species leaf material (control) would have a more constant chemical composition than single-species leaf material (both IAWS and NEWS).

The utilization of waste generated in the chemical industry for the purposes of sustainable forest management

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Keywords: sustainable forestry, waste management, heavy metals, post-agricultural land

Polish forestry traditionally operates on poorer quality land, while more fertile land is intended for agricultural purposes. If it is determined that the land is no longer suitable for food production, it can be designated for afforestation. Introducing the first generation of trees onto former agricultural land is fraught with a number of problems that are well described in the literature. Pre-fertilization of these lands with fertilizers containing phosphorus and calcium may be a significant factor improving soil properties and increasing the success of forest crops. Hence, there is a need for urgent research on the composition and form of the future alternative fertilizers, especially in the context of their impact on the growth of tree seedlings, selecting the appropriate dosage level and determining the migration of possible occurring pollutants, which will determine the potential use of fertilizer in afforestation process. For this reason, it is important to conduct field tests verifying the impact of fertilizers derived from waste raw materials on the growth of seedlings of forest-forming species. Economic, geopolitical and environmental reasons (a reduction in the amount of raw wood material available from abroad, the increase in the costs of obtaining and transporting raw wood material, the future increase of the acreage of protected forest areas) can reduce the availability of the wood for the wood industry sector. One of the ideas to mitigate this situation is to increase the afforestation success by the introduction of fertilizers based on the waste from the chemical industry, e.g. generated during the production of phosphoric acid, which contains essential elements such as phosphorus or calcium, but may also contain pollutants such as heavy metals.

The aim of the study was to assess the potential of Scots pine seedings for the short-term absorption of selected heavy metals from the soil fertilized with the waste material generated during the production of phosphoric acid. As a part of the task, soil and wood tissue samples were collected from tests to determine the impact of fertilizer addition on the condition of Scots pine seedlings after 5 months of pot cultivation. The ability of seedlings to accumulate heavy metals was also determined. The collected results allowed the preparation of an ongoing long-term test conducted in a larger scale.

This investigation received financial support from the Polish Ministry of Science and Higher Education (project no. S-5906-0-2023).

Peatland restoration correlates with *Sphagnum fuscum* morphology

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Keywords: peatland restoration, Sphagnum fuscum, carbon sequestration, peatland

Peatland restoration is recognised as an important part of climate change mitigation strategy. Peatlands not only have the potential to sequestrate great amounts of carbon and store huge amounts of water but also act as natural buffers for disasters. Pristine peatlands also host a unique biodiversity, protection of which is important in times of global biodiversity crisis. Despite the importance of the peatlands, vast areas of them have been – and still are – drained and mined every day.

Countless peatland rewetting projects have been introduced around the world with the aim of restoring the unique properties of a pristine peatland. However, the methods to assess the efficiency of peatland restoration success are lagging. Measures of restoration success are urgently needed to assess if and where the peatland functions, including valuable ecosystem services, are returning towards the natural state.

Here, I will discuss the use of *Sphagnum fuscum* functional traits analysis as a method to assess peatland restoration efficiency. *Sphagnum* are key species involved in peatland formation and biomass production. *Sphagnum fuscum* grows in drier parts of peatlands, forming hummocks. This suggests that it could be the first to recover during peatland restoration. Its functional traits were analysed from pristine, restored, and drained sites selected from southern Finnish peatlands.

We show that *Sphagnum fuscum* collected from restored peatlands showed a clear recovery trend, with most functional traits resembling the traits of the species from pristine rather than from drained peatlands. Our work indicates that peatland restoration can successfully recover not only *Sphagnum* coverage but also its traits, at least on hummock surfaces. This result is encouraging and suggests that peatland restoration can bring back ecosystem services, including important carbon sequestration abilities. Further, this study suggests that the use of plant functional traits can provide important information on peatland recovery after restoration. To assess the recovery of the plant functional diversity on peatlands after restoration in a wider perspective, more studies with more species are needed.

Genetic and clonal structure of the white poplar (*Populus alba*)

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Keywords: population genetics, dioecy, clonality, Populus alba, plant ecology

White poplar (*Populus alba* L.) is a dioecious and clonal species from the Salicaceae family, commonly found in river valleys and floodplains. These areas are characterized by unique environmental conditions that are crucial for the survival of white poplar and other riverside plants. White poplar is part of the biologically diverse poplar riparian forests (*Populetum albae*), which are increasingly rare in Europe due to drainage works and river regulation. This threat results in the spatial isolation of the white poplar population and limits the natural regeneration of the species. We need to understand the genetic resources of white poplar, including its variability and the factors that influence it, as it is crucial to protect these areas. Additionally, it is also important to consider the consequences of clonality in combination with the dioecious nature of white poplar.

Dioecy is a specific sexual system where distinct female and male plants exist within a population. This rare method of reproduction is present in only about 6% of angiosperms. Dioecy leads to noticeable differences between female and male plants, including variations in growth, sensitivity to stress, and preferred habitats, which can result in segregation based on gender. Clonality is the process of vegetative reproduction that gives rise to a new plant genetically identical to the parent plant unless somaclonal variability occurs, but potentially independent in terms of growth and reproduction. While clonality is widespread among plants, there is limited information on its ecological impacts in dioecious tree species.

Studying the interplay of dioecy and clonality is especially valuable among a species that occurs in its natural habitat. The project, including a doctoral dissertation, aims to investigate how clonality and dioecy impact the growth rate, response to climate, and spatial distribution of white poplar. The project includes three populations of poplar trees in Poland. By using nuclear microsatellite markers (SSRs), commonly used in population and conservation genetics, the study can assess the genetic structure of these populations and identify any sex-specific factors. The SSRs used in genotyping allow the identification of intra- and inter-population genetic variation.

Understanding the genetic structure of populations is essential for implementing effective conservation strategies to protect the biodiversity of unique forest ecosystems like riparian forests.

Mycobiome of *Alnus glutinosa* roots and rhizosphere in response to salinity

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Keywords: salt stress, fungal community, ectomycorrhizal fungi, black alder

Continuous changes of global climate conditions increase soil salinity and decrease water availability for plants. Afforestation is one of the most effective ways to combat the climate crisis, as it raises groundwater levels and reverses the process of soil salinisation. Pioneer tree species, like black alder (*Alnus glutinosa* Gaertn.), have proven to grow successfully in saline soil conditions.

The main objective of our study was to characterise the fungal microbiome on absorptive roots and in their rhizosphere, which may have a significant impact on the increased tolerance of black alder to high soil salinity levels during the growing season. We combined the use of the metagenomic approach to analyse the present mycobiome and absorptive root growth dynamics with Minirhizotron (MR) observations during three seasons (spring, summer, fall) at one non-saline and two saline sites in northern Poland.

Salinity decreased the total number of observed fine roots and caused lower production of roots during the growing season. While the production of absorptive roots was about 2.5 times higher during summer than in spring and fall at the control site, the lowest production was noted in summer in both saline soils. It was found that absorptive roots and rhizosphere fungal community differ significantly ($p = 0.001$) with significantly higher numbers of Amplicon sequence variants (ASV) and species diversity H' indices in the rhizosphere samples.

Thelephora, *Acephala* and *Phialocephala* sp. were more frequent in absorptive roots, while *Podila*, *Mortierella* and *Alternaria* sp. occurred more often in the rhizosphere. Relative abundance of EcM fungi increased in spring and summer and decreased in fall in saline soils compare to non-saline. The frequency of *Thelephora* sp. was higher in saline soils compared to non-saline, whereas in the case of *Lactarius*, *Tomentella* and *Russula* sp. the opposite tendency was observed. Salinity decreased the number of observed EcM root tips and morphotypes and also delayed root colonization during the growing season. At all sites the highest EcM tip production was recorded in fall.

In summary, soil salinity limits the growth of absorptive roots in black alder, resulting in changes of the generic structure of the mycobiome and the relative abundance of EcM fungi, as well as the occurrence of specific EcM morphotypes.

Growing silver birch (*Betula pendula*) on post-agricultural land for higher wood quality: a case study in north-western Poland

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Keywords: post-agricultural land, silviculture, tree development

Silver birch is one of the most widely spread hardwood tree species in Europe, especially in the Baltic countries. Birch is a pioneering species that adapts easily to difficult conditions with high productivity and rapid growth. Due to this, it is a preferred species for the afforestation of post-agricultural, mining and abandoned lands. As many studies showed, a history of land use may lead to improved growth of first-generation trees planted on post-agricultural land, and to harvesting large quantities of timber in a short period of time. The most intensive afforestation of post-agricultural land in Europe was observed after the Second World War. Although also in the 1990s several national programmes were established. In Poland, between 1995 and 2014, 274,300 ha of agricultural land was afforested this way.

Currently, various stands grown on post-agricultural land are reaching the cutting age. This is an opportunity to assess the effects of several decades of silviculture on such types of land, and to check the impact on tree development. This study aimed to analyse the influence of growing silver birch on post-agricultural land for higher wood quality, based on the selected morphological characteristics of the trees.

The study was conducted on 8 plots in the Łupawa and Polanów forest districts (north-western Poland, on post-agricultural and forest land). In each plot, trunk and crown morphological characteristics were measured for all birch trees. The length of the knotless trunk was calculated, and wood quality was assessed based on the visible trunk and wood defects.

The results show that trees which grow on post-agricultural land are characterised by greater diameter of tree trunk and height, with similar slenderness ratio and shorter crowns and longer trunks. For the assessment of wood quality, trees which grow on post-agricultural land develop longer knotless trunks, which means a higher visual wood quality. It shows that the potential for afforestation of post-agricultural land by silver birch is high. These findings highlight the importance of reestablishing afforestation on post-agricultural land, which could have beneficial implications from both a societal and wood production standpoint.

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