



## Review

## Forensic botany: who?, how?, where?, when?

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## ABSTRACT

Plants are a good source of biological forensic evidence; this is due to their ubiquity, their ability to collect reference material, and their sensitivity to environmental changes. However, in many countries, botanical evidence is recognised as being scientifically. Botanical evidence is not mostly used for perpetration, instead it tends to serve as circumstantial evidence. Plant materials constitute the basis, among others, for linking a suspect or object to a crime scene or a victim, confirming or not confirming an alibi, determining the post-mortem interval, and determining the origin of food/object. Forensic botany entails field work, knowledge of plants, understanding ecosystem processes, and a basis understanding of geoscience. In this study, experiments with mammal cadavers were conducted to determine the occurrence of an event. The simplest criterion characterising botanical evidence is its size. Therefore, macroremains include whole plants or their larger fragments (e.g. tree bark, leaves, seeds, prickles, and thorns), whereas microscopic evidence includes palynomorphs (spores and pollen grains), diatoms, and tissues. Botanical methods allow for an analysis to be repeated multiple times and the test material is easy to collect in the field. Forensic botany can be supplemented with molecular analyses, which, although specific and sensitive, still require validation.

## 1. Introduction

Plant material is an excellent source of forensic biological evidence in the justice system [1,2]. This can be attributed to the fact that individual plants tend to be fixed to one point. Furthermore, plants are ubiquitous, enable the collection of various reference material (e.g. herbarium, seed bank), and exhibit physiological responses that are reflective of the environmental conditions they inhabit. Modern technology offer more possibilities with regards to the analysis of an array of biological material, including taxonomic classification based on various characteristics, from macroscopic ones to those that are microscopic, and molecular detection. The identification of biological material, which has become a key aspect of court proceedings, currently depends equally on an expert witness's experience and competence and the continued development of forensic examination techniques [1–11].

This article highlights the possibility of using botanical materials in forensics with certain court cases used as examples. Literature that forms the basis of this study was selected through a traditional library query (books) and a literature search via online databases, mainly PubMed and Scopus. Literature items were searched based on appropriately selected phrases in the title, abstract, or keywords, in accordance with the diagram shown in Fig. 1.

## 1.1. Scientific evidence in a court

Attempts to use scientific evidence in courts, including botanical material, had already been undertaken by the end of the 19th century. These early efforts are captured by an Austrian criminologist, Hans Gross, the author of the textbook *Handbuch für untersuchungsrichter als system der kriminalistik*, which was written in 1893 [12]. Locard also described several criminal cases using botanical evidence [1]. In 1908, by comparing the botanical material from a victim's body, mud from the shoes, and vegetation from the crime scene (i.e., hawthorn and myrtle leaves, grass glumes, birch buds, and a bryophyte thallus), he linked the victim to the suspect. In another case, based on microscopic analysis, he confirmed that on the knife of a person suspected of cutting a large number of hops in one plantation, there were cells of this plant. One of the first highly publicised cases in which botanical material was used in an investigation was the case of the disappearance and murder of a child of the famous American pilot, Charles Lindberg [13]. Nonetheless, the way to recognise botanical material as fully admissible evidence in a case was long. Evidence, including botanical evidence, can be admitted as scientific evidence after it has met the relevant criteria, otherwise, just as in the case of Frye in 1923, such material cannot be admitted by the court (*Frye v. United States*, No. 3968, (D.C. Cir. 1923) [14]. The standards for the scientific nature of evidence/theory developed by the

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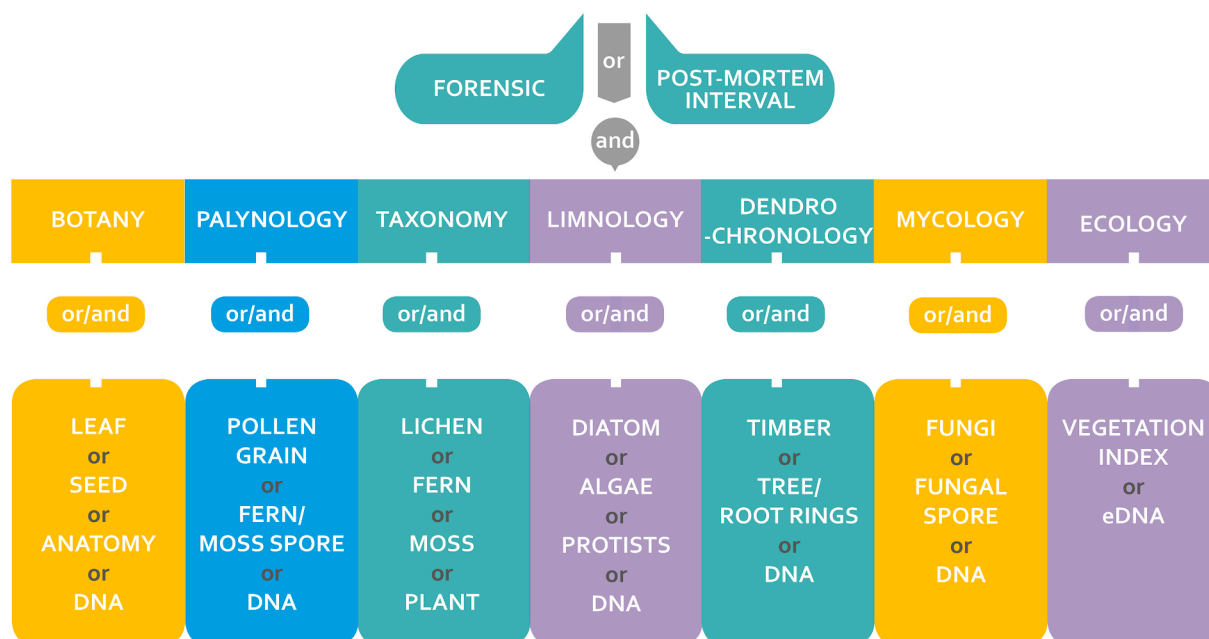
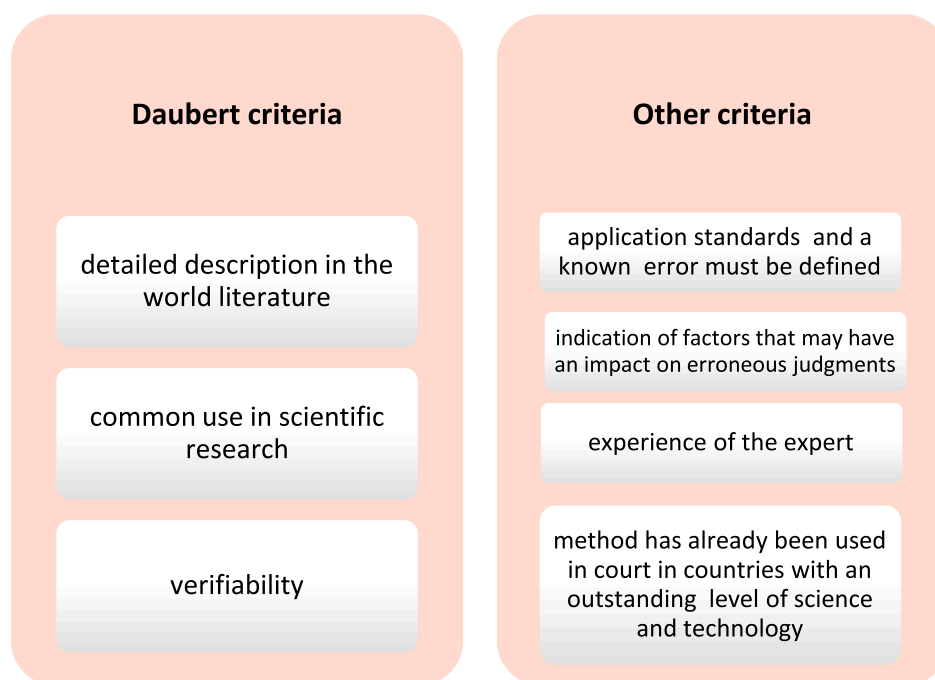


Fig. 1. Scheme of literature query in available databases.

Table 1

The criteria for recognizing scientific material or method as evidence in the court [6,15–17].



U.S. Supreme Court in 1993 (Daubert vs Merrell Dow Pharmaceuticals Inc.), as well as an additional requirement, are presented in Table 1. It should be emphasised that it is the court, not an expert witness from a particular field, that must make an independent assessment of the value of the evidence or examination technique and assess its suitability for specific court proceedings.

### 1.2. Forensic botany and botanical evidence

Forensic botany is an interdisciplinary science that combines an array of disciplines concerned with plant and fungal materials [2,13]. The application of this field within forensic science is rooted in an in-depth

understanding of flora (i.e., anatomy and physiology), as well as a broad knowledge of, among others, environmental biology, ecology, physiology, geobotany, geography, and geomorphology; but, above all, professional experience [5,18]. The potential of a more frequent application of botanical evidence (i.e., as scientific evidence), within the context of court proceedings, is high [4]. Botanical evidence can provide the basis for linking a suspect to a crime scene. It can do this by allowing for a linking objects found at an incident scene to a victim or suspect, determining the place and time of an incident, confirming or disconfirming an alibi (i.e., reducing a list of suspected persons), the “travel histories” of objects or people, the place of stay before death, determining the origin of food/drugs, etc. Throughout these applications, the analysis of

**Table 2**

The procedure with botanical evidences at the crime scene [3,5,6,19–21].

Report from the crime scene (also documented by photos)	<ul style="list-style-type: none"> <li>•the date and time, geographical coordinates,</li> <li>•a description of the landscape, dominant land use, land topography,</li> <li>•a type of soil/ soil disturbance,</li> <li>•a type of habitat,</li> <li>•the detailed description of vegetation,</li> <li>•any transient physical characteristics of plants (smell, color, degree of wilting).</li> </ul>
Collection of the evidences	<ul style="list-style-type: none"> <li>•should be sought in all possible places at a crime scene (e.g. soil, water), on objects, human body (e.g. stomach, upper airways, fecal matter),</li> <li>•control samples should be collected from the surroundings.</li> </ul>
Transport to the laboratory	<ul style="list-style-type: none"> <li>•appropriately packed (paper packaging; sterile conditions are often required) to minimize (cross)contamination,</li> <li>•each sample should be described in detail,</li> <li>•when collecting material from water, samples should be taken from its surface and from the bottom as well as disturbed water should be sampled.</li> </ul>



**Fig. 2.** Plant materials used in forensic (mentioned, among others, in [2,3,5,10,13,18,23,28–40]).

botanical evidence can be performed via a classical analysis of morphology and anatomy, or in some cases, via chemical and molecular screening techniques [5,10,17].

A detailed and clear expert report (i.e., regarding botanical evidence) is the main basis of its use in court proceedings. Specifically, the expert report should be detailed and supported by photographic documentation, especially when colour is a diagnostic feature relevant to a particular case. Additionally, evidence material should be sought in all

possible places at the scene of an incident, as well as on objects and a body [5,19,20]. The plant material should not be contaminated; therefore, special caution should be exercised during its collection. Subsequently, the material should be appropriately packed, described, and transported [3,6,21] (Table. 2).

Despite botanical materials and methods described in this paper having diagnostic value, in many countries they are not considered scientifically rigorous enough to be frequently applied (i.e., as a

standard approach) in court trials [17,22]. Their value is usually determined individually for a specific case and a decision concerning the admissibility of such evidence lies with the respective judicial body. Most frequently, botanical evidence is not the evidence of perpetration but serves as circumstantial evidence [22].

The simplest criterion for characterising botanical evidence is its size. Macroremains include whole plants and large fragments [4], whereas microscopic evidence includes palynomorphs, protists, diatoms, and plant tissues (Fig. 2) [5,18,23–25]. An excellent complement to traditional botanical methods is the combination of several analytical methods rooted in science (e.g., DNA analysis, toxicology, isotope analysis, spectroscopic methods, and machine learning) [10,17,23]. Scientific knowledge within the context of plant physiology, ecology, and geoscience suggests that it is worth applying to a greater extent and frequency in court cases [26,27].

## 2. Plant identification

The basis of a court botanist's work is the ability to identify plants [4]. Aquila et al. [51] described a case in which a victim was linked to a crime scene based on the occurrence of the species *Xanthium orientale* subsp. *italicum* on the victim's clothing. The botanist's efforts are particularly useful for identifying plants suspected to cause serious or fatal poisoning [2]. For example, confirmation that a plant is poisonous allows for the implementation of a range of medical procedures necessary for an affected human to survive. This knowledge can also be applied towards detecting illegal plant imports and properly identifying psychogenic or narcotic species [52,53]. It is worth mentioning that not all *Cannabis sativa* subspecies contain strong psychoactive substances and are narcotic. A good florist will identify plants that are not native to a given region's flora, ornamental plants, and those planted in urban green areas or home gardens. This skill was used in Taiwan to establish that a woman who had been found lying in a gutter had not been hit by a vehicle, but actually committed suicide. Fruit and stem fragments of a plant belonging to the genus *Solanaceae*, alien to the flora of the region, were found in her hair. Owing to a detailed study of the surrounding area, it was discovered that this plant was growing in one balcony of a high-rise building and that it was *Solanum nigrum* L. Therefore, the woman most likely fell from the building, with her body coming into contact with the plants as she fell [54].

In their examination efforts, botanists primarily focused on seed plants, but the lack of qualified specialists limits the possibility of using other plant groups in forensic science, such as hepatics, bryophytes, lichens, lycophytes, ferns, and horsetails. Although fungi are not phylogenetically related to plants, they are the subject of their study [4].

Some plants or fungi are characterised by specific habitat requirements; therefore, their presence in evidence can be an important clue regarding the location of a specific incident [5]. Mosses are good scientific evidence [4]. They easily attach to footwear and can remain there even after several hours of walking on dry and hard surfaces [55]. They can be useful in determining the time an act was committed or the time that would have passed between death and the body being found. Mosses with monopodial growth, where the apex of the stem continues to grow each year, are applicable in such cases because they allow for an analysis of the number of annual growth rings of the roots [28]. Not only *Bryum kapilarna* Hedw. and *Hypnum cupressiforme* Hedw. are characterised by this type of growth, but they are also ubiquitous, making them particularly useful in estimating the post-mortem interval (PMI). Lancia et al. [56] described a case in which the PMI of human skeletal remains was determined based on the growth rate of *Lepidodictyum riparium* as well the growth of monopodial moss. This botanical evidence supports the morphological and anthropological results of the forensic investigations. The authors underline that the analysis of bryophyte growth allows the determination of only the minimum time of death and not the correct time because mosses do not inhabit soft tissues, and that the qualitative spectrum of lichens occurring at the

scene of an incident can answer similar questions.

García et al. [29] indicated the potential of lichens as botanical materials in forensic science. Specifically, lichens can colonise bones (e.g. *Psiloparmelia distincta* and *Parmelis sativa*), with many of them growing annually if they have access of light and thus they can help forensic experts in answering the question 'when?'. According to Olech [57], the growth of lichens on bones is associated with microclimatic conditions and the retention of nutrients in the pores of the bone surface. *Thelenella antarctica*, *Lecanora antarctica*, and the genus *Caloplaca* sp. occur most frequently on the bones of cetaceans. Examples of lichen species that can be found on bones have mainly been recorded in environments such as the Arctic and Antarctica [57,58]. Lichens living in such environments are dependent on the substrate to a small extent, which allows them to colonise a bone substrate using strategies similar to those employed in colonising rocks.

Botanical evidence does not necessarily have to be physical. It can also be a visible, unambiguous reaction of a human or animal which is induced by previous contact with a plant or fungus, and many plants produce secondary metabolites that irritate the skin or eyes, inducing visible changes [3,6]. An example is a nettle that causes characteristic changes in the skin. Contact with Sosnowsky's hogweed is dangerous. Cases of phototoxic activity of many plants are known, including those of the families *Asteraceae* (chrysanthemum, dahlia), *Apiaceae* (celery, angelica), *Liliaceae* (garlic), *Hypericaceae* (St. John's wort), *Moraceae* (fig), and *Rutaceae* (citrus fruits). A reaction to contact with allergenic plants or plants with prickles or thorns is usually easy to notice [59].

The identification of a plant based on morphology is an efficient method of when performed by an experienced botanist. It is also inexpensive because it does not require expensive equipment or reagents, and samples identified in such a manner are less prone to contamination [4,6]. However, in many cases, the assessment of morphological features is not sufficient to determine the species or subspecies of a plant with certainty, and such information is often crucial in court proceedings. The accuracy of botanist identification can be confirmed by DNA analysis [10].

The method of collecting plant material from a crime scene for further DNA analysis is different from that used for generally collecting plant material. Within the context of DNA analysis, not a lot of material is required, but it must be well-protected against contamination with foreign genetic material. The approach to storage is also different. Samples can be placed in liquid nitrogen, a freezer, or silica gel. In this form, the material can be stored for a long time without being degraded, and plants stored in the herbarium are exposed to degradation by moulds, bacteria, and insects [30,60].

The development of DNA sequencing technology has created opportunities for a rapid and accurate identification of botanical evidence at the species level; a level of precision that is needed by forensic experts [60]. There is no ideal region for DNA barcoding. For plants, the four regions of plastid DNA for barcoding were used: ITS2, matK, rbcL, and trnL, as well as nuclear NDA, ITS, and ITS2 regions. The rbcL + trnH-psbA and matK regions are recommended for global plant databases, and matK, ITS2, trnL-F and rrn18 as alternative or complementary regions [30,60,61]. It should be emphasized that still more research is needed to validate and apply DNA barcoding in practice. Trace evidences from the crime scene are very often degraded or contaminated, hence the detection and identification of DNA may not give sufficiently reliable results, so in such cases, microRNA sequencing may be an alternative. It should be emphasized there is a need to elaborate validations of DNA barcoding techniques, in particular to ensure that protocols are robust enough for court procedures [10,60].

## 3. Plant remains

Forensic experts process plant fragments much more frequently than whole specimens [1,18,31,53,62–64]. With regards to the fragments, determining their parent plant is difficult depending on the size of the

material and the degree to which it is degraded [53]. Hall [32] provides examples when leaf fragments are scientific evidence in court cases, providing the basis for linking the perpetrator to the crime scene or the victim. In one of these cases, sticky *Petunia* leaves and flowers that had been found on the victim's hair and clothing were sufficient to link the perpetrator to this particular crime. A highly experienced botanist can identify plant material in vomit, stomach, intestines, or faeces [33,53,65].

### 3.1. Bark

Tree bark, since it does not degrade easily (i.e., bark can be easily stored without degrading), increasing its potential within the context of forensic evidence. Many of its morphological and anatomical features can be easily observed; and these features can be analysed multiple times [66]. Caccianiga et al. [34] described the bark of 16 tree species characteristic of Italy's dendroflora. Through these research efforts, they proved that the bark can be used as scientific evidence that enables a victim to be linked to a crime scene. In 2010, bark fragments recovered from the body of a man found in a forest in Italy were examined and the forensic experts, using a key prepared by themselves and reference material, identified it as *Robinia pseudoacacia* based on the phloem structure. This case confirmed that bark has a lot of potential within the context of species classification; however, bark reference collections remain scarce, whereas for forensic purposes, it would be important to have access to large collections of reference material that would include both native and exotic taxa [34].

### 3.2. Seeds and fruits

Research findings in the field of carpology have also found application in forensics. Experts in this field can match seeds/fruits to the parent plant species based on many characteristics. The diagnostic features that are used include morphological, anatomical, and organoleptic characteristics. Visual identification is based on the analysis of shape, colour, sheen, sculpture, and size [4]. Owing to optical tools, diagnostic features can be observed at a fine-scale, and these features include: walls of the fruit and seed coat, presence of oil cavities, and structure of the endosperm or cotyledons [67]. Seeds of plants of the family *Fabaceae* have a scar after the funiculus attaches a seed to the pod [13]. Fruits and seeds can also be distinguished by their flavour or smell (e.g. *Apiaceae* fruits).

Anemochorous seeds/fruits have structures that allow them to float in the air (seed fluff in *Taraxacum* sp., *Populus* sp.). Zoochorous species have structures that enable their attachment to the body of an animal [67]. Some seeds cling to a fabric so strongly that they remain on it even after washing; this allowed for the perpetrator of the murder of two children in 1997 to be identified [63]. Their bodies were found buried in a shaded wooded area near a cemetery, not long after they had been reported missing by their stepfather. When he became a suspect, biological material was retrieved from his clothing, including the seeds of *Geum canadense* Jacq. and *Galium aparine*. Both species were found at the gravesite, but they did not grow near his house, contrary to the suspect's explanation. The seeds were part of the evidence pointing to him being the perpetrator of the murder.

The footwear and wheels of a vehicle are good seed-dispersing vectors because seeds effectively attach to them, which provides the possibility of using seeds to determine the place where people or objects travelled [63,67]. This possibility was confirmed by a case conducted by Lipscomb and Diggs [13]. In 1995, they were requested to identify plant material in the case of abduction and sexual abuse of a two-year-old child. Owing to their expertise and experience, they identified the seeds recovered from the shoelaces of the suspect and linked them to the place where the child had been found. They established during the first examination that the seeds belonged to the family *Apiaceae*, whereas further examinations, including comparison with herbarium material,

allowed them to identify the species as *Torilis arvensis*, species native to southern Europe, and invasive to central and western Europe, and North America. Seeds of this plant, which exhibit ectozoochorous seed dispersal, are densely covered with hooked bristles that cling perfectly to the body of animals (fur, feathers), as well as to human clothing and footwear. Despite this precise identification, this was not the decisive evidence in this case, but was important enough to provide the basis, in conjunction with other evidence, for convicting the suspect. The authors of this paper indicate that many other plants can be similarly used as scientific evidence in court proceedings. For example, they provided grass species common in North Central Texas, USA, such as *Cenchrus*, *Nassella leucotrcha*, and *Stipa leucotrcha*.

Soil is an environmental element that is often collected by forensic teams. Samples were examined for seed bank content. Knowing the soil seed bank composition can help determine the scene of an incident, because there is a relationship between vegetation composition and soil seed bank composition. Nonetheless, this is not an ideal relationship because not all seeds are included in the seed bank, but the seed assemblage is also dependent on the seasonality, weather conditions, and animal activity [68]. However, the presence of seeds of rare, protected, or invasive plants is an important indicator of a given location and has great value for forensics [34,67,69]. The ability to discriminate between seeds is important when the stomach content is examined because some seeds remain intact in the alimentary tract for a long time. In 2001, an analysis of the stomach content of a dead child for the occurrence of biological material revealed the presence of seeds of the strongly toxic Calabar bean (*Physostigma venenosum* Balf.), whose seed coats have a characteristic appearance. The seed walls of this plant are thick and resistant to digestion, and they pointed to the possibility of the child having been poisoned [35].

In recent years, laboratories have begun to use specific DNA markers to identify seeds to species level. For example, within the context of seed identification, q-PCR has been shown to be an effective technique. One difficulty, however, is the extraction of DNA from seeds; it is often labourious and involves many steps that can also result in the loss of DNA as well as the introduction of impurities [70].

### 3.3. Litter

Plant litter has attributes that highlight its potential usefulness within the context of court proceedings. For example, it consists of a diverse range of biological material such as leaves, fungi, bark, plant pollen, and animals [36]. Leaves are identified based on their venation, shape, petiole, and leaf edge [4]; however, those found in plant litter are at varying stages of decomposition, which hinders their identification. Therefore, it is necessary that an expert witness not only has an extensive experience in plant identification, but also has access to a good reference herbarium. The analysis of leaf litter provided an important clue in searching for a missing person in the Sierra Nevada Mountains in 2002. Based on the qualitative and quantitative spectrum of identified leaves, the colour of the leaf litter, knowledge of the environmental requirements and the area of occurrence of the identified species, the leaf litter found in the murder suspect's car were linked to the altitude and slope at which the murder occurred [71].

### 3.4. Timber - dendrochronology and dendrochemistry

Forensic dendrology is a rapidly developing field of research. The ability to distinguish species based on analysis of wood alone is difficult, and hence, a full array of methods is used, from conventional macroscopic evaluation through microscopic methods (e.g., wood anatomical structure), and chemical or molecular methods. These methods differ significantly in terms of their required time investment and accuracy of the analyses [7,37,72–75]. Experts are assisted by modern automated sample identification techniques based on state-of-the-art IT solutions, that is, deep learning and, more specifically, Convolutional Neural



Networks (CNNs) [9]. Hermanson and Wiedenhoft [76] applied automated analysis (“machine vision”), and owing to the application of advanced image capture and processing algorithms, they were able to identify plants to species level using wood samples.

Wood was used as scientific evidence in criminal cases at the beginning of the 19th century. Arthur Koehler, an outstanding wood expert, proved that a wooden box in which a bomb had been found was made by analysing elm wood chips from the suspect’s carpenter workshop. His expertise and experience were used in one of the most famous cases concerning the kidnapping and murder of a child, the Lindbergh case of 1932. His task was to examine the wood from a ladder that had been used during kidnapping. By comparing the arrangement of the annual growth rings in the ladder, he showed that the ladder had been made using, among others, a flood board from the attic in the house of the person suspected of having committed kidnapping. Moreover, based on the anatomical structure of the wood, he demonstrated that the ladder was made of wood of gymnosperms, that is, North Carolina pine, ponderosa pine, Douglas fir, and birch [12,77].

Many tree species are included in the list of protected species that are prohibited from trade under the provisions of national and international treaties (CITES). The identification of species, particularly trunks devoid of leaves, frequently poses a difficulty in stopping illegal timber trade [72]. In forensics, conventional trees are identified based on their physical characteristics, such as colour or hardness, the anatomical structure of wood (sapwood and heartwood), pores, medullary rays, as well as an analysis of annual growth rings [78]. For proper species-level identifications using wood, it is necessary to refer to the reference material. Wood is a collection of heartwoods that are curated in xylaria, and their list (*the Index Xylariorum*) is kept by the Kew Gardens [37]. According to the forensic recommendations of the United Nations Office on Drugs and Crime (UNODC), it is recommended that the analysis of wood is used as a primary method, and only after such an analysis has been performed can one decide to use other methods [73,74]. Knowledge of wood anatomy can be useful for the forensic identification of charcoal originating from illegal logging. An experimental study carried out by Braga et al. [79] revealed that numerous elements of the wood anatomical structure are well-preserved in charcoal. In species identification, wood-linked diagnostic features include vessel groupings, vessel-ray pitting, perforation plates, axial parenchyma, ray cellular composition, storied structure, and secretory elements. The creation of a database with charcoal anatomy images for specific tree species could be a useful tool for institutions that supervise trade in valuable tree species [79,80].

Dendrochronology is a field that can help forensic science answer the question ‘when’, among other questions, including PMI determination [16,75]. In climatic regions with distinct seasons, the trunk and roots of woody plants are characterised by annual growth rings. Spring and autumn wood, produced in the second half of the growing season, can be distinguished in each growth ring. In spring, wood growth is more intense, and vessels transport water intensely owing to their large diameter. Vessels formed later predominantly perform mechanical functions. The width of the growth rings is also a reflection of prevailing environmental conditions, including deviations from these conditions [5,75]. The basic analysis of root and trunk growth in trees does not require complicated equipment and is not costly. To correctly determine the results based on the number of growth rings, a large piece of wood is necessary, preferably one containing a sequence of growth rings from the bark to the heartwood. Obtaining such a large sample can often be destructive to evidence materials. The precise determination of wood age depends on tree species [18]. Some tree species are characterised by inconsistencies in the sequential formation of growth rings; therefore, an appropriate database of specific wood samples of the same or related species from the same region or habitat must exist for comparison. These samples formed a reference chronology. Dendrochronology is also used in the case of roots that may grow above a buried body or change the growth direction, and each disturbance is visible in the growth rings; in

this way, it can be helpful in determining PMI. Pokines [81] noted that it is worth analysing the annual rings of not only the main root. However, even small roots can provide further evidence. Root dendrochronology is particularly useful when other biological methods such as forensic entomology cannot be applied. Trunk dendrochronology can also be useful in the case of illegal trade of valuable and critically endangered trees and illegal tree cutting, as it allows for a determination of the provenance of a tree [37]. An example of the use of this method to prove illegal cutting of *Pinus nigra* and *P. sylvestris* in Turkey was provided by Yaman and Akkemik [82]. The most frequently smuggled timber is of tropical origin, where there are no distinct seasons and therefore no distinct growth rings, greatly limiting the application of dendrochronology within the context of forensics [37].

The literature on the subject provides numerous examples of the application of dendrochronology to identify many wooden objects of historical value, among others violin artwork. Some of the world’s best violins are antiquities worth millions of dollars, and the theft of violin artworks or their forgery are offences, resulting in losses estimated at billions of dollars. The value of an instrument is determined not only by the skills of the instrument maker that has made it, but also by the quality of the wood used [83]. Dendrochronology finds good application in determining the age of the wood used to make an instrument, but unlike wood dating by the commonly used radiocarbon method, dendrochronology does not require taking wood samples from an instrument. Age can be estimated based on a comparison of the growth ring patterns with the patterns of other previously well-dated objects or trees growing in a particular area if we are certain where the trees used to make an instrument grew in a given region [84]. In Italy, spruce forests, commonly found in this country, were the main source of wood for workshops of well-known 17<sup>th</sup> and 18<sup>th</sup> century violin makers. Therefore, Tophan and McCornic [84] prepared a long reference sequence of growth rings and determined the age of 33 instruments with more than 60 % accuracy, including those manufactured in Stradivari’s workshops. However, it should be emphasised that one cannot determine unambiguously when an instrument is made because instrument makers could have used wood stored for many years or the outer rings could have been removed during wood treatment [83,84].

In forensic science, not only the number of growth rings can be used as evidence, but also their chemical composition, which is why dendrochemistry is not only a supplementary method, but also provides the possibility of answering more detailed questions [16]. It uses instrumental analysis, that is, mass spectrometry, near-infrared spectroscopy, stable isotopes, and radio-carbon, to address this issue [7,72]. The content and ratio of stable isotopes in a plant are correlated with its place of origin or cultivation, which is important from a forensic point of view for detecting the smuggling of protected plants [72]. Most often in plants and their products, secondary metabolites and isotopes of hydrogen, oxygen, carbon, and nitrogen are studied using continuous-flow isotope ratio mass spectrometry (CF-IRMS), which is considered highly accurate and sensitive [7]. An important application of forensic dendrochemistry is to solve problems associated with anthropogenic contamination, in particular, to determine the source of contamination, as well as the time and place of its emission [16]. Balouet et al. [75] described an example of the application of dendrochronology and dendrochemistry in courts to determine the time of occurrence of contamination of a property by trichloroethylene. The land in question was owned by two owners in succession and either of them claimed that the contamination had not occurred during his tenure. The analysis revealed asynchronous increases in the chlorine content of the tree rings which had occurred during both the first and second ownership. Based on the dating of the atypical increases and the determination of the chlorine concentration levels, it was possible to appropriately allocate the respective owners’ contributions to the cost of remediation of this land. Such methods can be used as independent scientific evidence or to supplement other evidence [16].

### 3.5. Plant anatomy

Dilcher [38] described cases in which the police used their knowledge of plant anatomy and morphology. In one case, the police asked for an expert report on the plant material retrieved from the trachea of a boy who, as his parents claimed, had fallen into a large stack of hay and suffocated. If this had happened, fragments of hay should have been in his airways – stems and leaves. The researcher did not find any fragments originating from grass but discovered the presence of corn starch grains. Corn bags were found at the scene of the incident, and hence, a presumption was made that the boy had them on his head right before his death. In another case, an expert witness was provided with crushed plant material and requested to confirm whether it was marijuana. The examiner selected the leaf fragments from the plant material. They are thin and contain numerous trichomes. He examined the cuticle in detail and compared the obtained results with the reference material, that is, *Cannabis* leaves kept in a herbarium. In effect, he rejected the thesis that marijuana was the plant material presented for analysis. Leaf epidermal features can be considered taxonomic for many plant species. In species of the family Piperales, some can be discriminated based on trichome structure as well as the types of pores and their arrangement. On this basis, it can be confirmed that a species with medicinal properties is sold [85]. Woodberg et al. [8] confirmed that knowledge of leaf micromorphology could be useful in the fight against illegal trade in plant species threatened with extinction. They demonstrated that protected cycad species can be identified based on the structure of the trichomes and stomata (density and dimensions of the stomatal band).

Trichomes are epidermal outgrowths [4] characterised by diversity of shape (e.g. glandular, peltate, hooked), structure (uni- or multicellular), and function (e.g. secretory, transpiration reduction), and sometimes they can have taxonomic value, for example, in the case of species of the genus *Ficus*. Bates et al. [86] describe a study analysed the botanical material found in the wreckage of an aircraft. The material was examined for the presence of trichomes. Hairs of plants from the families *Solanaceae* and *Malvaceae* were shown to be present in the engine, but a detailed analysis negated the premise that the penetration of plant material into the engines was the cause of the crash.

Cells of some plant species contain inclusions. Due to excessive mineral uptake by plants, mineral deposits called phytoliths are deposited in cells or intercellular spaces. These are calcium oxalate or silica (opal) crystals. In grasses and sedges, they are primarily deposited in epidermal cells, and in the palms and orchids in cells surrounding vascular bundles. Their size is several tens of micrometres. In many cases, phytoliths have diverse shapes with taxonomic values. They can be roundish, elliptical, longitudinal, rectangular, or saddle- or dumbbell-shaped. Due to the above reasons, investigation of soil and plant tissue content for the presence of phytoliths has application potential in forensic science [62,70,87].

Jakoby et al. [10] (2021) indicate the possibility of identifying plant genera based on the anatomical structure of the root, at the same time stressing that the procedure of sample preparation for analysis is time-consuming and identification itself requires experience. In their research, they focused only on unique morpho-anatomical features for a particular genus, i.e. root color, periderm structure and xylem location. On this basis, it was possible to identified root samples of *Pinus*, *Pistacia*, *Cupressus* genera. Analysis of fiber structure and secretory cavities, together with molecular marker analysis, requiring less botanical experience, enabled the identification of forensic samples to the species level.

In forensic procedures, remains of food found in vomit, faeces, and the digestive tract are often collected during autopsies [65,88,89]. In some cases, the food can also be found in the respiratory system (e.g., choking) [90]. Forensic scientists are interested in qualitative (i.e., what the victim ate) and quantitative aspects of a victim's food intake [65]. Larger fragments of plant tissues (e.g., parenchyma) are most often found in vomit and stomach contents, whereas in the gut (ileum and

colon), microremains with thick cell walls, such as fungal spores, plant pollen, fibres, and protoxylem elements, are preserved. These can be easily identified using a light microscope [53,89]. In a case described by Azzalini et al. [91], an autopsy showed the presence of toxic oleandrin and oleander leaf tissue in stomach. Since an accidental ingestion of an oleander leaves is unlikely; the presence of leaves in the digestive tract was indicative of self-poisoning. In another case, the presence of old grains was detected during an autopsy of the lung bronchi. Microscopic observations revealed that they originated from legumes [90].

As mentioned earlier, the quick identification of the factors causing serious poisoning is crucial for saving lives. In many cases in which it is not possible to identify a plant by its morphology or anatomy, poisoning is confirmed by the examination of toxins using chromatographic methods, such as liquid and gas chromatography [91]. For samples with different compositions, the use of these methods would require tedious purification procedures to remove interfering compounds and improve detection sensitivity. For such samples, qPCR can be used; the effectiveness of this method has been confirmed for the *Nerium oleander* plant, which is highly poisonous to humans and animals. Specifically, qPCR can facilitate the detection of this toxin in feed and food mixtures, even at low DNA concentrations. The suitability of the method for etched samples was confirmed, and the detection limit was 0.1 % for oleander in mixed samples [39]. This method was also used to distinguish between edible and poisonous plants by examining DNA from leaves and from samples digested in artificial gastric juice [92]. Research by Kim' [92] and Bai' [39] recognises q-PCR as a highly sensitive and rapid method which may be applied in forensic botany to prove poisoning.

### 4. Plant physiology

Plant fragments torn from their natural habitat undergo a series of morphological and physiological changes. Even if destroyed, these fragments can still be valuable in allows us to address questions relevant to forensics. However, the use of plant fragments tends to require a set of experiments; for example, to check how much time it takes for a broken branch of a specific species to wither or how quickly leaves of a particular species change their color if buried [32,51]. Palermo et al. [93] conducted a simulation of the changes of a leaf in a victim's hand. The experiment was conducted using leaves of several species, and half of the leaves were covered with aluminum paper, whereas the other half were adequately illuminated by sunlight. Each species was characterised by a different rate of chlorophyll reduction in the leaves, and the experiment demonstrated that, on this basis, it can be determined what time has elapsed from the detachment of a leaf from its parent plant or how many days it was in the dark. A similar experiment was described by Hall [32], who proved that lawn yellows and severe chlorosis occurred after a week of complete absence of light. Lichens can also change their colour under adverse conditions. Hawksworth and Wiltshire [41] performed an experiment using the cosmopolitan lichen, *Xanthoria parietina*. A well-illuminated thallus is yellow/orange with deeper orange sporophores; however, if buried, it changes its colour to green.

### 5. Forensic plant ecology

Knowledge of vegetation, plant community ecology, plant populations, and mechanisms affecting its changes has been used many times in forensic proceedings [94,95]. Easily noticeable changes depend on the growth cycle, which depends on the annual cycle [88]. The achievements in phenology (autophytophenology and synphytophenology) and knowledge of individual phytophenophases have been applied here [94,95]. Knowledge of the timing of some phenological phases and their sequence can help determine the date of an incident [88] and an example of application of knowledge regarding the emergence of seedling cotyledons and leafing is described by Hall [32].

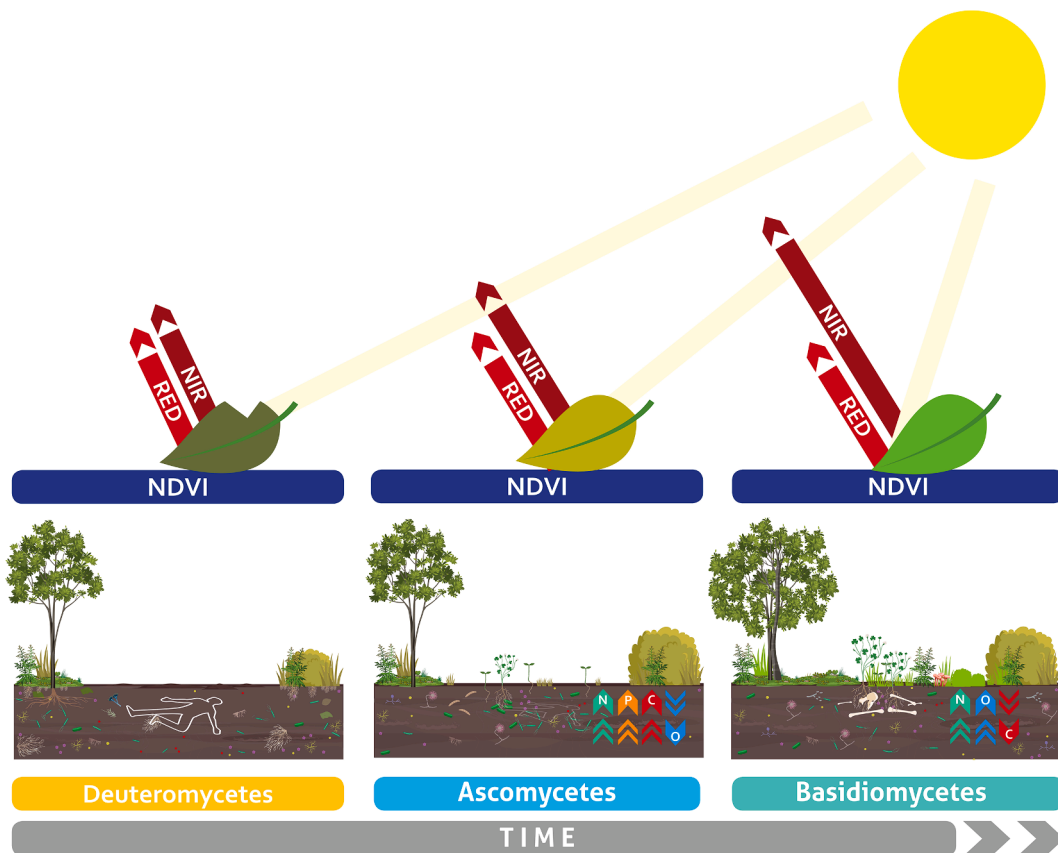


Fig. 3. Succession of vegetation growing on body decomposition island (based on [5,11,21,41–50]).

Phenology can also be used to prepare against bioterrorist attacks, which involve the purposeful introduction of crop pathogens and useful or forest-forming plants. Therefore, it is of key importance to determine the critical time in the growth cycle of plants when they are particularly exposed to pathogens, and to develop a model simulating infection spread time [96].

One of the easily noticeable changes that occur in the environment is the quantitative and qualitative change in a species assemblage. This can be noticed much more easily in plants and fungi because it is more difficult for them to avoid environmental stress. Bioindicators of environmental quality (soil and air), including contamination with heavy metals or radioactive contamination, are excellent examples of this. Lichens are the best bioindicators of air pollution [97,98]. For forensic experts, the analysis of the lichen biodiversity in crime scene can be used to confirm environmental pollution. A serious drawback of this method, however, is that the results of such tests are largely non-specific and cannot be attributed to a single pollutant with certainty [97]. The lichen transplantation method can also be employed to confirm pollution. Purvis et al. [40] used *Hypogymnia physodes* as a bioindicator. After 3 months of lichen exposure experiments, using inductively coupled plasma optical emission spectrometry (ICP-OES) and mass spectrometry, they detected the contamination of lichens by Cu. This method is discrete and relatively cheap, enabling the determination of pollutant deposition over short periods. Provided that there is sufficient background research and field and laboratory research procedures strictly adhered to, this can be a valuable qualitative tool in environmental forensics.

In cases of murder, war crimes, or mass murders, it is of key importance to find bodies. Hiding a corpse interferes with the environment and leaves traces. These traces can be manifested in the form of soil disturbances, root destruction, changes in plant growth orientation, etc. Fig. 3. During the decomposition of a human body, a specific ecosystem

forms, called a cadaver decomposition island. Anomalies in vegetation may include differences in species composition, which can also be seen in comparison with the surrounding undisturbed vegetation [5,21,42,43,99,100]. Changes in colour, density, and phenology can also be observed. Heat production and moisture release are short-term factors, whereas the release of nutrients from decomposing bodies, as well as better soil aeration and soil loosening during digging, have long-lasting effects [43]. The response of plants to serious environmental changes is conceptually well described; however, in practice, different plant responses are difficult to interpret. Plant growth is affected by the depth at which a body is buried, soil properties, and climate characteristics [21,42–44]. The timing of body deposition may also play an important role, especially in regions with distinct climatic differences [5,21]. Experiments have shown that the rate of decomposition of a human body is different and more variable than in the case of model animals; therefore, distinguishing human cadaver decomposition islands from cadaver decomposition islands of animals remains a challenge in practice [21,42,45].

During cadaver decomposition, an excessive outflow of biogenic elements (C,O,N,H,S, and P) into the soil results in increased chlorophyll production in plants, which causes a change in the spectrum recorded by a satellite. Vegetation, which has been considered an obstacle in finding corpses outdoors, can now become an advantage owing to remote sensing methods. Brabazon et al. [46] recommend the use of Unmanned Aerial Vehicles (UAVs). Technologies are becoming increasingly cheaper and more versatile owing to the effective use of biochemical, thermal vision, and hyperspectral sensors that allow the mapping of anomalies on the soil surface as well as in green vegetation and tree crowns. Using drones with modern technologies is particularly useful when graves are suspected to be located in difficult-to-access places, when search areas are vast, or where there are military conflicts [46,99,100].



Forensic ecology is still a developing field of knowledge, and there are doubts regarding its potential. Such expert reports have been used in the British court system; however, unlike many areas of court medicine, the procedures and protocols are not standardised [5].

## 6. Forensic mycology

### 6.1. Fungi

Mycological achievements are used more frequently in forensics. Fungal forensic evidence, similar to other botanical evidence, can help estimate the time and cause of an incident, link a suspect to a crime scene or a victim, determine their travel history, establish the causes of hallucination or poisoning, locate a buried corpse, establish that a biological weapon has been used, or provide evidence for smuggling or poaching [41,47,48,101–103]. Knowledge of fungi is important in the case of illegal trade in narcotic or hallucinogenic species, such as those of the genus *Psilocybe*, which have a wide geographic distribution [104].

Fungi occur almost everywhere, and they are often invisible to perpetrators who are frequently unaware that they have had contact with fungi. Fungi have very different environmental and nutritional requirements, and because of this, they are a good location indicator and tell much about the soil in which they live, such as in the previously described cadaver decomposition island [41,49,101,105].

#### 6.1.1. Fungi as indicators of burial sites and the time of death

Fungi are very good indicators of burial sites [49]. Two closely related chemoecological groups of fungi, ammonia fungi and post-putrefaction fungi, are indicators. Sporocarps have been observed in different forests worldwide and frequently mark grave sites. These groups of fungi provide visible markers for sites of cadaver decomposition and follow a repeated pattern of successional change as cadaver decomposition progresses: Deuteromycetes, Ascomycetes and Basidiomycetes. Ectomycorrhizal fungi occur in the final stages of cadaver decomposition. Species occurring in a specific phenophase also indicate burial sites, for example, common on many continents, *Hebeloma danicum* and *Laccaria bicolor*. They sporulate 1–4 years after the burial. At places where a corpse has been buried, other Basidiomycetes appear, including Agaricales of the genus *Hebeloma*: *H. vinosophyllum*, *H. spoliatum* and *H. radicosoides*. Fungi are generally geotrophic, but if their stalks are crooked or fruiting bodies are tilted, this indicates their mechanical disturbance or disturbance of the soil structure. We suggest that these phenomena can become useful tools for investigating crime scene [41,50,106].

Many studies have been conducted on the colonisation of livestock carcasses by fungi [50]. The most frequent model is the pig, but wild animals have also been selected for experiments, for example, the red-tailed hawk, mute swan, North American river otter, and bobcat. In the early stages of decomposition, *Filobasidium magnum*, *Trichoderma trimmonsii*, and *T. atroviride*, *Marasmius* sp. were observed, whereas *T. atroviride* and *T. simmonsii* were found on the skeleton of these wild animals. *Rhodotorula dairenensis* and *R. mucilaginosa* have only been isolated from bird skeletons. The author of an experiment [107] concluded that some fungal genera (*Filobasidium*, *Rhodotorula*, *Trichoderma*, *Umbelopsis*) could be considered important decomposers of wild animals. In the future, these discoveries may help establish living sites for illegally killed wild species populations.

It is also worth using forensic mycology when the time of death or deposition of a corpse is uncertain and fungal colonies are visible on human remains [108]. As proven by Hitosugi et al. [48], it is particularly important when entomological analysis cannot be applied and fungi can be treated as an independent line of evidence. In a case described by them, the presence of *Aspergillus terreus* and *Penicillium* sp. on the body of a deceased person allowed for a determination of the minimum interval since death. This is because it is known that after coming into contact with a deceased body, fungi of this genus develop colonies after 3–7

days. In their experiment on a mouse model, Metcalf et al. [109] also proved that microbial communities colonising a cadaver and their succession on decomposing remains can be successfully used to determine the PMI, and this method produces a smaller error than in the case of forensic entomology.

#### 6.1.2. Other applications of forensic mycology

Wet buildings and poor ventilation create specific ecological niches that promote the growth of fungi that are harmful to human health and life. These produce allergens, mycotoxins, and volatile compounds. This group of species includes, among others, *Apergillus* (*A. fumigatus*, *A. parasiticus*, *A. niger*, *A. clavatus*), *Penicillium*, *Cladosporium*, *Mucor*, *Trichoderma*, *Fusarium*, *Alternaria* (*A. alternata*), *Candida*, and *Serpula lacrymans*. The presence of *Stachybotrys chartarum* is evidence of the poor quality of buildings, and this fungus causes biodeterioration [110]. Analysis of fungal content in indoor environments can help to confirm the justification for health loss claims or claims for damage caused by a flood in a house or by a river flood [108,111].

Fungi can be dangerous tools for terrorists. Used as a weapon, they can be directed against people, livestock, or crops and can act directly through infection or indirectly through mycotoxins [112]. The use of plant pathogens as biological weapons can disrupt the economy of a given country. Among others, *Tilletia indica* and *Phakopsora pachyrhizi*, which cause diseases of the karnal bunt of wheat and Asian soybean rust, respectively, have potential in this respect [96,113]. Many fungi can produce toxins, and although most of them act over the long term, some can be mass-produced more quickly [114].

#### 6.1.3. Fungi DNA sequencing technologies in forensic

Fungal trace evidence is often small and degraded; hence, unambiguous taxonomic identification of fungal fragments based on morphological or biochemical features may often not be possible. Molecular methods, especially barcoding of the internal transcribed spacer (ITS) region, are often useful in circumventing such issues, as demonstrated by Lee et al. [115] in the identification of hallucinogenic mushrooms. The unknown sequence was compared with existing DNA databases, but very few fungi (<1%) have been sequenced for the ITS region. There are several selected databases dedicated to identifying ITS as well as other ribosome and protein coding sequences, but not a small percentage of all fungal sequences deposited in GenBank are based on misidentified material ITS [116]. Particularly valuable in forensics, are DNA sequencing technologies that allow for the identification of fungi obtained from environmental samples such as soil, litter, and house dust. For this purpose, environmental DNA (eDNA) metabarcoding is used, which allows criminologists to geolocate the tested sample based on the spectrum of fungi present in it. Attempts have also been made to identify humans without analysing their DNA, but based on their unique microbiome detected using metabarcoding [117].

### 6.2. Fungal spores

Fungal spores show great potential within the context of forensic evidence. This is because they are produced in large quantities, are very small (even several microns), are easily dispersed by wind, and often attach to a body, clothing, vegetation, and objects. Moreover, spores do not lose their taxonomic characters with the passage of time. The spores can also be easily observed and identified using an ordinary light microscope [49]. Their characteristic features include colour, size, dimensions, presence of septa, presence of a point of attachment, and branches.

Hawksworth and Wiltshire, and Hawksworth et al. [41,101] present many examples of how knowledge of mycology is used in forensic science. A frequently described case demonstrating the potential of court mycology is linking a suspect to the place where a dead body was hidden, based on the presence of spores of *Torula herbarum* and *Periconia*, characteristic only of nettle, in the clumps where the victim lay and in

the suspect's car. In another case, an assailant takes part in a shooting hid behind a tree next to which there was a hedge. In this drug-related shooting, the assailant killed his partner and hid himself behind the trunk of an oak growing close to the cypresses, forming a hedge. The cypress shrubs were infected with a fungus (*Pestalotiopsis funerea*) which sporulated abundantly at that time; hence, a substantial number of spores were found on the plant litter and on the suspect's body, clothes, and items belonging to him. Fungal spores provide information that is important during court proceedings.

Hawksworth et al. [101] provided a list of rare fungi species associated with a specific environment or host on which they parasitise. Finding spores as part of evidence is a valuable clue that allows forensic scientists to answer the question 'where?'. Spores of *Caryospora calli-carpa* have been found as evidence; it is a very rare species in Great Britain because no specimen of this fungus has been collected in Great Britain since 1865. This fungus should be considered rare because it was present in only seven out of 1100 samples examined over a period of 2 years [118]. The presence of spores of *Thecaphora frezzii*, a fungus that causes huge losses in peanut crops in South America, allowed for a confirmation that the imported peanut syrup originated from Argentina or Brazil. If the evidence includes *Curvularia inaequalis* spores, it would be indicative of the sample originating from North America, where it is a well-known parasite of *Triticum* and grasses [101].

Knowledge of the growth cycles of different fungal species and the types of spores produced by them can also help determine the time of an incident [108]. *Sarcoscypha* sp. with scarlet cups occur on fallen branches in early spring. When they are trodden on, spores can attach to shoes. Another potentially useful fungus is *Flammulina velutipes*, which only appears after the first ground frost. Both species are common in the Northern Hemisphere (NH). As Hawksworth and Wiltshire cited [41] knowledge of the growth cycle of *Phragmidium violaceum* was useful in determining the burial time of a young woman. Leaves of *Rubus fruticosus* with visible dark teliospores, produced in late summer and autumn, were found in the grave. The presence of an ergot in the gathered evidence can also help determine the time of an incident. *Claviceps purpurea* growing on cereals and other grasses is produced in autumn ergots that can attach to different surfaces because of their hooks. Knowledge of the rate of fungal colony growth is useful in determining the time of an incident [101]. For example, colonies of Mucorales fungi grow much faster than Ascomycetes [41].

A perfect knowledge of the fungal spore structure is of key importance to avoid the death of a patient with suspected fungal poisoning [41]. Mushroom poisoning can lead to organ damage after several dozen hours. Identification of fungal micro- and macroremains, including spores, found in vomit, gastric washing, or faeces assists in confirming such poisoning [89,101]. For an experienced diagnostician, spores of *Amanita phalloides* and *A. muscaria* differ in shape, colour, and size. Analysis of spores is an auxiliary examination that may confirm the consumption of fungi causing death cap syndrome, but the absence of fungal spores in material taken from a patient does not exclude poisoning [119–121].

Although it has been demonstrated that mycology provides useful forensic evidence in different ways, it is not frequently used. The most reliable method for fungal identification is a direct comparison with a correctly named reference material.

### 6.3. Other spores

Spores of bryophytes, lycophytes, or ferns belong to palynomorphs that have also found their application in forensics, and their presence in evidence allows the question 'where' to be answered [122]. They are produced in a much smaller amount than pollen grains or fungal spores and are heavier than the latter; hence, the phenomenon of long-distance dispersion is much less frequent. They occur in the air at low or very low concentrations [123,124], and their presence in evidence indicates close contact with the parent plants [22,122]. These are identified as species,

genera, or artificial taxa. Taxonomic characteristics include shape, dimension, colour, and morphology [125,126].

The fern spores have radial or bilateral symmetry. Another taxonomic characteristic is the aperture. Spores have a three-layered cell wall of varying thicknesses and chemical compositions. Elements of the middle layer and the exine (e.g. papillae, spines, reticulum, or clubs) are diagnostic features [125]. The ability to identify fern spores was invaluable in solving the murder of Joanne Nelson (the 'Valentine Girl'). Numerous pollen grains and fern spores, including *Polypodium*, rarely found in the area in question, were found in the soil collected from the car of a person suspected of having this murder. A comparison of the assemblage of palynomorphs with the vegetation assemblage indicated the potential place of burial [20]. The presence of *Lycopodium* spores in a sample of cocaine was very useful to estimate its origin, considering the time when sporangia were mature [127].

The presence of bryophyte and lycophyte spores in soil indicates that its layers belong to a particular area searched or can be an indicator for a specific habitat, as demonstrated by Bruce and Dettmann [128], as well as by Khokh and Shalaboda [129]. They also stressed that the presence of spores of lower plants (ferns, lycophytes, and bryophytes) in the soil is a reflection of their contribution to local vegetation. This was confirmed by conducting research on various types of forest communities in different climatic zones, in Australia and Belarus.

## 7. Forensic palynology

### 7.1. Pollen grains as objects of forensic investigations

Among the many types of palynomorphs, pollen grains have been used most frequently in forensic science [23,130–133]. They are produced in large numbers and can be identified as a genus/species based on their characteristic morphological structure. Their diagnostic features are primarily the number and type of apertures (pores and furrows), their arrangement, grain size and shape [131]. The cell wall is multi-layered, composed of sporopollenin, a substance very resistant to the effects of external factors, due to which pollen grains are preserved even in the most difficult environmental conditions. The outer layer, the exine, has a species-specific sculpture in the form of spines, papillae, reticulum, and striae. A small sample taken from the incident scene was sufficient to isolate a sufficient number of pollen grains. Such materials can be analysed repeatedly and sent for consultation or verification. The presence of pollen grains in the evidence allowed us to answer all the questions asked in the title of this article. In the forensic interpretation of palynological material, knowledge of pollination type, height of pollen release, and pollen dispersal capacity is important [23,131,134–136].

Many palynological studies underline that the qualitative and quantitative spectra of aeroplankton or pollen deposition largely reflect vegetation, and plant communities are characterized by specific palynoflora named 'pollen fingerprint' [137]. The spectrum of tree pollen reflects a region's vegetation. Pine pollen tends to be overrepresented, while the proportion of beech pollen is lower than the percentage of beech in a region's vegetation. Because pollen grains of herbaceous plants are released at much lower altitudes than those of trees, many of them are less likely to reach the upper atmosphere and spread far. These relationships should be considered when preparing an expert report for a court [23].

### 7.2. 'Pollen fingerprint' and the time and place of the crime

When an expert attempts to link an object or a person to a crime scene, a specific geobotanical region, or a plant community, he or she must have rich reference material, that is, a collection of reference microscopic slides and microscopic images [133,136]. The lack of a sufficient reference database will prevent an expert from providing a reliable opinion, as in the case of illegal imports of Persian rugs to the

USA. It was suspected that the rugs originated from Iran, where the import of goods was prohibited. A palynological analysis of samples taken from the rugs showed the presence of pollen grains from plants characteristic of both Egypt and Iran, with a predominance in the latter country; however, because of the lack of rich comparative material from the Near East, the experts did not have sufficient certainty to recommend banning these imports [130]. The lack of reference material from distant or war-affected regions, which are threatened with terrorism, is particularly acute. Warny et al. [138] proposed an innovative method to create a worldwide palynological database based on which geolocation could be carried out. They performed palynological analysis of pollen vacuumed from pelts of species characteristic of tens of mammals in different regions of Mexico. They showed that each region has a characteristic 'pollen fingerprint' and thus indicated the potential of this method in creating a metabase of reference material necessary for geolocation.

During the investigation of war crimes committed in Yugoslavia in 1995 in Srebrenica (Bosnia), the International Criminal Tribunal investigated mass graves of the massacred civilian population. Intelligence reported that war criminals, in order to hide the scale of executions, transported their bodies from their original graves to smaller ones in unknown locations. The purpose of this forensic examination was to link the secondary graves with the primary sites by comparing samples collected from the soil, bodies, and clothing, among others, in terms of their palynomorph content. Based on pollen analysis, in many cases, secondary sites were linked to execution sites. This case is thought to be the first in which such environmental evidence was used systematically in an investigation of serious war crimes [139].

Zavada et al. [140] showed that clothing is an excellent pollen trap. The thicker the fabric is, or if it has structural patterns, and the more frequently a piece of clothing is worn, the more pollen grains it contains. The pollen spectrum is representative of the vegetation of a particular region and season, which can help an expert determine the location of a human and the period when he or she is in a specific area. Interpretation of results can be difficult if a person visits many different places on the same day. Washing with water and detergent eliminates pollen from the fabrics. Webb et al. [141] verified existing assumptions. They showed that the qualitative and quantitative spectrum of pollen collected from clothing quickly changes over time because approximately 60 % of pollen is lost; therefore, material for analysis should be collected as quickly as possible. The type of fabric is also important because the pollen of wind-pollinated grasses attaches to cotton to a small degree. The experiments confirmed that washing clothes is not a method for the full removal of pollen, but the efficiency of this forensic method is greater than previously assumed, standing between 21 % and 95 %. The fact that plant pollen was preserved on clothing made it possible to link burglars to the crime scene in a court case conducted in New Zealand. Relatively large amounts of fresh *Hypericum* pollen were found on suspects' clothing, and the plant was present at the crime scene. In this case, plant pollen showed excellent trace evidence [134].

Some parts of the plants can act as natural traps for pollen grains. For instance, this is true in the case of fluff from infructescences of poplar or in the below-described example of tobacco leaves. Owing to the characteristic epidermis of the tobacco leaf, palynomorphs freely floating in the air easily attach to it. Cigarettes of a well-known American brand were examined for the presence of pollen grains, and it was demonstrated that the original cigarettes contained pollen of *Ambrosia* species, native to North America. None of the counterfeit cigarettes contained pollen of this genus or its percentage was minimal. Chan et al. [142] concluded that the lack of *Ambrosia* pollen is a strong premise that a given product does not originate in the United States.

The COVID-19 pandemic has drawn the attention of the general public to the need to maintain special hand hygiene. Guidelines designed to minimise the risk of viral infections have been developed. This inspired Hunt and Morawska [143] to undertake a study on the degree of pollen retention on the skin depending on the frequency of

hand washing and the observance of the relevant procedure. This study demonstrated that the number of pollen grains on hands was very small only if the WHO's recommendations in this respect were strictly adhered to. The authors concluded that when conventional hygiene is used, it is highly likely that pollen will remain in the hands of a suspected person for several days, which may help in court proceedings.

Mindelhall [144] described how palynology was employed to identify the origin of falsified (i.e. counterfeit) life-saving pharmaceuticals, particularly antimalarial ones. Under the auspices of the WHO and INTERPOL, in 2015, a team was appointed to gather evidence documenting this illegal activity. Palynological analysis was used to determine where the components were sourced from or where the counterfeit tablets were located. Cereal pollen was isolated from some samples, suggesting that the sources of some components were located near the agricultural areas. The pollen of *Artemisia* may have originated from *A. annua*, which is widespread in China. Other identified pollen grains originated from plants within a limited area in Southeast Asia. Spores of rarely found ferns and isoetopsida, such as *Stenochlaena* and *Isoetes*, were also identified; their range is limited to the coastal areas of Vietnam and southern China. A reliable analysis could be presented to the authorities, together with other environmental evidence, and it was possible to indicate the probable location for the production of illegal drugs.

The ability to isolate pollen from evidence material, high competence in pollen identification, and knowledge of pollen production phenology allows one to determine the period in which a sample comes. Palynological analysis of the material collected from a mass grave discovered in Magdeburg in 1994 revealed the presence of ribwort plantain and rye pollen, which corroborated one of the hypotheses that the victims had been Russian soldiers murdered by the KGB in the summer of 1953, the presence of pollen grains of alder and birch (*Alnus* sp. and *Betula* sp.), and flowering in early spring, that is, during the period when a shooting took place, on a suspect's weapon undermined the credibility of this suspect who claimed that he had not used this weapon for months [145].

### 7.3. Molecular palynology in forensics

Thus far, palynology largely entails time-consuming light microscopy, which requires a high level of expertise. To circumvent this issue, forensic laboratories are increasingly using non-microscopic techniques. Numerous studies indicate great potential for the identification of pollen grains using nondestructive spectroscopic methods (Raman, FTIR) [23,146,147]; however, there is no sufficient library of reference spectra to be successfully used in forensic science. After years of attempts, effective procedures for the isolation of pure genetic material from pollen grains have been developed; for many taxa, there is a good reference base; therefore, pollen grains can be identified based on both chloroplast and nuclear DNA [148–150]. However, one should remember the high genetic variability of plants between individuals depending on environmental conditions; therefore, it is currently impossible to distinguish plants based on variability in one locus. To date, only five different markers have been used to encode pollen DNA: *rbcl*, *matK*, *trnL*, *trnH-psbA*, and *ITS2*. The routine use of this method in forensic palynology is hindered by a few obstacles, the biggest of which is the impossibility of using this material for repeated analyses. Bell et al. [151] suggested methods that would allow for the subsampling of pollen samples. Although the stable isotope method is used in forensic botany, the determination of isotope content in pollen remains a challenge, especially in the context of pollen sampling, method validation, and time reduction [23].

## 8. Forensic limnology

### 8.1. Diatoms as objects of forensic investigations

The aquatic environment is difficult to investigate by forensic scientists because of its complex physical and chemical dynamics [152]. It is a place where diverse organisms live, including plankton, which have been most frequently examined by forensic scientists. Forensic limnology most often deals with diatoms [153]. These are photosynthesising algae with a silicate shell composed of structural, longitudinal (apical), and transverse (transapical) structures, the distribution of which is specific to individual taxa. Their ornamentation and size form the basis of diatom identification, which can often be performed using a light microscope. Owing to the specific silicate structure of the shell surrounding the cells, diatoms are resistant to the effects of physico-chemical factors and preserve their diagnostic features even after death. Thanks to this, it is possible to identify them both in modern materials and in materials preserved many years ago. Populations of algae, although variable, persist throughout the year. Owing to their microscopic size, they are more difficult to remove from places, clothing, or tools, and dirt or even wetting is sufficient to isolate them [154]. Scott et al. [155,156], Sharma et al. [157], and Levin et al. [158] experimentally confirmed that even short-term contact of clothing or footwear with water or soil is sufficient to transfer diatoms from their original environment. Diatoms persist on footwear even up to 168 h after contact with water, and on fabrics even for many weeks. Scott et al. [159] showed that diatoms are preserved even on clothing that has been substantially destroyed, for example, by fire.

Diatoms occur in large numbers in many habitats, and each has its own characteristic diatom biota. For instance, the presence of mainly benthic species in evidence suggests contact with the bed of a river, lake, or pond [153,159,160]. In their experiments, Bogusz et al. [154] demonstrated that objects submerged in aquatic habitats of different types are fully colonized after 4–6 weeks by the species living in the submerged environments. Limnological analysis should consider both the qualitative spectrum and analysis of the number of all species, as well as the community structure. Such analyses were undertaken in the case of a brutal battery of teenagers during fishing. Comparison of the evidence in terms of the presence of diatoms allowed the victims, suspects, and scene of the incident to be linked [160].

### 8.2. Microscopy techniques in diatoms identifications

Light microscopy is still the standard method used in limnological laboratories, but electron microscopy is increasingly used to identify diatoms, the advantage of which is high identification accuracy; however, from the point of view of a forensic laboratory, the equipment is expensive [161]. When evidence is scarce or fragmented, ultrasensitive, high-resolution tools are required. These tools include nanoscale and ultramicroscopic imaging methods such as atomic force microscopy (AFM). The lack of preparatory stages and the possibility of quick and non-destructive measurements in environments close to physiological conditions are advantageous for forensics [162]. Yadavalli and Ehrhardt [163] indicated the great potential of this method in forensic diatom imaging and identification; however, as in the case described above, the disadvantage is the cost of the equipment. The advantages of SEM and AFM techniques are the possibility of identifying an object at the genus level [164].

In addition, molecular techniques are increasingly used for diatom identification [164]. Li [165] analysed the morphology and 18S rDNA sequences of diatoms in one of the rivers in China. They identified q-PCR methods with high accuracy. They pointed out that diatom DNA barcoding could be a useful tool in forensic studies.

### 8.3. Diatoms test and drowning confirmation

Analysis of the diatom content in human tissues is helpful in determining whether a victim found died via drowning. During drowning, diatoms enter the lungs and subsequently penetrate the alveoli-capillary barrier and enter the organs and bone marrow. Diatoms have been shown to be smaller in liver and kidney tissues than in lung tissues and water samples and it has been demonstrated that pennate diatoms penetrate through the alveoli-capillary barrier more easily [164]. Studies on the presence of diatoms in corpses in water have been conducted for a long time. Sharma et al. [157] presented a historical outline indicating people's interest in this subject in the 17th century, whereas the first diatom test was performed at the beginning of the 20th century. Currently, the diatom test is conducted for all unnatural, unexpected, and suspected deaths when a body is found in water [166]. During an autopsy of a body found in water, the occurrence of diatoms in the lungs, organs, and bone marrow as well as at the potential scene of the incident is checked, which helps to establish whether drowning has taken place and where it has occurred [167]. Magrey and Raj [168] described examples of the practical application of the diatom test in India. Antemortem drowning was established based on the presence of *Cymbella*, *Nitzschia gracilis*, and *Navicula radiosa* in the bones of a 23-year-old woman found in a well. The same three diatom species were found in the water samples collected from the well.

The presence of diatoms in the lungs itself does not prove drowning because false-positive tests occur because diatoms can enter a corpse transported to water as a result of passive penetration under the influence of hydrostatic pressure [81,169]. Due to the unreliability of the diatom test, in some cases, an analysis of the presence of diatoms in many organs should be performed, and additional auxiliary tests should be carried out to confirm death by drowning [166,170,171]. Zhao et al. [169] proposed that, when preparing expert reports for a court, the ratio of the content of diatoms in a victim's body to their content in the water should be calculated; its value is higher in the case of drowning than in the case of post-mortem immersion. The absence of diatoms, on the other hand, is not the basis for excluding death due to drowning [172]. Cases of dry drowning have been reported when a violent laryngospasm occurs and water does not reach the lungs [173].

A recent study by Du et al. [174] questioned the reliability of the diatom test in indicating the site of drowning. They compared the results of their experiment by employing many chemometric methods (e.g. cluster analysis) which did not show an unambiguous linkage between the diatom assemblage from the lung tissue and the site of drowning.

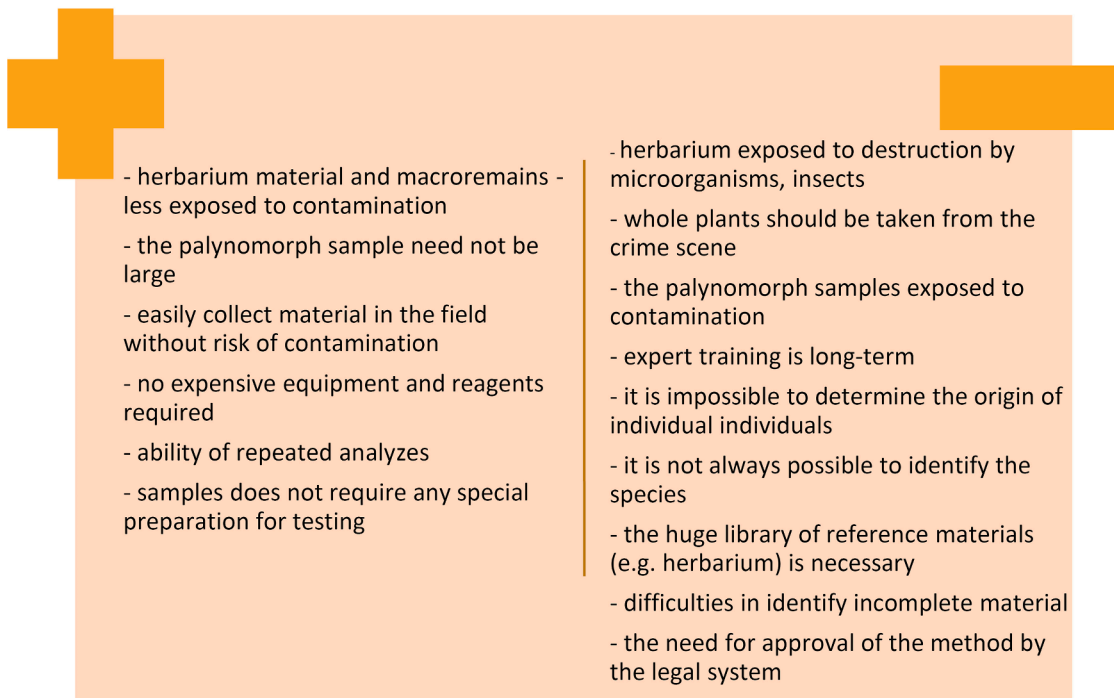
During the last decade, molecular methods have been included in the range of techniques used in diatom tests to confirm or deny drowning [175,176]. Yu [164] proved that SYBR Green real-time PCR is characterised by high specificity and sensitivity and can help to exclude some false-positive results but also confirm death by drowning more quickly. The detection range was at least 0.0001 ng. Since molecular analyses are highly sensitive, it was expected that barcoding would be extremely helpful in tests confirming drowning. Liu [176] experimentally demonstrated that microscopic observations were not highly correlated with identification using barcoding technology. The unsatisfactory efficiency of barcoding is due to the fact that the DNA reference sequence is insufficient and contains many errors. Therefore, it is necessary to build a reliable diatom DNA sequence database.

### 8.4. Diatoms and PMI estimations

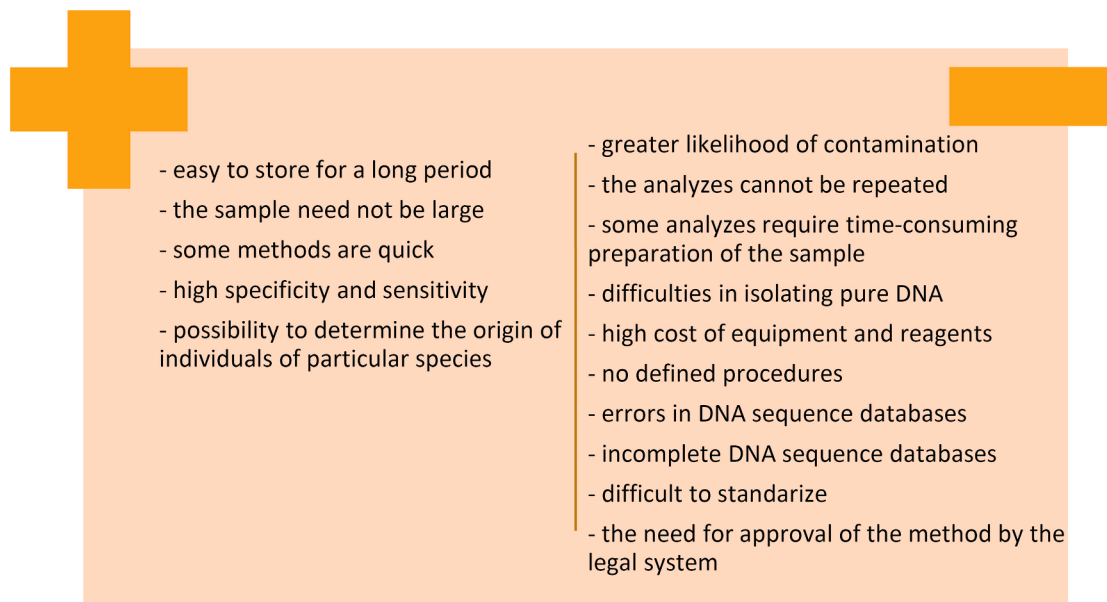
The biodiversity of diatoms colonising a corpse immersed in water changes in a certain order, suggesting that algae can be a good tool for PMI estimation. The number of diatom species decreased during the later stages of decomposition. Zimmerman and Wallace [177] believed that because PMI estimation based on aquatic insects is not accurate, applying semi-qualitative and qualitative approaches using aquatic plant models can increase the accuracy of estimates. Ishikawa et al.



### Morphological identification



### Molecular analysis



**Fig. 4.** Comparison of the morphological and molecular methods in terms of application in forensic botany (based on [3–6,10,17,20–23,30,37,39,53,60,64,65,70,115,116,130,136,147,151,161,164]).

[178] proposed a method to estimate the time since death at sea. This method combines water elemental analysis and examination of phytoplankton on teeth. They showed that the contents of typical elements in teeth, that is, Ca and P, decreased with time, whereas the Si, Mg, and K contents increased. After 30 days, a large part of the enamel was covered with phytoplankton, predominantly diatoms, whereas after 60 days, the entire enamel surface was covered. The PMI can also be estimated by

tracking the succession of diatom biotas on a cadaver. Diatoms isolated from soil are also good evidence and can be used to estimate the PMI [179].

#### 8.5. Other algae and protists in forensic investigations

Other organisms belonging to widely understood algae or protists are

also objects of forensic limnological studies [180]. Diaz-Palma et al. [181] detected phytoplankton other than diatoms in the bodies of drowning victims, including species belonging to Chlorophyta and Dinoflagellata. Based on their experiments, they presented convincing arguments regarding the need to supplement the diatom test with an analysis of the microalgae content in drowned bodies. Algae also colonise bones. In the polar climatic zone, the bones of cetaceans are colonised by Chlorophyta algae, including *Muriella terrestris*, or species of the genus *Chlorosarcina* sp. [57].

Blue-green algae, such as cyanobacteria, produce secondary metabolites that are harmful or even fatal to humans and animals [183]. Detection of such organisms and their proper identification are extremely important for the sake of life protection. Their presence in water decreases the landscape and tourism value of an area, causing severe economic losses. Such scientific evidence has already been used in court proceedings regarding environmental poisoning and related damages [182,183].

Organisms belonging to the Protista, such as amoebae (Rhizopoda), dinoflagellates, and chrysophytes, are also objects of forensic examination. In 1994, Siver et al. [160] showed that the presence of widely distributed *Mallomonas caudata* chrysophytes together with diatoms allowed the victims, suspects, and scene of the incident to be linked. Testate amoebae are used in PMI determination or help link a victim or suspect to a crime scene because the time of one generation is relatively short, usually several days under good conditions [179]. Szelecz et al. [184] revealed that, at the initial stage of cadaver decomposition, the number of these organisms distinctly decreases, while the process of biota renewal begins after approximately two months. Knowledge of these phenomena can be helpful in determining the time of death for bodies that have been forgotten for a long time. Swindles and Ruffell [185] described a case of analysis of evidence (soil, water, vegetation, and clothing samples) for their content of testate amoebae. A comparison of the species spectra made it possible to identify the location where the victim had been killed. They also confirmed the high evidence potential of these organisms because the researchers managed to isolate them from clothing 10 years after the incident and preserved their diagnostic features, which allowed the organisms to be discriminated with great accuracy, that is, to the genus *Arcella* spp. and even to the species *Centropyxis cassis*.

## 9. Summary

A botanist's expertise in a court can be an important tool in answering questions posed during forensic investigations. Evidence should be read in the context of the entire investigation process. A common standard is to evaluate the results of forensic investigations by defining two mutually exclusive hypotheses and establishing a likelihood ratio between them [186]. It serves as a quantitative measure of the strength of evidence [187]. The higher its value, the higher is the likelihood that the samples originate from the same environment [135,187,188]. The expert may be asked to comment on the importance of the evidence in the context of various competing claims: prosecution and defence [187]. The LR value is context-independent, but the expectations of the judges are different, as are the tolerances for possible error [188]. Sometimes it is proposed to use not only numerical LR values, but also their verbal interpretation and scale [188]; however, the latter is criticised as it does not express the precision of LR [189].

In the face of the continuous development of molecular biology and the use of sophisticated technology and equipment, the question arises as to whether traditional botany still has a future in the judiciary [10,60]. Both methods have advantages and disadvantages, as listed in Fig. 4. The methods differ with regards to collecting technique, material storage, cost and time consumption, specificity, and reliability. Unlike genetic materials, botanical materials (even microremains) can be analysed repeatedly, and expensive equipment and reagents are not required. Even a small amount of material was sufficient for genetic and

biochemical analyses. Despite the high specificity, in many cases, identifying plants based on their genetic material is not possible at species level. This is usually due to there being no reference material. Different techniques can and should be applied in forensic laboratories [10,37,60,92,162,176]. Forensic experts should also consider the possibility of conducting biochemical analyses at crime scenes. Mobile laboratories are becoming increasingly common in environmental research. Many analyses can be successfully conducted in the field using portable equipment such as FTIR, Raman spectroscopy, and gas chromatography–mass spectrometry. Small portable laboratories allow for rapid analysis, which is a desirable advantage in many forensic cases [190].

Although forensic botany is a recognised scientific method and botanical material meets the criteria of scientific evidence, it is routinely used in criminal investigations in a few countries. The reasons for this are, among others, the lack of qualified specialists, long training of experts, and lack of awareness of the evidence potential of plant material [17,20,64]. Introduction to modern forensic apparatuses and technologies is inevitable. Many instrumental methods are validated, but there are also those with great potential, but their routine application is not always possible. Undoubtedly, skills of highly qualified botanists/biologists experienced in field research involving plant morphology and ecology will still be needed [60]. The presented literature review indicates that even laboratories without highly specialised equipment can successfully investigate botanical evidence because the range of possibilities for assessing plant material is large.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## References

- [1] E. Locard, The analysis of dust traces, Part I, Am. J. Police Sci. 1 (1930) 276, <https://doi.org/10.2307/1147154>.
- [2] H. Miller Coyle, Introduction to forensic botany, in: H. Miller Coyle (Ed.), Forensic Botany: Principles and Applications to Criminal Casework, CRC Press, 2005, pp. 1–7.
- [3] H. Miller Coyle, Forensic Botany: Principles and Applications to Criminal Casework, CRC Press, 2004.
- [4] D.W. Hall, Plants as evidence, in: D.W. Hall, J.H. Byrd (Eds.), Forensic Botany: A Practical Guide, Wiley, 2012, pp. 12–44.
- [5] P.E.J. Wiltshire, Forensic ecology, in: P. White (Ed.), Crime Scene to Court: The Essentials of Forensic Science, 3rd ed., RSC Publishing, 2010, pp. 54–85.
- [6] D.W. Hall, J.H. Byrd, in: Forensic Botany: A Practical Guide, Wiley, 2012, pp. 45–78.
- [7] W. Meier-Augenstein, From stable isotope ecology to forensic isotope ecology — Isotopes' tales, Forensic Sci. Int. 300 (2019) 89–98, <https://doi.org/10.1016/j.forsciint.2019.04.023>.
- [8] W. Woodenberg, J. Govender, N. Murugan, S. Ramdhani, Serphen, Cycad forensics: leaflet micromorphology as a taxonomic tool for South African cycads, Plant Syst. Evol. 305 (6) (2019) 445–457.
- [9] F. Lens, C. Liang, Y. Guo, X. Tang, M. Jahanbanifard, F.S.C. da Silva, G. Ceccantini, F.J. Verbeek, Computer-assisted timber identification based on features extracted from microscopic wood sections, IAWA J. 41 (4) (2020) 660–680.
- [10] G. Jakoby, I. Rog, I. Shtein, I. Chashmonay, D. Ben-Yosef, A. Eshel, T. Klein, Tree Forensics: Modern DNA barcoding and traditional anatomy identify roots threatening an ancient necropolis, Plants People Planet. 3 (2021) 211–219, <https://doi.org/10.1002/ppp3.10165>.

- [11] B. Rocke, A. Ruffell, L. Donnelly, Drone aerial imagery for the simulation of a neonate burial based on the geoforensic search strategy (GSS), *J. Forensic Sci.* 66 (2021) 1506–1519, <https://doi.org/10.1111/1556-4029.14690>.
- [12] I. Burney, N. Pemberton, Making space for criminalistics: Hans Gross and fin-de-siècle CSI, *Stud. Hist. Philos. Biol. Biomed. Sci.* 44 (2013) 16–25, <https://doi.org/10.1016/j.shpsc.2012.09.002>.
- [13] B.I. Lipscomb, G.M. Diggs Jr, The use of animal-dispersed seeds and fruits in forensic botany, *SIDA, Contributions to Botany*. 18 (1998) 335–346, <https://fpamed.com/wp-content/uploads/2013/09/Frye-v-US-1923.pdf>.
- [14] B.A. Raum, Expert evidence, in: D.W. Hall, J.H. Byrd (Eds.), *Forensic Botany: A Practical Guide*, 1st ed., Wiley, Blackwell, UK, 2012: pp. 79–92. <https://doi.org/10.1002/9781119945734.ch3>.
- [15] J. Balouet, K. Smith, D. Vroblesky, G. Oudijk, Use of dendrochronology and dendrochemistry in environmental forensics: does it meet the Daubert criteria? *Environ.* 10 (2009) 268–276, <https://doi.org/10.1080/15275920903347545>.
- [16] T. Palmbach, G. Shutler, Legal consideration for acceptance of new forensic methods in court, in: H. Miller Coyle (Ed.), *Forensic Botany: Principles and Applications to Criminal Casework*, CRC Press, 2005, pp. 197–216.
- [17] D.W. Hall, W. Stern, Plant anatomy, in: D.W. Hall, J.H. Byrd (Eds.), *Forensic Botany: A Practical Guide*, 1st ed., Wiley, Blackwell, UK, 2012: pp. 119–126. <https://doi.org/10.1002/9781119945734.ch3>.
- [18] J.H. Bock, D.O. Norris, in: *Forensic Plant Science*, Elsevier, 2016, pp. 121–127.
- [19] P.E.J. Wiltshire, Forensic ecology, botany, and palynology, *Forensic Sci. Sem.* 6 (2016) 32–42.
- [20] C.J. Watson, S.L. Forbes, An investigation of the vegetation associated with grave sites in southern Ontario, *Can. Soc. Forensic Sci. J.* 41 (2008) 199–207, <https://doi.org/10.1080/00085030.2008.10757177>.
- [21] D.C. Mildenhall, Pollen plays part in crime-busting, *Foren. Focus* 11 (1992) 1–4.
- [22] M.O. Ezeogbo, Identifying the scene of a crime through pollen analysis, *Sci. Justice* 61 (2021) 205–213, <https://doi.org/10.1016/j.scijus.2020.12.002>.
- [23] L.K. Pramod, K.B. Kumar, A.M. Nasir, Role of forensic botany in crime scene investigation - a literature review, *J. Forensic Med. Sci. Law.* 30 (2021) 52–56, <https://doi.org/10.1111/1556-4029.12401>.
- [24] P.E.J. Wiltshire, Protocols for forensic palynology, *Palynology* 40 (2016) 4–24, <https://doi.org/10.1080/01916122.2015.1091138>.
- [25] R.M. Morgan, P.A. Bull, Forensic geoscience and crime detection. Identification, interpretation and presentation in forensic geoscience, *Minerva. Med. Leg.* 127 (2007) 73–89.
- [26] R.W. Fitzpatrick, L.J. Donnelly, An introduction to forensic soil science and forensic geology: a synthesis, *Geol. Soc. London Spec. Publ.* 492 (2021) 1–32, <https://doi.org/10.1144/SP492-2021-81>.
- [27] H.F.V. Cardoso, A. Santos, R. Dias, C. Garcia, M. Pinto, C. Sérgio, T. Magalhães, Establishing a minimum postmortem interval of human remains in an advanced state of skeletonization using the growth rate of bryophytes and plant roots, *Int. J. Leg. Med.* 124 (2010) 451–456, <https://doi.org/10.1007/s00414-009-0372-5>.
- [28] R. García, L. Magnin, L. Miotti, G. Barrientos, Lichens growing on human bone remains: a case study from continental Patagonia (Deseado Massif, Santa Cruz, Argentina), *J. King Saud Univ. Sci.* 32 (2020) 2219–2221, <https://doi.org/10.1016/j.jksus.2020.02.029>.
- [29] M.A. Gitzendanner, Use and guidelines for plant DNA analyses in forensics, in: D. W. Hall, J.H. Byrd (Eds.), *Forensic Botany: A Practical Guide*, 1st ed., Wiley, Blackwell, UK, 2012: pp. 93–106. <https://doi.org/10.1002/9781119945734.ch3>.
- [30] R.Y. Bhatia, S. Raghavan, K.V. Rao, V.N. Prasad, Forensic examination of leaf and leaf fragments in fresh and dried conditions, *J. Forensic Sci. Soc.* 13 (1973) 183–190, [https://doi.org/10.1016/s0015-7368\(73\)70794-5](https://doi.org/10.1016/s0015-7368(73)70794-5).
- [31] D.W. Hall, Case study in forensic botany, in: D.W. Hall, J.H. Byrd (Eds.), *Forensic Botany: A Practical Guide*, 1st ed., Wiley, Blackwell, UK, 2012, pp. 174–187, <https://doi.org/10.1002/9781119945734.ch3>.
- [32] J.H. Bock, D.O. Norris, Plant taxonomy cases, in: J.H. Bock, D.O. Norris (Eds.), *Forensic Plant Science*, Elsevier Inc., 2016: pp. 103–105. <https://doi.org/10.1016/B978-0-12-801475-2.00007-5>.
- [33] M. Caccianiga, C. Compostella, G. Caccia, C. Cattaneo, Contribution of plant anatomy to forensic investigation: tree bark morphology, *Forensic Sci. Int.* 318 (2021), 110598, <https://doi.org/10.1016/j.forsciint.2020.110598>.
- [34] A. Gunn, Plants, protists, fungi and microbes, in: *Essential Forensic Biology*, John Wiley & Sons, 2019, pp. 327–364.
- [35] Y. Marumo, Forensic examination of soil evidence, *Jpn. J. Sci. Tech. Iden.* 7 (2003) 95–111, <https://doi.org/10.3408/jasti.7.95>.
- [36] E.E. Dormontt, M. Boner, B. Braun, G. Breulmann, B. Degen, E. Espinoza, S. Gardner, P. Guillery, J.C. Hermanson, G. Koch, S.L. Lee, M. Kanashiro, A. Rimbawanto, D. Thomas, A.C. Wiedenhoef, Y. Yin, J. Zahnen, A.J. Lowe, Forensic timber identification: It's time to integrate disciplines to combat illegal logging, *Biol. Conserv.* 191 (2015) 790–798, <https://doi.org/10.1016/j.biocon.2015.06.038>.
- [37] D.L. Dilcher, Forensic botany: case studies in the use of plant anatomy, *Phytomorphology* 181 (2001) 181–184.
- [38] X. Bai, G. Wang, Y. Ren, J. Han, Detection of Highly Poisonous *Nerium oleander* Using Quantitative Real-Time PCR with Specific Primers, *Toxins* 14 (2022) 776, <https://doi.org/10.3390/toxins14110776>.
- [39] O.W. Purvis, B.J. Williamson, B. Spiro, V. Udachin, I.N. Mikhailova, A. Dolgoplova, Lichen monitoring as a potential tool in environmental forensics: case study of the Cu smelter and former mining town of Karabash, Russia, *Geol. Soc. London Spec. Publ.* 384 (2013) 133–146, <https://doi.org/10.1144/SP384.6>.
- [40] D.L. Hawksworth, P.E.J. Wiltshire, Forensic mycology: the use of fungi in criminal investigations, *Forensic Sci. Int.* 206 (2011) 1–11, <https://doi.org/10.1016/j.forsciint.2010.06.012>.
- [41] M. Caccianiga, S. Bottacin, C. Cattaneo, Vegetation dynamics as a tool for detecting clandestine graves, *J. Forensic Sci.* 57 (2012) 983–988, <https://doi.org/10.1111/j.1556-4029.2012.02071.x>.
- [42] C.J. Watson, M. Ueland, E.M.J. Schotsmans, J. Sterenberg, S.L. Forbes, S. Blau, Detecting grave sites from surface anomalies: a longitudinal study in an Australian woodland, *J. Forensic Sci.* 66 (2021) 479–490, <https://doi.org/10.1111/1556-4029.14626>.
- [43] M. Cholewa, M. Bonar, M. Kadej, Can plants indicate where a corpse is buried? Effects of buried animal tissues on plant chemistry: Preliminary study, *Forensic Sci. Int.* 333 (2022), 111208, <https://doi.org/10.1016/j.forsciint.2022.111208>.
- [44] Z. Knobel, M. Ueland, K.D. Nizio, D. Patel, S.L. Forbes, A comparison of human and pig decomposition rates and odour profiles in an Australian environment, *Aust. J. Forensic Sci.* 51 (2019) 557–572, <https://doi.org/10.1080/00450618.2018.1439100>.
- [45] H. Brabazon, J.M. DeBruyn, S.C. Lenaghan, F. Li, A.Z. Mundorff, D.W. Steadman, C.N. Stewart, Plants to remotely detect human decomposition? *Trends Plant Sci.* 25 (10) (2020) 947–949.
- [46] H. van de Voorde, P.J. Van Dijk, Determination of the time of death by fungal growth, *Z. Rechtsmed.* 89 (1982) 75–80, <https://doi.org/10.1007/BF02092372>.
- [47] M. Hitosugi, K. Ishii, T. Yaguchi, Y. Chigusa, A. Kurosui, M. Kido, T. Nagai, S. Tokudome, Fungi can be a useful forensic tool, *Leg. Med.* 8 (2006) 240–242, <https://doi.org/10.1016/j.legalmed.2006.04.005>.
- [48] M. Tibbett, D.O. Carter, Mushrooms and taphonomy: the fungi that mark woodland graves, *Mycologist* 17 (2003) 20–24, <https://doi.org/10.1017/S0269915X03001150>.
- [49] T. Fukuharu, K. Osaku, K. Iguhi, M. Asada, Occurrence of ammonia fungi on the forest ground after decomposition of a dog carcasses, *Nat. His. Rev.* 6 (2000) 9–14.
- [50] I. Aquila, F. Ausania, C. Di Nunzio, A. Serra, S. Boca, A. Capelli, P. Magni, P. Ricci, The role of forensic botany in crime scene investigation: case report and review of literature, *J. Forensic Sci.* 59 (2014) 820–824, <https://doi.org/10.1111/1556-4029.12401>.
- [51] J.H. Bock, D.O. Norris, Sources for Forensic Plant Science Evidence, in: J.H. Bock, D.O. Norris (Eds.), *Forensic Plant Science*, Elsevier, 2016: pp. 35–50. <https://doi.org/10.1016/B978-0-12-801475-2.00003-8>.
- [52] E.W.L. Brooks-Lim, S.A. Mérette, B.J. Hawkins, C. Maxwell, A. Washbrook, A. M. Shapiro, Fatal ingestion of *Taxus baccata*: English yew, *J. Forensic Sci.* 67 (2022) 820–826, <https://doi.org/10.1111/1556-4029.14941>.
- [53] H. Miller Coyle, C.-L. Lee, W.-Y. Lin, H.C. Lee, T.M. Palmbach, Forensic botany: using plant evidence to aid in forensic death investigation, *Croat. Med. J.* 46 (2005) 606–612. PMID: 16100764.
- [54] V. Virtanen, H. Korpeläinen, K. Kostamo, Forensic botany: usability of bryophyte material in forensic studies, *Forensic Sci. Internat.* 172 (2007) 161–163, <https://doi.org/10.1016/j.forsciint.2006.11.012>.
- [55] M. Lancia, F. Conforti, M. Aleffi, M. Caccianiga, M. Bacci, R. Rossi, The Use of *Leptodictyum riparium* (Hedw.) warnst in the estimation of minimum postmortem interval, *J. Forensic Sci.* 58 (2013) 239–242, <https://doi.org/10.1111/1556-4029.12024>.
- [56] M. Olech, Human impact on terrestrial ecosystems in west Antarctica, in: *Proceedings of the NIPR Symposium on Polar Biology* 9, 1996: pp. 299–306.
- [57] M.P. de Albuquerque, J. Putzke, A.L. Schünemann, F.C.B. Vieira, F.d.C. Victoria, A.B. Pereira, Colonisation of stranded whale bones by lichens and mosses at Hennequin Point, King George Island, Antarctica, *Polar Rec.* 54 (1) (2018) 29–35.
- [58] A. Nikiel, Przegląd surowców roślinnych o działaniu fotoczułym i fotokosmetycznym, *Kosmetologia Estetyczna*. 3 (2017) 231–238.
- [59] M.A. Spencer Forensic botany: time to embrace natural history collections, large scale environmental data and environmental DNA, *Emerg Top Life Sci.* 5(3) (2021) 475–485. <https://doi.org/10.1042/ETLS20200329>.
- [60] R. Cahyaningsih, L.J. Compton, S. Rahayu, J. Magos Brehm, N. Maxted, DNA Barcoding medicinal plant species from Indonesia, *Plants* 11 (2022) 1375, <https://doi.org/10.3390/plants11101375>.
- [61] Y. Marumo, H. Yanai, Morphological analysis of opal phytoliths for soil discrimination in forensic science investigation, *J. Forensic Sci.* 31 (1986) 1039–1049, <https://doi.org/10.1520/JFS111131>.
- [62] P.E.J. Wiltshire, D.L. Hawksworth, J.A. Webb, K.J. Edwards, Two sources and two kinds of trace evidence: enhancing the links between clothing, footwear and crime scene, *Forensic Sci. Int.* 254 (2015) 231–242, <https://doi.org/10.1016/j.forsciint.2015.05.033>.
- [63] M. Caccianiga, G. Caccia, D. Mazzarelli, D. Salsarola, P. Poppa, D. Gaudio, A. Cappella, L. Franceschetti, S. Tambuzzi, L. Maggioni, C. Cattaneo, Common and much less common scenarios in which botany is crucial for forensic pathologist and anthropologists: a series of eight case studies, *Int. J. Leg. Med.* 135 (2021) 1067–1077, <https://doi.org/10.1007/s00414-020-02456-0>.
- [64] D.O. Norris, J.H. Bock, Method for examination of fecal material from a crime scene using plant fragments, *J. Forens. Ident.* 51 (2001) 367–377.
- [65] S. Biswas, K. Gupta, S.N. Talapatra, A digitized database of bark morphology for identification of common tree species and literature study of bark phytochemicals and therapeutic usage, *World Scientific News* 42 (2016) 143–155.
- [66] K. Šumberová, M. Ducháček, Analysis of plant seed banks and seed dispersal vectors: Its potential and limits for forensic investigations, *Forensic Sci. Int.* 270 (2017) 121–128, <https://doi.org/10.1016/j.forsciint.2016.11.030>.
- [67] K.N. Hopfensperger, A review of similarity between seed bank and standing vegetation across ecosystems, *Oikos* 116 (2007) 1438–1448, <https://doi.org/10.1111/j.0030-1299.2007.15818.x>.
- [68] K. Šumberová, Z. Lososová, M. Ducháček, V. Horáková, M. Fabšičová, Distribution, habitat ecology, soil seed bank and seed dispersal of threatened



- Lindernia procumbens* and alien *Lindernia dubia* (Antirrhinaceae) in the Czech Republic, *Phyton* 52 (2012) 39–72.
- [70] J. Roth, I. Feine, O. Waiskopf, R. Gafny, M. Amiel, Application of a Forensic DNA Extraction System for Cannabis sativa Seed Identification. 2021, *J. Biomolecul. Tech.* 32 4 10.7171/3fc1f5fe.3e117784.
- [71] C. Ladd, H.C. Lee, The use of biological and botanical evidence in criminal investigations, in: H. Miller Coyle (Ed.), *Forensic Botany: Principles and Applications to Criminal Casework*, CRC Press, 2005, pp. 97–115.
- [72] K. Retief, A.G. West, M.F. Pfab, Can stable isotopes and radiocarbon dating provide a forensic solution for curbing illegal harvesting of threatened cycads? *J. Forensic Sci.* 59 (2014) 1541–1551, <https://doi.org/10.1111/1556-4029.12644>.
- [73] UNODC, Outcome of the expert group meeting on timber analysis (10–12 December 2014). Commission on Crime Prevention and Criminal Justice — World Crime Trends and Emerging Issues and Responses in the Field of Crime Prevention and Criminal Justice, Vienna, 2015.
- [74] UNODC, *Best practice guide on forensic timber identification*, United Nations Office for Drugs and Crime, Vienna, 2016.
- [75] C. Balouet, J. Burken, J. Martelain, J. Lageard, F. Karg, D. Megson, Dendrochemical forensics as material evidence in courts: How could trees lie?, *Environ.* (2021) 1–7. <https://doi.org/10.1080/15275922.2021.1940381>.
- [76] J.C. Hermanson, A.C. Wiedenhoef, A brief review of machine vision in the context of automated wood identification systems, *IAWA J.* 32 (2) (2011) 233–250.
- [77] A. Koehler, Techniques Used in Tracing the Lindbergh Kidnaping Ladder, *J. Crim. Law Criminol.* 27 (1937) 712–724.
- [78] M. Tomaszewska, Z. Włodarczyk, M. Szela, Soltyszewski, Ślady pochodzenia botanicznego w ekspertyzach kryminalistycznych, *Prob. Krym.* 241 (2003) 16–22, in Polish.
- [79] M.M. Braga Junior, T.S. Matos, G.M. de Andrade, L.d.J. dos Santos, A.L.M. Vieira, T.A.P. Gonçalves, S. Nisgoski, J. Pereira Motta, L.E.d.L. Melo, O. Osunkoya, Forestry control in the Brazilian Amazon: charcoal anatomy of tree species from protected areas, *Aust. J. Bot.* 70 (1) (2021) 13–31.
- [80] M.M. Braga Júnior, F.I.B. de Souza, L.E. de Lima Melo, Forestry control in the Brazilian Amazon II: charcoal anatomy of 21 species, *IAWA J.* 42 (3) (2021) 299–321.
- [81] J. Pokines, Two cases of dendrochronology used to corroborate a forensic postmortem interval, *J. Forensic Identif.* 68 (2018) 457–465.
- [82] B. Yaman, U. Akkemik, The use of dendrochronological method in dating of illegal tree cuttings in Turkey: a case study, *Balt. For.* 15 (2009) 121–126.
- [83] P. Cherubini, Tree-ring dating of musical instruments, *Science* 373 (2021) 1434–1436, <https://doi.org/10.1126/science.abj3823>.
- [84] J. Topham, D. McCormick, FOCUS: A Dendrochronological Investigation of Stringed Instruments of the Cremonese School (1666–1757) including “The Messiah” violin attributed to Antonio Stradivari, *J. Archaeol. Sci.* 27 (2000) 183–192, <https://doi.org/10.1006/JAS.1999.0516>.
- [85] J.-H. Song, S. Yang, G. Choi, Taxonomic implications of leaf micromorphology using microscopic analysis: a tool for identification and authentication of Korean piperaleas, *Plants* 9 (2020) 566, <https://doi.org/10.3390/plants9050566>.
- [86] D.M. Bates, G.J. Anderson, R.D. Lee, Forensic botany: trichome evidence, *J. Forensic Sci.* 42 (1997) 380–386.
- [87] C. Prychid, P. Rudall, M. Gregory, Systematics and Biology of Silica Bodies in Monocotyledons, *Bot. Rev.* 69 (2003) 377–440, [https://doi.org/10.1663/0006-8101\(2004\)069\[0377:SABOSB\]2.0.CO;2](https://doi.org/10.1663/0006-8101(2004)069[0377:SABOSB]2.0.CO;2).
- [88] D.W. Hall, Introduction to forensic botany, in: D.W. Hall, J.H. Byrd (Eds.), *Forensic Botany: A Practical Guide*, 1st ed., Wiley, Blackwell, UK, 2012, pp. 1–12, <https://doi.org/10.1002/9781119945734.ch3>.
- [89] P.E.J. Wiltshire, D.L. Hawksworth, K.J. Edwards Light microscopy can reveal the consumption of a mixture of psychotropic plant and fungal material in suspicious death *J. Forensic Legal Med.* 34 2015 73–80 <https://doi.org/10.1016/j.jflm.2015.05.010>.
- [90] L. Oertel, A. Gressel, M.C. Torte, C. Cannet, L. Berthelon, A. Aboubacar, J.S. Raul, A. Farrugia, Be careful with lentils!, About a forensic observation *Int. J. Legal. Med.* 135 (2021) 323–327, <https://doi.org/10.1007/s00414-020-02389-8>.
- [91] E. Azzalini, M. Bernini, S. Vezzoli, A. Antonietti, A. Verzeletti, A fatal case of self-poisoning through the ingestion of oleander leaves, *J. Forensic Leg. Med.* 65 (2019) 133–136, <https://doi.org/10.1016/j.jflm.2019.05.016>.
- [92] G.S. Kim, S.H. Oh, C.S. Jang, Development of molecular markers to distinguish between morphologically similar edible plants and poisonous plants using a real-time PCR assay, *J. Sci. Food Agric.* 101 (2021) 1030–1037, <https://doi.org/10.1002/jsfa.10711>.
- [93] A. Palermo, M.E. Mascaro, The application of evaluation of leaf pigments in forensic science, in: 113th Congress of the Italian Botanical Society - V International Plant Science Conference (IPSC), 2018.
- [94] J.H. Bock, D.O. Norris, Cases using evidence from plant anatomy, in: J.H. Bock, D. O. Norris (Eds.), *Forensic Plant Science*, Elsevier Inc., 2016, pp. 85–94, <https://doi.org/10.1016/B978-0-12-801475-2.00005-1>.
- [95] P.E.J. Wiltshire, Forensic ecology, botany, and palynology: some aspects of their role in criminal investigation, in: K. Ritz, L. Dawson, D. Miller (Eds.), *Criminal and Environmental Soil Forensics*, Springer, Netherlands, Dordrecht, 2009, pp. 129–149, [https://doi.org/10.1007/978-1-4020-9204-6\\_9](https://doi.org/10.1007/978-1-4020-9204-6_9).
- [96] J. Fletcher, C. Bender, B. Budowle, W.T. Cobb, S.E. Gold, C.A. Ishimaru, D. Luster, U. Melcher, R. Murch, H. Scherm, R.C. Seem, J.L. Sherwood, B.W. Sobral, S. A. Tolin, Plant pathogen forensics: capabilities, needs, and recommendations, *Microbiol. Mol. Biol. Rev.* 70 (2006) 450–471, <https://doi.org/10.1128/MMBR.00022-05>.
- [97] S. Loppi, May the diversity of epiphytic lichens be used in environmental forensics? *Diversity* 11 (2019) 36, <https://doi.org/10.3390/d11030036>.
- [98] J. Anderson, N. Lévesque, F. Caron, P. Beckett, G.A. Spiers, A review on the use of lichens as a biomonitoring tool for environmental radioactivity, *J. Environ. Radioact.* 243 (2022), 106797, <https://doi.org/10.1016/j.jenvrad.2021.106797>.
- [99] J.L. Silván-Cárdenas, A. Caccavari-Garza, M.E. Quinto-Sánchez, J.M. Madrigal-Gómez, E. Coronado-Juárez, D. Quiroz-Suarez, Assessing optical remote sensing for grave detection, *Forensic Sci. Int.* 329 (2021), 111064, <https://doi.org/10.1016/j.forsciint.2021.111064>.
- [100] B. Rocke, A. Ruffell, Detection of single burials using multispectral drone data: three case studies, *Forensic Sci.* 2 (2022) 72–87, <https://doi.org/10.3390/forensicsci2010006>.
- [101] D.L. Hawksworth, P.E.J. Wiltshire, J.A. Webb, Rarely reported fungal spores and structures: an overlooked source of probative trace evidence in criminal investigations, *Forensic Sci. Int.* 264 (2016) 41–46, <https://doi.org/10.1016/j.forsciint.2016.02.047>.
- [102] E. Hösikler, Z. Erkol, S. Petekkaya, V. Gündoğdu, H. Samurcu, Fungal growth on a corpse: a case report, *Rom. J. Leg. Med.* 26 (2018) 158–161, <https://doi.org/10.4323/rjlm.2018.158>.
- [103] S. Karadayı, Assessment of the link between evidence and crime scene through soil bacterial and fungal microbiome: a mock case in forensic study, *Forensic Sci. Int.* 329 (2021), 111060, <https://doi.org/10.1016/j.forsciint.2021.111060>.
- [104] G. Guzmán, J. Allen, J. Gartz, A Worldwide geographical distribution of the Neurotropic Fungi, an analysis and discussion, *Ann. Mus. Civ. Rovereto* 14 (1998) 189–280.
- [105] J. Samad, The use of fungi in the criminal investigation process in Iran, *Forensic Sci. Today* (2020) 024–025.
- [106] N. Sagara, T. Yamanaka, M. Tibbett, Soil fungi associated with graves and latrines: toward a forensic mycology, in: M. Tibbett, D.O. Carter (Eds.), *Soil Analysis in Forensic Tophonomy*, CRC Press, Boca Raton, Florida, USA, 2008, pp. 67–108.
- [107] R.F. Lakatos, Examining the potential use of fungi in forensic science, Master of Science thesis, Department of Botany & Plant Pathology West Lafayette, Indiana (2019) [https://hammer.purdue.edu/articles/thesis/Examining\\_the\\_Potential\\_Use\\_of\\_Fungi\\_in\\_Forensic\\_Science/9037235](https://hammer.purdue.edu/articles/thesis/Examining_the_Potential_Use_of_Fungi_in_Forensic_Science/9037235).
- [108] D.L. Hawksworth, P. Wiltshire, Forensic mycology: current perspectives, *RRFMS* 5 (2015) 75–83, <https://doi.org/10.2147/RRFMS.S83169>.
- [109] J.L. Metcalf, L. Wegener Parfrey, A. Gonzalez, C.L. Lauber, D. Knights, G. Ackermann, G.C. Humphrey, M.J. Gebert, W. Van Treuren, D. Berg-Lyons, K. Keppers, Y. Guo, J. Bullard, N. Fierer, D.O. Carter, R. Knight, A microbial clock provides an accurate estimate of the postmortem interval in a mouse model system, *ELife* 2 (2013) e01104.
- [110] M. Dylag, Fungi present in home and their impact on human health - A short review, *Insights Biol. Med.* 1 (2017) 016–025, <https://doi.org/10.29328/journal.hjbm.1001003>.
- [111] [http://orzeczenia.olsztyn.so.gov.pl/content/\\$N/150515000000503\\_I\\_C.000689\\_2010\\_Uz.2014-03-27.003](http://orzeczenia.olsztyn.so.gov.pl/content/$N/150515000000503_I_C.000689_2010_Uz.2014-03-27.003). (in Polish).
- [112] M.K. Klassen-Fischer, Fungi as Bioweapons, *Clin. Lab. Med.* 26 (2006) 387–395, <https://doi.org/10.1016/j.cll.2006.03.008>.
- [113] M. Wheelis, R. Casagrande, L.V. Madden, Biological attack on agriculture: low-tech, high-impact bioterrorism, *Bioscience* 52 (2002) 569–589, [https://doi.org/10.1641/0006-3568\(2002\)052\[0569:BAOALT\]2.0.CO;2](https://doi.org/10.1641/0006-3568(2002)052[0569:BAOALT]2.0.CO;2).
- [114] R.R.M. Paterson, Fungi and fungal toxins as weapons, *Mycol. Res.* 110 (2006) 1003–1010, <https://doi.org/10.1016/j.mycres.2006.04.004>.
- [115] J.C.I. Lee, M. Cole, A. Linacre, Identification of members of the genera *Panaeolus* and *Psilocybe* by a DNA test: a preliminary test for hallucinogenic fungi, *Forensic Sci. Int.* 112 (2–3) (2000) 123–133, [https://doi.org/10.1016/S0379-0738\(00\)00181-X](https://doi.org/10.1016/S0379-0738(00)00181-X).
- [116] H.A. Raja, A.N. Miller, C.J. Pearce, N.H. Oberlies, Fungal identification using molecular tools: a primer for the natural products research community, *J. Nat. Prod.* 80 (3) (2017) 756–770, <https://doi.org/10.1021/acs.jnatprod.6b01085>.
- [117] J.S. Allwood, N. Fierer, R.R. Dunn, The future of environmental DNA in forensic science, *App. Environ. Microbiol.* 86 (21) (2020) e01504–e2019, <https://doi.org/10.1128/AEM.01504-19>.
- [118] D.L. Hawksworth, J.A. Webb, P.E.J. Wiltshire, CARYOSPORA CALLICARPA: Found in archaeological and modern preparations – but not collected since 1865, *Field Mycol.* 11 (2010) 55–59, <https://doi.org/10.1016/j.fldmyc.2010.04.008>.
- [119] C.E. Becker, T.G. Tong, U. Boerner, R.L. Roe, A.T. Scott, M.B. MacQuarrie, F. Bartter, Diagnosis and treatment of Amanita phalloides-type mushroom poisoning: use of thioctic acid, *West. J. Med.* 125 (1976) 100–109.
- [120] M. Kapala, A. Nowacka, M. Kicka, M. Rakowski, Mushroom (fungi) poisonings investigated at the regional centre of acute poisoning, institute of occupational medicine and environmental health, sosnowiec, poland, problems of, *Forensic Sci.* 75 (2008) 282–293.
- [121] A. Wołoszyn, R. Kotłowski, A universal method for the identification of genes encoding amatoxins and phallotoxins in poisonous mushrooms, *Rocz. Panstw. Zakł. Hig.* 68 (2017) 247–251. PMID: 28895390.
- [122] A.R. Laurence, V.M. Bryant, Forensic Palynology, in: G. Bruinsma, D. Weisburd (Eds.), *Encyclopedia of Criminology and Criminal Justice*, Springer New York, New York, NY, 2014, pp. 1741–1754, [https://doi.org/10.1007/978-1-4614-5690-2\\_169](https://doi.org/10.1007/978-1-4614-5690-2_169).
- [123] I. Kasprzyk, Airborne pollen of entomophilous plants and spores of pteridophytes in Rzeszów and its environs (SE Poland), *Aerobiologia* 20 (2004) 217–222, <https://doi.org/10.1007/s10453-004-1185-0>.



- [124] W. Haratym, E. Weryszko-Chmielewska, M. Dmitruk, An analysis of the content of pteridophyta spores in aeroplankton of Lublin (2013–2014), *Acta Agrobot.* 67 (2014) 21–28, <https://doi.org/10.5586/aa.2014.041>.
- [125] E. Zenkter, Morphology and peculiar features of spores of fern species occurring in Poland, *Acta Agrobot.* 65 (2012) 3–10, <https://doi.org/10.5586/aa.2012.053>.
- [126] G.M.A. Lashin, U.Y. Abo-Salama, E.M. Abd El Halim, G.A.A. Hamouda, G.A.A. Hamouda, G.A.A. Hamouda, Spore morphology of some Bryophyta in Egypt, *Plant Arch.* 21 (2021) 110–119, <https://doi.org/10.51470/PLANTARCHIVES.2021.v21.no1.015>.
- [127] E.A. Stanley, Application in palynology to establish the province and travel history of illicit drugs, *Microscope* 40 (1992) 149–152.
- [128] R.G. Bruce, M.E. Dettmann, Palynological analyses of Australian surface soils and their potential in forensic science, *Forensic Sci. Int.* 81 (1996) 77–94, [https://doi.org/10.1016/S0379-0738\(96\)01973-1](https://doi.org/10.1016/S0379-0738(96)01973-1).
- [129] A. Khokh, V. Shalaboda, Forensic comparative analysis of soils by the method of spore-pollen analysis using projection to latent structures discriminant analysis, *Theor. and Pract. of Foren. Sci. and Crim.* 23 (2021) 212–224, <https://doi.org/10.32353/khrife.1.2021.16>.
- [130] V.M. Bryant, D.C. Mildenhall, Forensic palynology: a new way to catch crooks, in: V.M. Bryant, J.H. Wrenn (Eds.), *New Developments in Palynomorph Sampling, Extraction, and Analysis*, AASP Foundation, 1998, pp. 145–155.
- [131] K.A.J. Walsh, M. Horrocks, Palynology: its position in the field of forensic science, *J. Forensic Sci.* 53 (2008) 1053–1060, <https://doi.org/10.1111/j.1556-4029.2008.00802.x>.
- [132] V.M. Bryant, G.D. Jones, Forensic palynology: current status of a rarely used technique in the united states of america, *Forensic Sci. Int.* 163 (2006) 183–197, <https://doi.org/10.1016/j.forsciint.2005.11.021>.
- [133] A.R. Laurence, V.M. Bryant, Forensic palynology and the search for geolocation: factors for analysis and the baby doe case, *Forensic Sci. Int.* 302 (2019), 109903, <https://doi.org/10.1016/j.forsciint.2019.109903>.
- [134] D.C. Mildenhall, Hypericum pollen determines the presence of burglars at the scene of a crime: an example of forensic palynology, *Forensic Sci. Int.* 163 (2006) 231–235, <https://doi.org/10.1016/j.forsciint.2005.11.028>.
- [135] M. Horrocks, K.A.J. Walsh, Forensic palynology: assessing the value of the evidence, *Rev. Palaeobot. Palynol.* 103 (1998) 69–74, [https://doi.org/10.1016/S0034-6667\(98\)00027-X](https://doi.org/10.1016/S0034-6667(98)00027-X).
- [136] D.C. Mildenhall, P.E.J. Wiltshire, V.M. Bryant, Forensic palynology: why do it and how it works, *Forensic Sci. Int.* 163 (2006) 163–172, <https://doi.org/10.1016/j.forsciint.2006.07.012>.
- [137] J. Ochando, M. Munuera, J.S. Carrión, S. Fernández, G. Amorós, J. Recalde, Forensic palynology revisited: case studies from semi-arid Spain, *Rev. Palaeobot. Palynol.* 259 (2018) 29–38, <https://doi.org/10.1016/j.revpalbo.2018.09.015>.
- [138] S. Warny, S. Ferguson, M.S. Hafner, G. Escarguel, Using museum pelt collections to generate pollen prints from high-risk regions: a new palynological forensic strategy for geolocation, *Forensic Sci. Internat.* 306 (2020), 110061, <https://doi.org/10.1016/j.forsciint.2019.110061>.
- [139] A.G. Brown, The use of forensic botany and geology in war crimes investigations in NE Bosnia, *Forensic Sci. Int.* 163 (2006) 204–210, <https://doi.org/10.1016/j.forsciint.2006.05.025>.
- [140] M.S. Zavada, S.M. McGraw, M.A. Miller, The role of clothing fabrics as passive pollen collectors in the north-eastern United States, *Grana* 46 (2007) 285–291, <https://doi.org/10.1080/00173130701780104>.
- [141] J.C. Webb, H.A. Brown, H. Toms, A.E. Goodenough, Differential retention of pollen grains on clothing and the effectiveness of laboratory retrieval methods in forensic settings, *Forensic Sci. Int.* 288 (2018) 36–45, <https://doi.org/10.1016/j.forsciint.2018.04.010>.
- [142] T. Chan, G. Robinson, J. Liu, M. Kurti, Y. He, K. Lampe, Identifying counterfeit cigarettes using environmental pollen analysis: an improved procedure, *J. Forensic Sci.* 65 (2020) 2138–2145, <https://doi.org/10.1111/1556-4029.14540>.
- [143] C.O. Hunt, Z. Morawska, Are your hands clean? Pollen retention on the human hand after washing, *Rev. Palaeobot. Palynol.* 280 (2020), 104278, <https://doi.org/10.1016/j.revpalbo.2020.104278>.
- [144] D.C. Mildenhall, The role of forensic palynology in sourcing the origin of falsified antimalarial pharmaceuticals, *Palynology* 41 (2017) 203–206, <https://doi.org/10.1080/01916122.2016.1156587>.
- [145] R. Szibor, C. Schubert, R. Schöning, D. Krause, U. Wendt, Pollen analysis reveals murder season, *Nature* 395 (1998) 449–450, <https://doi.org/10.1038/26646>.
- [146] B. Zimmermann, A. Kohler, T.J. Baskin, Infrared spectroscopy of pollen identifies plant species and genus as well as environmental conditions, *PLoS* 9 (4) (2014) e95417.
- [147] J. Depciuch, I. Kasprzyk, E. Drzymała, M. Parlińska-Wojtan, Identification of birch pollen species using FTIR spectroscopy, *Aerobiologia* 34 (4) (2018) 525–538, <https://doi.org/10.1007/s10453-018-9528-4>.
- [148] Y. Matsuki, Y. Isagi, Y. Suyama, The determination of multiple microsatellite genotypes and DNA sequences from a single pollen grain, *Mol. Ecol. Notes* 7 (2) (2007) 194–198, <https://doi.org/10.1111/j.1471-8286.2006.01588.x>.
- [149] K. Kraaijeveld, L.A. de Weger, M. Ventayol García, H. Buermans, J. Frank, P. S. Hiemstra, J.T. den Dunnen, Efficient and sensitive identification and quantification of airborne pollen using next-generation DNA sequencing, *Mol. Ecol. Resour.* 15 (1) (2015) 8–16.
- [150] W. Sickel, M.J. Ankenbrand, G. Grimmer, A. Holzschuh, S. Härtel, J. Lanzen, et al., Increased efficiency in identifying mixed pollen samples by meta-barcoding with a dualindexing approach, *BMC Ecol.* 15 (1) (2015) 20, <https://doi.org/10.1186/s12898-015-0051-y>.
- [151] K.L. Bell, N. de Vere, A. Keller, R.T. Richardson, A. Gous, K.S. Burgess, B.J. Brosi, Pollen DNA barcoding: current applications and future prospects, *Genome* 59 (2016) 629–640, <https://doi.org/10.1139/gen-2015-0200>.
- [152] K.R. Scott, R.M. Morgan, V.J. Jones, A. Dudley, N. Cameron, Peter A bull the value of an empirical approach for the assessment of diatoms as environmental trace evidence in forensic limnology, *Arch. Environ. Forensic Sci.* 1 (1) (2016) 49–78, <https://doi.org/10.1558/aefs.32474>.
- [153] N.G. Cameron, The use of diatom analysis in forensic geoscience, *Geol. Soc. London Spec. Publ.* 232 (2004) 277–280, <https://doi.org/10.1144/GSL.SP.2004.232.01.25>.
- [154] I. Bogusz, M. Bogusz, J. Żelazna-Wieczorek, Diatoms from inland aquatic and soil habitats as indestructible and nonremovable forensic environmental evidence, *J. Forensic Sci.* 00 (2022) 1–15, <https://doi.org/10.1111/1556-4029.15017>.
- [155] K.R. Scott, R.M. Morgan, V.J. Jones, N.G. Cameron, The transferability of diatoms to clothing and the methods appropriate for their collection and analysis in forensic geoscience, *Forensic Sci. Internat.* 241 (2014) 127–137, <https://doi.org/10.1016/j.forsciint.2014.05.011>.
- [156] K.R. Scott, V.J. Jones, N.G. Cameron, J.M. Young, R.M. Morgan, Freshwater diatom persistence on clothing I: a quantitative assessment of trace evidence dynamics over time, *Forensic Sci. Int.* 325 (2021), 110898, <https://doi.org/10.1016/j.forsciint.2021.110898>.
- [157] M.K. Sharma, A. Sharma, A.K. Bhatt, Diatom, a phytoplanktonic community study from different water bodies of District Hamirpur, Himachal Pradesh: Role in forensic studies and biotechnological innovations, *IJLR* 14 (2021) 93–111.
- [158] E.A. Levin, R.M. Morgan, K.R. Scott, V.J. Jones, The transfer of diatoms from freshwater to footwear materials: an experimental study assessing transfer, persistence, and extraction methods for forensic reconstruction, *Sci. Justice* 57 (2017) 349–360, <https://doi.org/10.1016/j.scijus.2017.05.005>.
- [159] K.R. Scott, R.M. Morgan, V.J. Jones, A. Dudley, N. Cameron, P.A. Bull, The value of an empirical approach for the assessment of diatoms as environmental trace evidence in forensic limnology, *AEFS* 1 (2017) 49–78, <https://doi.org/10.1558/aefs.32474>.
- [160] P.A. Siver, W.D. Lord, D.J. McCarthy, Forensic limnology: the use of freshwater algal community ecology to link suspects to an aquatic crime scene in southern new england, *J. Forensic Sci.* 39 (1994) 847–853, <https://doi.org/10.1520/JFS13663J>.
- [161] P. Zhang, X. Kang, S. Zhang, C. Xiao, Y. Ma, H. Shi, Q. Xu, J. Zhao, L. Chen, C. Liu, The length and width of diatoms in drowning cases as the evidence of diatoms penetrating the alveoli-capillary barrier, *Int. J. Leg. Med.* 134 (2020) 1037–1042, <https://doi.org/10.1007/s00414-019-02164-4>.
- [162] T. Smijs, F. Galli, A. van Asten, Forensic potential of atomic force microscopy, *Forensic Chem* 2 (2016) 93–104, <https://doi.org/10.1016/j.forc.2016.10.005>.
- [163] V.K. Yadavalli, C.J. Ehrhardt, Atomic force microscopy as a biophysical tool for nanoscale forensic investigations, *Sci. Justice* 61 (1) (2021) 1–12, <https://doi.org/10.1016/j.scijus.2020.10.004>.
- [164] Z. Yu, Q. Xu, C. Xiao, H. Li, W. Wu, W. Du, J. Zhao, H. Liu, H. Wang, C. Liu, SYBR Green real-time qPCR method: diagnose drowning more rapidly and accurately, *Forensic Sci. Int.* 321 (2021), 110720, <https://doi.org/10.1016/j.forsciint.2021.110720>.
- [165] Z. Li, X. Liu, Y. Yanfang, H. Huang, X. Li, Q. Ji, K. Li, Y. Youjia, D. Li, Z. Mao, P. u. Yan, P. Chen, F. Chen, Barcoding for diatoms in the Yangtze River from the morphological observation and 18S rDNA polymorphic analysis, *Forensic Sci. Int.* 97 (2019) 81–89, <https://doi.org/10.1016/j.forsciint.2019.01.028>.
- [166] A. Khurshid, M.U. Shah, M. Khurshid, A. Sohail, G. Ali, Diatom-positive cadaver: drowning or homicide? *Cureus* 13 (2021) e18312.
- [167] G. Piegari, D. De Biase, I. d'Aquino, F. Prisco, R. Fico, R. Ilsami, N. Pozzato, A. Genoveso, O. Paciello, Diagnosis of drowning and the value of the diatom test in veterinary forensic pathology, *Front Vet. Sci.* 6 (2019) 404, <https://doi.org/10.3389/fvets.2019.00404>.
- [168] A.H. Magrey, M. Raj, Role of diatoms in forensic diagnosis of drowning cases from Jammu & Kashmir, India, *Biosci. Biotech. Res. Comm.* 7 (2014) 72–77.
- [169] J. Zhao, Y. Ma, C. Liu, J. Wen, S. Hu, H. Shi, L. Zhu, A quantitative comparison analysis of diatoms in the lung tissues and the drowning medium as an indicator of drowning, *J. Forensic Leg. Med.* 42 (2016) 75–78, <https://doi.org/10.1016/j.jflm.2016.05.021>.
- [170] Y. Zhou, Y. Cao, J. Huang, K. Deng, K. Ma, T. Zhang, L. Chen, J. Zhang, P. Huang, Research advances in forensic diatom testing, *Forensic Sci. Res.* 5 (2020) 98–105, <https://doi.org/10.1080/20961790.2020.1718901>.
- [171] X. Shen, Y. Liu, C. Xiao, C. Zheng, J. Huang, H. Shi, Q. Xu, J. Cheng, C. Liu, J. Zhao, Analysis of false-positive results of diatom test in the diagnosis of drowning — would not be an impediment, *Int. J. Leg. Med.* 133 (2019) 1819–1824, <https://doi.org/10.1007/s00414-019-02021-4>.
- [172] M.H.A. Piette, E.A. De Letter, Drowning: still a difficult autopsy diagnosis, *Forensic Sci. Int.* 163 (2006) 1–9, <https://doi.org/10.1016/j.forsciint.2004.10.027>.
- [173] J.V. Pachar, J.M. Cameron, submersion cases: a retrospective study —1988–1990, *Med. Sci. Law* 32 (1992) 15–17, <https://doi.org/10.1177/002580249203200105>.
- [174] Y. Du, J. Liu, Q. Li, H. Li, X. Kang, D. Zheng, H. Shi, Q. Xu, C. Liu, H. Wang, J. Zhao, Concordance analysis of diatom types and patterns in lung tissue and drowning medium in laboratory animal model, *Int. J. Leg. Med.* 136 (2022) 911–917, <https://doi.org/10.1007/s00414-021-02768-9>.
- [175] Z. Li, W. u. Bo, X. Cheng, W. u. Yunying, P. Zhang, H. e. Shi, D. Zheng, J. Cheng, C. Liu, J. Zhao, Evaluation of L/D ratio in a water-related case for the differentiation between drowning and postmortem immersion, *Forensic Sci. Int.: Synergy* 1 (2019) 68–70, <https://doi.org/10.1016/j.fsisy.2019.04.001>.

- [176] Y Liu, Y Zhao, Y Sun, P Li, S Wu, L Zhou, Ren, Comparative study on diatom morphology and molecular identification in drowning cases, *Forensic Sci. Int.* 317 (2020) 110552, <https://doi.org/10.1016/j.forsciint.2020.110552>.
- [177] K.A. Zimmerman, J.R. Wallace, The potential to determine a postmortem submersion interval based on algal diatom diversity on decomposing mammalian carcasses in brackish ponds in Delaware, *J. Forensic Sci.* 53 (2008) 935–941, <https://doi.org/10.1111/j.1556-4029.2008.00748.x>.
- [178] N. Ishikawa, Y. Miake, K. Kitamura, H. Yamamoto, A new method for estimating time since death by analysis of substances deposited on the surface of dental enamel in a body immersed in seawater, *Int. J. Leg. Med.* 133 (2019) 1421–1427, <https://doi.org/10.1007/s00414-019-02020-5>.
- [179] M. Wanner, E. Betker, S. Shimano, R. Krawczynski, Are soil testate amoebae and diatoms useful for forensics? *Forensic Sci. Int.* 289 (2018) 223–231, <https://doi.org/10.1016/j.forsciint.2018.05.027>.
- [180] M.K. Thakar, D. Luthra, J.S. Khattar, Forensic studies of phytoplankton ecology of two water bodies of Kurukshetra area of Haryana, State in India, *Egypt J. Forensic Sci.* 8 (2018) 38, <https://doi.org/10.1186/s41935-018-0068-4>.
- [181] P.A. Diaz-Palma, A. Alucema, G. Hayashida, N.I. Maidana, Development and standardization of a microalgae test for determining deaths by drowning, *Forensic Sci. Int.* 184 (2009) 37–41, <https://doi.org/10.1016/j.forsciint.2008.11.015>.
- [182] J.S. Rowan, S. Black, S.W. Franks, Sediment fingerprinting as an environmental forensics tool explaining cyanobacteria blooms in lakes, *Appl. Geogr.* 32 (2012) 832–843, <https://doi.org/10.1016/j.apgeog.2011.07.004>.
- [183] J.S. Metcalf, G.A. Codd, Co-occurrence of cyanobacteria and cyanotoxins with other environmental health hazards: impacts and implications, *Toxins* 12 (2020) 629, <https://doi.org/10.3390/toxins12100629>.
- [184] Szelec Can soil testate amoebae be used for estimating the time of death? A field experiment in deciduous forest, *Forensic Sci. Int.* 236 (2014) 90–98, <https://doi.org/10.1016/j.forsciint.2013.12.030>.
- [185] G.T. Swindles, A. Ruffell, A preliminary investigation into the use of testate amoebae for the discrimination of forensic soil samples, *Sci. Justice* 49 (2009) 182–190, <https://doi.org/10.1016/j.scijus.2008.11.002>.
- [186] M. Zieger, One number, two values? *Forensic Sci. Int.* 321 (2021), 110725, <https://doi.org/10.1016/j.forsciint.2021.110725>.
- [187] C.G.G. Aitken, Bayesian hierarchical random effects models in forensic science, *Front. Genet.* 9 (2018) 126, <https://doi.org/10.3389/fgene.2018.00126>.
- [188] K.A. Martire, R.I. Kemp, M. Sayle, B.R. Newell, On the interpretation of likelihood ratios in forensic scene evidence: presentation formats and the weak evidence effect, *Forensic Sci. Int.* 240 (2014) 61–68, <https://doi.org/10.1016/j.forsciint.2014.04.005>.
- [189] G.S. Morrison, E. Enzinger, What should a forensic practitioner's likelihood ratio be? *Sci. Justice* 56 (2016) 374–379, <https://doi.org/10.1016/j.scijus.2016.05.007>.
- [190] V. Spikmans, The evolution of environmental forensics: From laboratory to field analysis, *WIREs Forensic Sci.* 1 (2019) e1334.