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ORIGINAL RESEARCH PAPER

Mycological and palynological studies of early medieval cultural layers from strongholds in Pszczew and Santok (western Poland)

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Abstract

Cultural layers from early medieval strongholds in Pszczew and Santok have been examined for the presence of pollen grains and spores as well as residues of fungi. The presence of the following remains has been recorded: fossil hyphopodia of *Gaeumannomyces*, teliospores of *Puccinia*, spores of *Bipolaris*, *Thecaphora* and *Tilletia*, teliospores of the genus *Urocystis*, *Ustilago* and *Uromyces*, ascocarps (perithecium) of the Ascomycota or the pycnidium of Sphaeropsidales. A greater diversity and abundance of fungi spores sensu lato was recorded in Santok, as compared to Pszczew. Both early medieval sites recorded a significant proportion of cereal pollen, including *Secale cereale*. It remains an undisputed fact that the grains and other plants collected in both strongholds were strongly infected with fungi. The analysis of the cultural layers for the presence of fungi remains provides significant data on the presence of certain species of plants and their growth conditions in natural environments and in agriculture.

Keywords

fossil micro-remains; fungi; palynological analysis; cultural layers

This issue of Acta Mycologica is dedicated to Professor Maria Lisiewska and Professor Anna Bujakiewicz on the occasion of their 80th and 75th birthday, respectively.

Introduction

The subject of palynological research is primarily pollen grains and spores preserved separate from plants. In addition to plant micro-remains, samples of palynological material very often – and sometimes in abundance – contain other remains [1–4], with various systematic groups of fungi sensu lato [5] in several developmental stages of among them. Particularly intense development of the interest into the palaeomycological matters can be dated back to the second half of the 20th century [6–9]. Multidisciplinary research combining palynological and mycological analyses has been also carried out in Poland [2,10]. In this respect, the interdisciplinary mycological and palynological research not only completes the reconstruction of the palaeoenvironment, but it can also constitute an additional element pointing to, among other things, the hazards to the health of the plants from phytopathogenic fungi and the resulting consequences for the societies. Within the process of comprehensive reconstruction of early medieval living conditions, actions have therefore been taken to identify the fungi present in the respective cultural layers. The aim of the present paper is to answer the following question: What phytopathogenic fungi existed on agriculture plants in early medieval times?

Material and methods

Characteristics of the research area

The research material was obtained in the years 2009 through 2012 during excavations in the early medieval strongholds in Pszczew (φ 52°47′ N, λ 15°77′ E) and Santok (φ 52°74′ N, λ 15°42′ E), situated in the north-western part of the Wielkopolska Lowland.

Pszczew is situated in the Lubuska Highland inside the micro-region of Pagórki Świebodzińsko-Sulęcińskie [11]. The fieldwork was carried out in the south-eastern part of Lake Miejskie, also known as Lake Pszczewskie. The samples for the environmental studies were collected during an archaeological penetration of an early medieval stronghold (9/10th century) on the "Katarzyna" Peninsula.

The stronghold in Santok is located in the Gorzowska Valley on a small part of a fluvial terrace 25–26 meters above the sea level [11], not far from the place where the Noteć River connects with the Warta River. These rivers flow into the world's biggest ice marginal streamway, i.e., the Toruń–Eberswalde Pradolina (Toruń–Eberswalde ice-marginal streamway). In the excavation aimed at obtaining wood for dendrochronological and radiocarbon dating purposes [12], sediments were collected at settlement levels I (from the second half of the 8th century until the 9th century), II (first half of the 9th century) and III (from the third quarter of the 9th century until the turn of the third and the fourth quarter of the 9th century) for the analyses of pollen and fungal microscopic remains.

Research methodology

Sediments for pollen and mycological analyses were taken in iron boxes directly from the wall during the archaeological excavation. The specific nature of the cultural layer allowed for diversification of the sampling intervals (from 2 up to 7 cm). Finally, 22 samples from Pszczew and 13 samples from Santok sites were used for palynological interpretation. In the remaining sediment spectra the sporomorhs were either very badly preserved or totally destroyed. The samples collected for palynological analysis underwent standard chemical treatment using the Erdtman acetolysis method [13]. Organic micro-remains (pollen grains of trees and shrubs, herbaceous plants, all accompanying sporomorphs and other non-pollen palynomorphs such as *Botryococcus*, Bryales, *Filinia*, fungi, testate amoebae, *Trichuris* and Turbellaria) were identified using Ergaval (640×) and Olympus (600×) microscopes, with the use of necessary keys [14–18] and a collection of preparations for comparison.

The POLPAL computer program was used to juxtapose the data, perform the percentage calculations and create the diagrams [19].

Results

Pszczew

Three cores of sediments for palynological analysis were collected from the archaeological excavations at the site in Pszczew [20]. It was demonstrated that the pollen of *Pinus sylvestris* dominated there (up to 88%), with a significantly smaller pollen proportion (a few percent) of *Tilia*, *Ulmus*, *Corylus avellana*, *Betula*, *Quercus* and *Alnus*, and the occasional presence of *Fraxinus*, *Populus*, *Picea*, *Fagus sylvatica* and *Carpinus betulus*, i.e., in isolated samples and with values below 1% (Fig. 1).

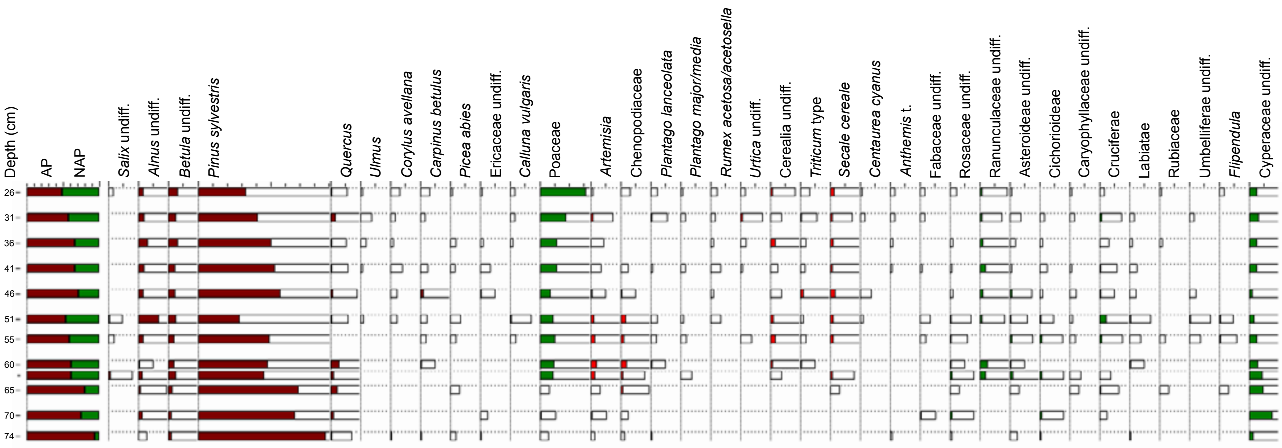

Fig. 1 Pszczew – core 2. Simplified percentage pollen diagram (analysis: Okuniewska-Nowalczyk 2012).

As far as the NAP plants (non-arborum pollen, pollen of non-trees, i.e., pollen of herbaceous plants and dwarf shrubs) are concerned, the analyzed spectra recorded higher quantities of the pollen of Poaceae (32%) as well as a large variety of pollen of plants that indicate human activity. They were represented here by Chenopodiaceae, *Artemisia*, *Plantago lanceolata*, *Plantago maior/media*, *Rumex acetosa/acetosella*, *Urtica*, *Centaurea cyanus*, *Secale cereale*, *Triticum* and other Cerealia (undetermined). The maximum frequency of grains of each taxon varied and ranged from 1.6% for *Urtica* (core 2) to 10% for Cerealia (core 3) and 13.5% for *Plantago lanceolate* (core 1).

During palynological analysis of the spectra, the presence of spores and endospores was determined, thus proving the existence of fungi among the organic remains (Fig. 2). Particularly important are the remains that point to the occurrence of phytopathogenic fungi, which are related to crops, especially cereals. It is concerned with identified hyphopodium that is probably produced by *Gaeumannomyces* (Fig. 2a). Another type of phytopathogens that were identified on the basis of characteristic over-wintering spores (teliospores) is fungi representing Uredinales, genus *Puccinia* (Fig. 2b).

Apart from rust fungi, the presence of other phytopathogenic species representing the Ustilaginales has been identified in the analyzed sediments. One of them is likely to be the genus *Thecaphora*, producing spore balls containing 1–25 cells, loosely or closely connected (Fig. 2c,d). Another representative of the Ustilaginales, which has been identified with high probability, are the spores (ustilospores) of the smut fungus from the genus *Tilletia*, which causes Karnal bunt. The ustilospores of *Tilletia* are the largest spores of all the Ustilaginaceae. Another distinctive feature of these spores is their net-like pattern of the surface, also visible in the preparation (Fig. 2e).

Santok

On the basis of the palynological analysis, domination of NAP taxa has been determined at all settlement levels in Santok (I–III). Predominating presence of the representatives of the following families has been shown: Chenopodiaceae, Fabaceae and Poaceae, including cereals, such as undetermined Cerealia and *Triticum*, but mostly *Secale cereale*. The largest volume of pollen of all aforementioned taxa was recorded for the end of the first settlement phase, probably at the beginning of the 9th century [12]. A higher proportion of weed pollen (Fig. 3) and endospores of Uredinales, among others, from the genus *Puccinia* (Fig. 4) was also associated with this period.

On level II, from the first half of the 9th century, the percentage values of cereals Cerealia (max. 13.5%), *Triticum* (max. 7.0%) and particularly *Secale cereale* (max. 22.0%) reached their maximum (Fig. 3). At the same time, the palynological spectra of this archaeological stratum recorded the largest variety of fungi (Fig. 4).

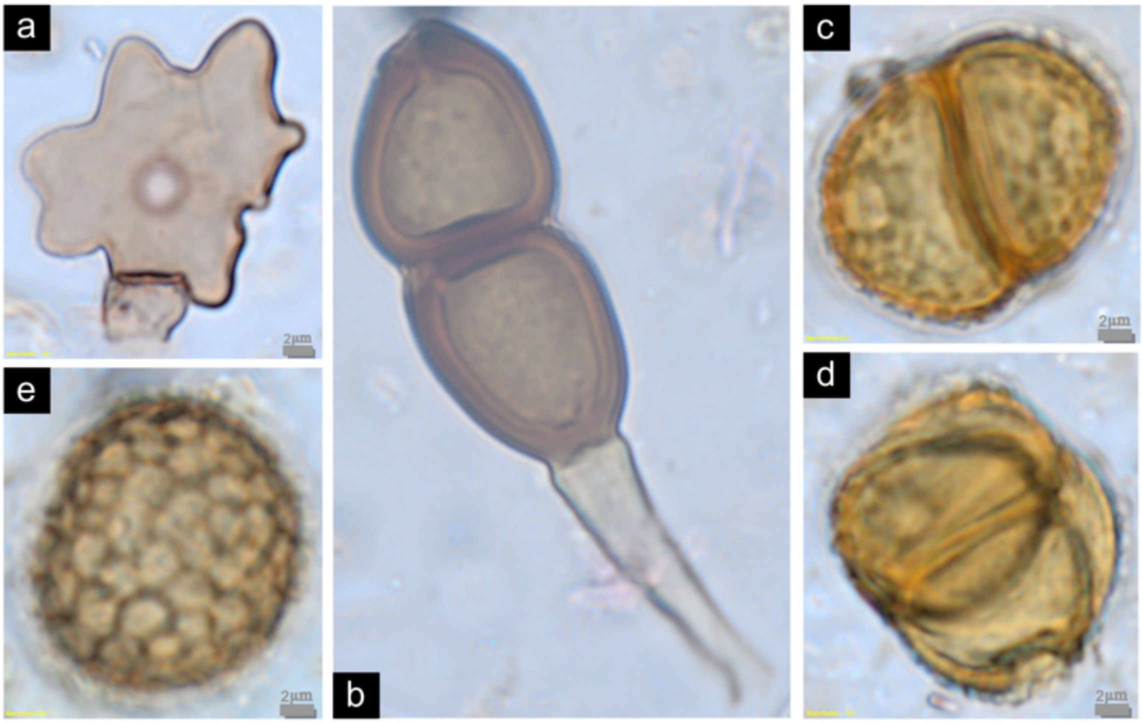

Fig. 2 Fungal spores and endospores identified in the organic material from the excavations in Pszczew. **a** Fossil hyphopodium of *Gaeumannomyces*. **b** Teliospore of *Puccinia*. **c,d** Spores of *Thecaphora*. **e** Ustilospore (chlamydospore) of *Tilletia*.

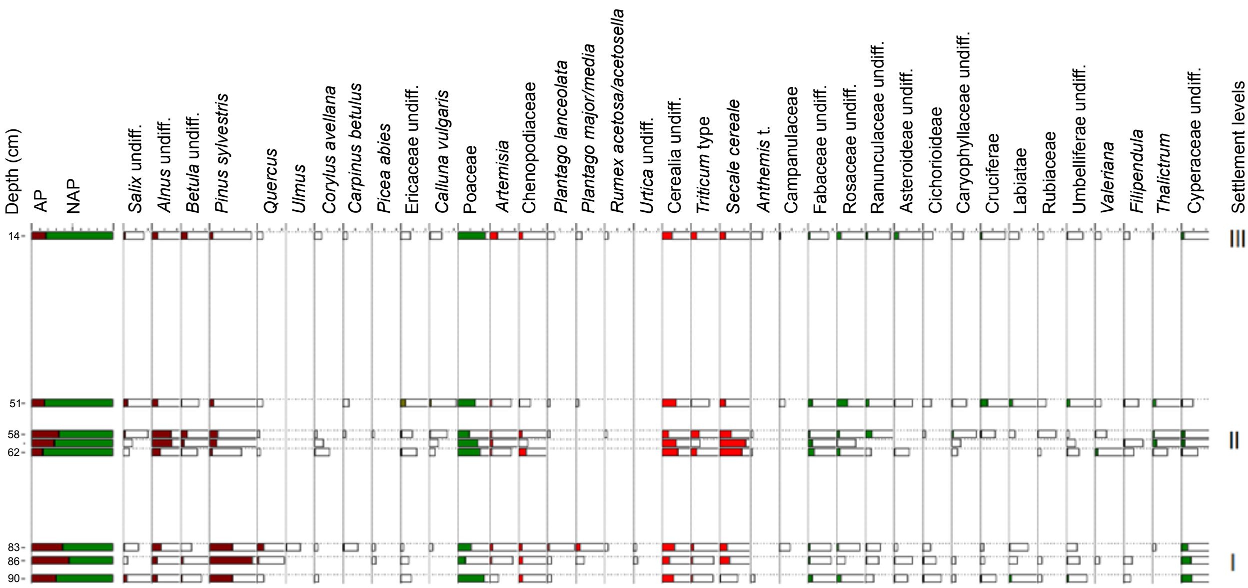

Fig. 3 Santok – site 1, excavation 2, northern section. Simplified percentage pollen diagram (analysis: Okuniewska-Nowaczyk 2012).

The most recent level (III, second half of the 9th century) was distinctive for the richness of taxa, including specimens that were not present in the settlement levels discussed previously. The diversity of plant species was enriched by such taxa as *Carduus typ*, *Centaurea jacea*, *Cirsium typ*, *Malva*, *Spergula arvensis*, *Xanthum* cf. and *Alisma plantago aquatica*.

Santok recorded both a greater diversity (10 taxa) and abundance of fungi spores sensu lato than Pszczew (4 taxa). Examples of identified fungi at both sites are presented on plate (Fig. 2, Fig. 5). As in Pszczew, structures resembling hyphopodium

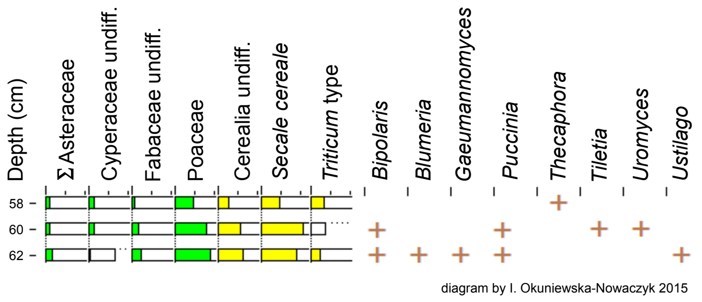

Fig. 4 Santok. Occurence of fungi genera in relation to selected NAP taxa (settlement level II).

of *Gaeumannomyces*, teliospores of *Puccinia* as well as spores of the genus *Tilletia* and *Tecaphora* were identified among the mycological remains (Fig. 2, Fig. 5). Most likely chlamydospores (ustilospores) of Ustilaginales other than *Tilletia* and *Urocystis*, presumably belonging to the genus *Ustilago* (Fig. 5f), were found among the organic remains in Santok. Other fungal taxa, not found in Pszczew but possibly present in Santok, are the genera *Bipolaris* and *Uromyces*, identified on the basis of preserved remains. Based on the fossil elements of fungal structures, it is difficult to identify fungi that produce morphological forms that are likely to be either the ascocarp (perithecium) of Ascomycota or the conidium (pycnidium) of Sphaeropsidales (Fig. 5c).

It is worth mentioning that what was richly represented in the remains from Santok were the spores (teliospores) characteristic of Uredinales. In addition to two-celled teliospores of the genus *Puccinia*, morphologically various shapes of single-celled, thick-walled spores were identified here, which are probably also teliospores of Uredinales from the genus *Uromyces* (Fig. 5h,i). As all Uredinales, they are known as obligate plant parasites [21,22].

Discussion

In the cultural layers of both strongholds, not all fungal remains were preserved well enough and sufficiently representative for their species or genus to be marked. The difficulty of unambiguously identifying various fossil fungi has been pointed out by many researchers, who use type and number to describe the collected fungal remains [5,7,23]. Nevertheless, there are also works in which ascopores, darkly tinted conidia or various fungal endospores constitute the basis for unambiguous designation. In the authors' own research presented here, the interpretation was applied only to specimens that could be most likely attributed to a genus by considering their basic characteristics, i.e., the size and the morphology of the observed forms. However, no account has been taken of a large number of palynological preparations in which the structures of the microscopic fungi were either fragmented or damaged. Macroscopic plant remains are the most commonly examined by archaeobotanists in archeological studies, which makes it then possible to identify larger fungal structures, such as *Claviceps* from the Develier-Courtételle site in north-western Switzerland, where even parts of the species of fungi of that type can be used for conclusive taxonomic designations [24]. Identifying the largest possible taxonomic diversity of fungi in cultural layers, especially phytopathogenic fungi, is of great importance in reconstructing the character of the communities of different eras. In the early medieval times, the scope of human agricultural activity was limited by the scale of danger to farm animals and crops. At the time, the most popular crops were the cereals [25,26]. This is confirmed by the palynological analyses of the cultural layers in Pszczew and Santok, indicating a significant proportion of grass pollen, among them cereals, including *Secale cereale* and *Triticum*. The low level of agriculture of the time certainly created favorable conditions for pests and phytopathogenic fungi to spread, which was indirectly confirmed by this research. What seems to be particularly important is – as determined in both sites – the presence of teliospores of the genus *Puccinia*, belonging to obligate parasites existing only on live flowering plants [21,22]. Since among Uredinales the genus *Puccinia* is currently most widely represented (ca. 4000 species), it can be assumed that these fungi were equally common in previous eras, when their host plants were present from the families: Asteraceae, Cyperaceae and Poaceae. This assumption is confirmed by the simultaneous presence of the pollen of the aforementioned plants and the teliospores of *Puccinia* in the analyzed material, e.g., on the settlement level II in Santok (Fig. 4). As far as farming is concerned, the

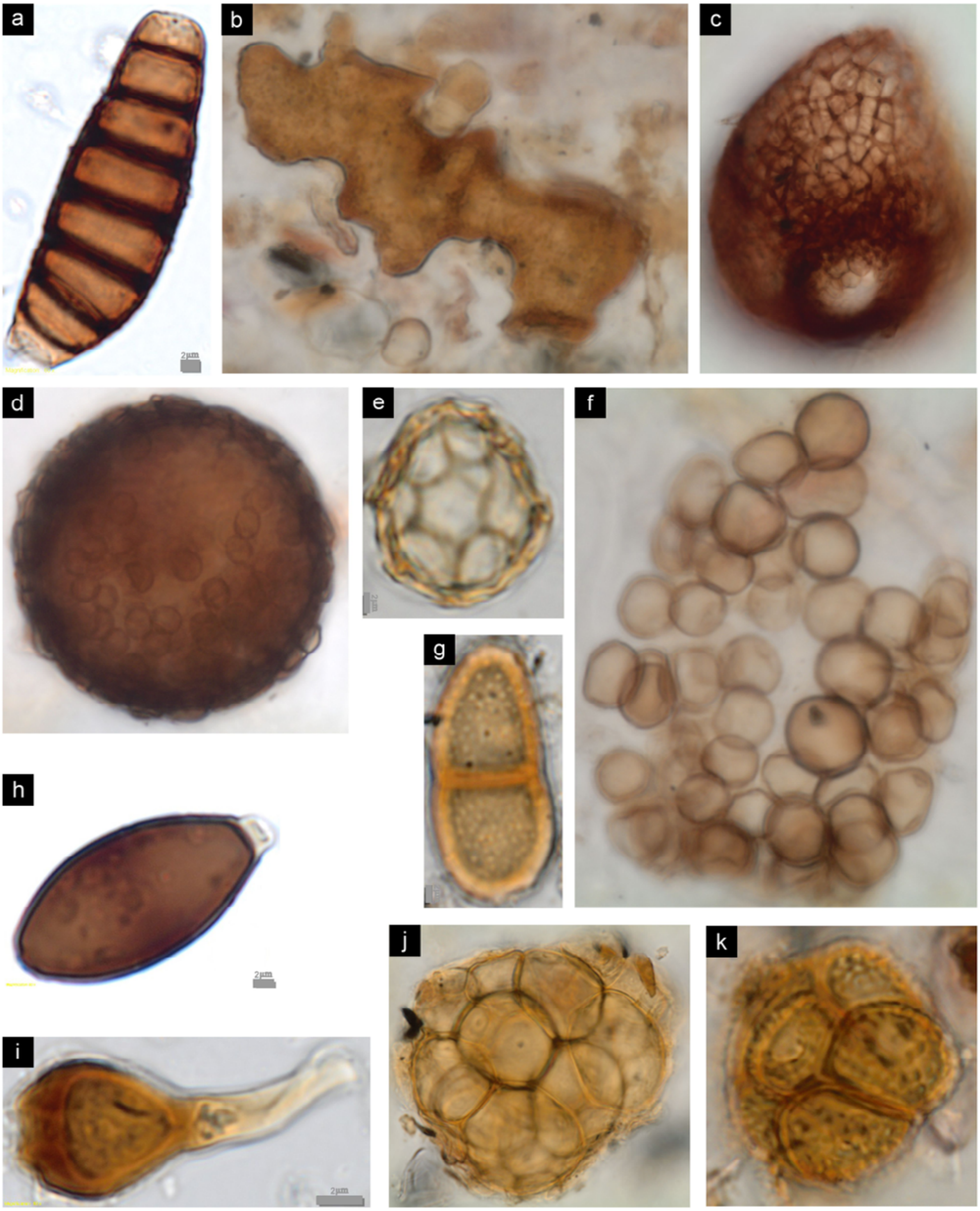

Fig. 5 Santok. Morphotic structures of fungi sensu lato. **a** Spore of the genus *Bipolaris*. **b** Hyphopodium of *Gaeumannomyces*. **c** Ascocarp: perithecium of Ascomycota or pycnidium of Spaheropsidales. **d** Teliospores of the genus *Urocystis*. **e** Chlamidospore of *Tilletia*. **f** *Ustilago*. **g** Teliospore of *Puccinia*. **h,i** Teliospores of *Uromyces*. **j,k** Spores of *Thecaphora*.

most important phytopathogenic species from the genus *Puccinia* are the species that attack grassy plants, particularly cereals, which were represented in the palynological remains primarily by the pollen of *Secale cereale* and *Triticum*. These fungi particularly attack the aboveground vegetative organs and during epiphytosis might reduce the yield by even 40–50% [27,28]. In addition to the genus *Puccinia*, representatives of other rust genera from the genus *Uromyces* were observed among the analyzed remains (albeit at the site in Santok). This is proved by numerous, morphologically varied yet always single-celled teliospores. Fungi of the genus *Uromyces* attack both monocots and dicots of the families Asteraeae, Euphorbiaceae, Liliaceae, Poaceae and Fabaceae. In this research, they most likely attacked host plants belonging to Poaceae or Fabaceae, since the presence of the pollen of these plants was identified (Fig. 4). The fact that it is plants of the family Fabaceae that are predominantly affected allows for an assumption that the specialization of the Uromyces pathogens might have been similar in the Middle Ages.

According to interpretations of many authors, connecting the occurrence of host plants and their phytopathogens is used to explain various environmental (e.g., palaeoecological and palaeoclimatic) changes. One such example might be found in the spores of the fungus type 10, whose high percentage values correlate with a high proportion of the pollen of Ericales [5]. This fungus occurs on the roots of *Calluna vulgaris*, which is associated with relatively dry moors. When the environment is humid, the proportion of these type of spores is low. According to van Geel [7], the presence of other spores, i.e., ascospores of *Neurospora* sp. (type 55C), points to local peat bog fires. Fungal infection might also cause changes in the dynamics of tree appearance, which is demonstrated in the decrease of the tree stands with linden as a result of infestation of *Kretzschmaria deusta* (*Ustulina deusta*) in the Middle Holocene in the area of Gołębiewo [10].

The presence of fungal remains was also recorded during the palynological analysis of sediments of different origin. Mycelium of fungi, sometimes in large quantities, were found in the analysis of the humus horizon. This fact can indirectly attest to the presence of certain genera and species of wild plants and plants cultivated by man. Such indicators are fossil hyphopodia *Gaeumannomyces* cf. *caricis*, which can serve to distinguish *Carex* among other vegetative remains [8]. Also in this research, hyphopodia characteristic of the genus *Gaeumannomyces* were found among the remains from the settlement layers. These fungi are currently represented by five common species that are parasites on the root and the stem base of grass and cereal in all regions of cultivation in temperate climates around the world. Phytopathogens of that kind cause a disease called stem base rot, which is particularly dangerous for wheat, which is grown in light and sandy soils [27,29,30]. It can be thus assumed that also in the early medieval times. *Gaeumannomyces* significantly contributed to infecting wheat, breaking its stems and consequently reducing its yield. Research has shown that the health and the yield of wheat was at risk, not only because of the infections of the vegetative parts of plants, but also because of the phytopathogens that would utterly destroy their generative parts, i.e., caryopses. At both sites (Pszczew and Santok), this is proved by the occurrence of spores of the Ustilaginales, genus *Tilletia*, which are phytopathogens associated mostly with grasses, also including cereals. The determined spores presumably belong to one of the species *Tilletia caries* or *Tilletia controversa*. *Tilletia caries* is a specialized wheat pathogen that causes Karnal bunt and completely destroys the endosperm in caryopses, leaving only the bran intact [17,31,32]; stunted wheat stems are also one of its effects. The other species, *Tilletia controversa*, causes dwarf bunt of wheat but also infects barley, rye and other grass species, whose seeds it also destroys. Other Ustilaginales that probably contribute to the reduction in the yield of cereals in the medieval period in question are also species from the genus *Ustilago*. All of the 17 species of these fungi described in the literature in Poland [33] infect only Poaceae. Some of them (*Ustilago tritici*) develop systemically and cause total destruction of caryopsis in the ear, i.e., total reduction of yield, particularly in wheat and barley. It is difficult to speculate which species has been identified in the study, since the chlamydospores (ustilospores) of different species of Ustilaginales (*Ustilago hordei*, *U. maydis*, *U. striformis*) are very similar to one another. The studies also determined the spores resembling *Tecaphora*, i.e., the smut fungi Ustilaginales occurring in weeds of the genus *Calystegia* [34], *Carduus* and also *Cirsium*, i.e., in the species present also in cereal crops [35]. The significant turnout of weed pollen might also reflect fallowing of fields [2].

Greater species diversity of phytopathogens at the site in Santok is confirmed by the presence of the spores resembling *Bipolaris* (*Helminthosporium*). This pathogen infects seedlings of cereals, particularly in cold and humid regions, which creates favorable conditions for over-extending the critical phase of seedling germination [36,37]. In warmer regions, helminthosporosis is the most common disease of oat leaves.

The phytopathogens, particularly cereal pathogens, identified in both strongholds not only confirm the universality of their occurrence but also indicate that as far as reconstructions of palaeoenvironment are concerned, the mycological studies, however still undervalued, are of great importance. For a variety of experts dealing with the past (archaeobotanists, palynologists) as well as humanists (archaeologists, historians, ethnologists), this is a rich source of information about the natural environment, economy and living conditions of ancient communities Undoubtedly, determining the occurrence of the remains of such different fossil fungi can explain the causes of crop failures and famine in the past ages. In this context, mycological research of cultural layers allows us to trace the time-line of the emergence of phytopathogens in crops and of their variability.

Remains of various fungi have been detected during palaeobotanical studies of lake sediments, peat deposits and samples from archaeological sites. Sometimes only one genus was preserved. However, quite a few of them were detected in the sediments from Pszczew and Santok, and particularly one genus was important for the reconstruction of human activity. Many palynologists either do not produce records of observed fungal remains at all or only describe them as indeterminate fungi. Nevertheless, based on our experience, the information concerning fungal remains is often very useful when studying material of different origin from archaeological sites, e.g., to determine or describe human activity in the area. Such an extensive mycological examination of medieval sites in western Poland has been done for the first time.

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