

Helicosporous aero-aquatic hyphomycete from the Neogene of the Gray Fossil Site, USA

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We report fungal conidia of helicosporous aero-aquatic hyphomycete found during a survey of palynological slides from samples collected from Neogene deposits at the Gray Fossil Site (Tennessee, USA). They differ from conidia of hitherto described fossil species of helicosporous fungi, therefore, a new fossil-species *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov. is described. *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov. can be compared to those modern genera of helicosporous fungi that are characterised by three-dimensional, barrel-shaped, hollow conidia and are classified as helicosporous aero-aquatic hyphomycetes. In particular, it resembles species belonging to the *Helicoon* and *Helicodendron* genera. The ecology of modern helicosporous aero-aquatic hyphomycetes and the presumable palaeoecology of the fossil *H. grayensis* G. Worobiec & E. Worobiec sp. nov. are discussed.

Key words: Fossil fungus, aero-aquatic hyphomycetes, palaeoecology, Early Pliocene, North America.

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Introduction

Fungi are one of the kingdoms of eukaryotic organisms. It is estimated that there are between 2.2 and 3.8 million species of fungi, the majority of which remain undescribed (Hawksworth and Lücking 2017). Ascomycota and Basidiomycota are the most diversified phyla of Fungi, both may reproduce sexually or asexually (Kirk et al. 2008). Sexual stage and asexual stage of a given species usually differ in respect of their morphology (pleomorphism). The sexual morphs in Ascomycota and Basidiomycota are also named teleomorphs, and asexual morphs are also named as anamorphs (and were previously called Fungi imperfecti or Deuteromycetes). Anamorphic fungi were divided into the artificial groups hyphomycetes and coelomycetes (Seifert et al. 2011). Most of the anamorphic fungi are asexual stages of Ascomycota and, rarely, of Basidiomycota, and comprise about 25 000 described modern species (Kirk et al. 2008). Saccardo et al., for his *Sylloge Fungorum* (1882–1931) series, proposed an artificial system for classifying fungal spores based mainly on spore septation and morphology: amerosporae, didymosporae, phragmosporae, dictyosporae, helicosporae, staurosporae, and scoleco-

sporae. Saccardoan conidial categories are as follows: ameroconidia, didymoconidia, phragmoconidia, dictyoconidia, stauroconidia, helicoconidia, and scolecoconidia (Seifert and Gams 2001). Currently, however, fungi are classified basically on their phylogenies inferred from DNA sequence analysis (Spatafora et al. 2017; Voigt et al. 2021; Wijayawardene et al. 2022). Unfortunately, in the case of fossil fungi, only morphological characteristics can be used to assess their taxonomy (Kalgutkar and Jansonius 2000; Le Renard et al. 2020) as DNA is not preserved in specimens estimated to be several million years old. Therefore, fossil fungal spores are still treated according to Saccardoan categories (Kalgutkar and Jansonius 2000).

Fossil conidia have been repeatedly recorded in sediments of various ages (Kalgutkar and Jansonius 2000; Taylor et al. 2015); fossil spores are commonly found during palynological investigations (Elsik 1996; Nuñez Otaño et al. 2021; Worobiec et al. 2022b, 2023). Among them, a unique group of fossils are conidia of helicosporous anamorphic fungi. Helicosporous fungi are easily identifiable due to the unique helicoid structure of their rolled-up conidia. Helicosporous conidia may involve multiple revolutions of fungal filament

and may coil in a single plane (two-dimensional) or form three-dimensional structure (Zhao et al. 2007; Taylor et al. 2015; Ma et al. 2024). The three-dimensional, barrel-shaped helicosporous conidia can trap air in the hollow of conidium body to float on the water surface (Michaelides and Kendrick 1982). Modern helicosporous fungi with this conidial morphology are classified as helicosporous aero-aquatic hyphomycetes (Michaelides and Kendrick 1982; Goos 1987; Goh and Hyde 1996; Wyatt et al. 2013; Yamaguchi 2023). Fossil representatives of helicosporous aero-aquatic hyphomycetes seem useful for the reconstruction of palaeoenvironment considering available data on the ecology of their modern counterparts (Goh and Hyde 1996; Zhao et al. 2007; Markovskaja 2012; Yamaguchi 2023).

During a survey of palynological slides from samples collected from the Elephant Pit at the Gray Fossil Site, Tennessee, USA (Worobiec et al. 2013) some fungal non-pollen palynomorphs (NPP), namely well-preserved three-dimensional helicosporous conidia were found. Our studies focus on the taxonomy and ecology of these fungal palynomorphs and consider the utility of fossil helicosporous aero-aquatic hyphomycetes as a palaeoecological proxy.

Institutional abbreviations.—EMA, Ernst-Moritz-Arndt-University Greifswald, Germany; HdV, Hugo de Vries Laboratory, University of Amsterdam, the Netherlands; KRAM P, Palaeobotanical collection, Herbarium KRAM of the W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland.

Other abbreviations.—GFS, Gray Fossil Site; NPP, non-pollen palynomorphs. Author names of fungal taxa are abbreviated according to MycoBank.

Nomenclatural acts.—This published work and the nomenclatural acts it contains have been registered in MycoBank (<https://www.mycobank.org>).

Geological setting

Lacustrine deposits of the Gray Fossil Site (GFS) (Fig. 1) in Washington County, Tennessee, USA (36°23'9.6" N, 82°29'52.8" W) were suggested to be multiple sinkholes/sub-basins that could represent asynchronous events (Zobaa et al. 2011; Worobiec et al. 2013). In the organic-rich, laminated sediments, a variety of fossils of animals, plants, and

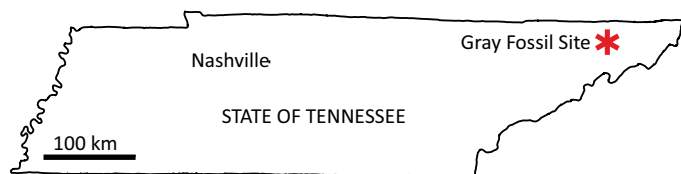


Fig. 1. Location map of the Gray Fossil Site in Washington County, Tennessee, USA. For more details see Shunk et al. (2006), Zobaa et al. (2011), and Ochoa et al. (2012).

fungi (e.g., Schubert and Wallace 2006; Liu and Jacques 2010; Zobaa et al. 2011; Mead et al. 2012; Ochoa et al. 2012, 2016; Worobiec et al. 2013; Huang et al. 2014, 2015) are preserved. The overall Gray Fossil Site fauna (Samuels et al. 2018) indicates an Early Pliocene age (latest Hemphillian to early Blancan, 4.5–4.9 Ma) for the Gray Fossil Site deposits. Palaeobotanical investigations revealed the presence of plant communities in open woodlands dominated by oaks and hickories, mixed with diverse vines and shrubs existing in a warm temperate to subtropical climate (Gong et al. 2010; Liu and Jacques 2010; Ochoa et al. 2012; Worobiec et al. 2013). So far, some fungal non-pollen palynomorphs have been reported from the Gray Fossil Site as well. Zobaa et al. (2011) found remains of *Callimothallus* sp., Ochoa et al. (2012) mentioned the presence of some fungal remains, and Worobiec et al. (2013) noted the occurrence of epiphyllous fungi. Later, Worobiec et al. (2018) reported *Cephalothecoidomyces neogenicus* G. Worobiec, F.H. Neumann & E. Worobiec, *Cenococcum* cf. *geophilum* Fr., and *Trichothyrites* cf. *padappakarensis* (R.K. Jain & R.C. Gupta) Kalgutkar & Janson., and finally, Worobiec et al. (2023) found conidia of *Mycoterolobium eccentricum* (R.K. Kar) G. Worobiec.

Material and methods

Palynological slides from samples collected at the Elephant Pit of the Gray Fossil Site (Worobiec et al. 2013) were re-examined for novel fungal forms. The samples were processed in the laboratory of the W. Szafer Institute of Botany, Polish Academy of Sciences, using Erdtman's acetolysis method (Moore et al. 1991) supplemented with treatment of the samples with hydrofluoric acid to remove mineral matter. The residuum was sieved through a 5 µm nylon mesh. The microscope slides were mounted in glycerine jelly and are stored at the W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland under catalogue numbers GFS/Elephant Pit (Worobiec et al. 2013). The holotype location on the slide is given using the England Finder coordinates. Microphotographs were taken with a Nikon Eclipse E400 microscope equipped with a Canon A640 digital camera.

Terminology for the morphology of helicosporous fungi follows Goos et al. (1985, 1986), Kalgutkar and Jansonius (2000), and Zhao et al. (2007).

Systematic palaeontology

Kingdom Fungi Bartling

Subkingdom Dikarya Hibbett, T.Y. James & Vilgalys

Division Ascomycota Caval.-Sm.

Class, order, and family uncertain

Fossil-genus *Helicoonites* Kalgutkar & Sigler

Type species: *Helicoonites goosii* Kalgutkar & Sigler; Early Eocene, Axel Heiberg Island, Northwest Territories, Canada.

Fossil-species *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov.

Fig. 2A–C.

Mycobank: MB 864238.

Etymology: The specific epithet *grayensis* refers to the Gray Fossil Site from which the fossil conidia were collected.

Type material: Holotype: slide KRAM P GFS/Elephant Pit 12(1), England Finder coordinate of specimen Q40/1. Paratypes: five specimens of conidia (slides KRAM P GFS/Elephant Pit 3(4), England Finder coordinate G35/3; KRAM P GFS/Elephant Pit 10(2), England Finder coordinate Z26/3; KRAM P GFS/Elephant Pit 10(7), England Finder coordinate Z42/2; KRAM P GFS/Elephant Pit 12(3), England Finder coordinate P39/1; KRAM P GFS/Elephant Pit 12(4), England Finder coordinate J40/4) from the type locality and horizon.

Type locality: Elephant Pit, Gray Fossil Site, Washington County, Tennessee, USA.

Type horizon: Early Pliocene (Hemphillian–Blancan transition, 4.9–4.5 Ma).

Material.—Type material only.

Description.—Conidia helicosporous, composed of distinctly septate filaments, smooth, 7.5–12.0 µm wide, cells forming a filament 9–20 µm long, filaments clockwise tightly coiled up to 8–9 times in three dimensions to form more or less ellipsoidal, hollow, doliiform conidium body, 70–80 µm long and 50–57 µm wide. Conidiophores and conidiogenous cells not observed.

Remarks.—The described fungal conidia from the Gray Fossil Site closely resemble the conidia of modern helicosporous aero-aquatic hyphomycetes. These fungi are characterised by the unique, three-dimensional, hollow, helicoid structure of their conidia (Michaelides and Kendrick 1982; Goh and Hyde 1996; Yamaguchi 2023). Conidia from the Gray Fossil Site are rather similar to those of the fossil-genus *Helicoonites* Kalgutkar & Sigler, and the fossil-species *Helicoonites goosii* Kalgutkar & Sigler, described from the Lower Eocene Iceberg Bay Formation on Axel Heiberg Island in the Canadian Northwest Territories. *Helicoonites goosii* is characterized by conidia with an ellipsoidal to barrel-shaped conidial body and a loosely to tightly coiled, three-dimensional conidial filament (Kalgutkar and Sigler 1995). According to the diagnosis of *H. goosii*, its conidial filament spirals are “made up of variable numbers of ascending coils or gyres” (Kalgutkar and Sigler 1995: 520). Therefore, the fossil-genus *Helicoonites* at first glance seems to differ from the helicosporous conidia from Gray Fossil Site, whose conidial filament appears to be more or less horizontally arranged. On the other hand, as can be seen in the holotype of *H. goosii* (Kalgutkar and Sigler 1995: fig. 4: 16), the base of the conidium is composed of parallel coils, whereas only in the upper part do the coils appear tilted. This may represent a mechanical deformation of the conidium, where the spore could have collapsed. Thus,

the conidial filaments of both *H. goosii* and the helicosporous conidia from the Gray Fossil Site can be considered to be arranged more or less horizontally. Nonetheless, the helicosporous conidia from the Gray Fossil Site differ in possessing conidial filaments 7.5–12.0 µm wide, compared with *H. goosii*, in which the filaments are approximately 5 µm wide. They also differ in conidial body size, which is larger (70–80 µm long and 50–57 µm wide) than that of *H. goosii* (50–75 µm long and 37–45 µm wide) (Kalgutkar and Sigler 1995). *Helicoonites goosii* is the only fossil-species of the fossil-genus *Helicoonites* described to date (Mycobank 2026). Given the morphological differences between the helicosporous conidia from Gray Fossil Site and *Helicoonites goosii*, we decided to describe a new fossil-species *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov.

Geographic and stratigraphic range.—Type locality and horizon only.

Discussion

Comparison of *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov. with fossil and modern representatives of helicosporous aero-aquatic hyphomycetes with barrel-shaped conidia.—Besides *Helicoonites goosii* from the Lower Eocene Iceberg Bay Formation on Axel Heiberg Island (Canadian Northwest Territories), Kalgutkar and Sigler (1995) described the helicosporous fossil-species *Helicosporiates pirozynskii* Kalgutkar & Sigler. According to the original diagnosis, the conidia of *H. pirozynskii* are “usually helically coiled in one plane or somewhat cochleate” (Kalgutkar and Sigler 1995: 520); thus, they differ from those of *Helicoonites grayensis* sp. nov., whose conidia are consistently three-dimensional and barrel-shaped; thus, they differ from those of *H. grayensis* G. Worobiec & E. Worobiec sp. nov., whose conidia are consistently three-dimensional and barrel-shaped. Moreover, *Helicosporiates pirozynskii* differs from *H. grayensis* G. Worobiec & E. Worobiec sp. nov. in having a considerably smaller conidial body (30–40 µm in diameter vs. 70–80 µm in length and 50–57 µm in width in *H. grayensis* G. Worobiec & E. Worobiec sp. nov.). Sherwood-Pike (1988) reported large helicoid spores with conidial filament tightly coiled in three dimensions, considered similar both to modern *Helicoon pluriseptatum* Beverw. and *Helicodendron giganteum* Glen-Bott, from the Early Miocene Clarkia locality in Northern Idaho, USA. Those conidia are similar to *H. grayensis* G. Worobiec & E. Worobiec sp. nov. as well. They differ, however, in that they possess a conidial filament coiled counter-clockwise (Sherwood-Pike 1988: pl. 2a), whereas *H. grayensis* G. Worobiec & E. Worobiec sp. nov. exhibits a filament that is coiled clockwise (Fig. 2C). Worobiec et al. (2022a) in the palynological profile from the Middle Miocene 1st mid-Polish lignite seam (MPLS-1) of the Drzewce lignite deposit (Konin region, cen-

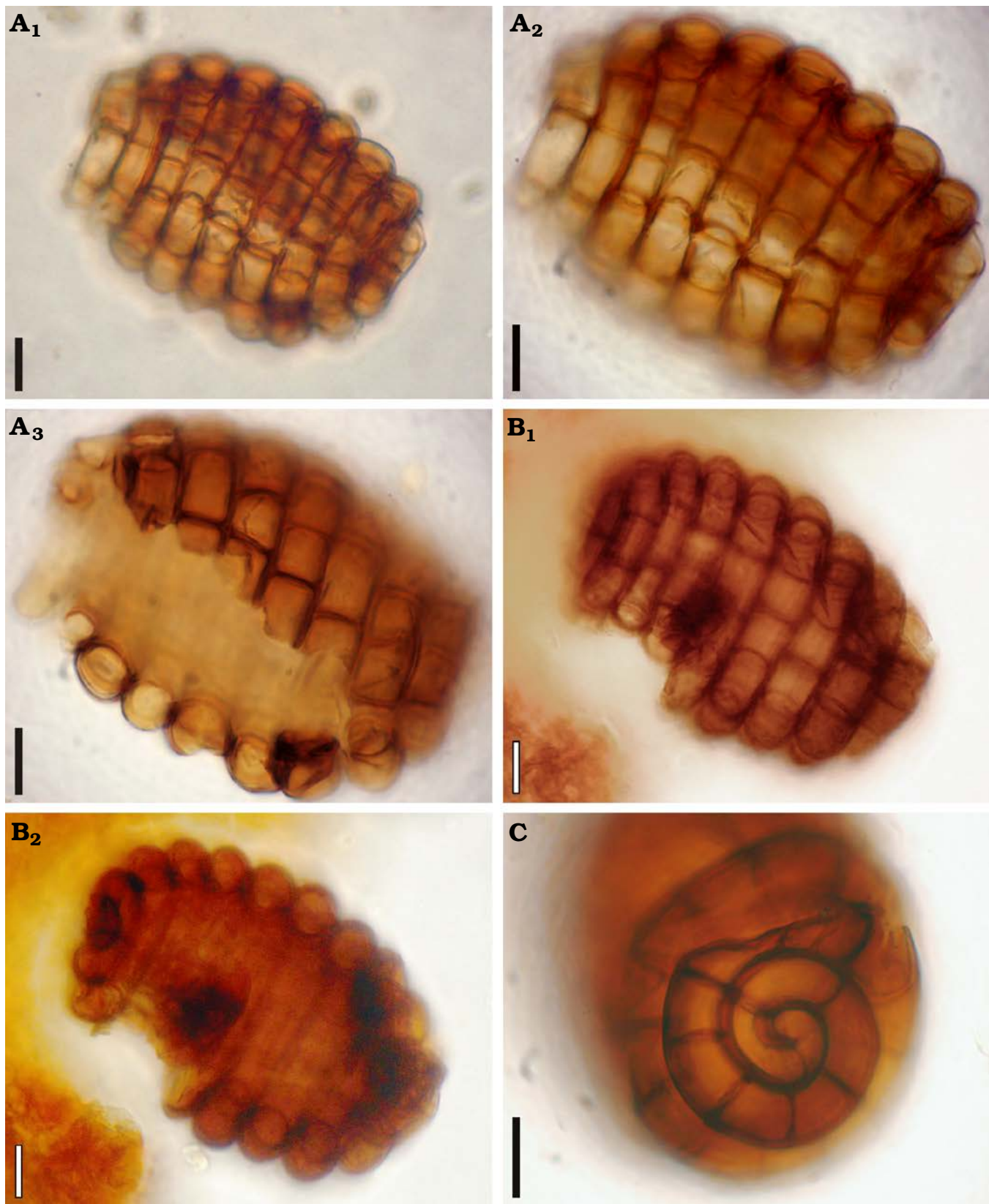


Fig. 2. Fossil conidia of ascomycotan fungus *Helicoonites grayensis* G. Worobiec & E. Worobiec, sp. nov., Gray Fossil Site, Tennessee, USA, Early Pliocene. **A.** Holotype, slide KRAM P GFS/Elephant Pit 12(1), conidium (**A**₁), details of the distal wall of the conidium formed by tightly coiled filament (**A**₂), details of the proximal, broken wall of the conidium formed by the tightly coiled filament (**A**₃). **B.** Paratype, slide KRAM P GFS/Elephant Pit 12(4), details of the proximal wall of the conidium formed by a tightly coiled filament (**B**₁), details of the internal hollow of the conidium (**B**₂). **C.** Paratype, slide KRAM P GFS/Elephant Pit 3(4), coiled conidial filament at the end of a conidium in polar view. Scale bars 10 µm.

tral Poland) encountered remains of helicosporous fungus (Worobiec et al. 2022a: fig. 4M) similar to both modern genera *Helicoon* Morgan and *Helicodendron* Peyronel. The conidium is similar to that of *H. grayensis* G. Worobiec & E. Worobiec sp. nov. as well, but it is fragmentarily preserved and therefore difficult to be definitively linked to *H. grayensis* G. Worobiec & E. Worobiec sp. nov.

From numerous Quaternary palynological profiles, helicosporous conidia were reported as non-pollen palynomorph HdV-30: *Helicoon pluriseptatum* (Van Geel 1978). Similarly to *H. grayensis* G. Worobiec & E. Worobiec sp. nov. their conidial filament is 8–9 times both clockwise and counter-clockwise tightly coiled in three dimensions, forming an ellipsoidal, hollow conidium body. However, those conidia are usually preserved in a broken state and therefore difficult to directly compare with *H. grayensis* G. Worobiec & E. Worobiec sp. nov. Additionally, in some cases (e.g., Filoc and Kupryjanowicz 2015: fig. 4B) fossil fungal remains referred to as *Helicoon pluriseptatum* in fact represent flat and coiled in one plane helicosporous conidia representing genera other than *Helicoon*. Goos et al. (1986) reported helicosporous conidium from a mediaeval archaeological site near Beetley, Norfolk, England (UK), which appeared similar to the modern species *Helicoon richonis* (Boud.) Linder. Finally, helicosporous conidia from modern surface samples collected in an alder carr in NE Germany were reported as the non-pollen palynomorph EMA-126 (*Helicoon/Helicodendron*) (Prager et al. 2012: pl. 4: 126A, B). However, the conidial filament of EMA-126 is rather fragmentarily preserved and displays only four coils. Therefore, this NPP cannot be confidently related to *H. grayensis* G. Worobiec & E. Worobiec sp. nov. It should be noted that spores of fossil helicosporous fungi with different morphologies have so far been reported in deposits ranging from the Eocene to the Holocene (Van Geel 1978; Kalgutkar and McIntyre 1991; Kalgutkar and Sigler 1995; Shumilovskikh et al. 2015; Romero et al. 2021; Saxena et al. 2021; Worobiec et al. 2022a, 2025).

Helicoonites grayensis G. Worobiec & E. Worobiec sp. nov. differs from such fossil helicosporous fungal genera, as *Circinoconites* R. Kar, Mand. & R.K. Kar, *Colligerites* K.P. Jain & R.K. Kar, *Elsikisporonites* P. Kumar, *Helicominutes* Barlinge & Paradkar, *Involutisporonites* R.T. Clarke, *Palaeocirrenalia* Ramanujam & Srisailam, *Paleoslimacomyces* Kalgutkar & Sigler, and *Retihelicosporonites* Ramanujam & K.P. Rao by conidial filament tightly coiled in three dimensions. Conidia of the aforementioned genera are generally two-dimensional, flat and coiled in one plane (Kalgutkar and Sigler 1995; Kalgutkar and Jansonius 2000; Saxena 2022) and are not classified as aero-aquatic hyphomycetes.

Helicoonites grayensis G. Worobiec & E. Worobiec sp. nov. from the Gray Fossil Site could be compared with the conidia of modern genera of helicosporous fungi with a barrel-shaped, hollow conidium body, classified as helicosporous aero-aquatic hyphomycetes. Most similar are conidia of species of the modern genera *Helicoon* and *Helicodendron*. The

same concerns the *Helicoon* species transferred to the genera *Helicoascotaiwania* Dayar., Maharachch. & K.D. Hyde, *Helicodendron*, *Helicorhoidion* S. Hughes, *Hyalohelicoon* Jian Ma, K.D. Hyde & Y.Z. Lu, *Magnohelicospora* R.F. Castañeda, Hern.-Restr., Gené & Guarro, *Pleohelicoon* Jayasiri, E.B.G. Jones & K.D. Hyde, and *Pseudohelicoon* Y.Z. Lu & K.D. Hyde (Goh and Kuo 2018; Ma et al. 2024). The conidia of the modern representatives of genera *Helicosingula* P.S. van Wyk, Marasas, Baard & Knox-Dav., *Inesiosporium* R.F. Castañeda & W. Gams, and *Rogergoosiella* A. Hern. Gut. & J. Mena, differ more or less in terms of their morphology compared to *H. grayensis* G. Worobiec & E. Worobiec sp. nov. The genus *Troposporium* Harkn. is distinct from *H. grayensis* G. Worobiec & E. Worobiec sp. nov. in non-septate conidial filament (Hernández-Gutiérrez and Mena Portales 1996). The taxonomy of modern helicosporous aero-aquatic hyphomycetes, however, is based not only on conidial characteristics, but also on the morphology of conidiophores and conidiogenous cells, and nowadays on phylogenetic analysis of DNA sequences (Zhao et al. 2007). While fossil helicosporous conidia are easily identifiable due to their distinctive twisted structure, however, in the fossil state dispersed conidia are usually preserved without their conidiophores and conidiogenous cells attached (Kalgutkar and Sigler 1995). Considering the fact that *H. grayensis* G. Worobiec & E. Worobiec sp. nov. preserved only its conidia, it is not possible to definitively relate *H. grayensis* G. Worobiec & E. Worobiec sp. nov. to any of the modern species of helicosporous aero-aquatic hyphomycetes with barrel-shaped conidia.

Modern helicosporous aero-aquatic hyphomycetes.—

Although fungi are primarily terrestrial organisms, some of them can be found in aquatic habitats, both lentic (ponds, lakes, swamps) and lotic (streams, rivers) (Dix and Webster 1995; Goh and Hyde 1996; Thormann 2006; Shearer et al. 2007; Wurzbacher et al. 2010). Freshwater anamorphic fungi, the most documented group of fungi found in aquatic environments, prefer to colonise woody substrates (Goh and Hyde 1996) and occur on submerged dead plant parts such as wood and leaves. Their main ecological role in aquatic ecosystems is the decomposition of dead plant material, including leaf litter, twigs and trunks, as well as flowers and fruits (Shearer 1992; Wong et al. 1998; Gessner et al. 2007; Kuehn 2016; Tsui et al. 2016; Gulis et al. 2019).

Aero-aquatic hyphomycetes, an ecological group of the freshwater anamorphic fungi are saprotrophs on decaying leaves, wood, and bark accumulated in small, shallow, stagnant or slow-running waters like woodland or swampy pools, ditches, and slowly running streams (Dix and Webster 1995; Goh and Hyde 1996; Webster and Weber 2007; Zhao et al. 2007; Markovskaja 2012; Yamaguchi 2023). Among them, helicosporous aero-aquatic hyphomycetes are a unique group of anamorphic fungi with tightly or loosely coiled, three-dimensional, hollow conidia (Michaelides and Kendrick 1982; Goos 1987; Goh and Hyde 1996; Wyatt et al. 2013; Yamaguchi 2023). There are various forms of

distinctively coiled, septate conidia that are modified multicellular hyphae (Kendrick 2003). Three-dimensional, barrel-shaped, coiled helicosporous conidia gain buoyancy from air trapped in the hollow of conidium body and are virtually unsinkable in water (Michaelides and Kendrick 1982). As with other helicosporous fungi, helicosporous aero-aquatic hyphomycetes are primarily saprotrophs on dead or decaying plant matter, such as plant litter, decaying twigs and rotten wood (Zhao et al. 2007; Ma et al. 2024). Helicosporous aero-aquatic hyphomycetes can degrade lignin and cellulose playing an essential role in carbon cycling in the aquatic environment (Abdullah and Taj-Aldeen 1989; Fisher et al. 1977, 1983; Yamaguchi 2023).

Species of the modern genera *Helicoon* and *Helicodendron* are a major group of modern helicosporous aero-aquatic hyphomycetes with three-dimensional, hollow, barrel-shaped conidia (Zhao et al. 2007). The genera *Helicoon* and *Helicodendron* are morphologically so similar to each other that it is impossible to distinguish between them based on conidium morphology alone. *Helicoon* is separated from *Helicodendron* by the conidia not being in chains, but produced singly (Linder 1929; Goos et al. 1985, 1986; Goos 1987; Zhao et al. 2007).

Modern helicosporous aero-aquatic hyphomycetes with three-dimensional, hollow structure of their conidia, form an ecological group without phylogenetic coherence that have evolved from multiple lineages within Ascomycota (Yamaguchi 2023). Species of the genus *Helicoon* are considered polyphyletic and arose convergently at least five times in different Ascomycota orders (Pleosporales, Pleurotheciales, Tubeufiales, Venturiales). Representatives of *Helicodendron* appeared to be polyphyletic as well although most representatives are classified within Leotiomyces (Tsui and Berbee 2006; Zhao et al. 2007; Goh and Kuo 2018; Yamaguchi 2023). Tsui and Berbee (2006) suggested that the barrel-shaped conidia of helicosporous aero-aquatic hyphomycetes, which form buoyant propagules capable of dispersal in water, have evolved multiple times independently as unrelated species adapted to aquatic dispersal.

The tentative palaeoecology of *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov.—Fossil fungi that have modern-day equivalents with limited habitat preferences could be used for palaeoenvironmental reconstruction (Ramanujam 1982; Kalgutkar 1993). Extant aero-aquatic anamorphic fungi including helicosporous ones are saprotrophs that colonize submerged, decaying plant material accumulated at the bottom of small, shallow, and typically stagnant water bodies, such as ponds and ditches, often surrounded by vegetation that contributes leaf litter, twigs, and other organic debris (Dix and Webster 1995; Goh and Hyde 1996; Webster and Weber 2007; Zhao et al. 2007; Markovskaja 2012; Yamaguchi 2023). Helicosporous aero-aquatic fungi like other aero-aquatic hyphomycetes, grow on submerged substrates, typically a few centimetres below the water surface, but sporulate only when the substrate becomes exposed

to air following lowering of the water level. This transition creates excellent aeration at the substratum-water interface, inducing helicosporous aero-aquatic hyphomycetes to sporulate vigorously when exposed to air. Their propagules trapped air inside and this enables them to float on the water surface. These non-wettable, buoyant propagules are subsequently dispersed by water upon renewed inundation, when the substrate becomes submerged again (Dix and Webster 1995; Goh and Hyde 1996; Webster and Weber 2007; Zhao et al. 2007; Markovskaja 2012; Yamaguchi 2023).

By analogy, the presence of helicosporous aero-aquatic hyphomycete *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov. at the Gray Fossil Site may indicate the existence of small, shallow water bodies characterized by fluctuating water levels and, at times, periodic desiccation. This interpretation is consistent with previous studies of the paleoenvironment of the Gray Fossil Site. The lacustrine deposits of Gray Fossil Site are interpreted to have formed in ponds that, at least in the case of smaller water bodies, were likely shallow, and possibly subject to episodic drying (Shunk et al. 2006; Gong et al. 2010; Liu and Jacques 2010; Zobaa et al. 2011; Ochoa et al. 2012; Worobiec et al. 2013; Keenan and Engel 2017). The abundance of lungless plethodontid salamander remains indicates that the areas surrounding these ponds maintained persistently high moisture levels, consistent with the ecological requirements of taxa inhabiting forested, humid environments (Boardman 2009; Boardman and Schubert 2011). The occurrence of *Pterocarya* and *Taxodium*, together with numerous remains of *Alligator* sp., provides further evidence for the presence of wetland vegetation associated with sinkhole pond systems (Schubert and Wallace 2006; Ochoa et al. 2012; Brandon 2013). Comparable modern analogues to the inferred habitat at the Gray Fossil Site are represented by isolated depressional wetlands of the southeastern USA, particularly those occurring within karst-related sinkhole systems. These wetlands develop in topographic depressions and are primarily sustained by precipitation-driven hydrological regimes. Water levels exhibit pronounced seasonal variability, with inundation typically occurring during winter and spring when precipitation exceeds evapotranspiration, followed by progressive drying during summer as higher temperatures and increased vegetation activity enhance water loss. Such hydrological dynamics are fundamental in maintaining the structural and functional diversity of these ecosystems (Kirkman et al. 2012). In these modern systems, as in the palaeoenvironment inferred for the Gray Fossil Site, *Taxodium ascendens* Brongn. (pond cypress) constitutes a characteristic component of the vegetation (Kirkman et al. 2012; Cartwright and Wolfe 2016). In addition, these wetlands provide important habitat for *Alligator mississippiensis* Daudin, 1801, further reinforcing their value as ecological analogues for the fossil assemblage (Kirkman et al. 2012). Recently, Worobiec et al. (2023) found at the Gray Fossil Site the fossil fungus *Mycoenterolobium eccentricum* (R.K. Kar) G. Worobiec whose modern relatives can be

categorized as aero-aquatic hyphomycetes (Goh and Hyde 1996). Its existence at the Gray Fossil Site reinforces the interpretation of small, shallow water bodies with pronounced seasonal fluctuations in water level.

Conclusions regarding the palaeoenvironment of the Gray Fossil Site, based on both previous palaeontological investigations and the presence of helicosporous aero-aquatic fungi, are consistent with the results of studies on the Early Miocene palaeoenvironment of the Clarkia locality in northern Idaho, USA, where similar helicosporous aero-aquatic fungi have been found. At Clarkia, helicosporous, three-dimensional, barrel-shaped conidia have been documented and interpreted as comparable to those of extant helicosporous aero-aquatic hyphomycete genera *Helicoon* and *Helicodendron* (Sherwood-Pike 1988). According to Sherwood-Pike (1988), the occurrence of these fungi at Clarkia is consistent with their known ecological preferences.

Based on the discussion above, fossil helicosporous aero-aquatic hyphomycetes may serve as useful palaeoenvironmental indicators, particularly when considered alongside other fossil evidence.

Concluding remarks

Well-preserved microremains of helicosporous aero-aquatic hyphomycete with three-dimensional barrel-shaped, hollow conidia were found during a survey of palynological slides from samples collected from Lower Pliocene deposits from the Elephant Pit at the Gray Fossil Site, Tennessee, USA. They differ from hitherto described fossil-species of helicosporous aero-aquatic hyphomycetes; therefore, a new fossil-species *Helicoonites grayensis* G. Worobiec & E. Worobiec sp. nov. has been described. Similarly to modern helicosporous aero-aquatic hyphomycetes, *H. grayensis* G. Worobiec & E. Worobiec sp. nov. may indicate the presence of small, shallow bodies of water that accumulated plant debris (decaying leaves, wood, and bark) and suggests fluctuations of water level or even the periodic drying of the water reservoir. This is in accordance with results of the previous studies of the palaeoenvironment of the Gray Fossil Site. Therefore, the results of our investigations confirm that fossil helicosporous aero-aquatic hyphomycetes are useful fungal non-pollen palynomorph proxies for reconstruction of a palaeoenvironment. The need for further research on fossil helicosporous aero-aquatic hyphomycetes should be emphasised to clarify their stratigraphic range, particularly the timing of their appearance, and to confirm their value as palaeoecological proxies.

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References

- Abdullah, S.K. and Taj-Aldeen, S.J. 1989. Extracellular enzymatic activity of aquatic and aero-aquatic conidial fungi. *Hydrobiologia* 174: 217–223.
- Boardman, G.S. 2009. *Salamanders of the Mio-Pliocene Gray Fossil Site, Washington County, Tennessee*. 89 pp. Unpublished M.S. thesis, East Tennessee State University, Johnson City.
- Boardman, G.S. and Schubert, B.W. 2011. First Mio-Pliocene salamander fossil assemblage from the southern Appalachians. *Palaeontologia Electronica* 14 (2): 16A.
- Brandon, S. 2013. *Discovery of Bald Cypress Fossil Leaves at the Gray Fossil Site, Tennessee and Their Ecological Significance*. 14 pp. Undergraduate Honors Theses, Paper 145. East Tennessee State University, Johnson City [available online, <http://dc.etsu.edu/honors/145>].
- Cartwright, J.M. and Wolfe, W.J. 2016. Insular ecosystems of the southeastern United States: A regional synthesis to support biodiversity conservation in a changing climate. *U.S. Geological Survey Professional Paper* 1828: 1–162.
- Dix, N.J. and Webster, J. 1995. *Fungal Ecology*. 549 pp. Springer-Science + Business Media, B.V., Dordrecht.
- Elsik, W.C. 1996. Fungi. In: J. Jansonius and D.C. McGregor (eds.), *Palynology: Principles and Applications 1*, 293–305. AASP Foundation, Dallas.
- Filóc, M. and Kupryjanowicz, M. 2015. Non-pollen palynomorphs characteristic for the dystrophic stage of humic lakes in the Wigry National Park, NE Poland. *Studia Quaternaria* 32: 31–41.
- Fisher, P.J., Davey, R.A., and Webster, J. 1983. Degradation of lignin by aquatic and aero-aquatic hyphomycetes. *Transactions of the British Mycological Society* 80: 166–168.
- Fisher, P.J., Sharma, P.D., and Webster, J. 1977. Cellulolytic ability of aero-aquatic hyphomycetes. *Transactions of the British Mycological Society* 69: 495–520.
- Gessner, M.O., Gulis, V., Kuehn, K.A., Chauvet, E., and Suberkropp, K. 2007. Fungal Decomposers of Plant Litter in Aquatic Ecosystems. In: C. Kubicek and I. Druzhina (eds.), *Environmental and Microbial Relationships. The Mycota 4*: 301–324. Springer, Berlin.
- Goh, T.K. and Hyde, K.D. 1996. Biodiversity of freshwater fungi. *Journal of Industrial Microbiology* 17 (5–6): 328–345.
- Goh, T.K. and Kuo, C.H. 2018. A new species of *Helicoon* from Taiwan. *Phytotaxa* 346 (2): 141–156.
- Gong, F., Karsai, I., and Liu, Y. 2010. *Vitis* seeds (Vitaceae) from the late Neogene Gray Fossil Site, northeastern Tennessee, U.S.A. *Review of Palaeobotany and Palynology* 162: 71–83.
- Goos, R.D. 1987. Fungi with a twist: the helicosporous Hyphomycetes. *Mycologia* 79: 1–22.
- Goos, R.D., Abdullah, S.K., Fisher, P.J., and Webster, J. 1985. The anamorph genus *Helicodendron*. *Transactions of the British Mycological Society* 84: 423–435.
- Goos, R.D., Abdullah, S.K., Fisher, P.J., and Webster, J. 1986. The anamorph

- genus *Helicoon*. *Transactions of the British Mycological Society* 87: 115–122.
- Gulis, V., Su, R., and Kuehn, K.A. 2019. Fungal Decomposers in Freshwater Environments. In: C. Hurst (ed.), *The Structure and Function of Aquatic Microbial Communities. Advances in Environmental Microbiology* 7: 121–155. Springer, Cham.
- Hawksworth, D.L. and Lücking, R. 2017. Fungal Diversity Revisited: 2.2 to 3.8 Million Species. *Microbiology Spectrum* 5 (4): 1–17.
- Hernández-Gutiérrez, A. and Mena Portales, J. 1996. A new helicosporous hyphomycete collected on *Roystonea regia* in Cuba. *Mycological Research* 100: 1483–1484.
- Huang, Y.J., Liu, Y., and Zavada, M. 2014. New fossil fruits of *Carya* (Juglandaceae) from the latest Miocene to earliest Pliocene in Tennessee, eastern United States. *Journal of Systematics and Evolution* 52: 508–520.
- Huang, Y.J., Liu, Y., Wen, J., and Quan, C. 2015. First fossil record of *Staphylea* L. (Staphyleaceae) from North America, and its biogeographic implications. *Plant Systematics and Evolution* 301: 2203–2218.
- Kalgutkar, R.M. 1993. Paleogene fungal palynomorphs from Bonnet Plume formation, Yukon Territory. In: L. Reynolds (ed.), *Contributions to Canadian Paleontology. Bulletin of the Geological Survey of Canada* 444: 51–105.
- Kalgutkar, R.M. and Jansonius, J. 2000. Synopsis of fossil fungal spores, mycelia and fructifications. *American Association of Stratigraphic Palynologists Contribution Series* 39: 1–428.
- Kalgutkar, R.M. and McIntyre, D.J. 1991. Helicosporous fungi and Early Eocene pollen, Eureka Sound Group, Axel Heiberg Island, Northwest Territories. *Canadian Journal of Earth Sciences* 28: 364–371.
- Kalgutkar, R.M. and Sigler, L. 1995. Some fossil fungal form-taxa from the Maastrichtian and Palaeogene ages. *Mycological Research* 99: 513–522.
- Keenan, S.W. and Engel, A.S. 2017. Reconstructing diagenetic conditions of bone at the Gray Fossil Site, Tennessee, USA. *Palaeogeography, Palaeoclimatology, Palaeoecology* 471: 48–57.
- Kendrick, B. 2003. Analysis of morphogenesis in hyphomycetes: new characters derived from considering some conidiophores and conidia as condensed hyphal systems. *Canadian Journal of Botany* 81 (2): 75–100.
- Kirk, P.M., Cannon, P.F., Minter, D.W., and Stalpers, J.A. 2008. *Ainsworth and Bisby's Dictionary of the Fungi*. 10th Edition. 771 pp. CAB International, Wallingford.
- Kirkman, L., Smith, L., and Golladay, S. 2012. Southeastern depression wetlands. In: D. Batzer and A. Baldwin (eds.), *Wetland Habitats of North America: Ecology and Conservation Concerns*, 203–216. University of California Press, Berkeley.
- Kuehn, K. 2016. Lentic and lotic habitats as templates for fungal communities: traits, adaptations, and their significance to litter decomposition within freshwater ecosystems. *Fungal Ecology* 19: 135–154.
- Le Renard, L., Firmino, A.L., Pereira, O.L., Stockey, R.A., and Berbee, M.L. 2020. Character evolution of modern fly-speck fungi and implications for interpreting thyrlotheal fossils. *American Journal of Botany* 107 (7): 1021–1040.
- Linder, D.H. 1929. A monograph of the helicosporous Fungi Imperfecti. *Annals of the Missouri Botanical Garden* 16 (3): 227–388.
- Liu, Y. and Jacques, F.M.B. 2010. *Sinomenium macrocarpus* sp. nov. (Menispermaceae) from the Miocene–Pliocene transition of Gray, northeast Tennessee, USA. *Review of Palaeobotany and Palynology* 159: 112–122.
- Ma, J., Hyde, K.D., Tibpromma, S., Gomdola, D., Liu, N.G., Norphanphoun, C., Bao, D.F., Boonmee, S., Xiao, X.J., Zhang, L.J., Luo, Z.L., Zhao, Q., Suwannarath, N., Karunarathna, S.C., Liu, J.K., and Lu, Y.Z. 2024. Taxonomy and systematics of lignicolous helicosporous hyphomycetes. *Fungal Diversity* 129 (1): 365–653.
- Markovskaja, S. 2012. Aero-aquatic fungi colonizing decaying leaves in woodland swampy pools of Aukštadvaris Regional Park (Lithuania). *Botanica Lithuanica* 18: 123–132.
- Mead, J.I., Schubert, B.W., Wallace, S.C., and Swift, S.L. 2012. Helodermitid lizard from the Mio-Pliocene oak-hickory forest of Tennessee, eastern USA, and a review of monstrosaurian osteoderms. *Acta Palaeontologica Polonica* 57: 111–121.
- Michaelides, J. and Kendrick, B. 1982. The bubble-trap propagules of *Beverwykella*, *Helicoon* and other aero-aquatic fungi. *Mycotaxon* 14: 247–260.
- Moore, P.D., Webb, J.A., and Collinson, M.E. 1991. *Pollen Analysis*. 216 pp. Blackwell, Oxford.
- Mycobank 2026. Available at <http://www.mycobank.org> (accessed June 2026).
- Núñez Otaño, N., Bianchinotti, M.V., and Saparrat, M.C.N. 2021. Palaeomycology: a modern mycological view of fungal palynomorphs. In: F. Marret, J. O'Keefe, P. Osterloff, M. Pound, and L. Shumilovskikh (eds.), *Applications of Non-pollen Palynomorphs: From Palaeoenvironmental Reconstructions to Biostratigraphy. Geological Society, London Special Publications* 511: 91–120.
- Ochoa, D., Whitelaw, M., Liu, Y., and Zavada, M. 2012. Palynology from Neogene sediments at the Gray Fossil Site, Tennessee, USA: Floristic Implications. *Review of Palaeobotany and Palynology* 184: 36–48.
- Ochoa, D., Zavada, M.S., Liu, Y., and Farlow, J.O. 2016. Floristic implications of two contemporaneous inland upper Neogene sites in the eastern US: Pipe Creek Sinkhole, Indiana, and the Gray Fossil Site, Tennessee (USA). *Paleobiodiversity and Palaeoenvironment* 96: 239–254.
- Prager, A., Theuerkauf, M., Couwenberg, J., Barthelmes, A., Aptroot, A., and Joosten, H. 2012. Pollen and non-pollen palynomorphs as tools for identifying alder carr deposits: A surface sample study from NE-Germany. *Review of Palaeobotany and Palynology* 186: 38–57.
- Ramanujam, C.G.K. 1982. Recent advances in the study of fossil fungi. In: D.C. Bharadwaj (ed.), *Recent advances in Cryptogamic Botany, Part II: Fossil Cryptogams*, 287–301. The Palaeobotanical Society, Lucknow.
- Romero, I.C., Núñez Otaño, N.B., Gibson, M.E., Spears, T.M., Fairchild, C.J., Tarlton, L., Jones, S., Belkin, H.E., Warny, S., Pound, M.J., and O'Keefe, J.M.K. 2021. First Record of Fungal Diversity in the Tropical and Warm-Temperate Middle Miocene Climate Optimum Forests of Eurasia. *Frontiers in Forests and Global Change* 4: 768405.
- Saccardo, P.A., Traverso, G.B., and Trotter, A. 1882–1931. *Sylloge fungorum omnium hucusque cognitorum. Vols. 1–25*. Imprint varies.
- Samuels, J.X., Bredehoeft, K.E., and Wallace, S.C. 2018. A new species of *Gulo* from the Early Pliocene Gray Fossil Site (Eastern United States); rethinking the evolution of wolverines. *PeerJ* 6: e4648.
- Saxena, R.K. 2022. Fossil Helicosporae: global diversity, morphological variations and distribution in time and space. *Geophytology* 51 (1–2): 87–108.
- Saxena, R.K., Wijayawardene, N.N., Dai, D.Q., Hyde, K.D., and Kirk, P.M. 2021. Diversity in fossil fungal spores. *Mycosphere* 12: 670–874.
- Schubert, B.W. and Wallace, S.C. 2006. Amphibians and reptiles of the Mio-Pliocene Gray Fossil Site and their paleoecological implications. *Journal of Vertebrate Paleontology* 26 (3): 122A.
- Seifert, K.A. and Gams, W. 2001. The taxonomy of anamorphic fungi. In: D.J. McLaughlin, E.G. McLaughlin, and P.A. Lemke (eds.), *The Mycota VII, Part A*, 307–347. Springer-Verlag, Berlin.
- Seifert, K., Morgan-Jones, G., Gams, W., and Kendrick, B. 2011. *The genera of Hyphomycetes. CBS Biodiversity Series* 9. 997 pp. CBS-KNAW Fungal Biodiversity Centre, Utrecht.
- Shearer, C.A. 1992. The role of woody debris. In: F. Bärlocher (ed.), *The Ecology of Aquatic Hyphomycetes. Ecological Studies (Analysis and Synthesis)* 94: 77–98. Springer, Berlin.
- Shearer, C.A., Descals, E., Kohlmeyer, B., Kohlmeyer, J., Marvanová, L., Padgett, D., Porter, D., Raja, H.A., Schmit, J.P., Thorton, H.A., and Voglmayr, H. 2007. Fungal biodiversity in aquatic habitats. *Biodiversity and Conservation* 16 (1): 49–67.
- Sherwood-Pike, M.A. 1988. Freshwater fungi: fossil record and paleoecological potential. *Palaeogeography, Palaeoclimatology, Palaeoecology* 62: 271–285.
- Shumilovskikh, L.S., Schlütz, F., Achterberg, I., Bauerochse, A., and Leuschner, H.H. 2015. Non-pollen palynomorphs from mid-Holocene peat

- of the raised bog Borsteler Moor (Lower Saxony, Germany). *Studia Quaternaria* 32: 5–18.
- Shunk, A.J., Driese, S.G., and Clark, G.M. 2006. Latest Miocene to earliest Pliocene sedimentation and climate record derived from paleosinkhole fill deposits, Gray Fossil Site, northeast Tennessee, U.S.A. *Palaeogeography, Palaeoclimatology, Palaeoecology* 231: 265–278.
- Spatafora, J.W., Aime, M.C., Grigoriev, I.V., Martin, F., Stajich, J.E., and Blackwell, M. 2017. The fungal tree of life: from molecular systematics to genome-scale phylogenies. *Microbiology Spectrum* 5 (5): 1–32.
- Taylor, T.N., Krings, M., and Taylor, E.L. 2015. *Fossil Fungi*. 398 pp. Elsevier, Academic Press, Amsterdam.
- Thormann, M.N. 2006. The role of fungi in boreal peatlands. In: R.K. Wieder and D.H. Vitt (eds.), *Boreal Peatland Ecosystems. Ecological Studies* 188: 101–123.
- Tsui, C.K.M. and Berbee, M.L. 2006. Phylogenetic relationships and convergence of helicosporous fungi inferred from ribosomal DNA sequences. *Molecular Phylogenetics and Evolution* 39: 587–597.
- Tsui, C.K.M., Baschien C., and Goh T.K. 2016. Biology and ecology of freshwater fungi. In: D.W. Li (ed.), *Biology of Microfungi*, 285–313. Springer International Publishing, Cham.
- Van Geel, B. 1978. A palaeoecological study of Holocene peat bog sections in Germany and the Netherlands, based on the analysis of pollen, spores and macro- and microscopic remains of fungi, algae, cormophytes and animals. *Review of Palaeobotany and Palynology* 25: 1–120.
- Voigt, K., James, T.Y., Kirk, P.M., de A. Santiago, A. L. C. M., Waldman, B., Griffith, G.W., Fu, M., Radek, R., Strasser, J.F.H., Wurzbacher, C., Jerônimo, G.H., Simmons, D.R., Seto, K., Gentekaki, E., Hurdeal, V.G., Hyde, K.D., Nguyen, T.T.T., and Lee, H.B. 2021. Early-diverging fungal phyla: taxonomy, species concept, ecology, distribution, anthropogenic impact, and novel phylogenetic proposals. *Fungal Diversity* 109 (1): 59–98.
- Webster, J. and Weber, R.W.S. 2007. *Introduction to Fungi*. Third edition. 867 pp. Cambridge University Press, Cambridge.
- Wijayawardene, N.N., Hyde, K.D., Dai, D.Q., Sánchez-García, M., Goto, B.T., Saxena, R.K., Erdoğan, M., Selçuk, F., Rajeshkumar, K.C., Ap-troot, A., Błaszowski, J., Boonyuen, N., da Silva, G.A., de Souza, F.A., Dong, W., Ertz, D., Haelewaters, D., Jones, E.B.G., Karunarathna, S.C., Kirk, P.M., Kukwa, M., Kumla, J., Leontyev, D.V., Lumbsch, H.T., Maharachchikumbura, S.S.N., Marguno, F., Martínez-Rodríguez, P., Mešić, A., Monteiro, J.S., Oehl, F., Pawłowska, J., Pem, D., Pfliegler, W.P., Phillips, A.J.L., Pošta, A., He, M.Q., Li, J.X., Raza, M., Sruthi, O.P., Suetrong, S., Suwannarach, N., Tedersoo, L., Thiagaraja, V., Tibpromma, S., Tkalčec, Z., Tokarev, Y.S., Wanasinghe, D.N., Wijesundara, D.S.A., Wimalaseana, S.D.M.K., Madrid, H., Zhang, G.Q., Gao, Y., Sánchez-Castro, I., Tang, L.Z., Stadler, M., Yurkov, A., and Thines, M. 2022. Outline of fungi and fungus-like taxa—2021. *Mycosphere* 13: 53–453.
- Wong, M.K., Goh, T.K., Hodgkiss, I.J., Hyde, K.D., Ranghoo, V.M., Tsui, C.K., Ho, W.H., Wong, W.S.W., and Yuen, T.K. 1998. Role of fungi in freshwater ecosystems. *Biodiversity and Conservation* 7 (9): 1187–1206.
- Worobiec, E., Liu, Y., and Zavada, M.S. 2013. Palaeoenvironment of late Neogene lacustrine sediments at the Gray Fossil Site, Tennessee, USA. *Annales Societatis Geologorum Poloniae* 83: 51–63.
- Worobiec, E., Widera, M., and Worobiec, G. 2022a. Palaeoenvironment of the middle Miocene wetlands at Drzewce, Konin region, central Poland. *Annales Societatis Geologorum Poloniae* 92: 201–218.
- Worobiec, E., Worobiec, G., and Widera, M. 2025. Pollen and non-pollen palynomorphs as tools for interpreting palaeoenvironmental changes in the Miocene lignite from the Chłapowo Cliff (northern Poland). *Annales Societatis Geologorum Poloniae* 95: 161–178.
- Worobiec, G., Worobiec, E., and Liu, Y.C. 2018. Fungal remains from late Neogene deposits at the Gray Fossil Site, Tennessee, USA. *Mycosphere* 9: 1014–1024.
- Worobiec, G., Worobiec, E., and Liu, Y.C. 2023. Taxonomy and palaeoecology of the fossil anamorphic fungus *Mycoenterolobium eccentricum* (R.K. Kar) G. Worobiec, n. comb. *Comptes Rendus Palevol* 22: 585–594.
- Worobiec, G., Worobiec, E. and Widera, M. 2022b. Middle Miocene wetland fungi from the Adamów Lignite Mine, central Poland. *Historical Biology* 34: 841–856.
- Wurzbacher, C.M., Bärlocher, F. and Grossart, H.P., 2010. Fungi in lake ecosystems. *Aquatic Microbial Ecology* 59: 125–149.
- Wyatt, T.T., Wösten, H.A., and Dijksterhuis, J. 2013. Fungal spores for dispersion in space and time. *Advances in Applied Microbiology* 85: 43–91.
- Yamaguchi, K. 2023. Recent studies on aero-aquatic fungi, with special reference to diversity of conidial morphology and convergent evolution. *Mycoscience* 64 (5): 128–135.
- Zhao, G.Z., Liu, X.Z., and Wu, W.P. 2007. Helicosporous hyphomycetes from China. *Fungal Diversity* 26: 313–524.
- Zobaa, M.K., Zavada, M.S., Whitelaw, M.J., Shunk, A.J., and Oboh-Ikuenobe, F.E. 2011. Palynology and palynofacies analyses of the Gray Fossil Site, eastern Tennessee: Their role in understanding the basin-fill history. *Palaeogeography, Palaeoclimatology, Palaeoecology* 308: 433–444.