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With 3 figures

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Abstract: The various types of spherical microfossils collectively termed fossil fungal "sporocarps" exhibit basic congruities in morphology that have been used to suggest they all may belong to the same higher taxonomic category. Both the Ascomycota and zygomycete fungi have been discussed in this respect, but features that precisely delimit the nature and taxonomic position of these fossils have not been documented. Here, we present two new specimens of the Pennsylvanian "sporocarp" *Sprocarpon asteroides* from the Lower Coal Measures of Great Britain. Both provide evidence that a spore with a multi-layered wall was formed in this structure by blastic inflation of a hyphal tip. The outer spore walls appear to be continuous with the wall of the subtending hypha, while the inner wall (the spore wall proper) more likely developed de novo. *Sporocarpon asteroides* is interpreted as a unisporic sporocarp with a pseudoparenchymatous peridium, likely with affinities to the Glomeromycota. This discovery supports the notion that the fossil fungal "sporocarps" include several biologically different structures.

Key words: fossil fungal "sporocarp", Glomeromycota, Lower Coal Measures, Pennsylvanian, pseudoparenchymatous investment, spore wall

1 Introduction

Various types of small spherical structures often collectively termed fossil fungal "sporocarps"[1] are commonly found in Pennsylvanian coal balls and certain chert deposits (surveyed by STUBBLEFIELD et al. 1983; TAYLOR et al. 2015). All consist of a central cavity bounded by a contiguous perimeter wall and additionally surrounded by a prominent investment composed of loosely entwined or tightly interwoven hyphae. "Sporocarps" occur singly or in small clusters; hardly any of them provide information on the systems on or in which they were borne. Several different genera, including *Dubiocarpon* S.A. HUTCH., *Mycocarpon* S.A. HUTCH., *Sporocarpon* WILL., and *Traquairia* CARRUTH., have been erected to accommodate these fossils, which are distinguished from one another based primarily on the construction of the investment. While fossil fungal "sporocarps" are mostly known from the Pennsylvanian, there are also several forms described from the Mississippian (SCOTT 1911; TAYLOR et al. 1994; KRINGS et al. 2010b) and Triassic (WHITE & TAYLOR 1988, 1989a, 1991; TAYLOR & WHITE 1989), and one from the Lower Devonian Rhynie cherts (KRINGS et al. 2014). What relationship, if any, exists between the "sporocarps" and a number of other, structurally similar but generally much smaller Rhynie chert fossils referred to as mantled fungal reproductive units (e.g., KRINGS & TAYLOR 2014, 2015a, 2015b; KRINGS et al. 2016; KRINGS & HARPER 2017, 2020) remains uncertain.

Fossil fungal "sporocarps" in general have been attributed to the Ascomycota based on specimens containing one or more spheres believed to represent asci that in turn contain small spherules interpreted as ascospores (HUTCHINSON & WALTON 1953; HUTCHINSON 1955; STUBBLEFIELD et al. 1983). According to this idea, the "sporocarp" would be a cleistothecium. Another hypothesis, however, uses specimens that contain a single, large sphere to suggest affinities to the zygomycete fungi (PIROZYNSKI 1976; TAYLOR & WHITE 1989). The sphere is interpreted as a zygospore, while the "sporocarp" would be the zygosporangium enveloped in a hyphal investment equivalent to that seen in certain extant Endogonales (Mucoromycotina). Smaller spheres in some cases present in the large sphere are regarded as intrusive parasites. There is circumstantial evidence to corroborate the latter hypothesis (e.g., KRINGS et al. 2010b, 2011a, 2011d; KRINGS & TAYLOR 2012), but structural

[1]The term sporocarp, used in mycology to refer to a multicellular structure in which spores or spore-producing entities are formed, is put in quotation marks because it may not be applicable to all of these fossils (for details, see KRINGS et al. 2011d).

features that precisely delimit the nature and taxonomic position of the fungal “sporocarps” have not been documented to date.

Every new specimen that displays features not hitherto seen can provide critical new information on these enigmatic fossils, and thus deserves thoughtful consideration. Here, we present two specimens of *Sporocarpon asteroides* WILL. from the Lower Coal Measures of Great Britain that both contain a structure suggestive of the formation of a spore with a multi-layered wall in this “sporocarp” species. The outer walls appear to be continuous with the wall of a subtending hypha, while the inner wall more likely developed de novo.

2 Material and Methods

The specimens of *Sporocarpon asteroides* described in this study are present in a single thin section (c. 60 µm thick) that was prepared from a coal ball from the Lower Coal Measures of Great Britain. The coal ball was collected sometime in the 1970s by John HOLMES, and comes from the Union Seam at Rowley Tip, Burnley (Lancashire), which is Westphalian A or Langsettian (Bashkirian/Lower Pennsylvanian) in age. The Union Seam and its stratigraphic equivalent in Great Britain, the Halifax Hard Seam, together with the contemporaneous Bouxharmont Seam in Belgium and the Finefrau-Nebenbank Seam in the Netherlands and Germany, represent the source strata of the richest European coal ball floras (for details, refer to GALTIER 1997).

The thin section was prepared according to a standard procedure in which a piece of the coal ball was cemented to a glass slide and then ground with abrasive until it was sufficiently thin to be examined with transmitted light (HASS & ROWE 1999). John HOLMES had already marked the *Sporocarpon* specimens on a drawing of the cut faces and labelled them as “curious spores 400 µm”. The coal ball, offcuts, and slide are deposited in the Collections de Paléobotanique, Université de Montpellier, France, under accession numbers B07 and B07A1aT 01. Fossils were analysed using normal transmitted light microscopy equipment. Digital images were captured with a Leica DFC-480 camera and gently processed in Adobe Photoshop CS4 for brightness and contrast.

3 Results

Coal ball B07 contains an accumulation of permineralized stigmarian rootlets, fragments of arborescent lycophyte axes, microphylls, megaspores, and *Lepidocarpon* sp., calamite remains, and several fern rachides, including *Botryopteris hirsuta* (WILL.) SCOTT and *B. ramosa* (WILL.) SCOTT, all embedded in a clear matrix interspersed with abundant organic debris, scattered fungal hyphae, and small propagules. Six three-dimensionally preserved specimens of *Sporocarpon asteroides* in different sectional planes occur in thin section B07A1aT 01. All, except two are, as far as we can see, typical examples of *S. asteroides* (for details on the morphology of this “sporocarp” species, refer to STUBBLEFIELD et al. 1983). The two atypical specimens (denoted I and II in Fig. 1A), which are detailed in the paragraphs below, are ideally cut and provide excellent median longitudinal section views of the fossils. They occur in close proximity to a third specimen (denoted III in Fig. 1A) sectioned slightly off center, and a fourth one (denoted IV in Fig. 1A) of which only a part of the outer surface can be seen because it is located in a different plane.

Specimens I and II correspond in size and overall appearance to typical *Sporocarpon asteroides*, but differ from all previously described individuals in regard to the outer boundary of the cavity and the cavity contents (Figs. 1B, C, E, 2A–D). Both specimens exhibit the characteristic, irregularly lobed pseudoparenchymatous investment enveloping a (near-)spherical cavity approximately 300 µm in diameter. A contiguous wall that extends along, and is closely appressed to, the inner surface of the investment, as it occurs in typical *S. asteroides*, is present in neither specimen (Figs. 1E, 2C, D). Instead, each of the two cavities contains a single large sac-like vesicle (sv in Figs. 1B, C_1, E, $2A_1$, B_1, C, D) with a thin, wrinkled, and finely granulose wall (e.g., Fig. 2C); the position of the vesicle within the cavity is eccentric. The vesicle is c. 230 µm high and 215 µm wide in specimen I and c. 260 µm high and 230 µm wide in specimen II. A similar, but distinctly smaller and bulb-shaped vesicle is present in specimen III (arrow in Fig. 1D). What appear to be short fragments of narrow filament- or fiber-like structures of some kind are recognizable here and there in the space between the investment and the vesicle wall, particularly where the two structures are in close proximity to each other (fs in Fig. 1E).

Located within the confines of the vesicle in specimens I and II is a compound structure that comprises a basal, squat-columnar component (less than 40 µm high and 20 µm wide), which is hollow in specimen II (sce in Figs. $1C_1$, E, $2A_1$) but apparently massive in specimen I (sce in Figs. 1B, $2B_1$). Attached laterally to the columnar component are the remains of at least two walls (w1 and w2 in Figs. 1B, C_1, $2A_2$, B_2) of which, unfortunately, only fragments are preserved. Moreover, at the tip of the columnar component is a walled sphere, which is more or less intact and c. 155 µm in diameter in specimen I (cs in Figs. 1B, $2B_1$), but collapsed and mostly disintegrated (but still recognizable) in specimen II (cs in Figs. $1C_1$, $2B_1$). The wall of this sphere appears to be layered (csw in Fig. 2D). As to whether the sphere was physically connected to the tip of the columnar component or developed freely (de novo) within the confines of the innermost surrounding wall (w2) cannot be determined. The latter seems to be more likely, however, based on the position of the collapsed sphere in specimen II (Fig. $1C_1$). Specimen II also suggests that the columnar component was connected to, or continued into, some structure on the outside of the "sporocarp" (arrows in Fig. $1C_2$). Unfortunately, this structure is either not preserved, or not located in the portion (slice) of the "sporocarp" present in the thin section. The vesicle in the cavity of specimen III is empty (Fig. 1D). Tiny, dot-shaped inclusions, which occur in large numbers in the spaces between the individual wall layers (Fig. 2D), are probably remains of the decayed parts of the compound structure (but see below).

4 Discussion

Fossil fungal "sporocarps" range among the most extensively studied pre-Cretaceous fungal fossils, and their morphology is well understood today (KRINGS et al. 2014). Nevertheless, the biological nature and affinities of these structures have remained a matter of controversy since they were first brought to the attention of the scientific community by CARRUTHERS (1873) and WILLIAMSON (1878, 1880, 1883). The fact that virtually all "sporocarp" specimens documented to date appear to be fully developed (mature) structures, in tandem with the lack of information on other life cycle stages of the organisms that produced them, and the inconsistency that exists with regard to the cavity contents are the main obstacles to a more complete understanding of these fossils (TAYLOR et al. 2015).

4.1 The sac-like vesicle

The most recent taxonomic revision of the "sporocarp" genus *Sporocarpon* by STUBBLEFIELD et al. (1983) recognizes three species, *S. asteroides*, *S. cellulosum* WILL., and *S. leismanii* STUBBLEFIELD et al., all of them characterized by a prominent, cohesive investment that is pseudoparenchymatous. The species differ primarily in the organization of the investment, which is irregularly lobed in *S. asteroides*, constructed of radiating files of cells in *S. cellulosum*, and prolonged into narrow, conical rays in *S. leismanii*. The cavities of the *Sporocarpon* specimens figured in literature are either empty or contain small spheres, or they contain a single large sphere, which is either empty or contains one or more smaller spheres (WILLIAMSON 1880; MCLEAN 1922; HUTCHINSON 1955; BAXTER 1960, 1975; DAVIS & LEISMAN 1962; STUBBLEFIELD et al. 1983). However, structures corresponding to the ones present in the fossils described in this study have not been documented previously in *Sporocarpon*, with the exception of the sac-like vesicle. STUBBLEFIELD et al. (1983) found that the wall lining the inner surface of the investment (i.e. the cavity perimeter wall) in *S. asteroides* is multi-layered and granulose on the outer surface of the innermost layer. The sac-like vesicles in the specimens described here (sv in Fig. 1B, C, arrow in Fig. 1D) all possess a finely granulose wall (sv in Figs. 1E, 2C), suggesting that the vesicle actually is the innermost layer of the perimeter wall that has separated from the outer layers. The outer layers, in turn, no longer exist. They appear to have disintegrated, except for remnants occurring in the form of short filament- or fiber-like fragments in the space between the inner surface of the investment and the vesicle (fs in Fig. 1E). If this is accurate, then the question arises why the vesicle is so much smaller than the cavity? One possible explanation could be that the perimeter wall in vivo was of considerable thickness. However, a thick perimeter wall has not been recorded for any fossil fungal "sporocarp" to date, which renders this hypothesis improbable. More likely, based on the wrinkled wall, is that the vesicle has shrunk during fossilization (see below). An alternative, albeit highly speculative, interpretation of the filament- or fiber-like structures in *S. asteroids*, and likewise of the dot-shaped inclusions visible in the spaces between the individual wall layers (Fig. 2D), is that they are endosymbionts, perhaps bacteria comparable to the endobacteria found in fungi today (BONFANTE & DESIRÒ 2017), including Endogonales (DESIRÒ et al. 2015) and many species of Glomeromycota (e.g., DESIRÒ et al. 2014; TOOMER et al. 2015; VENICE et al. 2021).

4.2 The compound structure

The compound structure that occurs in the vesicle in specimens I and II is evidence that a spore of some kind (i.e. the central sphere) was formed inside these "sporocarps," and that this spore had a complex wall comprised of a wall proper (i.e. the layered wall of the central sphere; csw in Fig. 2D) and the two outer walls labelled w1 and w2 in Figs. 1B, C_1, $2A_2$, and B_2. A spore-like body bounded by a multi-layered wall has previously been documented solely in the Mississippian "sporocarp" *Roannaisia bivitilis* T.N. TAYLOR et al. from France (TAYLOR et al. 1994: pl. II, 1). What appear to be concentrically arranged walls or wall layers are also visible in several Pennsylvanian *Traquairia* specimens from Great Britain and the United States (SCOTT 1911: textfig. 4; STUBBLEFIELD & TAYLOR 1983; KRINGS et al. 2011d: pl. I, 1). It has been suggested, however, that these layers have formed from the splitting of the cavity perimeter wall during fossilization (STUBBLEFIELD & TAYLOR 1983). By contrast, KRINGS et al. (2011d) believe that they represent an artefact which came into being during the permineralization process as a result of several successive phases of shrinking of the spore-like body in the cavity. Walls w1 and w2 in *Sporocarpon asteroides* originate from the base of the compound structure, where they arise from the squat-columnar component, which we interpret as the tip of a subtending hypha. Accordingly, the physical connection between w1 and w2 and the columnar component would imply that these walls were continuous with the wall of the subtending hypha. The wall bounding the central sphere probably formed de novo.

4.3 Affinities to Glomeromycota

Based on the preceding considerations, we entertain the possibility that *Sporocarpon asteroides* was a unisporic sporocarp with a pseudoparenchymatous peridium that enclosed a spore with a multi-layered wall (Fig. 3). The cavity perimeter wall is viewed as a product of the innermost layer of the peridium, rather than as a part of the spore. The origin of the peridium remains elusive. No evidence of paired or single gametangia (and suspensors) has been found, suggesting that the spore developed asexually and by blastic expansion of a hyphal tip, and thus that *S. asteroides* may have been a member of the Glomeromycota. This concurs with a major counterargument to the interpretation of fossil fungal “sporocarps” in general as zygosporangia containing zygospores, namely the total lack of evidence of gametangia in these structures (KRINGS & TAYLOR 2012; KRINGS et al. 2013a). Even if the gametangia, as argued elsewhere (KRINGS & HARPER 2020; KRINGS 2022), were small and embedded in the investment, if they have existed, then at least some would have been detected, given the large number of “sporocarp” specimens examined to date. Several bona fide zygomycete fungi that also have been described from the Carboniferous and Triassic show zygosporangia with attached gametangia, and investments similar to the ones seen in fossil fungal “sporocarps” (WHITE & TAYLOR 1989b; KRINGS et al. 2012, 2013b).

Glomeromycota are soil-borne fungi that enter into mycorrhizal associations with plants. They produce large (up to more than 800 μm in diameter) spores with multi-layered walls on non-septate hyphae (REDECKER & RAAB 2006; WALKER et al. 2018). The oldest bona fide fossil glomeromycotan spores are Early Devonian in age (e.g., STUBBLEFIELD & BANKS 1983; HARPER et al. 2020; WALKER et al. 2021; LALICA & TOMESCU 2022), and there is also documented evidence of Glomeromycota from several of those Carboniferous deposits that have yielded “sporocarp” fossils (e.g., WAGNER & TAYLOR 1981, 1982; STUBBLEFIELD et al. 1985; KRINGS et al. 2011c). In addition, there are various types of mantled fungal reproductive units from the Lower Devonian Rhynie chert that are morphologically similar to Carboniferous fungal “sporocarps” and demonstrably borne on a simple tubular subtending hypha. These fossils have also been considered to belong to the Glomeromycota (KRINGS & TAYLOR 2014; KRINGS & HARPER 2017, 2020). Unfortunately, the continuation of the subtending hypha on the outside of *Sporocarpon asteroides* remains unknown. There is, in fact, very little documented evidence of the structures that gave rise to any of the fossil fungal "sporocarps." One putative subtending structure occurs in a specimen of *Mycocarpon cinctum* M. KRINGS et al. from the Mississippian of France in the form of an inflated appendage extending from the outer surface of the investment (KRINGS et al. 2010b: pl. 1, fig. 1a, pl. 2, fig. 4). This structure appears to interface the "sporocarp" with its source organism, and is perhaps congruent with the bulbous bases seen in present-day species of the glomeromycotan order Gigasporales (KHADE 2011; WALKER et al. 2018).

4.4 Possible objections

The interpretation of *Sporocarpon asteroides* as a member of the Glomeromycota could be countered by pointing out that the putative spore is considerably smaller than the cavity bounded by the investment. The question therefore is what gave shape to the spherical cavity in this fossil? A peridium typically precedes spore formation or develops concomitantly with the spores, encloses the spores, and expands as the spores grow larger (GIOVANNETTI et al. 1991; MEIER & CHARVAT 1992, and references therein). We believe that the size difference between the cavity and spore in *S. asteroides* is a preservation artefact resulting from shrinkage of the spore due to plasmolysis caused by the physico-chemical properties of the depositional environment and the permineralization process (see SCOTT & REX 1985). A similar effect can be seen in present-day glomeromycotan spores after they have been exposed to certain acidic mounting media. It is known, for instance, that spores embedded in PVLG (polyvinyl-lacto-glycerol) may shrink considerably or collapse with plasmolysis of the spore contents (GAMPER et al. 2009). An ensuing question is, why is there not a spore in every specimen of *S. asteroides*? We speculate that the pseudoparenchymatous investment may have been durable and remained (largely) intact for an extended period of time after the spore had germinated or become non-viable for some other reason and its walls decayed. If so, then the majority of *S. asteroides* fossils would be empty investments.

It might also be argued that cohesive, pseudoparenchymatous investments like that of *Sporocarpon asteroides* do not occur in present-day Glomeromycota (e.g., FURRAZOLA et al. 2016; GUPTA 2017; JOBIM et al. 2019; YAMAMOTO et al. 2019). It is basically correct that the fossil differs from all present-day members of the Glomeromycota producing spores enclosed in an evidently differentiated hyphal peridium. However, there are certain unisporic sporocarps that, in section view, display a relatively high level of organization in the hyphal coverings (e.g., MEIER & CHARVAT 1992: fig. 8). Moreover, we have to consider the possibility that fossil lineages of glomeromycotan fungi, as well as fossil representatives of present-day lineages, have existed that were characterized by structural features unknown in any present-day representative.

Finally, an interesting specimen of *Sporocarpon asteroides* has been figured by McLean (1922: pl. VIII, fig. 9) and described as "containing a spherical structure apparently dehiscing and extruding a mass of what appear to be small spores united by filaments." It is more likely, however, that the spherical structure and the alleged spores do not belong together. The former appears to correspond to the sac-like vesicle seen in the specimens described here, whereas the latter probably represent an intrusive organism that had entered *S. asteroides* and produced its own thallus and reproductive units. Support for this view comes from abundant occurrences of spore-like bodies interconnected by filaments in Carboniferous plant parts preserved in coal balls and chert (Dotzler et al. 2011; Krings et al. 2010a, 2011b; Strullu-Derrien et al. 2021). Moreover, there is evidence of mycoparasitism in *Dubiocarpon* which demonstrates that Carboniferous fungal "sporocarps" were invaded by other organisms and used as a habitat (Krings et al. 2011a).

5 Conclusions

Fossil fungal "sporocarps" continue to invite speculation as to their biological nature and systematic affinities. Although there are morphological differences that have been used to classify the "sporocarps" into a number of fossil genera and species, there are also basic similarities, which could mean that they all belong to the same higher taxonomic category. Both the Ascomycota and zygomycete fungi have been discussed in this respect, but neither of these attributions has received undivided approval. The specimens presented in this study strongly suggest that the "sporocarp" species *Sporocarpon asteroides* was a unisporic glomeromycotan sporocarp. This does not necessarily mean, however, that the other *Sporocarpon* species, let alone the other "sporocarp" genera, were also Glomeromycota. Rather, we consider it likely that the assemblage of fossils subsumed under fossil fungal "sporocarps" is heterogeneous and includes a range of biologically different structures. Our knowledge of these structures in general continues to be incomplete, and there is need for additional specimens to be discovered, thoroughly investigated, and documented. The present study demonstrates that new features of fossil fungal "sporocarps" can still be found, and can be used to refine current interpretations of these common but as yet enigmatic fossils, and eventually take us to the core of their biological nature and affinities.

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6 References

BAXTER, R.W. (1960): *Sporocarpon* and allied genera from the American Pennsylvanian. – Phytomorphology, **10**: 19–25.

BAXTER, R.W. (1975): Fossil fungi from American Pennsylvanian coal balls. – University of Kansas Paleontological Contributions, **77**: 1–6.

BONFANTE, P. & DESIRÒ, A. (2017): Who lives in a fungus? The diversity, origins and functions of fungal endobacteria living in Mucoromycota. – ISME Journal, **11**: 1727–1735.

CARRUTHERS, W. (1873): On *Traquairia*, a radiolarian rhizopod from the coal-measures. – Report of the 42nd Meeting of the British Association for the Advancement of Science; held at Brighton in August 1872, p. 126; London, UK (John Murray).

DAVIS, B. & LEISMAN, G.A. (1962): Further observations on *Sporocarpon* and allied genera. – Bulletin of the Torrey Botanical Club, **89**: 97–109.

DESIRÒ, A., SALVIOLI, A., NGONKEU, E., MONDO, S., EPIS, S., FACCIO, A., KAECH, A., PAWLOWSKA, T. & BONFANTE, P. (2014): Detection of a novel intracellular microbiome hosted in arbuscular mycorrhizal fungi. – ISME Journal, **8**: 257– 270.

DESIRÒ, A., FACCIO, A., KAECH, A., BIDARTONDO, M.I. & BONFANTE, P. (2015): *Endogone*, one of the oldest plant-associated fungi, host unique Mollicutes-related endobacteria. – New Phytologist, **205**: 1464–1472.

DOTZLER, N., TAYLOR, T.N., GALTIER, J. & KRINGS, M. (2011): *Sphenophyllum* (Sphenophyllales) leaves colonized by fungi from the Upper Pennsylvanian Grand-Croix cherts of central France. – Zitteliana A, **51**: 3–8.

FURRAZOLA, E., TORRES-ARIAS, Y., THOEN, D., BERBARA, R.L.L., JOBIM, K. & GOTO, B.T. (2016): *Glomus segmentatum*, rediscovery of a rare epigeous sporocarpic fungus to Cuba. – Current Research in Environmantal & Applied Mycology, **6**: 143–149.

GALTIER, J. (1997): Coal-ball floras of the Namurian-Westphalian of Europe. – Review of Palaeobotany and Palynology, **95**: 51–72.

GAMPER, H.A., WALKER, C. & SCHÜßLER, A. (2009): *Diversispora celata* sp. nov.: molecular ecology and phylotaxonomy of an inconspicuous arbuscular mycorrhizal fungus. – New Phytologist, **182**: 495–506.

GIOVANNETTI, M., AVIO, L. & SALUTINI, L. (1991): Morphological, cytochemical, and ontogenetic characteristics of a new species of vesicular-arbuscular mycorrhizal fungus. – Canadian Journal of Botany, **68**: 161–167.

GUPTA, M.M. (2017): Differential response of arbuscular mycorrhizal sporocarps in long-term trap culturing. – Phytomorphology, **67**: 1–11.

HARPER, C.J., WALKER, C., SCHWENDEMANN, A.B., KERP, H. & KRINGS, M. (2020): *Archaeosporites rhyniensis* gen. et sp. nov. (Glomeromycota, Archaeosporaceae), from the Lower Devonian Rhynie chert – a fungal lineage morphologically unchanged from more than 400 million years. – Annals of Botany, **126**: 915–928.

HASS, H. & ROWE, N.P. (1999): Thin sections and wafering. – In: T.P. JONES & N.P. ROWE (Eds.): Fossil Plants and Spores: Modern Techniques, p. 76–81; London, UK (The Geological Society).

HUTCHINSON, S.A. (1955): A review of the genus *Sporocarpon* Williamson. – Annals of Botany, **19**: 425–435.

HUTCHINSON, S.A. & WALTON, J. (1953): A presumed ascomycete from the Upper Carboniferous. – Nature, **172**: 36–37.

JOBIM, K., BŁASZKOWSKI, J., NIEZGODA, P., KOZŁOWSKA, A., ZUBEK, S., MLECZKO, P., CHACHUŁA, P., ISHIKAWA, N.K. & GOTO, B.T. (2019): New sporocarpic traxa in the phylum Glomeromycota: *Sclerocarpum amazonicum* gen. et sp. nov. in the family Glomeraceae (Glomerales) and *Diversispora sporocarpica* sp. nov. in the Diversisporaceae (Diversisporales). – Mycological Progress, **18**: 369–384.

KHADE, S.W. (2011): New characteristics for morphotaxonomy of *Gigaspora* species belonging to arbuscular mycorrhizal fungi. – Journal of Plant Development, **18**: 71–80.

KRINGS, M. (2022): An unusual specimen of the enigmatic fungal reproductive unit *Windipila spinifera* from the Lower Devonian Rhynie cherts of Scotland. – Zitteliana, **96**: 145–152.

KRINGS, M. & HARPER, C.J. (2017): A mantled fungal reproductive unit from the Lower Devonian Windyfield chert, Scotland, with prominent spines and otherwise shaped projections extending out from the mantle. – Neues Jahrbuch für Geologie und Paläontologie, Abhandlungen, **285**: 201–211.

Krings, M. & Harper, C.J. (2020): Morphological diversity of fungal reproductive units in the Lower Devonian Rhynie and Windyfield cherts, Scotland: A new species of the genus *Windipila*. – PalZ, **94**: 619–632.

Krings, M. & Taylor, T.N. (2012): Fungal reproductive units enveloped in a hyphal mantle from the Lower Pennsylvanian of Great Britain, and their relevance to our understanding of Carboniferous fungal "sporocarps". – Review of Palaeobotany and Palynology, **175**: 1–9.

Krings, M. & Taylor, T.N. (2014): A mantled fungal reproductive unit from the Lower Devonian Rhynie chert that demonstrates Carboniferous "sporocarp" morphology and development. – Neues Jahrbuch für Geologie und Paläontologie, Abhandlungen, **273**: 197–205.

Krings, M. & Taylor, T.N. (2015a): A fungal reproductive unit from the Lower Devonian Rhynie chert (Aberdeenshire, Scotland) that demonstrates an unusual hyphal investment pattern. – Scottish Journal of Geology, **51**: 131–139.

Krings, M. & Taylor, T.N. (2015b): Mantled fungal reproductive units in land plant tissue from the Lower Devonian Rhynie chert. – Bulletin of Geosciences, **90**: 1–6.

Krings, M., Dotzler, N., Taylor, T.N. & Galtier, J. (2010a): A fungal community in plant tissue from the Lower Coal Measures (Langsettian, Lower Pennsylvanian) of Great Britain. – Bulletin of Geosciences, **85**: 679–690.

Krings, M., Dotzler, N., Taylor, T.N. & Galtier, J. (2010b): Microfungi from the upper Visean (Mississippian) of central France: Structure and development of the sporocarp *Mycocarpon cinctum* nov. sp. – Zitteliana A, **50**: 127–135.

Krings, M., Dotzler, N. & Taylor, T.N. (2011a): Mycoparasitism in *Dubiocarpon*, a fungal sporocarp from the Carboniferous. – Neues Jahrbuch für Geologie und Paläontologie, Abhandlungen, **262**: 241–245.

Krings, M., Taylor, T.N., Dotzler, N. & Galtier, J. (2011b): Fungal remains in cordaite (Cordaitales) leaves from the Upper Pennsylvanian of central France. – Bulletin of Geosciences, **86**: 777–784.

Krings, M., Taylor, T.N., Taylor, E.L., Dotzler, N. & Walker, C. (2011c): Arbuscular mycorrhizal-like fungi in Carboniferous arborescent lycopsids. – New Phytologist, **191**: 311–314.

Krings, M., Taylor, T.N. & White, J.F. (2011d): Fungal sporocarps from the Carboniferous: An unusual specimen of *Traquairia*. – Review of Palaeobotany and Palynology, **168**: 1–6.

Krings, M., Taylor, T.N., Dotzler, N. & Persichini, G. (2012): Fossil fungi with suggested affinities to the Endogonaceae from the Middle Triassic of Antarctica. – Mycologia, **104**: 835–844.

Krings, M., Taylor, T.N. & Dotzler, N. (2013a): Fossil evidence of the zygomycetous fungi. – Persoonia, **30**: 1–10.

Krings, M., White, J.F., Dotzler, N. & Harper, C.J. (2013b): A putative zygomycetous fungus with mantled zygosporangia and apposed gametangia from the Lower Coal Measures (Carboniferous) of Great Britain. – International Journal of Plant Sciences, **174**: 269–277.

Krings, M., Taylor, T.N., Taylor, E.L., Kerp, H. & Dotzler, N. (2014): First record of a fungal “sporocarp” from the Lower Devonian Rhynie chert. – Palaeobiodiversity and Palaeoenvironments, **94**: 221–227.

Krings, M., Taylor, T.N., Dotzler, N. & Harper, C.J. (2016): Morphology and ontogenetic development of *Zwergimyces vestitus*, a fungal reproductive unit enveloped in a hyphal mantle from the Lower Devonian Rhynie chert. – Review of Palaeobotany and Palynology, **228**: 47–56.

Lalica, M.A.K. & Tomescu, A.M.F. (2022): The early fossil record of glomeromycete fungi: New data on spores associated with early tracheophytes in the Lower Devonian (Emsian; c. 400 Ma) of Gaspé (Quebec, Canada). – Review of Palaeobotany and Palynology, **298**: ID104590.

McLean, R.C. (1922): On the fossil genus *Sporocarpon*. – Annals of Botany, **36**: 71–90.

Meier, R. & Charvat, I. (1992): Peridial development in *Glomus mosseae* (Glomaceae). – American Journal of Botany, **79**: 928–936.

Pirozynski, K.A. (1976): Fossil fungi. – Annual Review of Phytopathology, **14**: 237–246.

Redecker, D. & Raab, P. (2006): Phylogeny of the Glomeromycota (arbuscular mycorrhizal fungi): recent developments and new gene markers. – Mycologia, **98**: 885–895.

Scott, A.C. & Rex, G. (1985): The formation and significance of Carboniferous coal balls. – Philosophical Transactions of the Royal Society of London B: Biological Sciences, **311**: 123–137.

Scott, R. (1911): On *Traquairia*. – Annals of Botany, **25**: 459–467.

Strullu-Derrien, C., Gèze, M., Spencer, A.R.T., De Franceschi, D., Kenrick, P., Selosse, M.A. & Knoll, A.H. (2021): An expanded diversity of oomycetes in Carboniferous forests: Reinterpretation of *Oochytrium lepidodendri* (Renault 1894) from the Esnost chert, Massif Central, France. – PLoS ONE, **16**: e0247849.

STUBBLEFIELD, S.P. & BANKS, H.P. (1983): Fungal remains in the Devonian trimerophyte *Psilophyton dawsonii*. – American Journal of Botany, **70**: 1258–1261.
STUBBLEFIELD, S.P. & TAYLOR, T.N. (1983): Studies of Paleozoic fungi. I. The structure and organization of *Traquairia* (Ascomycota). – American Journal of Botany, **70**: 387–399.
STUBBLEFIELD, S.P., TAYLOR, T.N., MILLER, C.E. & COLE, G.T. (1983): Studies in Carboniferous fungi. II. The structure and organization of *Mycocarpon*, *Sporocarpon*, *Dubiocarpon*, and *Coleocarpon* (Ascomycotina). – American Journal of Botany, **70**: 1482–1498.
STUBBLEFIELD, S.P., TAYLOR, T.N. & MILLER, C.E. (1985): Studies of Paleozoic fungi. IV. Wall ultrastructure of fossil endogonaceous chlamydospores. – Mycologia, **77**: 83–96.
TAYLOR, T.N. & WHITE, J.F. Jr. (1989): Fossil fungi (Endogonaceae) from the Triassic of Antarctica. – American Journal of Botany, **76**: 389–396.
TAYLOR, T.N., GALTIER, J. & AXSMITH, B.J. (1994): Fungi from the Lower Carboniferous of central France. – Review of Palaeobotany and Palynology, **83**: 253–260.
TAYLOR, T.N., KRINGS, M. & TAYLOR, E.L. (2015): Fossil Fungi. 1st Edition – xv + 382 pp.; Amsterdam, Boston, Heidelberg, London (Elsevier/Academic Press Inc.).
TOOMER, K.H., CHEN, X., NAITO, M., MONDO, S.J., DEN BAKKER, H.C., VANKUREN, N.W., LEKBERG, Y., MORTON, J.B. & PAWLOWSKA, T.E. (2015): Molecular evolution patterns reveal life history features of mycoplasma-related endobacteria associated with arbuscular mycorrhizal fungi. – Molecular Ecology, **24**: 3485–3500.
VENICE, F., CHIALVA, M., DOMINGO, G., NOVERO, M., CARPENTIERI, A., SALVIOLI DI FOSSALUNGA, A., GHIGNONE, S., AMORESANO, A., VANNINI, C., LANFRANCO, L. & BONFANTE, P. (2021): Symbiotic responses of *Lotus japonicus* to two isogenic lines of a mycorrhizal fungus differing in the presence/absence of an endobacterium. – The Plant Journal, **108**: 1547–1564.
WAGNER, C.A. & TAYLOR, T.N. (1981): Evidence for endomycorrhizae in Pennsylvanian age plant fossils. – Science, **212**: 562–563.
WAGNER, C.A. & TAYLOR, T.N. (1982): Fungal chlamydospores from the Pennsylvanian of North America. – Review of Palaeobotany and Palynology, **37**: 317–328.
WALKER, C., HARPER, C.J., BRUNDRETT, M.C. & KRINGS, M. (2018): Looking for arbuscular mycorrhizal fungi (AMF) in the fossil record – an illustrated guide. – In: M. KRINGS, C.J. HARPER, N.R. CÚNEO & G.W. ROTHWELL (Eds.): Transformative Paleobotany: Commemorating the Life and Legacy of Thomas N. Taylor, pp. 481–518; London (Elsevier/Academic Press Inc.).

WALKER, C., HARPER, C.J., BRUNDRETT, M. & KRINGS, M. (2021): The Early Devonian fungus *Mycokidstonia sphaerialoides* from the Rhynie chert is a member of the Ambisporaceae (Glomeromycota, Archaeosporales), not an ascomycete. – Review of Palaeobotany and Palynology, **287**: ID104384.

WHITE, J.F. Jr. & TAYLOR, T.N. (1988): Triassic fungus from Antarctica with possible ascomycetous affinities. – American Journal of Botany, **75**: 1495–1500.

WHITE, J.F. Jr., TAYLOR, T.N. (1989a): An evaluation of sporocarp structure in the Triassic fungus *Endochaetophora*. – Review of Palaeobotany and Palynology, **61**: 341–345.

WHITE, J.F. Jr. & TAYLOR, T.N. (1989b): Triassic fungi with suggested affinities to the Endogonales (Zygomycotina). – Review of Palaeobotany and Palynology, **61**: 53–61.

WHITE, J.F. Jr. & TAYLOR, T.N. (1991): Fungal sporocarps from the Triassic peat deposits in Antarctica. – Review of Palaeobotany and Palynology, **67**: 229–236.

WILLIAMSON, W.C. (1878): On the organization of the fossil plants of the coal-measures. Part IX. – Philosophical Transactions of the Royal Society of London, **169**: 319–364.

WILLIAMSON, W.C. (1880): On the organization of the fossil plants of the coal-measures. Part 10 – including an examination of the supposed radiolarians of the Carboniferous rocks. – Philosophical Transactions of the Royal Society, London, **B171**: 493–539.

WILLIAMSON, W.C. (1883): On the organization of the fossil plants of the coal-measures: – Part XII. – Philosophical Transactions of the Royal Society of London, **174**: 459–475.

YAMAMOTO, K., TAKASHIMA, Y., SUYAMA, M., MASAKI, T. & DEGAWA, Y. (2019): A sporocarpic species of Glomeromycotina, *Glomus radiatum* new to Japan. – Truffology, **2**: 6–9.

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Figure captions

Fig. 1. New specimens of the Carboniferous fungal "sporocarp" *Sporocarpon asteroides* from the Lower Coal Measures of Great Britain. **A.** Cluster of four specimens (labelled I–IV) in coal ball matrix rich in organic debris; bar = 300 µm. **B, C$_1$.** Cavities of specimens I and II in higher magnification (in the median longitudinal section view), showing contents; sv = sac-like vesicle, sce = squat-columnar component, w1 and w2 = outer walls, cs = central sphere bounded by a layered wall proper; bars = 100 µm. **C$_2$.** Lower part of specimen II, same magnification as Fig. 1C$_1$ but slightly different focal plane, showing squat-columnar component apparently continuing into some structure on the "sporocarp" outside (arrows); bar = 100 µm. **D.** Cavity of specimen III, containing an empty sac-like vesicle (arrow); bar = 100 µm. **E.** Detail of Fig. 1C$_1$, focusing on proximal portion of investment and compound structure; fs = layer of filament- or fiber-like structures between investment and vesicle wall (sv); sce = squat-columnar component; bar = 20 µm.

Fig. 2. New specimens of the Carboniferous fungal "sporocarp" *Sporocarpon asteroides* from the Lower Coal Measures of Great Britain. **A, B.** Proximal portions of cavities of specimens I and II (in longitudinal section views) in normal (A$_1$, B$_1$) and, in a slightly different focal plane, inverted (A$_2$, B$_2$) light, showing sac-like vesicle (sv) and compound structure, the latter comprised of squat-columnar component (sce), two outer walls (w1, w2), and central sphere (cs) bounded by layered wall proper; bars = 50 µm. **C.** Detail of Fig. 1B, showing finely

granular wall of sac-like vesicle (sv) and portion of investment (in); bar = 20 µm. **D.** Detail of Fig. 1B, focusing on layered wall of central sphere (csw), sac-like vesicle (sv), and investment (in); bar = 30 µm.

Fig. 3. *Sporocarpon asteroides* in vivo, reconstruction. Median longitudinal section, showing the characteristic pseudoparenchymatous investment (in), the cavity perimeter wall with its finely granular innermost layer (cpw), two spore walls w1 and w2 that are continuous with the wall of the subtending hypha, and a spore wall proper (csw) that formed de novo.