Zitteliana 96, 2022, 145–152 | DOI 10.3897/zitteliana.96.86327

SNSB Bayerische Staatssammlung für Paläontologie und Geologie

ZITTELIANA
An International Journal of Paleontology and Geobiology

An unusual specimen of the enigmatic fungal reproductive unit *Windipila spinifera* from the Lower Devonian Rhynie cherts of Scotland

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https://zoobank.org/B4312F0C-F88C-4F61-999B-D5E3FD553A99

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Academic editor: Alexander Nützel ♦ **Received** 9 May 2022 ♦ **Accepted** 26 July 2022 ♦ **Published** 8 August 2022

Abstract

Windipila spinifera from the Rhynie cherts is a spheroidal microfossil enveloped in a hyphal mantle from which extend prominent spines and otherwise shaped projections. It is believed to be a reproductive unit of a fungus in the Glomeromycota or zygomycetes, but features to determine the systematic affinities have not hitherto been documented. This study describes a new specimen of *W. spinifera* that contains a single spherical structure from which a hypha arises that extends outside and terminates in what appears to be a sporangium. The specimen is reminiscent of germinated zygospores of the germ-sporangial type, and thus may suggest affinities of *W. spinifera* to the zygomycetes. However, the interior sphere and its outgrowth could also be a part of another organism that had invaded *W. spinifera*.

Keywords

fossil fungi, Glomeromycota, intrusive organisms, zygomycetes, zygospore germination

1. Introduction

The Lower Devonian Rhynie cherts (including the Rhynie and Windyfield cherts) of Scotland give detailed insights into fungal diversity in a terrestrial ecosystem c. 410 Ma ago (Taylor et al. 2015; Krings et al. 2017a; Strullu-Derrien et al. 2019). One important proxy indicator of this diversity is the morphological variety of fungal propagules and microscopic reproductive units, both of which are present in virtually every thin section of the cherts. However, dealing with these fossils is difficult because they mostly occur detached from the systems on or in which they were produced, and thus do not provide an inclusive comparison with present-day equivalents to determine their systematic affinities (Krings and Harper 2020).

Several types of fungal reproductive units have been described from the Rhynie cherts that all possess an ancillary covering in the form of a hyphal investment or mantle. Mantle morphology varies (considerably) among the different types, and thus renders them easy to distinguish from one another (Krings and Taylor 2013, 2014, 2015a, 2015b; Krings et al. 2014, 2016, 2017b; Krings and Harper 2017, 2018, 2020). One of these fossils is *Windipila spinifera* (Fig. 1A), a walled spheroid less than 150 µm in diameter that is enveloped in circumferential hyphae from which extend prominent, thin-walled spines and otherwise shaped projections (Krings and Harper 2017, 2018).

Most mantled fungal reproductive units from the Rhynie cherts, including *Windipila spinifera*, are believed to belong to either the Glomeromycota or the zygomycete fungi based on similar features in modern lineages known to produce spores or sporangia with hyphal mantles (e.g., Krings et al. 2016, 2017a; Krings and Harper 2020). The occurrence of several specimens on what appear to be simple, tubular subtending hyphae has been used to suggest that *W. spinifera* developed asexually by blastic inflation and thickening of a hyphal tip, rather than sexually and following gametangial fusion, and for this reason might be a glomoid chlamydospore (Krings and Harper 2017). However, structural features to more precisely delimit the nature and systematic affinities of this fossil have not been documented to date.

This study describes a new specimen of *Windipila spinifera* from the Windyfield chert that contains a single spherical structure from which an unbranched hypha is given off that evidently passes through the wall and mantle and terminates in what appears to be a sporangium with a distal discharge papilla. While this specimen does also not clarify the affinities of *W. spinifera*, it resembles a germinated zygospore or azygospore with a tubular sporangiophore and distal germ sporangium, and thus is suggestive of affinities to the zygomycetes.

2. Geological setting

The Rhynie chert Lagerstätte is located in the northern part of the Rhynie outlier of Lower Old Red Sandstone in Aberdeenshire, Scotland, in an extensive sequence of sedimentary and volcanic rocks. The cherts occur in the Rhynie Block of the Dryden Flags Formation located northwest of the village of Rhynie. The Lagerstätte is made up of fossiliferous beds containing lacustrine shales and chert that have been interpreted as a series of ephemeral pools within a geothermal wetland, with alkali-chloride hot springs that were part of a complex hydrothermal system in a region affected by volcanic activity (Rice et al. 2002; Rice and Ashcroft 2004; Trewin and Fayers 2016; Channing 2017). The Windyfield chert site occurs ~700 m to the northeast of the original Rhynie chert site (Trewin and Rice 1992; Fayers 2003; Garwood et al. 2020). It was deposited in an area of hot-spring feeder activity based on the hydrothermally altered nature of its associated fluvio-lacustrine shales and sandstones; paleoenvironments ranged from terrestrial, vegetated aprons of laminated and brecciated sinter to low-temperature pools and marginal aquatic settings (Rice et al. 2002; Fayers 2003; Fayers and Trewin 2004). The Rhynie and Windyfield cherts are regarded as broadly coeval, i.e., 411.5±1.3 to 407.6±2.6 million years old according to Mark et al. (2011, 2013) and Parry et al. (2011), Pragian-earliest Emsian according to Wellman (2006, 2017) and Wellman et al. (2006). For details on the geology and development of the Rhynie chert Lagerstätte, as well as on the paleontology, refer to Trewin and Kerp (2017), Garwood et al. (2020), and the Rhynie chert volume edited by Edwards et al. (2018).

3. Material and methods

The specimen described below was identified in a thin section prepared by cementing a wafer of the Windyfield chert to a microscope slide, and then grinding the rock slice until it was sufficiently thin (i.e. c. 60–80 μm thick) to transmit light (for details on thin section preparation, refer to Hass and Rowe 1999). The slide is deposited in the SNSB-Bayerische Staatssammlung für Paläontologie und Geologie (SNSB-BSPG) in Munich, Germany, under accession number SNSB-BSPG 2016 VII 117. The specimen was studied using normal transmitted light microscopy; digital images were captured with a Leica DFC-480 camera and processed in Adobe Photoshop CS5. For comparison, another specimen of *Windipila spinifera* from the holdings of the SNSB-BSPG (accession number SNSB-BSPG 2016 VII 82) has also been illustrated.

4. Results

The specimen (Fig. $1B_1$) is located in the chert matrix, within silicified litter comprised predominantly of fragmented and mostly heavily degraded land plant axes, dispersed land plant spores, unidentifiable debris, and fungal hyphae. The reproductive unit is spherical, roughly 145 μm in diameter (including mantle but excluding projections), and composed of a central cavity bounded by a non-hyphal wall up to 2 μm thick. No evidence has been found of pores or otherwise shaped orifices in the wall. The reproductive unit is enveloped in a hyphal mantle composed of 1–3 layers of tightly interlaced hyphae extending along the outer surface of the non-hyphal wall. Prominent projections, irregularly distributed over the entire surface, are given off from the circumferential hyphae in an outward direction. However, there are fewer projections in this specimen as compared to the specimens described previously (Fig. $1A_{1,2}$; Krings and Harper 2017, 2018). The structure in vivo was probably positioned terminally on a lateral branch or outgrowth of a tubular hypha (see below). However, the attachment site is obscured by the mantle hyphae and most of the hyphal nexus is not recognizable due to the plane of the section through the specimen.

The reproductive unit contains a single, more or less spherical structure (henceforth called interior sphere) that is c. 55 μm in diameter, lacks structured contents, and is delineated by a smooth wall up to 0.75 μm thick. Its position within the cavity is highly eccentric in such a way that it comes into contact with, and in vivo perhaps adhered to, the inner surface of the non-hyphal wall of the reproductive unit. The interior sphere is somewhat flattened where it is in physical contact with this wall. No evidence of a subtending hypha or any other kind of parental structure has been found. However, the cavity of the reproductive unit contains shrivelled fragments of thin-walled hyphae or filaments (Fig. $1B_3$). Arising from roughly the center of the flattened portion of the interior sphere is a narrow hypha 1–2 μm wide (arrows in Fig. 1C), which evidently passes through the non-hyphal wall of the repro-

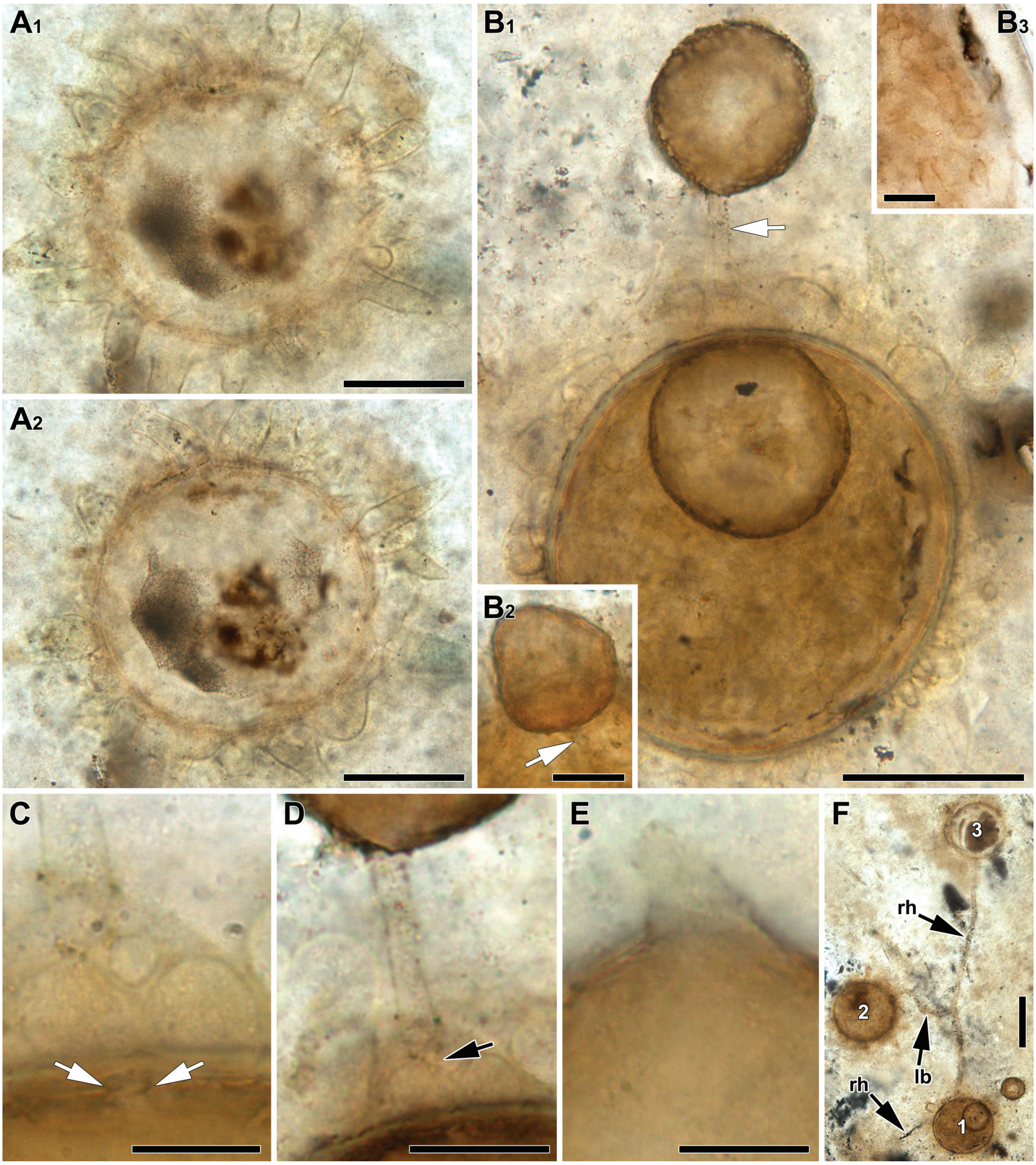

Figure 1. *Windipila spinifera* from the Lower Devonian Rhynie cherts (Windyfield chert). **$A_{1,2}$.** Well preserved specimen in two different focal planes (A_1: median optical section, A_2: section off center), with prominent spines and otherwise shaped projections extending from mantle (focal plane A_1 previously published by Krings and Harper 2018: fig. 1A). Slide SNSB-BSPG 2016 VII 82. **B–F.** New specimen. Slide SNSB-BSPG 2016 VII 117. **B_1.** Overview (median optical section), showing interior sphere in highly eccentric position and stalk-like hypha (arrow) terminating in exterior sphere. **B_2.** Second exterior structure in different plane; note attached fragment of putative parental hypha (arrow). **B_3.** Shrivelled hyphae or filaments in cavity of reproductive unit. **C.** Hypha arising from interior sphere (arrows). **D.** Stalk-like hypha becoming wider in periphery of mantle (arrow), and extending in outward direction. **E.** Papilla-like, distal putative discharge apparatus of exterior sphere. **F.** Hypha (rh) with three attached *W. spinifera* reproductive units (labeled 1–3), of which the one in the lower right of the image (labeled 1) is magnified in Fig. $1B_1$; specimen 2 occurs on a lateral branch (lb) and 3 at the tip of the hypha. Scale bars: 10 µm (**B_3, C, E**); 20 µm (**B_2, D**); 50 µm (**A_1, A_2, B_1**); 100 µm (**F**).

ductive unit and proceeds on the outside into the mantle. Unfortunately, the exact path of the hypha through the wall and mantle is not traceable. The hypha reappears in the periphery of the mantle where it expands to up to 6.5 µm wide (arrow in Fig. 1D) and becomes tubular (stalk-like), and from there runs outwards c. 40 µm into the ambience (arrow in Fig. $1B_1$). The hypha is unbranched and terminates in another spherical structure (henceforth called exterior sphere), which is 46 μm in diameter, bounded by a smooth wall, and does also not have structured contents. The exterior sphere is characterized by a single, distal, thin-walled papilla-like projection 8.5 μm high (Fig. 1E). A second exterior structure, c. 38 μm in diameter and somewhat asymmetrical or distorted, occurs in close proximity to the exterior sphere in a different plane of the specimen (Fig. $1B_2$). This structure, which is partially embedded in the mantle of the reproductive unit, is subtended by a short fragment of what appears to be a hypha (arrow in Fig. $1B_2$). As to whether this hypha also originates from the interior sphere cannot be determined.

The hypha (rh in Fig. 1F) that gives rise to the specimen described above (labeled 1 in Fig. 1F) bears two further *Windipila spinifera* reproductive units (labeled 2 and 3 in Fig. 1F), one of which occurs on a lateral branch (lb in Fig. 1F), while the other (partially degraded) appears to be positioned at the tip of the hypha itself. Because these latter specimens do not exhibit any new or unusual features, they will not be discussed here any further.

5. Discussion

Eight morphologically different types of mantled fungal reproductive units have so far been described from the Rhynie cherts, of which *Windipila spinifera* with its prominent mantle spines certainly is one of the more peculiar ones (Krings and Harper 2017, 2018). A similar, albeit less impressive mantle occurs in the little-known *W. pumila* (Krings and Harper 2018). A third type attributed to *Windipila*, *W. wimmervoecksii*, is characterized by mantle hyphae with vesicle-like inflations from which extend needle-like processes that connect the inner mantle tier with an outer tier of irregularly inflated and interwoven hyphae (Krings and Harper 2020).

Windipila has been suggested to have affinities to either the Glomeromycota or the zygomycete fungi (Krings and Harper 2020). One of the key features distinguishing these two groups of fungi is the sexual stage of the life cycle, which occurs in zygomycetes as a result of zygosporogenesis following gametangial fusion, but has not been observed in Glomeromycota (Benjamin 1979; Benny et al. 2001, 2012). Mature zygosporangia or zygospores with attached gametangia and/or suspensors are therefore the most important component of the zygomycete life cycle that can be used to positively identify a fossil member in this group of fungi (Krings et al. 2012, 2013). However, none of the *Windipila* species can presently be positively identified as a zygomycete because there is no evidence of gametangia and/or suspensors. Rather, several specimens of *W. spinifera* (Krings and Harper 2017: fig. 2) and *W. wimmervoecksii* (Krings and Harper 2020: figs 2d, 3a) appear to have developed on simple, tubular subtending hyphae. Moreover, hyphal anastomoses and H-branching, which are common in several lineages of glomeromycotan fungi (Walker et al. 2018) but believed to be lacking or rare in zygomycetes (Gregory 1984; Glass and Fleissner 2006; Ivarsson et al. 2015), have been documented in *W. wimmervoecksii* (Krings and Harper 2020: figs 4a, b, 5).

The most interesting structural element of the only known specimen of *Windipila pumila* is a walled spherical structure that occurs in the cavity of this fossil (Krings and Harper 2018: fig. 2). Similar spheres have not hitherto been recorded in *W. spinifera* or *W. wimmervoecksii*, but they have been described in other Rhynie chert mantled fungal reproductive units, including *Scepasmatocarpion fenestrulatum* (Krings and Taylor 2015a: figs 1e, 2c) and *Zwergimyces vestitus* (Krings et al. 2016: pl. III, 2–8). The interior spheres have been used to speculate that these reproductive units could be sporangia or sporangiola, perhaps comparable to the zygosporangia of zygomycetes or the sporangial or sporangiolar layer(s) of certain glomeromycotan spore walls (see Walker et al. 2021: fig. 1), and the interior spheres either the zygospore or the sporangiospore proper (e.g., Krings and Harper 2018).

The fossil presented in this study provides the first evidence of the occurrence of interior spheres also in *Windipila spinifera*. Moreover, it exhibits several structural features pertaining to the interior sphere that have not been documented in any other Rhynie chert mantled reproductive unit. Said interior sphere gives off a hypha that exits the reproductive unit and terminates in another spherical structure, which is characterized by a papilla-like protrusion that most likely functioned as a discharge apparatus. There is a certain level of morphological correspondence between the fossil and certain zygomycetes of the order Mucorales in which zygospore germination entails the formation of a germ sporangium (e.g., Brefeld 1872; De Bary 1884; Blakeslee 1906; Gauger 1961: figs 1–3; Hocking 1967). During the sexual stage of the life cycle, these fungi produce zygosporangia (zspo in Fig. 2) containing single zygospores (zsp in Fig. 2) following gametangial fusion, and sometimes azygosporangia containing azygospores, which are similar to zygospores but form parthenogenetically without gametangial fusion (e.g., Benjamin and Mehrotra 1963; O'Donnell et al. 1977). The zygosporangium wall eventually ruptures and the zygospore germinates. If germination is of the germ-sporangial type (sensu Guo and Michailides 1998), then the zygospore produces a tube-like structure termed a sporangiophore or promycelium (sph in Fig. 2), on the tip of which a germ sporangium develops (spo in Fig. 2). After maturation of the spores (called germ sporangiospores), the germ sporangium ruptures and liberates the spores, which, if they fall on a suitable substrate, germinate and develop into a new mycelium (e.g., Cerdá-Olmedo 2001: fig. 1). It is conceivable that the *W. spinifera* specimen described above represents a zygosporangium or azygosporangium containing a zygospore or azygospore (i.e. the interior sphere), which has germinated and produced a sporangiophore (i.e. the stalk-like hypha) with a terminal germ sporangium (i.e. the exterior sphere). The second exterior

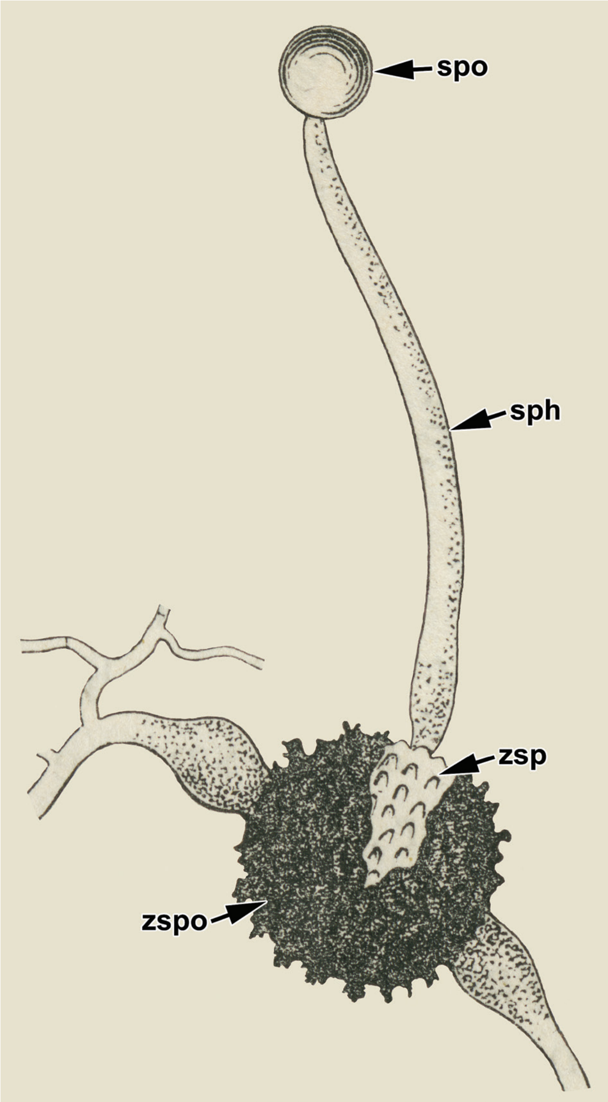

Figure 2. Germinated zygospore of *Mucor mucedo* (Mucorales); zspo = zygosporangium, zsp = zygospore, sph = sporangiophore or promycelium, spo = germ sporangium (modified from De Bary 1884: fig. 71C).

structure, which occurs partially embedded in the hyphal mantle of the reproductive unit, could be another germ sporangium. Germinated zygospores with two sporangiophores, each bearing a terminal sporangium, have also been reported in certain present-day zygomycetes (e.g., Michailides and Spotts 1988).

One might counter this comparison by pointing out that no evidence of gametangia and suspensors has been found in any of the *Windipila spinifera* specimens. It is possible, however, that these structures were small, inconspicuous, and entirely embedded in the mantle. For example, the zygosporangia of certain present-day *Mortierella* (Mortierellaceae, Mucorales) species are surrounded by extensive hyphal coverings arising from the suspensors or from hyphae at the base of the suspensors (Brefeld 1881: pl. V, figs 12, 14, 15; Kuhlman 1972; Ansell and Young 1983) that render the gametangia and suspensors very difficult to recognize. It can also be argued that the interior sphere in *W. spinifera* is too small to be a zygospore or azygospore because zygosporogenesis involves the development of a complex, multi-layered wall (i.e. the combined sporangium and spore wall) in which newly deposited wall layers follow exactly the shape of the previously deposited layers (e.g., Brefeld 1872: pl. II, fig. 20; Hawker and Gooday 1968; Hawker and Backett 1971; O'Donnell et al. 1978). Rather, the interior sphere is somehow reminiscent of the single oil globules that are visible in the zygospores of, for instance, certain representatives of the mucoralean genera *Radiomyces* (Radiomycetaceae; e.g., Embree 1959: figs 12, 13; Benny and Benjamin 1991: figs 8, 16, 18, 29h, i) and *Dichotomocladium* (Lichtheimiaceae; e.g., Benny and Benjamin 1993: fig. 4b). However, it seems unlikely that oil globules would become preserved in a recognizable form. It can also not be ruled out that the spore proper in *W. spinifera* has shrunken during germination, and again during the silicification process. One must furthermore take into account the possibility that entirely fossil lineages of zygomycete fungi, as well as fossil representatives of present-day lineages, have existed that were characterized by traits unknown in any present-day representative. For example, the germ sporangium in present-day Mucorales ruptures at maturity to release the spores, while the fossil spores likely were liberated through a distal discharge papilla.

An alternative hypothesis views the interior sphere in *Windipila spinifera* as a part or life cycle stage of some other fungus or fungus-like organism that had invaded and colonized the reproductive unit, and reproduced by producing a sporangium (or sporangia?) outside of the host. Two lines of evidence seem to support this hypothesis. First, co-occurring with the interior sphere in the cavity of the *W. spinifera* specimen are fragments of hyphae or filaments, which indicate that, at some point, another fungus has been present in this structure, unless the mantle hyphae had invaded the cavity from the circumference (see below). Second, several different types of small spherules occurring singly or in groups, and probably representing the reproductive units of intrusive fungi, have been reported in the cavity of *Zwergimyces vestitus* (Krings et al. 2016), and thus indicate that mantled fungal reproductive units in the Rhynie paleoecosystem have been invaded by other microorganisms and used as a habitat. On the other hand, there is no evidence of a physical connection between the interior sphere in *W. spinifera* and any system on which it could have developed. This argues against the hypothesis that the sphere belongs to an intrusive organism. It is also possible that the *W. spinifera* specimen had already fulfilled its function, namely to produce a spore that subsequently germinated, and, consequently, was in the process of natural decay at the time of fossilization. If this is the case, then the hyphae inside the cavity could belong to saprotrophic fungi or even be outgrowths from the mantle hyphae that invaded the senescing reproductive unit and participated in its degradation.

6. Conclusions

The fossil record rarely gives comprehensive insights into the life history of fungi, which is unfortunate because life history stages can provide valuable information to assess the systematic affinities of fossil fungi in the absence of molecular data (Taylor et al. 2015). The affinities of the organism that produced *Windipila spinifera* mantled reproductive units remain conjectural, in spite of the fact that the fossil detailed above shares certain features with germinated zygospores of the germ-sporangial type. I hope that specimens showing these features in greater detail become available as exploration of the Rhynie cherts continues, and that these fossils will clarify the nature of the interior sphere and reveal hitherto unknown aspects, which can then be used to determine the systematic position of *W. spinifera*.

Acknowledgments

Funding was received from the National Science Foundation (DEB-1441604 subcontract S1696A-A). I thank H. Martin and S. Sónyi (both Munich, Germany) for technical assistance, and Nora Dotzler (Munich, Germany) for insightful comments and suggestions on the manuscript.

References

Ansell PJ, Young TWK (1983) Light and electron microscopy of *Mortierella indohii* zygospores. Mycologia 75: 64–69. https://doi.org/10.1080/00275514.1983.12021638

Benjamin RK (1979) Zygomycetes and their spores. In: Kendrick B (Ed.) The Whole Fungus. II. The Sexual-Asexual Synthesis [Proceedings of the 2nd International Mycological Conference held at the Environmental Sciences Centre of the University of Calgary Kananaskis, Alberta, Canada]. CAN: National Museum of Natural Sciences, National Museums of Canada, and the Kananaskis Foundation, Ottawa, 573–621.

Benjamin RK, Mehrotra BS (1963) Obligate azygospore formation in two species of *Mucor* (Mucorales). Aliso 5: 235–245. https://doi.org/10.5642/aliso.19630503.02

Benny GL (2012) Current systematics of Zygomycota with a brief review of their biology. In: Misra JK, Tewari JP, Deshmukh SK (Eds) Systematics and Evolution of Fungi. CRC Press, Boca Raton, FL, 55–105.

Benny GL, Benjamin RK (1991) The Radiomycetaceae (Mucorales: Zygomycetes). III. A new species of *Radiomyces*, and cladistic analysis and taxonomy of the family; with a discussion of evolutionary ordinal relationships in Zygomycotina. Mycologia 83: 713–735. https://doi.org/10.1080/00275514.1991.12026077

Benny GL, Benjamin RK (1993) Observations on Thamnidiaceae (Mucorales). VI. Two new species of *Dichotomocladium* and the zygospores of *D. hesseltinei* (Chaetocladiaceae). Mycologia 85: 660–671. https://doi.org/10.1080/00275514.1993.12026318

Benny GL, Hamber RA, Morton JB (2001) Zygomycota: Zygomycetes. In: McLoughlin DJ, McLoughlin EG, Lemke PE (Eds) The Mycota. VIIA. Systematics and Evolution. Springer Verlag, Berlin, 113–146. https://doi.org/10.1007/978-3-662-10376-0_6

Blakeslee AF (1906) Zygospore germination in the Mucorineae. Annales Mycologici 4: 1–28.

Brefeld O (1872) Botanische Untersuchungen über Schimmelpilze. I. Heft: *Mucor Mucedo*, *Chaetocladium Jones'ii*, *Piptocephalis Freseniana*. Zygomyceten. Verlag von Arthur Felix, Leipzig, viii + 64 pp.

Brefeld O (1881) Botanische Untersuchungen über Schimmelpilze. [Untersuchungen aus dem Gesammtgebiete der Mykologie]. IV. Heft: 1. Culturmethoden zur Untersuchung der Pilze. 2. *Bacillus Subtilis*. 3. *Chaetocladium Fresenianum*. 4. *Pilobolus*. 5. *Mortierella Rostafinskii*. 6. *Entomophthora Radicans*. 7. *Peziza tuberosa* und *P. Sclerotiorum*. 8. *Pyenis sclerotivora*. 9. Weitere Untersuchungen von verschiedenen Ascomyceten. 10. Bemerkungen zur vergleichenden Morphologie der Ascomyceten. 11. Zur vergleichenden Morphologie der Pilze. Verlag von Arthur Felix, Leipzig, vi + 191 pp.

Cerdá-Olmedo E (2001) *Phycomyces* and the biology of light and color. FEMS Microbiology Reviews 25: 503–521. https://doi.org/10.1111/j.1574-6976.2001.tb00588.x

Channing A (2017) A review of active hot-spring analogues of Rhynie: environments, habitats and ecosystems. Philosophical Transactions of the Royal Society London, Series B 373: 20160489. https://doi.org/10.1098/rstb.2016.0490

De Bary A (1884) Vergleichende Morphologie und Biologie der Pilze, Mycetozoen und Bacterien. Verlag von W. Engelmann, Leipzig, xvi + 558 pp. https://doi.org/10.5962/bhl.title.42380

Edwards D, Dolan L, Kenrick P [Eds] (2018) The Rhynie cherts: our earliest terrestrial ecosystem revisited. Philosophical Transactions of the Royal Society of London B 373: 1–201. https://doi.org/10.1098/rstb.2016.0489

Embree RW (1959) *Radiomyces*, a new genus in the Mucorales. American Journal of Botany 46: 25–30. https://doi.org/10.1002/j.1537-2197.1959.tb06977.x

Fayers SR (2003) The biota and palaeoenvironements of the Windyfield Chert, Early Devonian, Rhynie, Scotland. Ph.D. thesis, University of Aberdeen, Aberdeen, Scotland, UK, 549 pp.

Fayers SR, Trewin NH (2004) A review of the palaeoenvironments and biota of the Windyfield chert. Transactions of the Royal Society of Edinburgh, Earth Sciences 94: 325–339. https://doi.org/10.1017/S0263593300000729

Garwood RJ, Oliver H, Spencer ART (2020) An introduction to the Rhynie chert. Geological Magazine 157: 47–64. https://doi.org/10.1017/S0016756819000670

Gauger WL (1961) The germination of zygospores of *Rhizopus stolonifer*. American Journal of Botany 48: 427–429. https://doi.org/10.1002/j.1537-2197.1961.tb11662.x

Glass NL, Fleissner A (2006) Re-wiring the network: understanding the mechanism and function of anastomosis in filamentous ascomycete fungi. In: Kües U, Fischer R (Eds) The Mycota, I. Growth, Differentiation and Sexuality. Springer Verlag, Berlin, Heidelberg, 123–139. https://doi.org/10.1007/3-540-28135-5_7

Gregory PH (1984) The fungal mycelium – an historical perspective. In: Jennings DH, Rayner ADM (Eds) The Ecology and Physiology of the Fungal Mycelium. Cambridge University Press, Cambridge, UK, 1–22.

Guo LY, Michailides TJ (1998) Factors affecting the rate and mode of germination of *Mucor piriformis* zygospores. Mycological Research 102: 815–819. https://doi.org/10.1017/S0953756297005650

Hass H, Rowe NP (1999) Thin sections and wafering. In: Jones TP, Rowe NP (Eds) Fossil Plants and Spores: Modern Techniques. The Geological Society, London, 76–81.

Hawker LE, Beckett A (1971) Fine structure and development of the zygospore in *Rhizopus sexualis* (Smith) Callen. Philosophical Transactions of the Royal Society of London, Series B, Biological Sciences 18: 71–100. https://doi.org/10.1098/rstb.1971.0110

Hawker LE, Gooday MA (1968) Development of the zygospore wall in *Rhizopus sexualis* (Smith) Callen. Journal of General Microbiology 54: 13–20. https://doi.org/10.1099/00221287-54-1-13

Hocking D (1967) Zygospore initiation, development and germination in *Phycomyces blakesleeanus*. Transactions of the British Mycological Society 50: 207–220. https://doi.org/10.1016/S0007-1536(67)80031-7

Ivarsson M, Peckmann J, Tehler A, Broman C, Bach W, Behrens K, Reitner J, Böttcher ME, Norbäck Ivarsson L (2015) Zygomycetes in vesicular basanites from Vesteris Seamount, Greenland Basin – a new type of cryptoendolithic fungi. PLoS ONE 10(7): e0133368. https://doi.org/10.1371/journal.pone.0133368

Krings M, Harper CJ (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. https://doi.org/10.1127/njgpa/2017/0677

Krings M, Harper CJ (2018) Additional observations on the fungal reproductive unit *Windipila spinifera* from the Windyfield chert, and description of a similar form, *Windipila pumila* nov. sp., from the nearby Rhynie chert (Lower Devonian, Scotland). Neues Jahrbuch für Geologie und Paläontologie, Abhandlungen 288: 235–242. https://doi.org/10.1127/njgpa/2018/0736

Krings M, Harper CJ (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. https://doi.org/10.1007/s12542-019-00507-5

Krings M, Taylor TN (2013) *Zwergimyces vestitus* (Kidston et W.H. Lang) nov. comb., a fungal reproductive unit enveloped in a hyphal mantle from the Lower Devonian Rhynie chert. Review of Palaeobotany and Palynology 190: 15–19. https://doi.org/10.1016/j.revpalbo.2012.11.008

Krings M, Taylor TN (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. https://doi.org/10.1127/0077-7749/2014/0423

Krings M, Taylor TN (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. https://doi.org/10.1144/sjg2014-026

Krings M, Taylor TN (2015b) Mantled fungal reproductive units in land plant tissue from the Lower Devonian Rhynie chert. Bulletin of Geosciences 90: 1–6. https://doi.org/10.3140/bull.geosci.1519

Krings M, Taylor TN, Dotzler N, Persichini G (2012) Fossil fungi with suggested affinities to the Endogonaceae from the Middle Triassic of Antarctica. Mycologia 104: 835–844. https://doi.org/10.3852/11-384

Krings M, Taylor TN, Dotzler N (2013) Fossil evidence of the zygomycetous fungi. Persoonia 30: 1–10. https://doi.org/10.3767/003158513X664819

Krings M, Taylor TN, Taylor EL, Kerp H, Dotzler N (2014) First record of a fungal “sporocarp” from the Lower Devonian Rhynie chert. Palaeobiodiversity and Palaeoenvironments 94: 221–227. https://doi.org/10.1007/s12549-013-0135-7

Krings M, Taylor TN, Dotzler N, Harper CJ (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. https://doi.org/10.1016/j.revpalbo.2016.01.005

Krings M, Harper CJ, Taylor EL (2017a) Fungi and fungal interactions in the Rhynie chert: A review of the evidence, with the description of *Perexiflasca tayloriana* gen. et sp. nov. Philosophical Transactions of the Royal Society of London B 373: 20160500. https://doi.org/10.1098/rstb.2016.0500

Krings M, Walker C, Harper CJ, Martin H, Sónyi S, Kustatscher E, Taylor TN (2017b) Unusual fungal reproductive units from the Lower Devonian Rhynie chert. Zitteliana 89: 29–37.

Kuhlman EG (1972) Variation in zygospore formation among species of *Mortierella*. Mycologia 64: 325–341. https://doi.org/10.1080/00275514.1972.12019267

Mark DF, Rice CM, Fallick AE, Trewin NH, Lee MR, Boyce A, Lee JKW (2011) $^{40}Ar/^{39}Ar$ dating of hydrothermal activity, biota and gold mineralization in the Rhynie hot-spring system, Aberdeenshire, Scotland. Geochimica et Cosmochimica Acta 75: 555–569. https://doi.org/10.1016/j.gca.2010.10.014

Mark DF, Rice CM, Trewin NH (2013) Discussion on ‘A high-precision U-Pb age constraint on the Rhynie Chert Konservat-Lagerstätte: time scale and other implications’ [Journal, Vol. 168, 863–872]. Journal of the Geological Society of London 170: 701–703. https://doi.org/10.1144/jgs2011-110

Michailides TJ, Spotts RA (1988) Germination of zygospores of *Mucor piriformis* (On the life history of *Mucor piriformis*). Mycologia 80: 837–844. https://doi.org/10.1080/00275514.1988.12025732

O’Donnell KL, Ellis JJ, Hesseltine CW, Hooper GR (1977) Azygosporogenesis in *Mucor azygosporus*. Canadian Journal of Botany 55: 2712–2720. https://doi.org/10.1139/b77-310

O’Donnell KL, Flegler SL, Hooper GR (1978) Zygosporangium and zygospore formation in *Phycomyces nitens*. Canadian Journal of Botany 56: 91–100. https://doi.org/10.1139/b78-009

Parry SF, Noble SR, Crowley QG, Wellman CH (2011) A high precision U-Pb age constraint on the Rhynie chert Konservat-Lagerstätte: time scale and other implications. Journal of the Geological Society of London 168: 863–872. https://doi.org/10.1144/0016-76492010-043

Rice CA, Ashcroft WA (2004) The geology of the northern half of the Rhynie Basin, Aberdeenshire, Scotland. Transactions of the Royay Society Edinburgh, Earth Sciences 94: 299–308. https://doi.org/10.1017/S0263593300000705

Rice CM, Trewin NH, Anderson LI (2002) Geological setting of the Early Devonian Rhynie cherts, Aberdeenshire, Scotland: An early terrestrial hot spring system. Journal of the Geological Society of London 159: 203–214. https://doi.org/10.1144/0016-764900-181

Strullu-Derrien C, Kenrick P, Knoll AH (2019) The Rhynie chert. Current Biology 29: R1218–R1223. https://doi.org/10.1016/j.cub.2019.10.030

Taylor TN, Krings M, Taylor EL (2015) Fossil Fungi, 1st edn. Elsevier/Academic Press Inc., Amsterdam, Boston, Heidelberg, London, xv + 382 pp. https://doi.org/10.1016/B978-0-12-387731-4.00001-3

Trewin NH, Fayers SR (2016) Macro to micro aspects of the plant preservation in the Early Devonian Rhynie cherts, Aberdeenshire, Scotland. Earth and Environmental Sciences Transactions of the Royal Society of Edinburgh 106: 67–80. https://doi.org/10.1017/S1755691016000025

Trewin NH, Kerp H (2017) The Rhynie and Windyfield cherts, Early Devonian, Rhynie, Scotland. In: Fraser NC, Sues HD (Eds) Terrestrial Conservation Lagerstätten. Windows into the Evolution of Life on Land. Dunedin Academic Press, Edinburgh, UK, 1–38.

Trewin NH, Rice CM (1992) Stratigraphy and sedimentology of the Devonian Rhynie chert locality. Scottish Journal of Geology 28: 37–47. https://doi.org/10.1144/sjg28010037

Walker C, Harper CJ, Brundrett MC, Krings M (2018) Looking for arbuscular mycorrhizal fungi (AMF) in the fossil record: An illustrated guide. In: Krings M, Harper CJ, Cúneo NR, Rothwell GW (Eds) Transformative Paleobotany: Papers to Commemorate the Life and Legacy of Thomas N. Taylor. Elsevier/Academic Press Inc., London, San Diego, CA, Cambridge, MA, Oxford, 481–517. https://doi.org/10.1016/B978-0-12-813012-4.00020-6

Walker C, Harper CJ, 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: 104384. https://doi.org/10.1016/j.revpalbo.2021.104384

Wellman CH (2006) Spore assemblages from the Lower Devonian 'Lower Old Red Sandstone' deposits of the Rhynie outlier, Scotland. Transactions of the Royal Society of Edinburgh, Earth Sciences 97: 167–211. https://doi.org/10.1017/S0263593300001449

Wellman CH (2017) Palaeoecology and palaeophytogeography of the Rhynie chert plants: further evidence from integrated analysis of *in situ* and dispersed spores. Philosophical Transactions of the Royal Society of London B 373: 20160491. https://doi.org/10.1098/rstb.2016.0491

Wellman CH, Kerp H, Hass H (2006) Spores of the Rhynie chert plant *Aglaophyton* (*Rhynia*) *major* (Kidston and Lang) D.S. Edwards, 1986. Review of Palaeobotany and Palynology 142: 229–250. https://doi.org/10.1016/j.revpalbo.2006.04.009