

ACAULOSPORA EXCAVATA SP. NOV. - AN ENDOMYCORRHIZAL FUNGUS FROM COTE D'IVOIRE

K. INGLEBY¹ C. WALKER² & P.A. MASON¹

¹*Institute of Terrestrial Ecology, Bush Estate,
Penicuik, Midlothian, Scotland, U.K. EH26 0QB*

²*Forestry Commission, The Forestry Authority,
Northern Research Station, Roslin, Midlothian,
Scotland, U.K. EH25 9SY*

SUMMARY

A newly discovered endomycorrhizal fungus, *Acaulospora excavata*, is described and illustrated.

INTRODUCTION

As part of a study of spore populations of arbuscular mycorrhizal fungi in Côte d'Ivoire (Wilson *et al.* 1992), soil samples were collected from beneath *Terminalia superba* Engl. & Diels and *T. ivorensis* A. Chev. in the Mopri Forest Reserve, in October 1990. Among the spore types extracted was a previously undescribed member of the genus *Acaulospora* Gerdemann and Trappe. This fungus is described herein as *Acaulospora excavata* sp. nov.

MATERIALS AND METHODS

Spores were extracted from soil by sucrose centrifugation (Walker *et al.* 1982) and suspended in water for initial examination under a dissecting microscope (x 6-50). Illumination was by incident light from a quartz-iodine fibre-optic source with a colour temperature of 3200°K. Spore colour was described from freshly extracted spores suspended in water in plastic Petri dishes and illuminated by the same light source used for examining the spores. Colour was determined by comparison with a standard colour chart (Anon. 1969), the numbers following the colour descriptions refer to those given in the colour chart. Colour matching of structures viewed with transmitted light under a compound microscope was more difficult than with a dissecting microscope, so for such observations colours (for example, of individual walls in the description) were not precisely matched to a chart, and only generalised colour descriptions are used (Walker *et al.* 1993).

Specimens were mounted in polyvinyl alcohol lacto-glycerol (PVLG) (Koske & Tessier 1983) or distilled water (Spain 1990) with or without Melzer's reagent (Morton 1986a) for further study under a compound microscope (x 150-1500) with brightfield and Nomarski differential interference contrast illumination.

Wall structures were described using the standardized terminology and micrographs of Walker (1983) and Morton (1986b). So far, attempts to establish this species in pot culture have failed.

ACAULOSPORA EXCAVATA Ingleby and Walker sp. nov. (Figs 1 & 2)

Sporae singillatim in terra enatae, lateraliter gestae in sacculo sporifero, pallide ochraceae, ochraceae vel aurantiae, globosae vel subglobosae, 115-200 x 100-165 μm . Sporarum tunicae 3 in turmis tribus: Turma A cum tunica singula, colorata, 8-11 μm crassa, foveis interdum leviter angularibus, 4-20 x 4-16 μm in diam et 2-6 μm profundis; turma B cum tunica singula, hyalina, membranacea, 0.5-1.0 μm crassa; turma C cum tunica singula, amorpha, baccata, 1-2.5 μm crassa, in solutione Melzeri carnea vel purpurea.

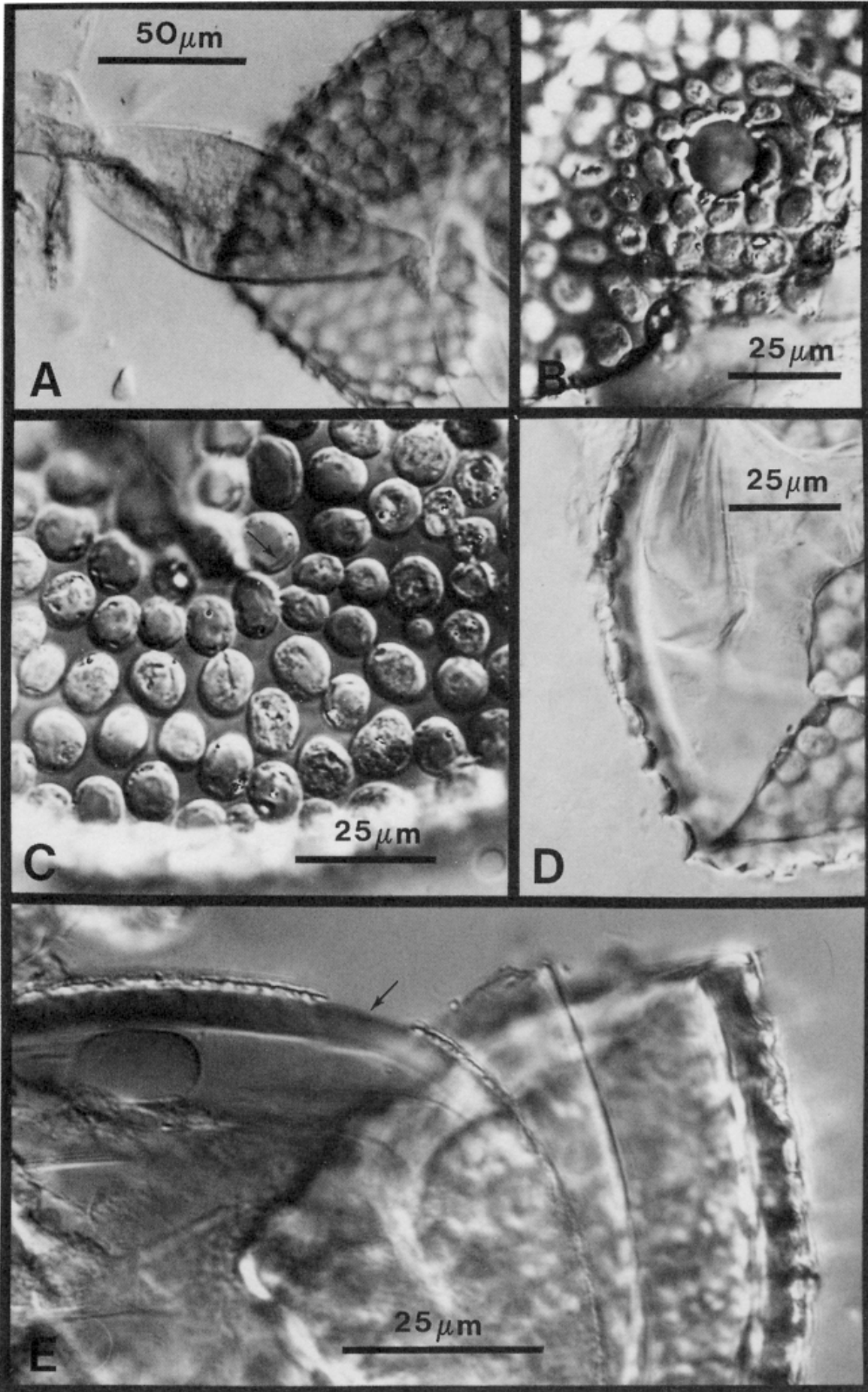
SPORES borne singly in the soil laterally on the neck of a hyaline sporiferous saccule that collapses after the spores mature; spores pale ochraceous (6) to ochre (9) to orange (48); globose to subglobose, 115-200 x 100-165 μm .

SACCULE neck at point of attachment to the spore 9-19 μm in diameter with walls 1-2 μm thick (Figs. 1A, 1B). Dimensions of saccule not determined.

WALL GROUP A of a single, pale yellow, laminated wall, 8-11 μm thick, not reacting to Melzer's reagent. Outer surface ornamented by circular to subcircular to elliptical pits, 4-20 x 4-16 μm in diameter and 2-6 μm in depth sometimes with a slightly angular outline (Figs. 1C, 1D). These pits appear flat-bottomed with the basal area 3-15 x 3-12 μm in diameter (Fig. 1C, arrowed). Pits adjacent or up to 9 μm apart.

Fig. 1. Light photomicrographs of spores of *Aucaulospora excavata*.

- A. Saccule neck still attached to spore.
- B. Saccule scar. Note the smaller pits immediately surrounding the scar.
- C. Surface view of ornamented outer wall showing pit size, shape and distribution. These pits are flat-bottomed and in some cases the basal perimeter is in focus (arrowed).
- D. A fractured spore showing the pits in cross-section.
- E. A fractured spore showing all three walls. Note the beaded inner wall 3 which has expanded in PVLG and the area where the beaded deposition is absent (arrowed).



SPORE WALL STRUCTURE of three walls (1-3) in three wall groups (A-C) (see Murograph Fig. 2).

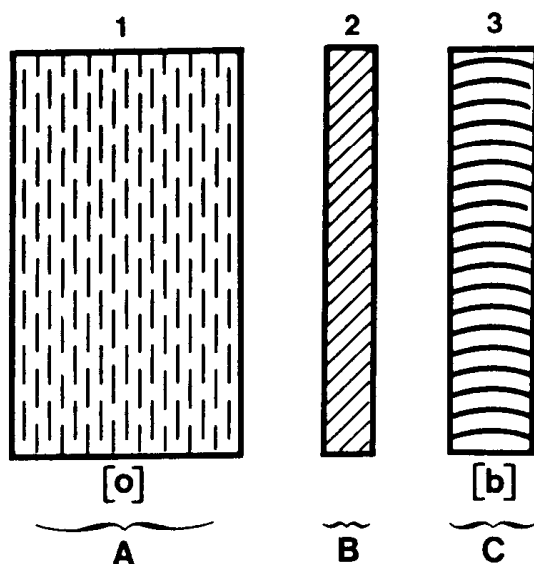


Fig. 2 Murograph (after Walker 1983) of *Acaulospora excavata*.

WALL GROUP B of a single, hyaline, membranous wall, 0.5-1.0 μm thick, not reacting to Melzer's reagent (Fig. 1E).

WALL GROUP C of a single 'beaded', amorphous wall, 1-2 μm thick (including beaded ornamentation on outer surface), which reacts strongly in Melzer's reagent turning pink or purple within minutes of crushing the spores. This wall is plastic in PVLG and, when crushed, may become up to 9 μm thick (Fig. 1E).

ETYMOLOGY: Latin *excavata* - referring to the appearance of the pits on the outer wall.

DISTRIBUTION AND HABITAT: Known only from a single soil sample collected from the Mopri Forest Reserve. The sampling area was one of natural forest which had been manually cleared and burnt in 1989 before replanting with *T. ivorensis*. This disturbance had resulted in marked changes in soil chemistry with increases in pH (from 6.5 to 7.5), organic matter and available nutrients; at the sample point where *A. excavata* was found the pH was particularly high (8.2). Changes in vegetation also occurred after the clearance. In addition to planting with *T. ivorensis* the plot was rapidly colonised by a wide range of herbaceous species, most notably *Chromolaena odorata* (L.) King & Robinson.

MYCORRHIZAL ASSOCIATIONS: Not known. Baiting out the species was attempted by both single-spore and multi-spore isolation and open-pot culture (Gilmore 1968) using *Plantago lanceolata* L. and *Vigna unguiculata* (L.) Walp. as host plants. None of these attempts succeeded.

HOLOTYPE: Cote d'Ivoire: South West of Tissage - Mopri Forest Reserve under *Terminalia ivorensis*, E; isotype OSC (Walker 1674. 30 X 90).

DISCUSSION

Spores of *A. excavata* possess a distinctly ornamented outer wall surface of densely crowded, flat-bottomed pits. Six other species of *Acaulospora* have been described so far which possess a distinctly pitted outer wall, namely *Acaulospora foveata* Trappe and Janos (Janos & Trappe 1982), *A. cavernata* Blaszkowski (Blaszkowski 1989), *A. lacunosa* Morton (Morton 1986b), *A. paulinae* Blaszkowski (Blaszkowski 1988), *A. undulata* Sieverding (Sieverding 1988) and *A. scrobiculata* Trappe (Trappe 1977).

Spores of *A. foveata* can be distinguished from *A. excavata* by their generally larger size (185-310 x 215-350 μm) and colour (yellow/brown to red/brown to black/brown). Spores of *A. paulinae* are smaller (60-95 μm) and those of *A. scrobiculata* are a paler colour (hyaline to olive to brown) while those of *A. undulata* are both smaller (55-85 μm) and are a paler colour (hyaline) than those of *A. excavata*. We have not been able to examine spores of *A. cavernata*, but from their description, they are similar in size and colour to those of *A. excavata*.

From their descriptions, *A. cavernata*, *A. lacunosa*, *A. paulinae* and *A. scrobiculata* have smaller pits than *A. excavata* (always < 6 μm diam compared with 4-20 x 4-16 μm) and thinner outer walls (< 6 μm thick compared with 8-11 μm). Pits of *A. lacunosa*, *A. paulinae* and *A. scrobiculata* are also more widely spaced and irregularly shaped. Additionally spores of *A. cavernata* are described as having a second wall in wall group A and a coriaceous wall in wall group C while those of *A. lacunosa* have a second wall in wall group B. Spores of *A. undulata* possess an even thinner ornamented wall (1-1.5 μm thick) consisting of depressions separated by ridges 1 μm wide. They are also described as having an outer evanescent wall, though examination of specimens from the type culture does not show this phenomenon. Although *A. excavata* and *A. foveata* have similar sized pits, the smaller spore size and more densely crowded pits of *A. excavata* combine to give the surface of the outer wall a completely different appearance to that of *A. foveata*.

Wall group C of *A. excavata* is described as a single beaded wall, though it can appear as two walls in PVLG mounts. The depositions on the outer surface of this amorphous wall clearly do not constitute a distinct wall, though

in places they are so crowded as to seemingly form a separate layer. In other places, however, the beads may be absent, possibly due to detachment during crushing (Figure 1E, arrowed).

ACKNOWLEDGEMENTS

This project was funded by the UK Overseas Development Administration and the Commonwealth Development Corporation, and was contracted to ITE by the International Forest Science Consultancy. Work using imported soil was carried out under DAFS licence number IP/MISC/28/90 issued under the Plant Health (Great Britain) order 1987. We wish to thank Dr J.M. Trappe, Oregon State University, Department of Forest Science for preparing the latin diagnosis and for his helpful review of the manuscript.

LITERATURE CITED

- ANON, 1969. *Flora of British fungi colour, identification chart*. Royal Botanic Garden, Edinburgh, HMSO.
- BLASZKOWSKI, J. 1988. Three new vesicular-arbuscular mycorrhizal fungi (*Endogonaceae*) from Poland. *Bull. Pol. Acad. Sci: Biol. Sci.* **36**: 271-275.
- BLASZKOWSKI, J. 1989. *Acaulospora cavernata* (Endogonales) - a new species from Poland with pitted spores. *Crypt. Bot.* **1**: 204-207
- GILMORE, A.E. 1968. Phycomycetous mycorrhizal organisms collected by open-pot culture methods. *Hilgardia* **39**: 87-105.
- JANOS, D.P. & TRAPPE, J.M. 1982. New *Acaulospora* species from America. *Mycotaxon* **15**: 515-522.
- JENKINS, W.R. 1964. A rapid centrifugal-flotation technique for separating nematodes from soil. *Plant Disease Report* **48**: 692.
- KOSKE, R.E. & TESSIER, B. 1983. A convenient permanent slide mounting medium. *Mycological Society of America's Newsletter*, **34**(2): 59.
- MORTON, J.B. 1986A. Effects of mountants and fixatives on wall structures and Melzer's reaction in spores of two *Acaulospora* species (*Endogonaceae*). *Mycologia* **78**: 787- 794.
- MORTON, J.B. 1986B. Three new species of *Acaulospora* (*Endogonaceae*) from high aluminium, low pH soils in West Virginia. *Mycologia* **78**: 641-648.
- SIEVERDING, E. 1988. Two new species of vesicular arbuscular mycorrhizal fungi in the *Endogonaceae* from tropical highlands of Africa. *Agnew. Botanik* **62**: 373-380.
- SPAIN, J.L. 1990. Arguments for diagnoses based on unaltered wall structures. *Mycotaxon* **38**: 71-76.
- TRAPPE, J.M. 1977. Three new *Endogonaceae*: *Glomus constrictus*, *Sclerocystis clavispora* and *Acaulospora scrobiculata*. *Mycotaxon* **6**: 359-366.
- WALKER, C. 1983. Taxonomic concepts in the *Endogonaceae*: spore wall characteristics in species descriptions. *Mycotaxon* **18**: 443-455.

- WALKER, C., MIZE, C.W. & McNABB, H.S. 1982. Populations of endogonaceous fungi at two locations in central Iowa. *Canadian Journal of Botany* **60**: 2518-2529.
- WALKER, C., GIANINAZZI-PEARSON, V. & MARION-ESPINASSE, H. 1993. *Scutellospora castanea*, a newly described arbuscular mycorrhizal species. Submitted to *Cryptogamie*.
- WILSON, J., INGLEBY, K., MASON, P.A., IBRAHIM, K.D. & LAWSON, G.J. 1992. Long-term changes in VA mycorrhizal spore populations in *Terminalia* plantations in Côte d'Ivoire. In: *Mycorrhizas in Ecosystems*. D.J. Read, D.H. Lewis, A.H. Fitter and I.J. Alexander (Eds.) C.A.B. International, Wallingford, U.K.