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THE DEVELOPMENT AND LIBERATION OF THE CONIDIA OF *XYLOSPHAERA FURCATA*

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(With Plate 17 and 4 Text-figures)

The stroma of *Xylosphaera furcata*, found growing in the nests of the termite *Macrotermes natalensis*, bears conidiophores each with two types of conidia that are violently discharged from the conidiophore to a distance of 0·5 mm. by means of a sudden rounding-off of the wall between the conidiophore and the conidium.

During an investigation in West Africa of fungi associated with termite nests (Dixon, 1959), *Xylosphaera furcata* (Fr.) Dennis was commonly found. The young stromata produced a conidial phase with conidia of two distinct shapes and these were apparently violently discharged.

MATERIALS AND METHODS

Xylosphaera furcata grows in the nests of termites of the group Macrotermitinae. These termites build an extensive series of sponge-like brood chambers from masticated vegetable fragments and the fungus grows vegetatively in the material of the 'sponge'. When the brood chamber is deserted the fungus produces long thin stromata which penetrate the soil, and the fertile regions of the fruit body develop either above ground level if the nest is hypogeal or on the surface of the nest if it is epigeal.

The growth of the stroma was observed by collecting 'sponge' from the epigeal nests of *Macrotermes natalensis* Havilland in the Botanic Garden, University of Ghana, Accra. The nests were opened to expose the central sponge area, portions of which were broken off, the young termites shaken out of the interstices and the 'sponge' broken into approximately 5 cm. cubes. These were placed either directly on a layer of moist filter-paper in glass troughs fitted with glass lids and kept in the light at 30° C.; or in polythene bags and brought to England where they were similarly treated. The time interval between collection in Ghana and setting up in London varied from 24 to 48 hr.

Large numbers of stromata developed from the surface of the 'sponge' and reached maturity after 16–20 days. During development the stroma bears a dense terminal cluster of conidia. The conidia germinated readily on yeast extract cellulose agar (Ingold & Dring, 1957) giving a slow-growing colony which produced normal stromata. Observations of growth and development were made on both sources of stromatal material. The development and liberation of the conidia was observed by transferring a small portion of 'sponge' bearing a single conidial stroma 4–5 mm. long to a Perspex micromanipulation chamber (Dixon, 1958). In this the conidiophores could be micromanipulated when necessary by means of a fine glass needle introduced from the side. By laying the stroma in a horizontal position the horizontal distance of spore discharge could be measured.

Cytological preparations were made from fresh unfixed material. A small stroma 3–4 mm. long was placed in a drop of freshly prepared 2 % orcein (Gurr's synthetic) in 60 % propionic acid, sealed with a no. 0 coverglass and squashed by gently heating and tapping until the constituent hyphae had spread out into a single flattened layer. The nuclei were clearly stained and differentiated from the cytoplasm and the preparation remained usable for 3–4 days.

THE CONIDIAL PHASE

The stroma arises from the mycelium submerged in the substratum. The initial consists of a fascicle of parallel hyphae forming a small vertical white strand 0·5 mm. diam. Growth is apical and the apex is loosely organized, in contrast to the hyphae behind the apex. The conidia are produced by the peripheral hyphae of the apex and as the central core continues to grow the conidiophores become displaced laterally, forming a zone of conidial development immediately behind the tip.

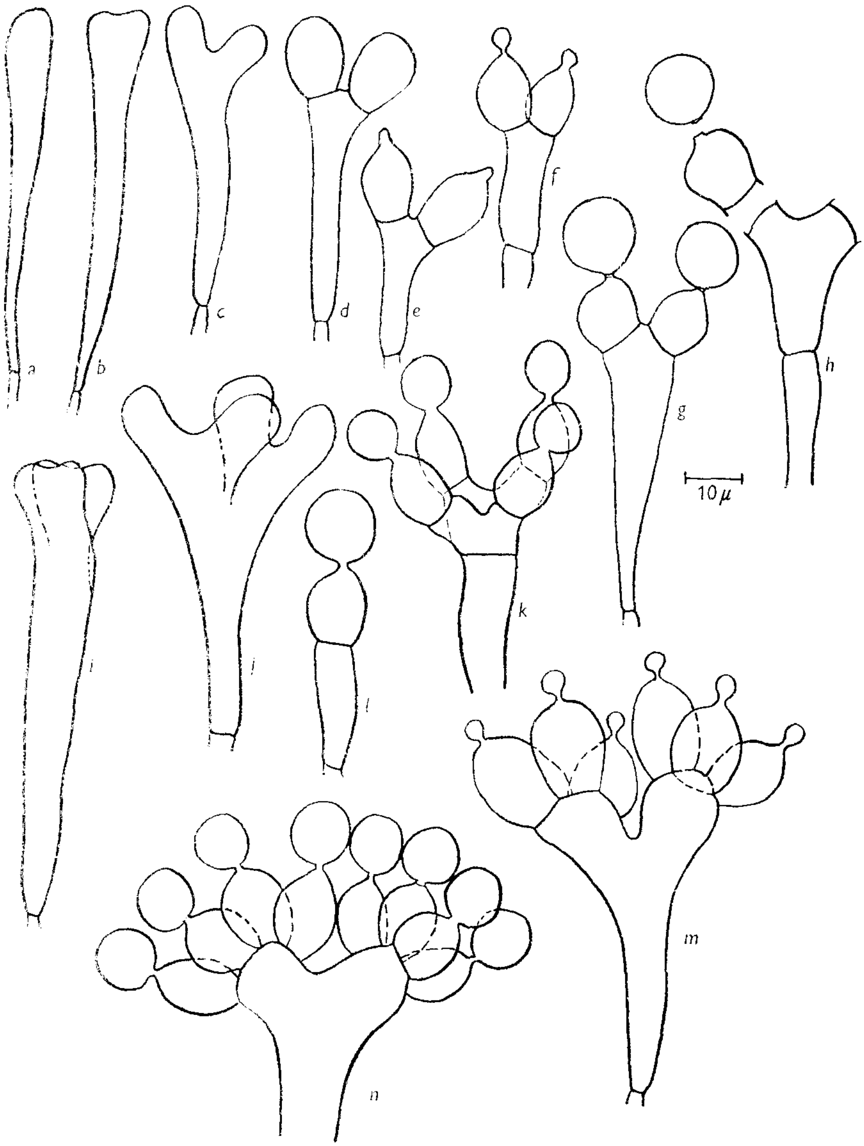
Conidiophore initials develop from a terminal cell 30–70 μ long (Text-fig. 1 *a*). This cell branches dichotomously to give two short divergent arms (Text-fig. 1 *b*, *c*). Cross-walls laid down at the base of the branches of the dichotomy separate distal subspherical cells from the proximal conidiophore (Text-fig. 1 *d*). On each of the distal cells an apical papilla is formed (Text-fig. 1 *e*) and from this a terminal spherical cell is budded off (Text-fig. 1 *f*, *g*). At maturity both terminal and subterminal cells separate from each other and from the conidiophore to form conidia of two distinct shapes, a spherical conidium 10–12 μ diam., with a small basal collar, and an apiculate conidium 10–15 μ long with a large basal ring 6–8 μ across (Pl. 17; Text-fig. 1 *h*).

The first dichotomy may branch again, giving a head of four branches (Text-fig. 1 *i*, *j*). These are cut off by cross-walls and eventually form a head bearing four terminal spherical conidia (Text-fig. 1 *k*). The majority of conidial heads conform to one or other developmental pattern but variations are found.

The conidiophore may fail to branch and produce only a single spherical conidium (Text-fig. 1 *l*). Branching in excess of the double dichotomy may occur giving conidial heads bearing 12 or 16 conidia (Text-fig. 1 *m*, *n*).

Cytology of conidial development

The terminal cell of the conidiophore contains 50–70 nuclei concentrated near the apex (Text-fig. 2 *a*). With the formation of the dichotomy the nuclei are distributed in approximately equal numbers in each arm (Text-fig. 2 *b*), and the cross-wall isolates 20–40 nuclei from the conidiophore (Text-fig. 2*c*). The papilla from which the spherical conidium is budded forms an isthmus (Text-fig. 2*d*) and through this narrow passage 10–20 nuclei squeeze (Text-fig. 2*e*, *f*). Before the formation of a cross-wall

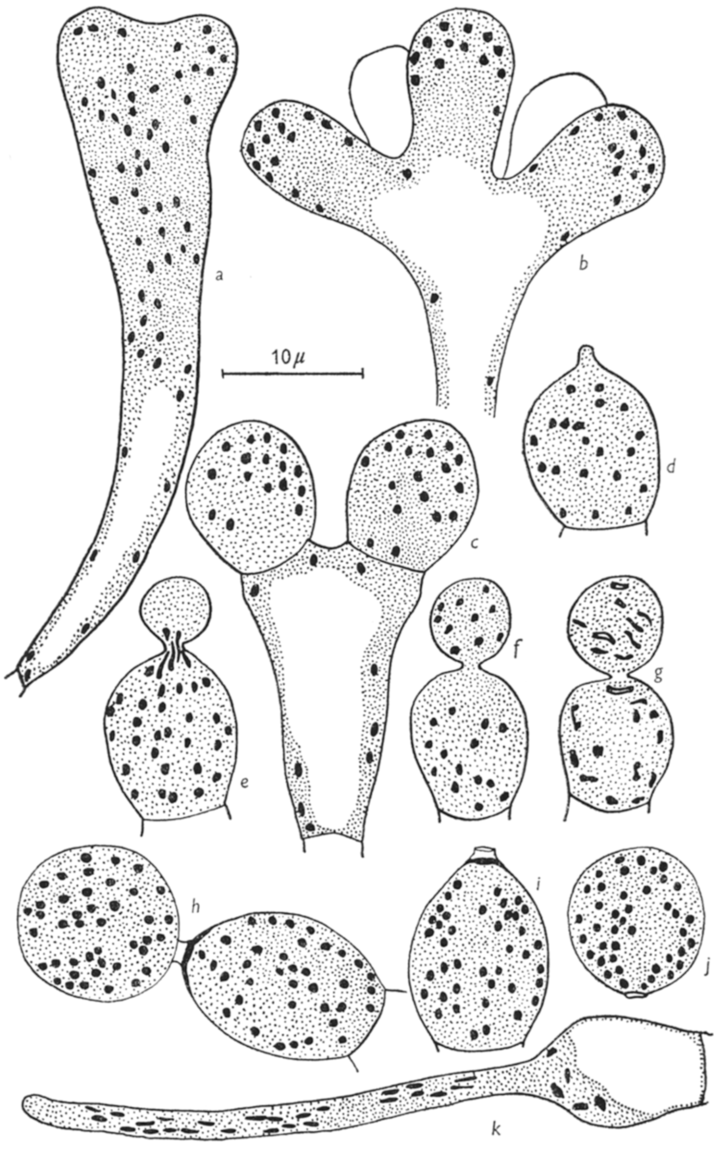


Text-fig. 1. Development of conidiophore and conidia.

the nuclei in the two maturing conidia divide (Text-fig. 1*g*) and at maturity 40–60 nuclei are present (Text-fig. 2*h*). This number remains constant until germination (Text-fig. 2*k*).

THE MECHANISM OF SPORE LIBERATION

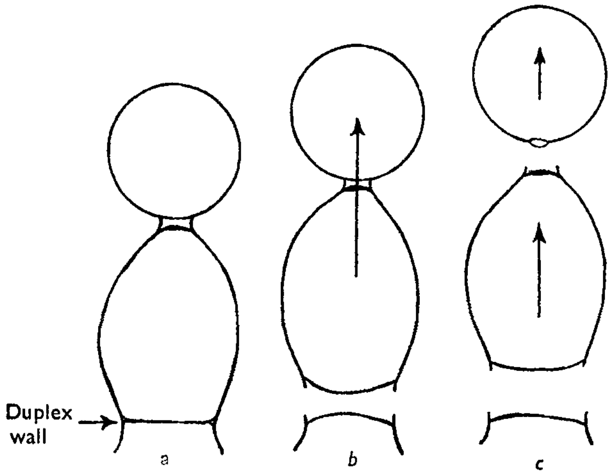
The subterminal conidium is separated from the conidiophore by a cross-wall which bulges slightly into the terminal cell of the conidiophore. This cross-wall is duplex, one layer belonging to the spore wall and the other to the apex of the conidiophore (Text-fig. 3*a*). At the moment of release rupture occurs on a circular line where the spore joins the conidio-



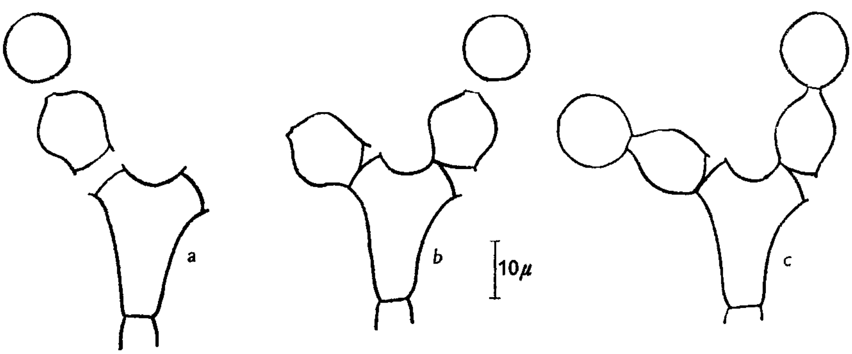
Text-figs. 2. Cytology of conidial development.

phore, and the incurved wall becomes rapidly convex, shooting off the two conidia (Text-fig. 3*b*). The sharp movement is sufficient to break the collar between the two conidia and they are dispersed separately (Text-fig. 3*c*).

The horizontal distance of discharge is small. The maximum distance measured from a sample of ten stroma was 0·6 mm. though the distance for the majority was 0·4–0·5 mm.



Text-fig. 3. Mechanism of conidial release.



Text-fig. 4. Complete, partial and unsuccessful examples of conidial release.

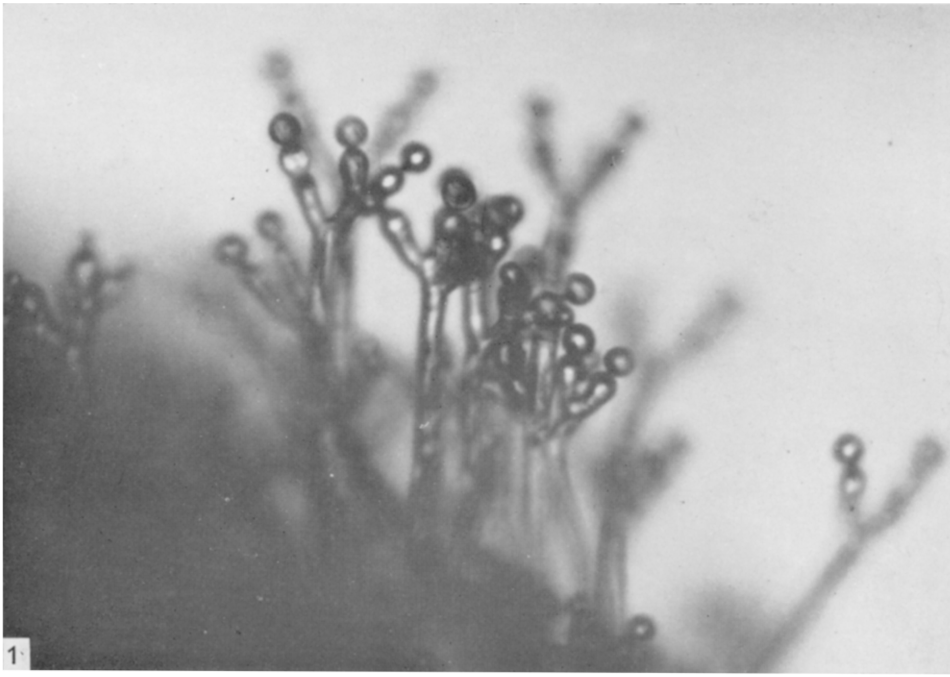
The collar joining the two conidia is very delicate. It was possible to remove the spherical conidium from its position with a micro needle without either dislodging the apiculate broadly attached conidium or triggering the discharge mechanism.

A spore deposit surrounding a horizontally placed stroma is composed of a mixture of the two conidia as single units. From the observations made it appeared that if the two conidia were discharged separation always occurred (Text-fig. 4*a*). Partial or total failures of the discharge mechanism were found. Sometimes the release of the apical conidium had been effected but the circular rupture was incomplete and the subapical conidium remained partially attached (Text-fig. 4*b*), or else the rupture mechanism had not taken place with sufficient force to liberate either of the conidia, but merely displaced the pair to one side (Text-fig. 4*c*).

DISCUSSION

The conidial state of *X. furcata* differs from that described for other members of the genus. *X. polymorpha* (Pers. ex Mérat) Dumort. (Brefeld, 1891) has a branched septate conidiophore bearing an apical cluster of ovoid radula spores and Tulasne's (1863) illustrations of *X. pedunculata* Dicks., *X. carpophila* (Pers.) Dumort. and *X. hyphoxylon* (L.) Dumort. show a similar type. Dennis (1962) placed *X. furcata* in the subgenus *Pseudoxylaria*, basing this separation on the minute ascospore and the absence of a germ slit in the ascospore wall. The distinctive conidial phase of *X. furcata* suggests the possibility that this may be another characteristic feature of the subgenus. An investigation of the conidia of *X. nigripes*, placed in the same subgenus, would be of interest.

The rounding-off mechanism of conidial release in *X. furcata* is paralleled by the mechanism of conidial discharge in *Sclerospora philippinensis* West. (Weston, 1923) and basidiospore discharge in *Gymnosporangium nidus-avis* Thaxt. (Prince, 1943). In all three, the conidia are released by the separation of two walls in plane contact, each of which rounds off to a small extent; and the distance of discharge is small, ranging from 0·5 mm. in *X. furcata* to 1–2 mm. in *S. philippinensis*. This mechanism is distinct from the rounding-off in *C. villosus* Martin (*Entomophthora coronata* (Cost.) Kev.); the apex of the conidiophore projects into a deep basal invagination of the terminal conidium and at the moment of discharge the invagination abruptly everts throwing the conidium off to a distance of up to 4 cm. (Ingold, 1939). The dual nature of the reproductive unit, involving a pick-a-back arrangement of two conidia, appears to be a unique feature. The subapical conidium can be regarded as a motor spore and the passive apical conidium forms a passenger spore. A similar mechanism occurs in conidial release in *Basidiobolus ranarum* Eidam (Ingold, 1953) where the conidiophore bursts along a transverse line of weakness and the apical part, above the line of rupture, contracts. This contraction forces the conidiphore sap backwards and the apical portion bearing the terminal conidium is rocketed forwards. The conidiophore apex acts as the motor and the conidium represents the passenger. In this case, however, only one component acts as a unit of reproduction, whereas in *X. furcata* both components can act as reproductive units.



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EXPLANATION OF PLATE 17

Figs. 1, 2. *X. furcata*. Apex of a 4-day-old stroma with clusters of conidiophores and conidia.

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