*Journal of General Microbiology* (1972), **73**, 45–54
*Printed in Great Britain*



# Conidiophore and Spore Development in *Aspergillus nidulans*

By P. T. P. OLIVER
*Institute of Genetics, University of Glasgow, Glasgow, G11 5JS*

(*Received* 10 *February* 1972; *revised* 31 *May* 1972)

SUMMARY

Conidiophore development was studied in wild-type and structurally defective mutants of *Aspergillus nidulans*. Cytoplasmic vesicles were found concentrated at growing apices. Plasmalemmasomes occurred frequently in the subapical and basal cytoplasm of growing sterigmata and near their developing septa; after septum formation they were flattened against the septa and the wall of the older cell and were associated with lomasomes. At this stage the wall developed a superficial lamina; the basipetally budding spore chain was invested from its initiation by a continuation of this lamina. The lamina was also associated with a dense network in the medium around the basal mycelium. The inner, secondary wall of the conidiophore became more electron-lucent with age; the continuity between this wall and the single wall of the sterigmata suggested this structure to be the site of action of the *bristle* locus. Pigmentation of wild-type and mutant conidiophores was associated with an electron-dense modification of the outer, primary wall. Stratification of the spore wall was indicated only by the similar development of a layer of electron-dense material. Cytoplasmic microfilaments were observed in all parts of the colony.

## INTRODUCTION

A survey of mutants of *Aspergillus nidulans* affected in conidiation (asexual sporulation) showed that very few genetic loci controlled defined stages in the development of the conidiospore-bearing appartus (Clutterbuck, 1969*a*). It is therefore of interest to investigate further the development of structures affected by these loci.

The dimensions of the conidial apparatus and the sequence of development have been described by Clutterbuck (1969*b*). Structural studies on conidiation in *Aspergillus giganteus* (Trinci, Peat & Banbury, 1968) and in Penicillium species (Fletcher, 1971) have been reported; this report considers particularly the distribution of cytoplasmic organelles and the maturation of extracellular structures.

## METHODS

All strains were derivatives of the Glasgow stock strain *biA1* and included: *biA1*, with wild-type colour and morphology; *bristle* mutant *brlA1*, which forms colourless sterile conidiophore initials of indefinite length; *brlA9*, which forms similar 'bristles' which are lightly pigmented and occasionally branched; *ivory* mutant *ivoA1*, which forms colourless conidiophores of normal morphology; *white-spore* mutant *wA9*; *dark-spore* mutant *drkA1*. The mutants and growth media were described by Clutterbuck (1969*a*); gene symbols are as given in Clutterbuck & Cove (1972).

Colonies were grown on agar plates at 37 °C. In colonies growing normally the radial succession of developmental stages was not shown precisely by the conidiophores, though zones were distinguished containing predominantly the earlier and the later 4 h of their development. To induce synchrony for study of a particular stage mycelium was grown

from a uniformly spread inoculum under cellophane for 24 h. The cellophane was removed and the incubation continued; conidiation then proceeded synchronously to within approximately 1 h of development within a wide area of the colony (A. J. Clutterbuck, unpublished). Samples were collected with minimum disturbance at $\frac{1}{2}$ h intervals, fixed and then examined to select the one nearest the stage required.

For fixation of radially growing or synchronized colonies a chosen square of agar about $20 \times 20 \times 3$ mm was immersed in glutaraldehyde (2·5%, w/v, in 0·05 M-phosphate buffer, pH 6·8; or in liquid minimal medium, at 37 °C) under a water vacuum pump and shaken until wetted; it was left to stand in a change of the glutaraldehyde solution for an hour, washed (in the buffer or distilled water, at room temperature) and osmicated (1% $OsO_4$ in the same medium as the wash, 3 h). Before or during dehydration the desired region was selected and the agar cut down to near $5 \times 2 \times 1$ mm for dehydration in ethanol and flat embedding in Araldite. Silver ultrathin sections were examined, unstained or stained with Reynolds' lead citrate, in a Siemens Elmiskop IA. One-micron sections of the same block were examined under phase contrast and correlated with studies of the living and fixed colony.

## RESULTS

*Wall development.* The conidiophores had a two-layered wall (Fig. 1 to 3); narrow, single-walled filaments occurred at the frequency predicted for sterile aerial hyphae. At the foot of the conidiophore the outer layer (primary wall) was continuous with the wall of the basal mycelium (Fig. 1, 2*c*); the inner layer (secondary wall) lined the side and ends of the foot, except in the longest feet where it might taper away irregularly. In the *bristle* mutants the 'bristle' walls were always two-layered, supporting their identification (Clutterbuck, 1969*a*) as undeveloped conidiophore initials. At stalk apices in both wild-type and mutant strains the two layers could not be distinguished, though the wall attained a greater thickness than the primary wall well in advance of the differentiation between the two layers 15 to 30 $\mu$m behind the apex (Fig. 1*b*).

The secondary wall was markedly more electron-lucent than the primary wall except near the apex. On a dark print of a transversely (not obliquely) sectioned wall the primary wall, like any hyphal wall, had a 'grey' electron-density similar to that of the embedding resin as had the newly formed secondary wall, while the secondary wall of a mature stalk or 'bristle' appeared white. The 'grey' density persisted at the dome of vesicles which carried phialides, showing an abrupt transition to the 'white' modification of the wall which tapered away inwards over the lower half of the vesicle (Fig. 1*d*). The metulae emerged through interruptions in the primary wall of the vesicle; the continuity of the conidiophore secondary wall with the single wall of the sterigmata (both metulae and phialides) was similar to that shown by Trinci *et al.* (1968), while the spore (conidium) wall arose as a new layer lining the open collarette at the phialide apex (cf. Trinci *et al.* 1968; Fletcher, 1971).

The superficial dense lamina, 0·02 $\mu$m thick, which invests the spore chains was continuous with one investing the entire conidiophore. The formation of this commenced on the lower part of the stalk and then extended over the stalk and vesicle after metula formation. Its development on the sterigmata coincided with the development within them of lomasomes after septum formation (Fig. 4, 5*c*). The apices of second and later phialides emerged through the lamina, but new spore buds carried the lamina as they were added to the basipetally extending spore chain (Fig. 1*e*, 4).

Within the agar medium electron-dense material was arranged in a loose network

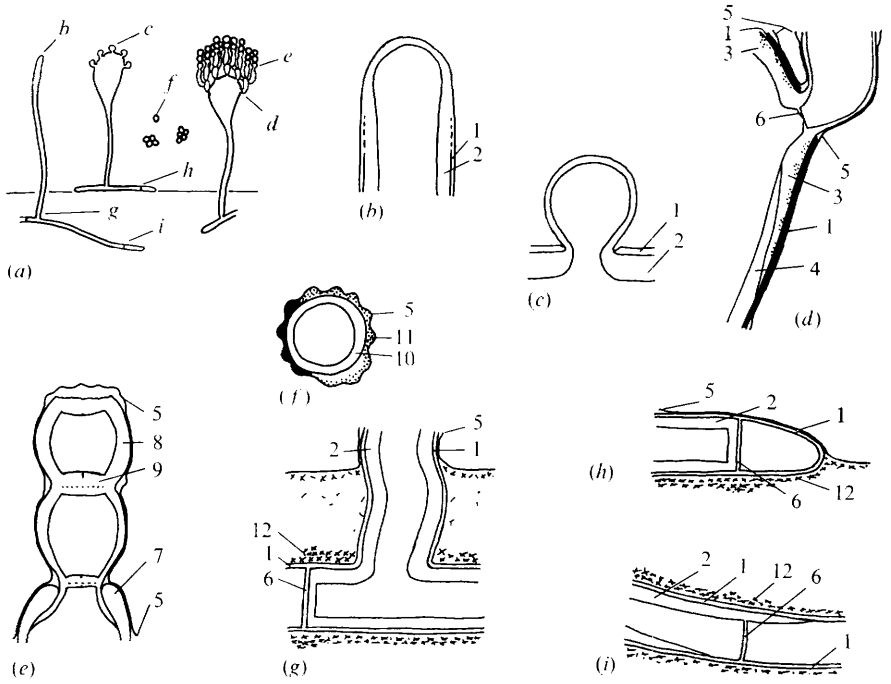


Fig. 1. Schematic diagrams of the arrangement of wall layers in the conidiophore and conidiospore at selected stages of development (not to scale). (*a*) Whole conidiophores as in a section of a colony, with key to diagrams (*b*) to (*i*); (*b*) apex of 'bristle' or of conidiophore initial; (*c*) origin of metula; (*d*) mature conidiophore; (*e*) conidiospore development; (*f*) mature conidiospore; (*g*) stalk and foot below agar surface; (*h*) foot and hypha at agar surface; (*i*) remote end of foot.

Key to numbering of wall features in Fig. 1 to 5: (1) primary wall, (2) secondary wall, (3) persistent 'grey' secondary wall, (4) secondary wall with 'white' contrast, (5) superficial lamina, (6) septum, (7) collarette, (8) side wall of spore, (9) cross wall of spore, (10) mature spore wall, (11) pigmented material of spore, (12) electron-dense network around basal mycelium.

around older hyphae (Fig. 1, 2*c*). At the agar surface this was replaced by a lamina which overlay both the hyphae and the agar; this and the conidiophore lamina were continuous, while the portions of the stalk within the agar eventually developed a network (Fig. 1*g–i*).

*Conidiophore wall pigmentation.* At about the time the spores were first being formed the wild-type conidiophores became a dark brown colour; exhausted sterigmata also appeared brown. In spore-bearing conidiophores the primary wall had become entirely electron-dense (Fig. 2*a*). Wild-type conidiophores during pigmentation were compared with those in *ivory* mutant *ivoA1* (in which the stalk is colourless while the vesicle dome and sterigmata may eventually become light brown), with the colourless 'bristles' of mutant *brlA1* and with the light brown 'bristles' of mutant *brlA9*; conidiophores were sampled up to 24 h old, when they would be fully developed and their cytoplasm only recently autolysed.

Development of pigment was thus correlated with the deposition of electron-dense material, first at the two surfaces of the primary wall (Fig. 2*b*) and then progressively throughout the structure of that layer. All *ivoA1* stalks and most *brlA1* 'bristles' developed the superficial dense lamina but no other dense material. The vesicles in *ivoA1* and a number of *brlA1* 'bristles' showed rudimentary development of the electron-dense material

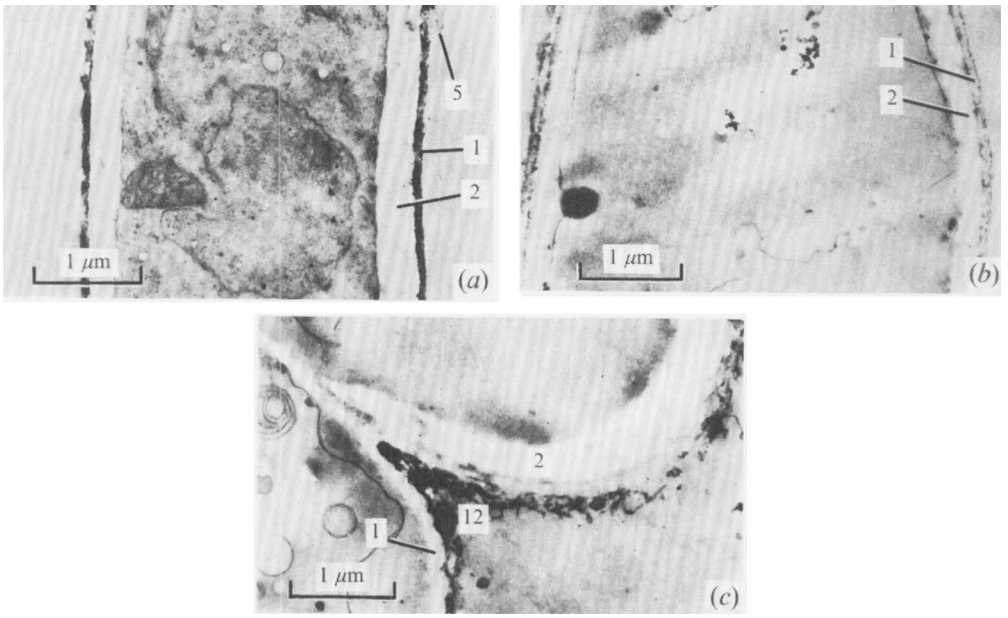


Fig. 2. Sections of conidiophores 12 to 24 h old (with autolysing cytoplasm) showing distribution of electron-dense wall material. (*a*) Wild-type stalk with primary wall fully pigmented; (*b*) *brlA9* 'bristle' with pigment deposited at the two boundaries of the primary wall; (*c*) foot (right) and adjoining hypha (left) of *ivoA1* showing external dense network.

more often than brown pigment was visible in the light microscope. The 'bristles' in *brlA9* showed a varied degree of development of the electron-dense material.

The secondary wall of pigmented conidiophores was not modified where it had matured to the 'white' contrast. The vesicle dome, where the 'grey' contrast had persisted, showed irregular invasion of dense material at the outer surface of the secondary wall adjacent to the pigmented primary wall (Fig. 1 *d*); an invasion of dense material occasionally appeared also at the surface of older sterigmata below the superficial lamina.

*Spore wall maturation and pigmentation.* Newly delimited spores had a thin side wall. The cross wall developed as a single layered, rapidly thickening septum which divided at the future plane of abscission before the next bud developed below it. In the next one or two spores above the new one the side wall and cross walls thickened, apparently independently, so that the cross wall was more or less biconvex until the spore finally matured (Fig. 1 *e*). A pore canal, 0·05 μm wide, persisted in the thickened cross wall, as shown by Fletcher (1971). No subdivision of the electron-lucent wall was observed in immature or mature spores.

The newest five or so spores (the lowest) in each chain of the spore cluster were white and not pigmented green. In embedded colonies the fifth and sixth spores were only occasionally retained on the chains, but these always appeared similar to most of the free-lying spores. In mature spores thus distinguished the surface lamina was a prominent, markedly corrugated envelope with the corrugations filled by electron-dense material, either continuous or formed into a network of fibrils permeated by the embedding resin (Fig. 1 *f*). The dense material could be associated with the green pigment by comparison with the *white-spore* mutant *wA9*, in which it was reduced though not absent, and with the *dark-spore* mutant *drkA1*, containing more alkali-extractable pigment, in which the dense

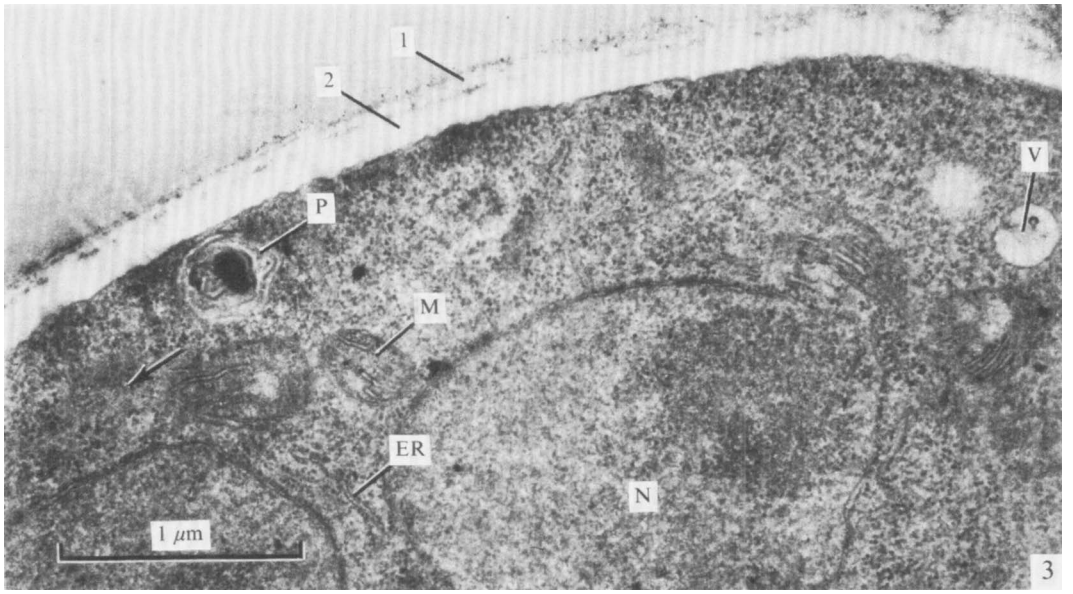


Fig. 3. Section of a growing conidiophore stalk showing double wall and dense cytoplasm. Arrow, microtubule; ER, endoplasmic reticulum; M, mitochondrion; N, nucleus; P, plasmalemmasome; V, vacuole.

material was especially prominent (C. Mucci, unpublished). This material probably constitutes the pattern of rodlets seen in surface view by Hess & Stocks (1969), especially as they note that an overlying sheet can be revealed after vacuum etching which could be identified with the superficial lamina. The electron-lucent wall of the mature spore was thinner than in immature spores (0·20 to 0·28 $\mu$m with a mode at 0·23 compared with 0·32 for the third and fourth spores from the phialide) and had a more sharply defined surface, being more 'white' in contrast with the resin. Irregular zones occasionally occurred between the electron-dense material and the electron-lucent wall or between this and the plasmalemma; the content of these zones was indistinguishable from the embedding resin and they are therefore not interpreted as additional wall layers.

*Organelle distribution.* In the growing conidiophore stalk the organelles were arranged similarly to those in mycelial hyphae (Fig. 3, 4). The apex showed no defined organization except groups of microvesicles (McClure, Park & Robinson, 1968; Grove & Bracker, 1970; the term 'microvesicle' is used here to distinguish these cytoplasmic bodies from the vesicular head of the conidiophore itself). Between the apex and the nearest nuclei was a densely packed zone of ribosomes and mitochondria. Vacuolation developed in the older regions of the stalk in association with bodies morphologically similar to autophagic vacuoles (De Duve & Wattiaux, 1966). Plasmalemmasomes and lomasomes (Heath & Greenwood, 1970) were sparsely and non-specifically distributed.

During the swelling of the conidiophore apex to form the vesicle the apical microvesicles were occasionally scattered or in clusters throughout the entire volume (Fig. 4). The next stage was sharply distinguished by apical microvesicles occurring in concentrated clusters at the surface of the vesicle dome, immediately adjacent to conical pits in the inner surface of the secondary wall, over each of which the primary wall was stretched as a shallow superficial papilla; this stage was rarely observed, as if it involved rapid development.

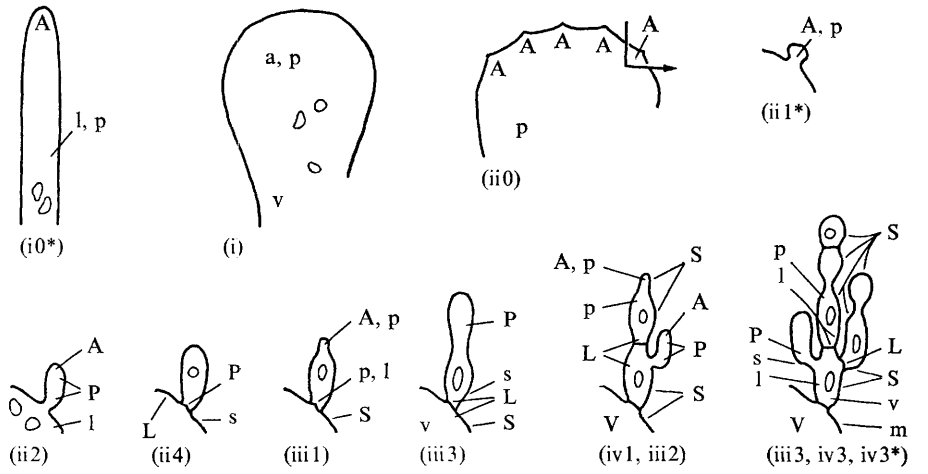


Fig. 4. Organelle distribution summarized in schematic diagrams of selected stages with cytoplasm and nuclei outlined (not to scale). * Stages corresponding to Fig. 1; (i) to (iv) development of vesicle, metulae, phialides and spores respectively; (0) to (4) corresponding stages of budding cycles; a, apical microvesicles; l, lomasomes; m, melanization of primary wall; p, plasmalemmasomes; s, superficial lamina; v, vacuolation of cytoplasm; capital lettering shows frequent (60 to 100 %) occurrence.

Metulae were first seen as small spherical buds (0·8 to 1·5 $\mu$m) furnished with apical microvesicles or plasmalemmasomes and invested by a thin wall; spores and first phialides arose as similar small spheres whereas later phialides were never seen shorter than 2 $\mu$m. The development of metulae, phialides and spores was effected by a budding cycle in which a uninucleate growing cell became cut off from the adjacent cell which then matured autonomously (Fig. 4). Nuclear division did not take place until buds were fully enlarged; the first three budding cycles were synchronous on each conidiophore, although synchrony later deteriorated. Metula formation requires approximately three mitoses in immediate succession in the vesicle to match the number of buds (Clutterbuck, 1969*b*); in vesicles with new buds 4 to 5 $\mu$m long the nuclei contained bundles of tubules similar to those observed in hyphal spindles by Robinow & Caten (1969), terminating at a diffuse area of the nuclear membrane, not at a centriole. Nucleated buds always had a forming or completed cross-septum; the event of nuclear migration has not been observed in living or fixed cells, except where apparently retarded in a bud restricted by its neighbours.

The apical budding pattern which formed the sterigmata involved the maintenance of microvesicles at the growing apex aligned against it as well as, or instead of, being clustered near to it (Fig. 5*a*). Plasmalemmasomes were common at the near apical and near basal shoulders of these buds; in full-size buds they were frequently near or attached to the newly forming cross septum (Fig. 5*b*) or flattened against it after its formation (Fig. 5*c*). In the older cell cut off by the new septum plasmalemmasomes, lomasomes and a spectrum of intermediary forms occurred flattened against any part of the cell wall (Fig. 4); in the vesicle at this stage plasmalemmasomes were also associated with the autophagic vacuoles.

The interstitial (basipetal) budding pattern of sporogenesis involved no microvesicles or plasmalemmasomes within the enlarging spore bud, though they occurred there when it was first formed. Plasmalemmasomes occurred in the phialide near the collarette, especially near the end of each budding cycle. After delimitation the spore plasmalemma was

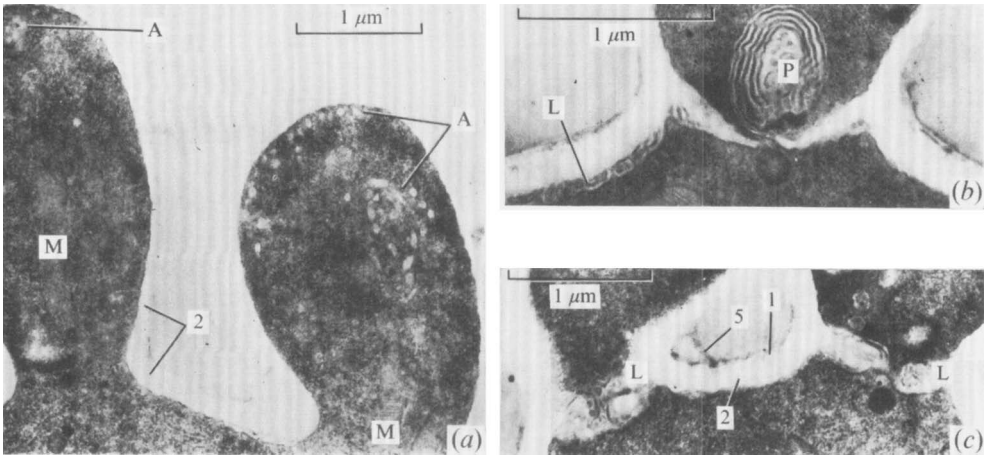


Fig. 5. Sections of metulae during development. (*a*) Varied arrangement of apical microvesicles (A); (*b*) base when newly delimited (cell length 4·6 μm) showing plasmalemmasome (P) attached to septal pore and, in the vesicle, commencement of lomasome formation (L); (*c*) bases during phialide formation (length to growing apex 8·3 μm) showing plasmalemmasome/lomasomes (L) at septa; numbering as in Fig. 1; M, mitochondrion.

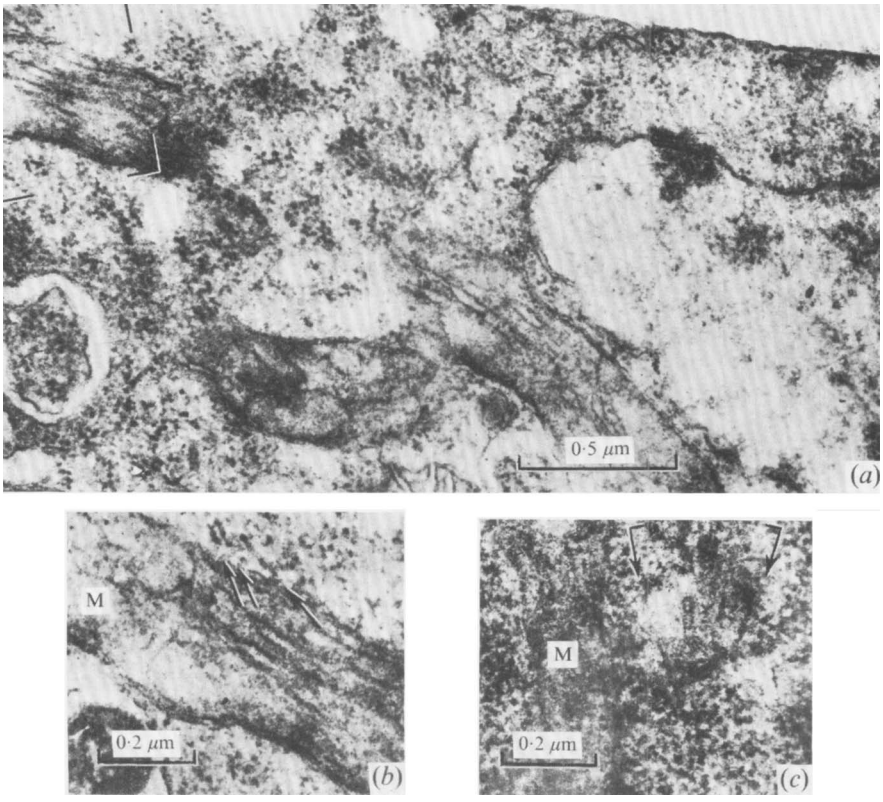


Fig. 6. (*a*) Subapical cytoplasm showing arrangement around organelles; marked area overlaps with (*b*). (*b*) Cytoplasmic filaments (arrowed) next to a mitochondrion (M). (*c*) Cytoplasmic filaments (arrowed sector) around a microtubule parallel to a mitochondrion (M).

commonly convoluted as if by exocytosis, but the membranes there were not as dense as in the plasmalemmasomes. A variety of multimembrane bodies were observed at this stage, including mitochondria within autophagic vacuoles and, occasionally, lomasome-like bodies resembling those described by Weisberg & Turian (1971). In mature spores normal organelles were found, suggesting that autophagy in the immature spore had reduced the number of organelles without eliminating any type.

*Cytoplasmic streaming.* In the living conidiophores and hyphae an elaborate pattern of streaming and pulsation of organelles was observed; the layout of organelles in fixed material reflected this phenomenon (Fig. 6*a*). It was noted that cytoplasmic microtubules were sparsely distributed and occurred neither in bundles nor in any other pattern that could be related to morphogenesis or organelle distribution. More generally found were filaments around 10 nm in thickness and up to 150 nm in length; the corresponding near-transverse sections could not be distinguished from the ribosomes. The longer filaments could be recorded on repeat micrographs and at different planes of focus; they varied gradually in sharpness through focus, unlike the sharp changes in contrast that would have occurred had they been phase contrast effects. These filaments were aligned parallel or at a low angle to microtubules and to elongated nuclei, mitochondria and cell surfaces (Figs. 6*b*, *c*), and were either tangential or near normal to rounded organelles. They occurred, for example, parallel to the cell membrane of the conidiophore vesicle between the bases of new buds, and normal to the cell membrane of a growing spore.

## DISCUSSION

Conidiation appears to involve the formation of at least three types of wall, their morphological distinction presumably reflecting differences in composition. The first type is the continuation of the hyphal wall which extends up to the vesicle, the second extends from the conidiophore foot to the collarette, forming a secondary wall in the conidiophore and the single wall of the sterigmata, and the third is the spore wall which arises within the collarette. The cycle is completed by a new wall layer being formed at germination.

In these terms the action of the *bristle* locus, in some respects analogous to a 'hypha-to-yeast' morphological transition, involves the termination of formation of the first type of wall, while the second breaks through independently. The observation that the *bristle* locus affects the sterigmata as well as the vesicle, as shown by the autonomy of alleles in heterokaryotic conidiophores and by the properties of 'leaky' alleles (Clutterbuck, 1969*a*), suggests that the second type of wall is an important characteristic of this stage of conidiophore development. The initiation of the basipetal budding pattern of the spores, affected by the *abacus* and *medusa* loci (Clutterbuck, 1969*a*), involves the termination of the second type of wall together with the initiation of the third. The eventual coincidence of surface lamina formation with the apex of the second wall appears to mark this transition to basipetal budding.

At the apex of the growing conidiophore stalk the two wall layers appeared to be initially unified. This can be interpreted in terms of the wall materials maturing from an initially plastic secretion with at least one component common to both layers; in young (24 h) hyphae of *Neurospora crassa* (Hunsley & Burnett, 1970) the substructure of the wall is similarly absent at the apex and develops subapically.

During spore maturation the spore wall is reduced in thickness, which could indicate extensive dehydration during the reorganization of the wall. Alternatively it could indicate that the observed single wall was composed of two layers, unified when immature as at

the conidiophore apex, one giving rise during pigmentation to the external electron-dense material and the other to the persistent electron-lucent wall. This would be similar to the development observed in Penicillium (Fletcher, 1971), where the sculptured surface is derived from an outer wall layer; this layer is apparent after permanganate treatment but is much less distinct in the parallel preparations with glutaraldehyde/osmium, in which the wall is more similar to the single layer seen in Aspergillus (using no added wetting agent). As permanganate is known to be a degradative agent (Bradbury & Meek, 1960) the suggested comparison could be studied further by comparing untreated walls with those degraded with specific reagents such as used by Hunsley & Burnett (1970).

Because in hyphae the plasmalemmasomes are non-specifically distributed there has been some doubt as to their function. Girbardt (1969) distinguishes them and the lomasomes from the cytoplasmic vesicles by their cytochemical similarity to the plasmalemma, and concludes they are independent of the exocytotic activity of the vesicles. In the sterigmata, where age is much more precisely defined by bud size, an initial suggestion of interdependence becomes resolved into the strictly apical localization of the microvesicles versus the subapical and basal localization of the plasmalemmasomes. This and their later association with lomasomes strongly supports the hypothesis of Heath & Greenwood (1970) that the plasmalemmasomes arise from the incorporation of apical microvesicle membranes into the cell surface, necessarily in excess of the increase in wall surface area; they may be a collection of this excess membrane which becomes sequestered later. Considering the association of plasmalemmasomes with the septal pore, such collection of the membrane could be associated with septum formation if the limits of this activity determined the position of the septum, especially if two separate surface regions were redistributing their newly expanded membrane, each centrifugally and therefore in opposed directions.

The development of brown pigment in the primary wall of the conidiophore is preceded by the formation of the superficial electron-dense lamina; observations on the localization of free sulphydryl groups (to be published) indicate an association between these two events and lomasome formation. This association implies transmission of materials through the secondary wall, in which pigment formation appears to be excluded where this layer matures to the 'white' modification. The development of green pigment in the spore is similarly restricted to an exterior site under the superficial lamina following cytoplasmic maturation. In the basal mycelium pigmentation (melanization) of the hyphal wall is preceded by the coloration of the medium and it is therefore interesting to note that the electron-dense network developed around older unpigmented hyphae is continuous with the conidiophore lamina; in studies of hyphal melanization (e.g. Bull, 1970) this network could have been excluded by washing from the samples taken from liquid culture. The aerial regions of the conidiophore differ from the basal mycelium in that aqueous excretion products must accumulate at the surface instead of dispersing into the medium; this restricted environment could determine the relatively early pigmentation of the conidiophore, which is well in advance of that in the mycelium even though this comprises already older hyphae.

The association of cytoplasmic streaming with filaments is now well established in a variety of tissues (Wessels *et al.* 1971) but has not to my knowledge been previously reported in filamentous fungi. In many tissues 4 nm filaments are associated with the cell surface and 10 nm filaments with deeper seated sites, but subsurface filaments ranging from 4 to 10 nm have also been reported (Perry, John & Thomas, 1971; Tucker, 1971), while here approximately 10 nm filaments are common to both regions. Identification of filaments is easiest where they form extensive parallel arrays associated with large-scale

movements. The observation that they are not so arrayed here is consistent with the involved pattern of movements seen in living hyphae and conidiophores, which may require great changes of direction over distances of less than 1 $\mu$m to follow the contours of organelles. Taking into account the consequent requirement that the filaments should be short and should lie in varied directions, there is scope for further study of their influence on more general, less unidirectional organelle motility.

It is important to consider cytoplasmic streaming, in addition to regulation of the rate of growth, as a factor controlling morphogenesis through selective placing of organelles. Discussion has usually concentrated on the unipolar extension of a yeast cell or a cylindrical hypha, where this consideration need not apply once the pole has been determined. In the conidiophore, however, the *bristle* locus controls an acceleration of unipolar growth to form the vesicle, followed by a switch to multipolar clustering of microvesicles giving rise to the metulae. Not only has the rate of growth been regulated, but also the position at which it takes place.

I am grateful to Dr A. J. Clutterbuck for his continued interest and encouragement, and to other colleagues for discussion. I thank Mr C. Mucci for skilled and patient technical assistance.

## REFERENCES

BRADBURY, S. & MEEK, G. A. (1960). A study of potassium permanganate 'fixation' for electron microscopy. *Quarterly Journal of Microscopical Science* **101**, 241–250.

BULL, A. T. (1970). Chemical composition of wild-type and mutant *Aspergillus nidulans* cell walls. *Journal of General Microbiology* **63**, 75–94.

CLUTTERBUCK, A. J. (1969*a*). A mutational analysis of conidial development in *Aspergillus nidulans*. *Genetics* **63**, 317–327.

CLUTTERBUCK, A. J. (1969*b*). Cell volume per nucleus in haploid and diploid strains of *Aspergillus nidulans*. *Journal of General Microbiology* **55**, 291–299.

CLUTTERBUCK, A. J. & COVE, D. J. (1972). The genetic loci of *Aspergillus nidulans*. In *Handbook of Microbiology*. Edited by H. Lechevalier. Cleveland: Chemical Rubber Company. (In the press).

DE DUVE, C. & WATTIAUX, R. (1966). Functions of lysosomes. *Annual Review of Physiology* **28**, 435–492.

FLETCHER, J. (1971). Conidium ontogeny in Penicillium. *Journal of General Microbiology* **67**, 207–214.

GIRBARDT, M. (1969). Die Ultrastruktur der Apicalregion von Pilzhyphen. *Protoplasma* **67**, 413–441.

GROVE, S. N. & BRACKER, C. E. (1970). Protoplasmic organisation of hyphal tips among fungi: vesicles and Spitzenkörper. *Journal of Bacteriology* **104**, 989–1009.

HEATH, I. B. & GREENWOOD, A. D. (1970). The structure and formation of lomasomes. *Journal of General Microbiology* **62**, 129–137.

HESS, W. M. & STOCKS, D. L. (1969). Surface characteristics of *Aspergillus* conidia. *Mycologia* **61**, 560–571.

HUNSLEY, D. & BURNETT, J. H. (1970). The ultrastructural architecture of the walls of some hyphal fungi. *Journal of General Microbiology* **62**, 203–218.

MCCLURE, W. K., PARK, D. & ROBINSON, P. M. (1968). Apical organisation in the somatic hyphae of fungi. *Journal of General Microbiology* **50**, 177–182.

PERRY, M. M., JOHN, H. A. & THOMAS, N. S. T. (1971). Actin-like filaments in the cleavage furrow of newt egg. *Experimental Cell Research* **65**, 249–253.

ROBINOW, C. F. & CATEN, C. E. (1969). Mitosis in *Aspergillus nidulans*. *Journal of Cell Science* **5**, 403–431.

TRINCI, A. P. J., PEAT, A. & BANBURY, G. H. (1968). Fine structure of phialide and conidiospore development of *Aspergillus giganteus* Wehmer. *Annals of Botany* **32**, 241–249.

TUCKER, J. B. (1971). Microtubules and a contractile ring of filaments associated with a cleavage furrow. *Journal of Cell Science* **8**, 557–571.

WEISBERG, S. H. & TURIAN, G. (1971). Ultrastructure of *Aspergillus nidulans* conidia and conidial lomasomes. *Protoplasma* **72**, 55–67.

WESSELS, N. K., SPOONER, B. S., ASH, J. F., BRADLEY, M. O., LUDUENA, M. A., TAYLOR, E. L., WRENN, J. T. & YAMADA, K. M. (1971). Microfilaments in cellular and developmental processes. *Science, New York* **171**, 135–143.