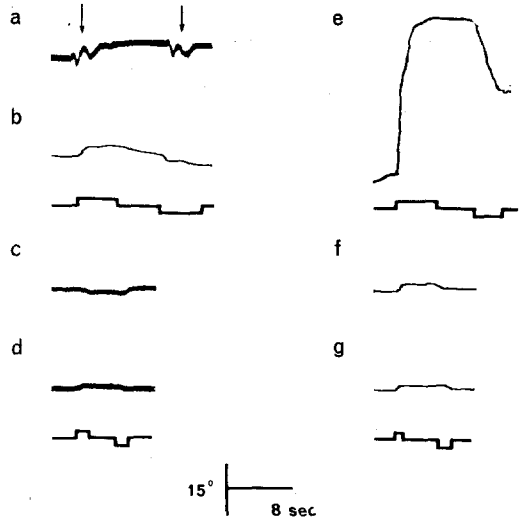
back to its original position as either a black or white stripe passed across the eye.

A further difference in response was seen when a single black-white edge was passed across the left eye. Figure c) and d) show the response of a female *Pieris* to small movements of a white-black edge (white anterior) and the black-white edge (black anterior). Although the female's head followed the movement of the black-white edge in both directions, the head turned in the opposite direction when the white-black edge was moved. The males followed the motion of both edges (Figure f and g).



The head movement responses of male and female *Pieris* to movement of 90° stripes and single edges. Upward deflection in both head movement and drum marker traces indicates movement to the right. Vertical calibration shows degrees of head turning. Speeds for large movements of broad stripes were approximately 90°/sec and for small movements of single edges 8°/sec. a–d) responses of female; a, b) to movement of 90° stripes. Arrows point to oscillations induced by the broad stripes. c) Response to white-black edge and d) black-white edge. e–g) Responses of male *Pieris*; e) to movement of 90° stripe, f) to movement of white-black edge and g) black-white edge.

Black-white edge (both directions)		White-black edge (both directions)	
Males (%)	Females (%)	Males (%)	Females (%)
72.5	100.0	70.5	2.3
97.5	64.0	66.6	0.0
91.0	83.5	93.5	0.0
84.0	86.4	88.5	12.8
	98.8		56.7

For each animal data from approximately 40 tests with each edge were expressed as the percentage of movements where the head moved in the direction of drum rotation (Table).

Tests where the head did not move at all were excluded (up to 1 in 6 trials). Therefore 0% indicates a 100% reversal. A χ^2 test on the pooled data shows a significant difference at the 0.1% level between responses of the male and female to the white-black edge and between responses of the female to black-white and white-black edges. No difference was found between male and female responses to black-white edge or male responses to black-white edge. In the female it is presumably the reversal of responses to one edge that causes the head to oscillate when very broad stripes are passed before one eye.

To demonstrate that the reversal response of the female to the black-white edge was indeed an optomotor response either edge was placed in front of the eye in the dark. The head movement was noted when the light was switched on. In the 3 females tested (10 trials with each edge) there was usually no movement (only 1 trial in 5). No preference could be seen for either black or white areas. Therefore there was no phototactic preference.

In the courtship behaviour of *Pieris* the male may pursue either flying males or females. The female exhibits no such behaviour, only alighting immediately after accepting a pursuing male[2]. The male, for accuracy of pursuit, may have different visual requirements from the female, particularly with respect to motion analysis. The difference between the sexes in optomotor responses also reflects a difference in the visual system. This may be either a structural or functional difference in the optic lobes where motion perception is thought to take place[4].

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Zusammenfassung. Männliche und weibliche Kohlweisslinge, *Pieris rapae L.*, zeigen unterschiedliche optomotorische Reaktionen zu einer bewegten Schwarz-weiss-Kante. Es wird angenommen, dass dies auf unterschiedlicher neuraler Integration im Zentralnervensystem der beiden Geschlechter beruht und dass die spezifische optomotorische Reaktion des Männchens bei seinem visuell orientierten Geschlechtsverhalten von Bedeutung ist.

[4] K. MIMURA, Z. vergl. Physiol. *73*, 105 (1971).

Ultrastructure of Epihyphal Rhizoids in *Allomyces arbusculus*

Fungal rhizoids can be defined as non-nucleated thin tubes formed in the basal extension of the main trunk-like hyphae. In aquatic fungi, such basal rhizoids are usually polarly emitted either from zoospores of zygotes[1]. In extensively grown mycelia of *Allomyces*, other rhizoids also appear laterally from the main hyphae and can thus be designated as epihyphal. Their extrusion through the hyphal walls can be highly increased under certain environmental conditions, like the presence of sublethal concentrations of copper ions in the medium[2] or, as recently found and applied in the present study, under the semi-anaerobic conditions of submerged liquid cultivation.

[1] R. EMERSON, Lloydia *4*, 77 (1941).
[2] G. TURIAN, Bull. Soc. bot., Genève *74*, 241 (1964).

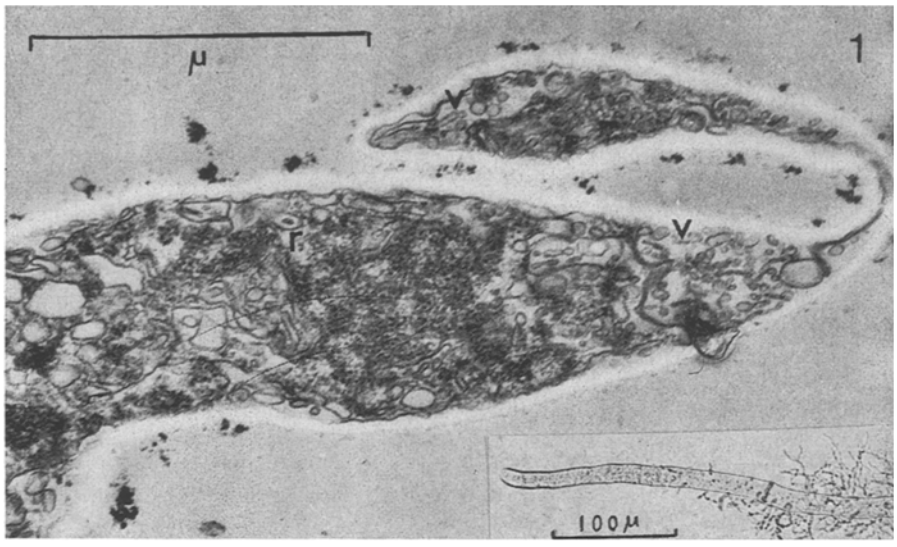

Fig. 1. An epihyphal rhizoid showing vesicula (v) in its tapering, apical portion and ribosomes (r) in the more basal zone. Insert, a rhizogenic hypha.

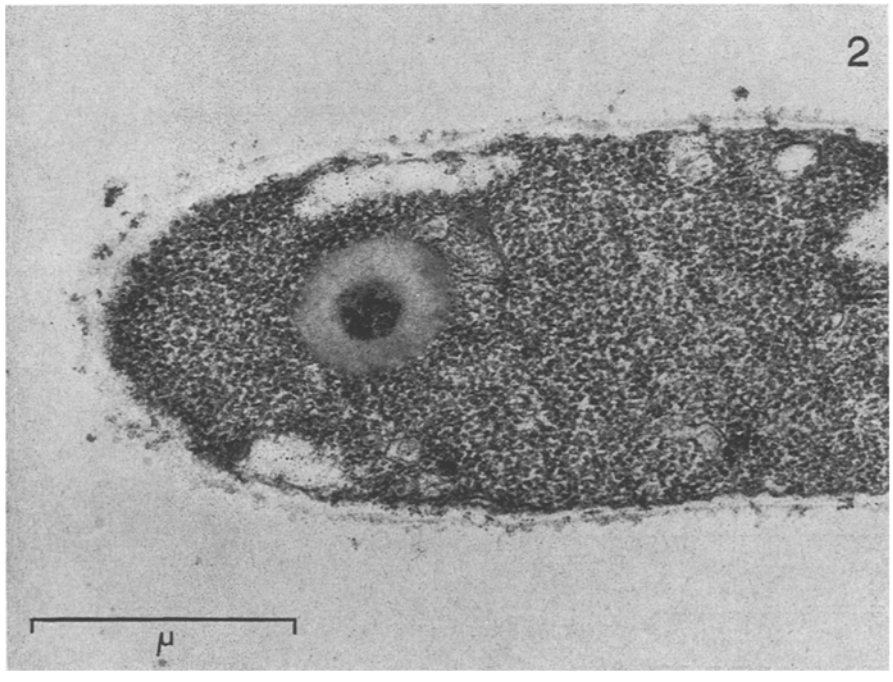

Fig. 2. Apex of a young rhizoid, densely packed with ribosomes.

Reports on ultrastructure of fungal rhizoids are still few and concern only the basal types in aquatic fungi, such as *Rhizophlyctis rosea*[3] and *Blastocladiella emersonii*[4]. In a general ultrastructural study of *Allomyces*[5], we had made only an incidental notice of the epihyphal rhizoids. This has induced us to pursue a more detailed study of such 'organs', which are of great interest because they grow by apical elongation like hyphae but under remote nuclear control, i.e., the nucleus which controls this growth process is present in the supporting hypha.

Allomyces arbusculus has been grown at 25 °C from meiospores inoculated in glucose-casein hydrolysate-yeast extract. 60 ml of this liquid medium[6] was dispensed into 200 ml flasks which induced the formation of small mycelial balls of gametophytic mycelium after 3 days of mild agitation on the shaker.

These balls were washed with distilled water, partially dried with absorbent paper and fixed for 30 min in glutaraldehyde 3% in veronal buffer pH 7.2. After repeated washings in buffer, the material was postfixed in 1% OsO4 in phosphate buffer for 30 min, washed, and stained in uranyl-nitrate. Sectioning was done on a Porter-Blum ultramicrotome and observations were made with the Hitachi electron microscope, model HS-7S.

In a longitudinal section of epihyphal rhizoids, a basal zone more densely populated with ribosomes, and an apical zone containing mainly small vesicula, can be distinguished (Figure 1). Such a topocytological distinction did not appear in the only partially longitudinal sec-

[3] T. C. CHAMBERS and L. G. WILLOUGHBY, J. R. microsc. Soc. *83*, 355 (1964).
[4] P. E. LESSIE and J. S. LOVETT, Am. J. Bot. *55*, 220 (1968).
[5] G. TURIAN and N. OULEVEY, Cytobiologie *4*, 250 (1971).
[6] G. TURIAN, Devel. Biol. *6*, 61 (1963).

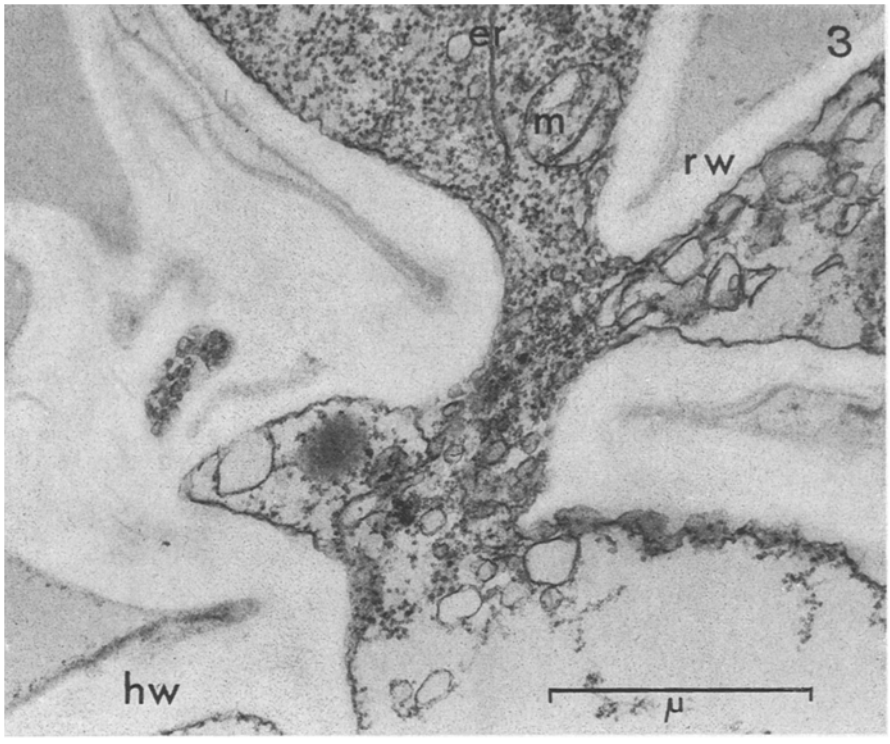

Fig. 3. Rhizoidal outgrowth from hypha with a thick hyphal wall (hw) and thinner rhizoidal wall (rw); element of endoplasmic reticulum (er) and a mitochondrium (m) in the rhizoidal base.

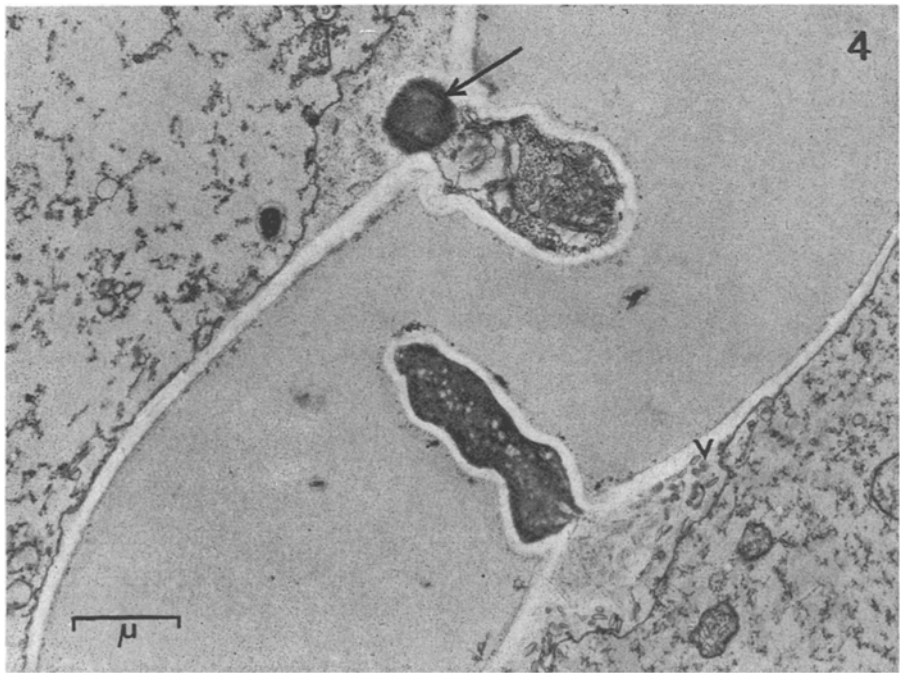

Fig. 4. Short rhizoids plugged (arrow) at their vesicula-rich (v) insertion point on older hyphae.

tion incidently shown in a previous ultrastructural study of *Allomyces*[5].

Maximal density of ribosomal packing is evident in short, young rhizoidal elements (Figure 2) generally showing a few large granules with a dense core. These presumably correspond to the metachromatic, polyphosphate-containing granules previously detected in an optical microscope study[7].

Elements of typical endoplasmic reticulum were rare and mainly present in the basal zone of rhizoidal outgrowth, in which a few poorly cristated mitochondria can be seen (Figure 3). The living characteristics of the rhizoids should be emphasized, especially their high ribosomal content, in sharp contrast with the senescing aspect of their relatively empty supporting hyphae. In the distal rhizogenic region, note the thick wall and a sparse, decreased content of cytoplasmic elements. Short, ribosome-containing rhizoids outgrown from the older,

[7] G. Turian, Rev. Cytol. Biol. vég. *19*, 241 (1958).

more distal portions of such hyphae, can be separated from the autolysed hyphal content by a dense plug isolated behind a restrictive zone containing only small vesicula (Figure 4). The survival of rhizoids, suggested from their still dense, ribosomes-containing cytoplasm is interesting from a physiological point of view.

[8] The support of the Swiss National Research Fund is gratefully acknowledged.

Résumé. Les rhizoïdes d'*Allomyces arbusculus*, émis d'hyphes végétatifs et en conséquence nommés épihyphaux, sont riches en ribosomes quand ils sont jeunes; ils se remplissent par contre de vésicules et ne contiennent plus que quelques mitochondries peu développées dans les plus âgés.

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The Growth of Cell Membranes During Synchronized Cell Division of *Saccharomyces cerevisiae*

The study of biosynthesis of complex structures such as cellular membranes presents several problems like the growth of these membranes in dividing cells. In the present paper, the distribution of radioactivity in the cytoplasmic membranes of mature and young cells labelled by pulse chase technique has been investigated.

Buds and mature cells of yeast *Saccharomyces cerevisiae* strain IMI 140428 cultivated in Edinburgh Minimal Medium[1] with L-Leucine-^{3}H (10^5 cpm/ml) added during the last 30 min of incubation were used. Cells (about 30 g wet weight) taken from the culture in mid log phase were collected by centrifugation and washed twice with the same broth free from radioactivity. Sucrose density centrifugation (tube 80 ml capacity) according to MITCHINSON and VINCENT[2] method was employed to obtain synchronously growing cultures. The top layer ($\sim 5\%$) of cells was collected and incubated in new nonradioactive EMM broth. All the operations were performed at 25°C. Samples of the synchronized cell suspensions were collected on $0.025M$ cyanide during the first division cycle at 60, 75 and 90 min of incubation (in the phase preceding nuclear division) and at 115 min (after nuclear division; doubling time 125 min). The cells collected by centrifugation were drastically shaken in a Waring blendor for 30 sec. In this way, buds are detached from parent cells with a yield ranging from 10 to 25% (Table). The sample containing buds and parent cells was layered on sucrose gradient[2]. After centrifugation, the top layers containing the buds were separated from the other layers containing both the budding cells and large cells. Cells were checked for size distribution by phase contrast microscopy.

Membrane isolation was carried out both on buds and mature cells. For this purpose, the cells were transformed into protoplasts by the EDDY and WILLIAMSON[3] method, using a citrate phosphate buffer $0.005M$ pH 5.8 containing $0.55M$ rhamnose and 1.0 mg/ml freeze-dried snail enzyme (*Helix pomatia*). Membranes were then purified as described by MATILE[4]. Controls for the absence of hexo-

[1] J. M. MITCHINSON and P. R. GROSS, Expl Cell Res. *37*, 259 (1965).
[2] J. M. MITCHINSON and W. S. VINCENT, Nature, Lond. *205*, 987 (1965).
[3] A. A. EDDY and D. H. WILLIAMSON, Nature, Lond. *179*, 1252 (1957).
[4] Ph. MATILE, *Membranes Structure and Function*; FEBS Symposium *20*, 39 (1970).

Specific radioactivity of protein and lipids of purified membranes obtained from mature cells and buds of syncronized cultures of *Saccharomyces*

	Time (min)	Cell recovery[a] (% of total cells layered on the gradient)	Total protein (mg)	Total radioactivity (cpm)	Specific radioactivity (cpm/mg)	Relative specific radioactivity
			Leucine-^{3}H experiments			
Inoculum			3.92	17560	4479	1.00
Top layer (buds)	60	10	0.54	1670	3092	0.69
	75	16	1.09	2878	2641	0.59
	90	19	1.37	3390	2474	0.55
	115	25	1.94	4408	2272	0.51
Other layers (mature cells)	60	90	5.33	15936	2989	0.66
	75	84	5.38	14426	2681	0.59
	90	81	5.66	14619	2582	0.57
	115	75	6.22	14057	2259	0.50
			$^{32}PO_4^{\equiv}$ experiments			
Inoculum			3.65	1270	348	1.00
Top layer (buds)	90	19	1.46	284	195	0.56
Other layers (mature cells)	90	81	5.50	1105	201	0.58

Mature cells and buds were separated by centrifugation in sucrose density gradient. The data are the average of 3 experiments. [a] Cell count in haemocytometers.