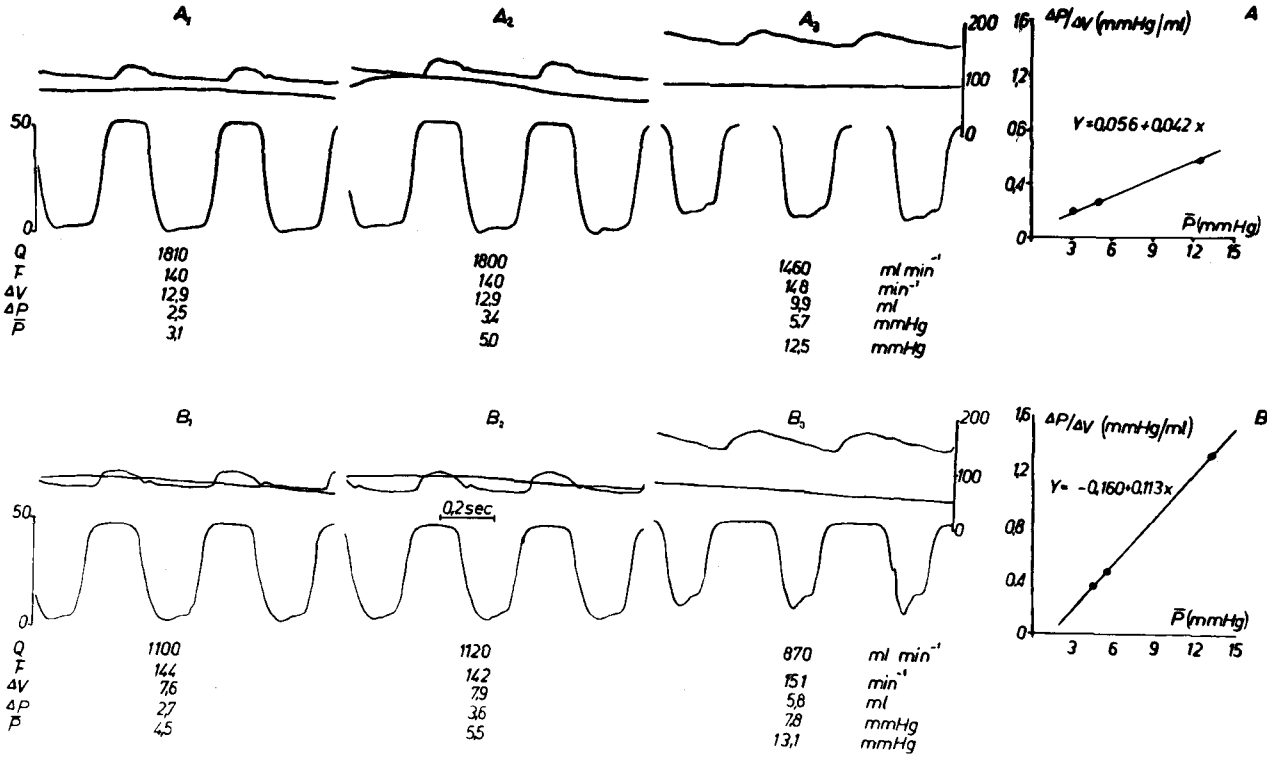


Left ventricular wall stiffness before (A) and after (B) noradrenaline infusion (experiment 3–2, 16 kg, 24 h after coronary ligation). Left: Aortic and left ventricular diastolic pressures at 3 different afterloads (fragments of thermodilution curves also can be seen on the records). Values of cardiac output (Q), heart rate (F), stroke volume ($\Delta$V), intraventricular pressure increase during diastole ($\Delta$P) and mean left ventricular diastolic pressure ($\bar{P}_D$) are in each panel. Right: Plot of the $\Delta P/\Delta V$ ratio againt $\bar{P}_D$. Passive elastic modulus of the left ventricle (slope of the regression line) increased from 0.042 to 0.113.

influence of the sympathetic transmitter. These findings suggest that the increased wall stiffness seen after catecholamine administration is a manifestation of same pathologic pattern induced by myocardial hypoxia. The exact mechanism underlying this pattern remains to be elucidated.

*Summary.* Infusion of noradrenaline (1.0 µg/kg/min body weight, i.v.) brings about an increase of the diastolic wall stiffness in the ischaemic canine heart. Similar effect is not elicitable in the normal heart.

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## Chemical Transfer of Learned Behaviour: No Specific Effect Observed in Rats Trained to Swim Either of two Mazes

The mechanism by which intellectual information is stored in the brain has remained obscure despite many attempts to resolve this important question. One theory holds that brain chemicals might serve as specific memory carriers. Consequently, interanimal transfer of acquired behaviour should be feasible using brain extracts. This has been a controversial issue since the time of the first allegedly successful experiment[1]. Two behaviour-inducing peptides have been isolated from the brains of trained rats; one elicits fear of darkness[2] while the other is sound habituating[3]. It has been argued that both these effects might be non-specific, simply being brought about by alterations in the state of alertness of the recipient animals. Such an argument would not be suitable to invalidate a recent report claiming the successful transfer in mice of detailed spatial information about a maze[4].

The importance of this latter finding, if reproducible, prompted us to repeat the experiment under slightly altered conditions. In particular, a swimming maze with rats was used to minimize the possibility that the animals might exploit odour cues for orienting in the maze. As in the original experiment, 2 different mazes were used to test the specificity of the transferred information, but the 2 mazes were set up as mirror images to facilitate

[1] E. Glassman, A. Rev. Biochem. *38*, 605 (1969). – W. W. Stewart, Nature, Lond. *238*, 202 (1972).
[2] G. Ungar, D. M. Desiderio and W. Parr, Nature, Lond. *238*, 198 (1972).
[3] G. Ungar and S. R. Burzynski, Fedn. Proc. *32*, 367 (1973).
[4] G. J. Radcliffe, jr. and J. W. Shelton, Experientia *30*, 1284 (1974).

comparison. Briefly, 3 groups of donor rats were trained in either maze 1 or maze 2 (Figure 1) or were kept as passive controls without training. Donor brains were extracted and injected into naive recipients. Recipients of each type of extract were divided into 2 subgroups and tested in either maze 1 or maze 2. Rates of learning in recipients were then compared.

*Methods.* All rats were males of the SIV-50 strain weighing 350–400 g. Potential donors (110 animals) were subdivided into 3 groups. Animals of the first group (30 animals) received no training and were used as passive control donors. The other 2 groups (40 animals each) were allowed to adapt to the swimming maze by experiencing the 6 standard pretraining problems of the Hebb-Williams test (3 problems a day, 3 trials per problem)[5] and were then trained in either maze 1 or maze 2 (Figure 1). After the last trial, the 30 animals scoring best (all of them consistently swimming the maze in less than 20 sec) were selected from each of the two trained groups and were used as the maze 1 and maze 2 donors respectively. They were decapitated and their brains were immediately homogenized[6] in the cold with 2 vols. distilled water. The homogenate was stirred for 3 h, centrifuged for 30 min at 25,000 *g*, and the supernatant was frozen until used for injection 3 days later. Potential recipients (100 animals) received the same standard pretraining as the donors. The 90 animals with the best scores were selected and were subdivided into 3 equal recipient groups. Recipient animals were injected i.p. with 0.8 brain equivalent each of either of the 3 extracts (from passive control, maze 1 and maze 2 donors). Each recipient group was subdivided into 2 subgroups which, beginning on the following day, were tested in either maze 1 or maze 2 (3 trials a day for 6 days; maximum duration of trial, 3 min). Animals that by the end of day 3 had not developed proper maze swimming behaviour but instead clung to the partition walls were eliminated (a total of 15 animals, 1–5 rats per subgroup). Testing consisted of measuring the time spent in the maze. All tests were performed double-blind.

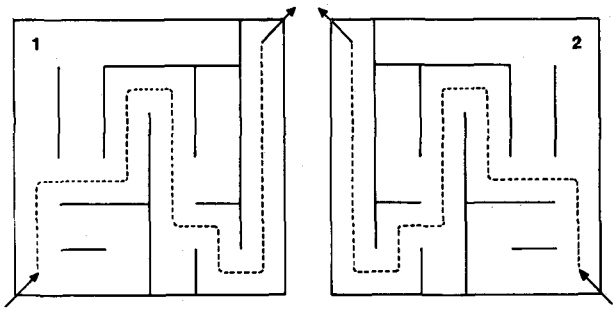


Fig. 1. The two swimming mazes (150 cm × 150 cm) used for training and testing. The arrows denote entrances and exits respectively. The shortest paths are indicated by broken lines.

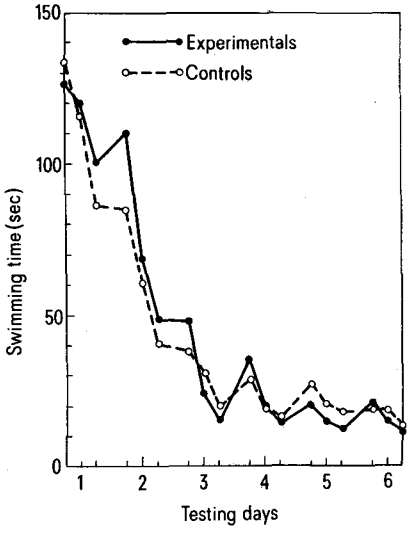


Fig. 2. Graphical representation of pooled learning rates in recipient rats injected with various brain extracts. Swimming time refers to the average time spent by the animals in the swimming maze during testing (ordinate). Recipients were tested in 3 daily trials on 6 consecutive days, day 1 being the first post-injection day (abscissa). Experimentals ($N = 24$) are pooled recipients of homologous extracts ($1 \rightarrow 1$, $2 \rightarrow 2$), while controls ($N = 51$) are pooled recipients of heterologous extracts ($C \rightarrow 1$, $C \rightarrow 2$, $1 \rightarrow 2$, $2 \rightarrow 1$). No significant differences exist between the two plots.

*Results.* In neither of the 6 recipient subgroups ($C \rightarrow 1$, $C \rightarrow 2$, $1 \rightarrow 2$, $2 \rightarrow 1$, $1 \rightarrow 1$, $2 \rightarrow 2$; the first symbol refers to the source of the donor extract and the second to the maze in which the recipient was tested; C = passive control, 1 = maze 1, 2 = maze 2) and at no time during testing did learning rates differ significantly from each other, or from those previously registered with the trained donors. Analysis of variance then showed that intergroup and intragroup variations were indistinguishable statistically. It thus became possible to pool all control pairings ($C \rightarrow 1$, $C \rightarrow 2$, $1 \rightarrow 2$, $2 \rightarrow 1$) and compare them with the pooled experimental pairings ($1 \rightarrow 1$, $2 \rightarrow 2$). This is shown in Figure 2. Learning curves of pooled experimentals and controls were closely similar, and at no point did statistical analysis reveal a significant difference.

*Discussion.* In an attempt to duplicate the reportedly successful transfer of spatial information by brain extracts in mice[4], we were unable to obtain evidence of such a phenomenon occurring in rats. In fact, our experiments completely failed to show any behavioural effect of the brain extracts injected. This raises the question as to whether previous positive findings might not have been attributable to the persistence of odour traces left in the maze by the trained donors. The use of a swimming maze in the present study prevented the build-up of any such cues. This might explain the discrepant results of the two studies. Alternative explanations include a species difference and an increased task difficulty prevailing in the present work. Finally, uncontrollable factors might have influenced either of the two studies giving rise to the contradictory results.

*Summary.* There has been a recent report claiming the successful transfer of detailed spatial information about a maze by injecting brain extracts of trained mice into naive recipients. We have repeated this experiment with rats

[5] K. BAETTIG, Psychopharmacologia, Berl. *18*, 68 (1970).

[6] Glass-teflon homogenizer, 840 rpm, 3 × 1 min; all biochemical operations were carried out at 2–4 °C and were based on a protocol devised by G. UNGAR (personal communication).

using a swimming maze as a precaution against odour cues. No evidence for information transfer has been obtained under these conditions.

*Zusammenfassung.* Kürzlich wurde über ein Experiment berichtet, in dem es gelungen sein soll, die spezifische räumliche Repräsentation eines Labyrinths durch Gehirnextrakte von dressierten Mäusen auf Empfängertiere zu übertragen. Wir haben das Experiment unter Ausschluss von Geruchsspuren (Schwimmlabyrinth) an Ratten wiederholt und dabei keine Informationsübertragung nachweisen können.

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## Fine Structural Characterization of Microbodies and Woronin Bodies in *Trichophyton mentagrophytes*

*Trichophyton mentagrophytes* is a dermatophytic fungus of considerable medical importance. In spite of this, research regarding its ultrastructural morphology[1-4] has revealed only its general aspects, the usual techniques used to date being inadequate to bring to light the finer structural details.

An improved fixation, and new knowledge regarding the substructure of the cell wall[5], were obtained by using a prefixative containing *tris*-1-aziridinyl-phosphine oxide (TAPO), a compound recently introduced with success in biological electron microscopy[6-10].

In this study, we report that, using basically the same technique, a better preservation of the internal structures of the fungus may also be obtained. In particular, it is noted that in the hyphal cells, 2 types of organelles surrounded by a single unit membrane are present, i.e. microbodies and Woronin bodies. These terms are widely accepted and have been morphologically characterized.

Woronin bodies were already identified in foregoing ultrastructural studies in *Trichophyton* spp. and other dermatophytes, not because of their morphology which presented certain ambiguities, but on the basis of their position near the septum, and therefore also called septal or peripheral granules[1-4,11,12]. On the contrary, microbodies have not been identified in dermatophytic fungi.

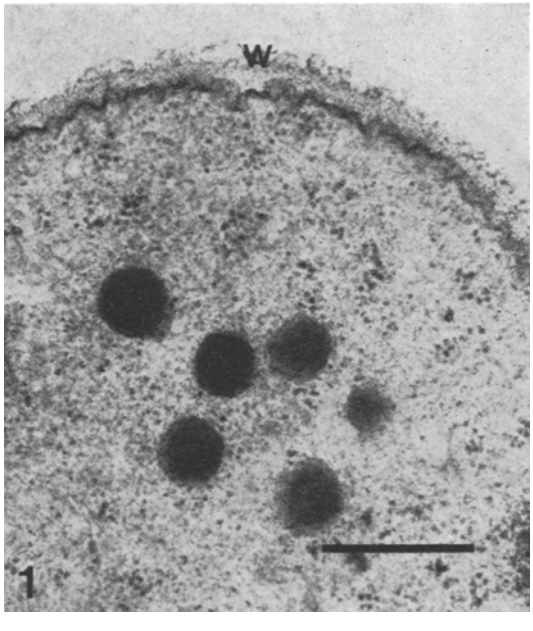


Fig. 1. Cross section through a young hyphal cell of *Trichophyton mentagrophytes* CBS 560.66 showing several Woronin bodies. M, mitochondrion; Mb, microbody; S, septum; W, hyphal wall; Wb, Woronin body. In all the electron micrographs the length of the bar corresponds to 0.5 μm.

*Methods. Trichophyton mentagrophytes*, strain No. 560.66 (Centraal Bureau voor Schimmelcultures, Baarn, The Netherlands), was grown on a Sabouraud maltose agar medium, at 28 °C, on a thin sheet of cellophane, as recently described[13]. Electron microscopic observations were carried out on the youngest hyphae harvested from the outside portion of the cultures in logarithmic phase. The specimens were fixed in a mixture of 6% glutaraldehyde (Eastman Kodak Company, Rochester, N.Y.) and 1% TAPO (*tris*-1-aziridinyl-phosphine oxide; K and K Laboratories Inc., Plainview, N.Y.) in a 0.1 *M* phosphate buffer (pH 6.2), at 4 °C for 2 h. After a brief washing, the samples were postfixed in 1% $OsO_4$ in the same buffer for 1 h, at room temperature, dehydrated in acetone and embedded in Durcupan ACM. Ultrathin sections, mainly cut longitudinally to the hyphal strands, were obtained with a LKB Ultrotome III ultramicrotome and then stained with uranyl acetate and lead citrate and observed through a Jeol JEM-T7 at 60 Kv.

*Results and discussion.* The young hyphal cells of *T. mentagrophytes* contain 2 types of organelles that show the characteristic and distinctive aspects of the Woronin bodies and microbodies only when a prefixative containing a glutaraldehyde-TAPO mixture is used, followed by an osmium postfixation.

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[9] A. CASSONE, Experientia *29*, 1303 (1973).
[10] W. DJACZENKO and C. CALENDA CIMMINO, J. Cell Biol. *57*, 859 (1973).
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[12] G. L. VANNINI and G. DALL'OLIO, Ann. Univ. Ferrara. Sez. IV-Bot. *4*, 127 (1973).
[13] G. L. VANNINI, G. DALL'OLIO and A. BONORA, Experientia *30*, 203 (1974).