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ADMIRALTY MATERIALS LABORATORY, HOLTON HEATH, POOLE

Applied Biology Division

H.M.S. HERMES - FUNGAL GROWTHS IN HYDRAULIC SYSTEM

by

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S U M M A R Y

Growth taken from the hydraulic suction auto-clean strainer of H.M.S. HERMES were found to be a mixed flora of hyphomycetes and ascomycetes; the former was identified as species of Aspergillus while the latter proved to be a sporogenous yeast.

Tests carried out with various concentrations of "Panacide" Sodium showed that a concentration of 1 in 1000 was effective in killing the growths. Recommendations are made for cleaning the system and for adding and maintaining the required concentration of fungicide in the hydraulic fluid.

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H.M.S.HERMES - FUNGAL GROWTHS IN HYDRAULIC SYSTEM1. INTRODUCTION

A sample of a biological growth taken from the forward hydraulic suction auto-clean strainer was received from C.D.L., H.M. Dockyard, Portsmouth, for examination. Subsequently, a visit by a member of H.M.L. staff was made to H.M.S.HERMES and samples of the growth were taken from the miscellaneous hydraulic tank and from the arrester gear tank. The latter was heavily contaminated with oil.

2. THE PATHOGENIC ORGANISMS

On collection, the material consisted of slimy felts of a pale greyish brown colour, which on drying had a cheese-like consistency. Microscopical examination showed it to be a mixture of the vegetative stages of mycelial fungi and a yeast.

Streak cultures of the material were made on wort agar and incubated at 25°C for 14 days. Frequent sub-cultures were made and finally pure isolates of the mycelial species were obtained which were identified as Aspergillus candidus Link and Aspergillus flavipes (Bainier and Sartory) Thom and Church.

Aspergillus candidus, when pure, produces pale buff coloured colonies which have paler margins. The mould grows vigorously and quickly forms dense hyphal mats, which when wetted give rise to the slimy felts found in the auto-clean strainer.

From the hyphal mat, numerous conidiophores arise which bear globular white masses of microscopic spores. These spores are extremely minute and the slightest air current is capable of spreading them over considerable distances. Fig. 1 shows several fruiting bodies of Aspergillus candidus, each of which would be capable of producing many thousands of spores. Aspergillus flavipes is somewhat similar in appearance but produces columnar heads of spores, particularly in old cultures. The colour of the colony is buff or greyish-buff, often tinted with pink or brown.

The yeast element of the flora is probably a wild saccharomyces and gives the material its sliminess. This organism, shown in Fig. 2, is unicellular and the cells are broadly elliptical, 5-7 $\mu$  long, 2-5 $\mu$  wide and usually slightly broader at one end. Wild yeasts are common in Nature and it would be impossible to determine the origin of the infection. As the yeast constituted a very small proportion of the total bulk of material, its specific determination was not attempted.

3. EXPERIMENTAL

Previous research on a similar problem of mould growth in the hydraulic system of H.M.S.CENTUR (H.M.L. Report No.B/15(C) December 1960), showed that of the several fungicides examined, "Panacide" Sodium (sodium salt of 5:5' dichloro 2:2' dihydroxy diphenyl ether) was likely to solve the problem. Recommendations were therefore made that "Panacide" Sodium should be added to the glycerine/water mixture of H. S.CENTUR so that the resultant strength should be about 0.1%. Examination of the system 7 months after the initial addition indicated that this treatment had been successful.

Since, however, the system in H.M.S.CENTUR had been infected with a mixed flora of Aspergillus spp. quite different from that found in H.M.S.HERMES, experiments were carried out to determine whether, and at what concentration, "Panacide" Sodium would inhibit growth of the Aspergillus candidus/yeast flora.

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For this purpose, "Panacide" Sodium was added to tubes of glycerine/water hydraulic fluid to give concentrations of 1-5%, 1-10%, 1-20%, 1-50%, 1-10,000, 1-20,000, 1-50,000, 1-100,000 respectively. Six 25 ml replicates of each were prepared. Controls without the addition of the fungicide were also set up. Spores from a mixed culture of the Candida candida/yeast flora were transferred to the surface of 2 cm. diameter cellulose acetate membrane filters and one filter was floated on the surface of the liquid in each tube. The tubes were incubated at 30°C for one month.

Visual observation before the filter membranes were removed from the tubes of hydraulic fluid showed that heavy fungal growth had developed in all tubes containing concentrations of the "Panacide" Sodium below 1-50%. (Fig. 3). In the tubes having the higher concentrations of toxic substance the filter membranes were removed and carefully washed free from "Panacide" with sterile distilled water on a sintered glass filter. They were then plated out on petri dishes of wort-agar and incubated for 14 days at 30°C.

After 7 days, positive growth was visible on the filters from the tubes containing 1-20% and 1-50% "Panacide" Sodium. Filter membranes from the higher concentrations of 1-100 and 1-500 showed no fungal development.

The results after 14 days are shown in Fig. 4., from which it will be seen that the 1 in 1000 concentration prevented mould growth but that the concentration of 1 in 20% was not sufficiently effective.

#### 4. CONCLUSIONS AND RECOMMENDATIONS

The elimination of the mould will depend upon the thorough cleansing of the whole hydraulic system and the addition of "Panacide" Sodium in sufficient concentration (1-1000) to the glycerine/water fluid.

The tanks, pumps, filters, etc., and such other parts of the system that are accessible should be brushed down with a solution made by adding 3 or 4 oz. of the 40% solution of "Panacide" Sodium (as supplied by the manufacturers) to 2 gallons of fresh water (1 teacup to 1 bucket of water). When all visible traces of the mould have been removed, the system should be filled with fresh water to which "Panacide" Sodium has been added (2 pints of the 40% "Panacide" Sodium as supplied to 100 gallons of water). The mixture should remain in the system for 2 or 3 days during which time it should be circulated occasionally by operating the pumps. Experience has shown that it is important to remove as much growth as possible so that the chances of re-infection are reduced to the minimum.

The system should then be drained and filled in the usual manner with the 40/60 hydraulic fluid and "Panacide" Sodium to give a 0.1% solution; (2 pints of "Panacide" Sodium 40%, as supplied by the makers to 100 gallons is a convenient measure for practical purposes).

Experience in H.M.S.CENTUR has shown that minute traces of mould growth begin to re-appear after 3 or 4 months if less "Panacide" is used. These traces, however, might originate from particles that have become dislodged from inaccessible parts of the system by the operation of the pumps and have so escaped the original cleaning down.

In order to eradicate the infection completely it is recommended that the hydraulic fluid, together with the "Panacide" Sodium, should be changed every 3 months or, if this is not convenient, that a further 1 pint of "Panacide" per 100 gallons of hydraulic fluid should be added, if traces of mould are observed.

"Panacide" Sodium is obtainable from Messrs. British Drug Houses Ltd., Poole, Dorset.

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FIG. 1. *Aspergillus candidus* Link, showing conidiophores with spores, taken from the suction auto-clean strainer of the hydraulic system.  
 $\times 300$

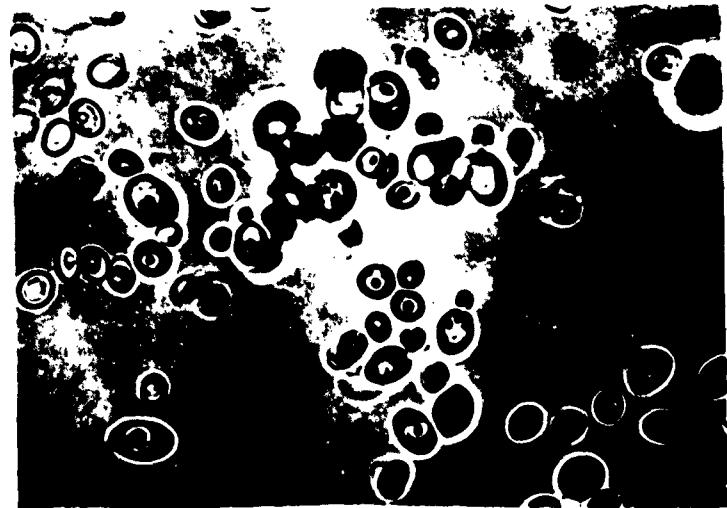
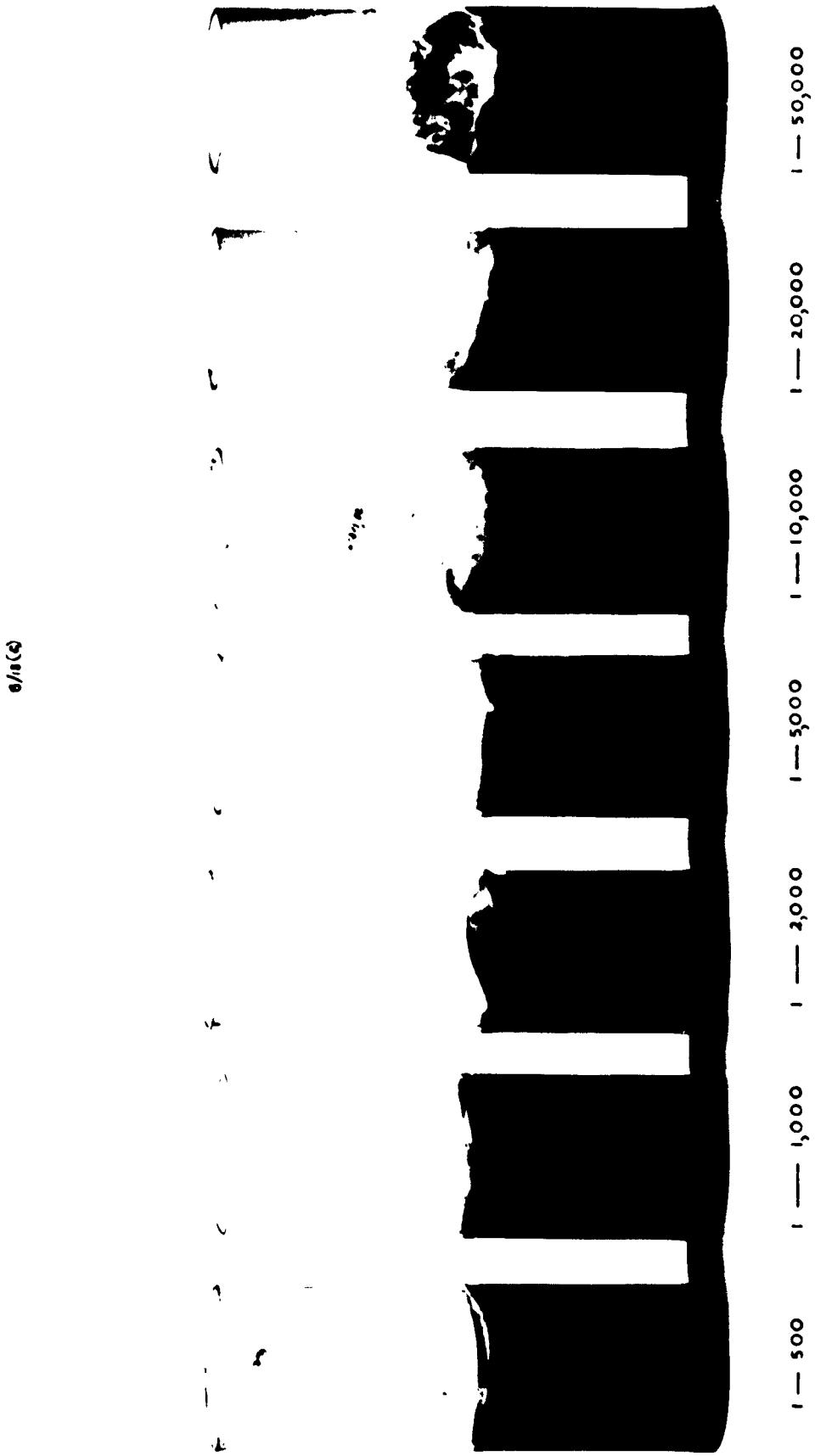


FIG. 2. Yeast cells found living with the *Aspergillus* in the hydraulic fluid.  
 $\times 1500$



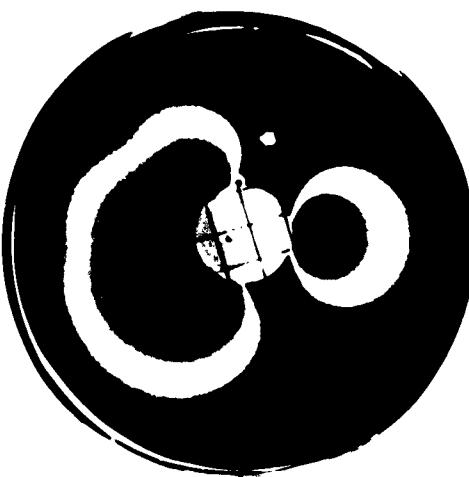
**FIG. 2.** *Aspergillus*/yeast flora growing on the surface of a glycerine/water mixture with various concentrations of "Parasicide" Sodium. Concentrations of 1-500 and 1-1000 showed no growth. 1-2000 and 1-5000 showed very slight growth, below 1-5000 showed heavy fungal growth.



I — 500



I — 1,000



I — 2,000



I — 5000

FIG. 4. Membrane filters on wort agar plates showing growth of *Aspergillus* sp. after being immersed in hydraulic fluid containing "Panacide" Sodium for 1 month. Concentrations higher than 1-1000 completely kill the spores, whereas at 1-2000 or below the spores remain viable and eventually develop.



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