EXPERIMENTAL MADUROMYCOSIS
I. INFECTION OF MICE WITH PHIALOPHORA JEANSELMEI

by
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ABSTRACT

Infection with pure culture of Phialophora jeanselmei was achieved in cortisone-treated mice but not in the untreated group. The peritoneal and subcutaneous lesions 2 to 5 mm. in diameter and 1 to 5 in number, were studied at intervals from 30 days to 85 days by routine histological and histochemical methods and confirmed by culture. The late (85 day) lesions were different from the initial (30 day) lesions in such a manner as to suggest the onset of hypersensitivity at some point of time after 30 days. The suggestive features present in the late and conspicuously absent in the early lesions were (a) marked leucocytic infiltration (b) massive necrosis (c) formation of "grains" by confluence of several necrotic microcolonies (d) fibroblastic proliferation. These findings are of interest to the understanding of clinical mycetoma since the hypersensitivity herein postulated might explain (a) the pathogenesis of human mycetoma (b) the morphogenesis of the "grains" so characteristic of the human lesion. Specific hypersensitivity is yet to be demonstrated and further experiments are in progress.

INTRODUCTION

Maduromycosis, a form of mycetoma characterised by chronic granulomatous inflammation in the feet and the hands, is commonly caused by the species Madurella mycetomi (LAVERN), M. grisea (MACKINNON, FERRADA & MONTEMeyer) and less frequently by Phialophora jeanselmei (LANGERON) according to the reports from India (ANDLEIGH, 1959; KANDHAR et al., 1964; PADHYE & THURMALACHAR, 1966; Klokke et al., 1968), Africa (EMMONS, 1945; ABBOTT, 1956; MURRAY et al., 1964; LYNCH, 1964; EMMONS, 1965) and South America (MACKINNON, 1954).

There was only one report in the literature of successful experimental lesion in rabbits with P. jeanselmei (SYMMERS, 1945). Many attempts, with some success, have been reported with the commoner species namely M. mycetomi on mice by MURRAY et al. (1960 to 1962). Experimental lesions had also been claimed by earlier workers in pigeons (NICOLLE & PINOY, 1908), rabbits (RADELLI, 1911) and mice (BORELLI, 1957). However, the results had not been either uniform or reproducible with regard to the technique or the host.

There is need for a simple and reproducible technique of produc-

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ing experimental lesions by these species of fungi. Such an experimental model would not only aid the understanding of the pathogenesis and the tissue and immunological response but also facilitate evaluation of new antifungal antibiotics such as Hamycin (Thirumalachar et al. 1961). Murray & Colichon (1962) were able to use such lesions caused by M. mycetomi as models to assess the response to antibiotics.

**Materials and Methods**

**The organism**

From the stock culture\(^1\), the fungus was grown on several plates of Sabouraud's agar at pH 5.4 and containing chloramphenicol 0.5 mg/ml of medium. In order to facilitate harvesting of the growth free from the medium the following modification of the technique reported by Murray et al. (1960) was used.

Whatman No 1 filter paper circles of 5 cm diameter with a central hole of 0.5 cm were sterilised and placed on the agar so that the central hole surrounded the seeded grain of the fungus. The fungus grew profusely on the upper and under surface of the filter paper as well as in the medium. For harvesting, the filter papers were lifted off on the tenth day and under sterile conditions the growth was gently scraped off into a container, weighed, ground in mortar and pestle and suspended in 0.9 % saline so that 5 mg of fungus was contained in 0.5 ml of the suspension.

**Injections**

0.5 ml of the suspension was injected intraperitoneally as well as subcutaneously in the left groin.

**Animals**

25 young albino mice from our stock colony, each weighing 15 to 20 g at 5 to 6 weeks of age were used. Group I and II of 10 animals in each were treated similarly with the fungus except that those in Group I received 2.5 mg of hydrocortisone acetate subcutaneously 24 hours before and weekly after the injection of the fungus suspension. 5 animals in Group III served as controls.

3 animals in Group I died in the first week and showed no lesions. The remaining 7 animals were sacrificed at arbitrary intervals at day 30 (2 animals) 45 (1), 55 (1) and 85(2). 10 animals in Group II were sacrificed, 4 at day 85 and 2 each at the other intervals as in Group I.

The lesions were studied by inspection of the size, colour and number. Confirmation by culture was obtained in each case. Slide cultures on corn meal agar were also studied. For histological studies the lesion including the surrounding tissue was fixed in acetic al-

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