

Experimental infection with *Basidiobolus haptosporus*

A. L. Bittencourt¹, W. L. C. dos Santos¹, Z. A. Andrade¹ & M. G. S. Araujo²

¹ Department of Pathology, University of Bahia, School of Medicine, Salvador, Bahia, Brazil

² Centro para Estudo e Controle de Doenças Transmissíveis (LACEN), Salvador, Bahia, Brazil

Abstract

The authors tried to reproduce experimentally the *Basidiobolus haptosporus* infection. Culture forms of the fungus were inoculated in 26 adult hamsters, in two newborn hamsters and in two marmosets. Oral, intratesticular, intrahepatic, intraperitoneal and subcutaneous inoculations were made. Bethametasone was given prior to inoculation in a group of animals.

The lesions produced were only of the foreign body type and there was no development of the fungus in the animal tissues. The AA concluded that an experimental model for the *B. haptosporus* infection has not yet been found.

Introduction

The finding of several cases of subcutaneous infection due to *B. haptosporus* in Brazil led us to attempt to reproduce the infection experimentally for further studies of its immunopathology and mechanisms of transmission (1, 2).

Since Coremans-Pelseneer (3) reported the experimental reproduction of this infection in the hamster, we decided to use this animal as an experimental model for this infection, employing the same method of inoculation for our own studies.

Material and methods

The methods described by Coremans-Pelseneer (3) were used for 26 hamsters with weights varying from 80 to 120 grams, two 13-day old hamsters (*Mesocricetus auratus*) and two marmosets (*Callithrix jacchus*). Inocula were prepared from cultures from two patients according to previously described technique (3). Three oral, three intratesticular, two intrahepatic, five intraperitoneal, and 13

subcutaneous inoculations were made in the hamsters. Bethametasone, 0.001 mg/g, daily, was given two weeks before inoculation up to two weeks afterwards to six of the hamsters inoculated subcutaneously and to two of the hamsters inoculated intraperitoneally. In two hamsters, simultaneous inoculation was made with live and heat-killed (100 °C for 30 min) suspensions. In the marmosets only subcutaneous inoculations were made. Subcutaneous inoculations were evaluated by excision of nodules 5 to 40 days later. Animals inoculated by other routes were autopsied 5 to 50 days following inoculation. The nodules and the organs of the autopsied animals were submitted to histopathologic examination. The sections were stained with hematoxylin-eosin and with the method of periodic-acid Schiff (P.A.S.). Silver impregnation (Grocott's technique) was performed on all sections. Cultures of the subcutaneous and hepatic lesions were taken in 15 instances at periods varying between five and 36 days.

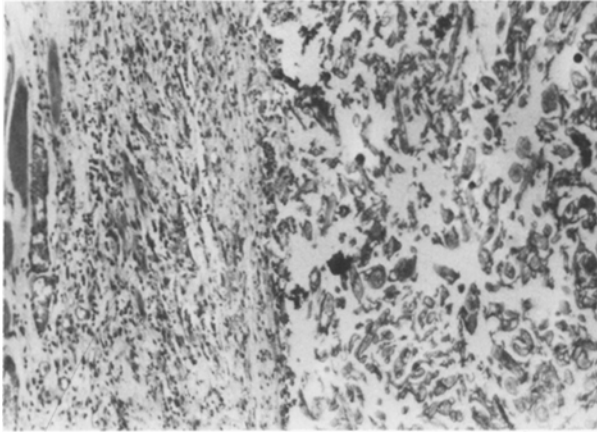


Fig. 1. Hamster. After 11 days of inoculation. See a cavity containing large numbers of the inoculated culture forms of the fungus. The wall exhibited fibrosis. HE. $\times 70$.

Results

No lesions were found in animals inoculated orally or intratesticularly. Nodules varying from 0.5 to 2 cm in diameter appeared at the sites of subcutaneous inoculations. After about six days these nodules reached their maximal size, opened and discharged a yellow, pasty material. Nodules in the animals treated with corticosteroids and in the newborns were identical in appearance to those in the other animals. Nodules with diameters of up to 1.5 cm and with central cavities containing yellow pasty material were found adhering to intestinal loops in the animals inoculated intraperitoneally. In animals injected intrahepatically, autopsy at 9 and 28 days showed rare yellow miliary lesions with maximum diameters of 0.2 cm on the surface of the liver. Two animals which were reinoculated subcutaneously after the disappearance of the primary lesions developed nodules identical to those of the primary inoculation. Injection of heat-killed culture forms resulted in nodules with the same characteristics but smaller than those produced by live cultures. The cultures were positive in two instances (taken 13 and 18 days after inoculation). Histologic examination of the lesions showed cavities containing large numbers of culture forms of the fungus in state of degeneration (Fig. 1). The fibrous wall of these cavities contained infiltrates of neutrophils and mononuclear cells and an intense reaction with

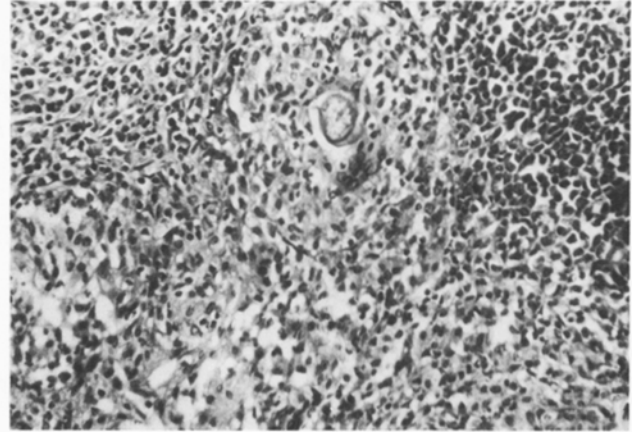


Fig. 2. Hamster. After 9 days of inoculation. Section of liver showing a zygospore within a granuloma. HE. $\times 140$.

macrophages and occasional giant cells. Hyphae were not observed in the walls of the cyst even in cases in which corticosteroids had been administered. In the sections of liver, granulomata containing degenerating zygospores were observed (Fig. 2) in addition to the features present in the other lesions.

Discussion

B. haptosporus produces in man a progressive lesion consisting of a dense infiltrate of mononuclear cells and eosinophils, granulomatous reactions, and hyphae surrounded by PAS-positive eosinophilic material (Splendore phenomenon). But the lesions produced in the present study were inflammatory reactions of the foreign body type. There was no development of the fungus in the tissues. Only degenerated culture forms of the fungus were seen. Furthermore, the inoculation of heat-killed cultures forms provoked the formation of nodules with similar histopathologic aspects, except for their smaller size.

We conclude that an experimental model for *B. haptosporus* infection has not yet been found.

References

1. Bittencourt, A. L., Londero, A. T., Araujo, M. G. S., Mendonça, N. & Bastos, J. C. A., 1979. Occurrence of subcutaneous zygomycosis caused by *Basidiobolus haptosporus* in Brazil. *Mycopathologia* 68: 101-104.
2. Bittencourt, A. L., Araujo, M. G. S. & Paes, M. S. F., 1980. Occurrence of subcutaneous zygomycoses caused by *Basidiobolus haptosporus* with pulmonary involvement. *Mycopathologia* 71: 155-158.
3. Coremans-Pelsener, J., 1974. Biologie des champignons du genre *Basidiobolus*. *Eidam* 1886. Saprophytisme et pouvoir pathogène. *Acta Zoologica e Pathologica* 60: 7-143.