Experimental model of intra-abdominal candidiasis in Swiss mice

Couto, F.M.M.1, Nascimento, S.N.2, Pereira, S.F.1, Silva, V.K.A.1, Leal, A.F.G.3, Neves, R.P.1

Abstract
The objective of this study was to develop an experimental model of intra-abdominal candidiasis in Swiss mice. Adult male albino Swiss mice, weighing approximately 30 g, were obtained from the Department of Antibiotics, Federal University of Pernambuco (UFPE). A single injection of 0.5 mg/kg of dexamethasone was administered intraperitoneally for three consecutive days and then at four-day intervals for the rest of the experiment. Seven days after initiation of immunosuppression, the mice (n=6) were infected by intraperitoneal injection of 0.2 ml of C. albicans (10^7 cells/mL in saline). The mycological diagnosis was made by collecting the liver, spleen and kidneys. The organs were fragmented into pieces and inoculated onto culture medium contained in Petri dishes. It was possible to isolate the fungus from the liver (all animals), spleen (three animals) and kidneys (three animals). This experimental model of the intra-abdominal candidiasis in Swiss mice can be used to evaluate the pathology of fungi as well as the evaluation of new drugs.

Introduction
Fungi can infect virtually any organ or structure in the abdomen. Intra-abdominal fungal infections can be divided in two groups according to their clinical presentation: localized, following surgery, trauma, or placement of foreign device; and disseminated, in critical ill or immunocompromised patients. It is important to recognize these infections early since serious complications, such as intestinal obstruction, bleeding, perforation, peritonitis, sepsis and death, can occur (Rebolledo et al. [2011]). Among fungal agents that can cause intra-abdominal infection, Candida species are the most common (Bassetti et al. [2006]).

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Material and Methods
Adult male albino Swiss mice, weighing approximately 30 g, were obtained from the Department of Antibiotics, Federal University of Pernambuco (UFPE). The animals were treated in accordance with the established experimental procedures after approval by the institution’s ethics committee. We used the immunosuppression protocol from Medeiros et al. [2010]. A single injection of 0.5 mg/kg of dexamethasone (Teuto, Brazil) was administered intraperitoneally for three consecutive days and then at four-day intervals for the rest of the experiment. The experiment was performed in two steps: 1) in the first, the animals were observed for mortality for 14 days; 2) in the second stage, the animals were sacrificed after 7 days of infection for the mycological diagnosis. Seven days after initiation of immunosuppression, the mice (n=6) were infected by intraperitoneal injection of 0.2 ml of C. albicans (10^7 cells/mL in saline). The concentration of C. albicans was adjusted using a spectrophotometer (Thermo Scientific Genesys™ 10S UV-Vis). The animals were observed daily for clinical signs and mortality for 14 days. The animals were euthanized in a CO2 chamber.

The mycological diagnosis was made by collecting the liver, spleen and kidneys. The organs were fragmented into pieces and inoculated onto culture medium contained in Petri dishes. The culture medium used was Sabouraud dextrose agar (SDA) (Difco) supplemented with chloramphenicol (50 mg/mL). The plates were incubated at 37 °C for up to 72 h. Grown yeast colonies were purified, isolated and identified through morphological and physiological characteristics (Barnett et al. [1983]) to confirm the species involved in the infection.

To evaluate the therapeutic efficacy of PO, fungal growth (FG) in culture media was scored according Leal et al. [2015] as follows: 0 – no fungal growth in culture medium; and 1- fungal growth in culture medium.

Results
In the first stage of the experiment, the in vivo mortality rate was six dead animals. This evaluation took place 14 days after infection with yeast. In the second stage of the experiment, the experi-
mental model was repeated and the animals were sacrificed after 7 days. It was possible to isolate the fungus from the liver (all animals), spleen (three animals) and kidneys (three animals).

Discussion and Conclusion
Invasive candidiasis is a frequent and life-threatening complication in critically ill surgical patients (Tissot et al. [2013]). According to Tissot et al. [2013], some 30 to 40% of episodes of recurrent gastrointestinal tract perforation or acute necrotizing pancreatitis are complicated by intra-abdominal candidiasis. Some clinical studies have reported that the morbidity and mortality of intra-abdominal candidiasis vary from 52 to 63% (Tissot et al. [2013]; Dupont et al. [2002]). Treatment of this type of infection is a major challenge because of the difficulty of diagnosis, limited number of antifungals and emergence of strains resistant to the available drugs (Yu et al. [2011]). This experimental model of the intra-abdominal candidiasis in Swiss mice can be used to evaluate the pathology of fungi as well as the evaluation of new drugs.

References