

CONSEQUENCES OF ORAL INFECTION WITH CANDIDA ALBICANS ON THE HISTOARCHITECTONICS OF ORGANS IN MALE RATS

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Abstract

Experiments were conducted to evaluate the immunosuppression's effect on the development of oral candidiasis in Wistar rats. Animals were submitted to immunosuppression in order to facilitate the establishment and persistence of *Candida albicans* infection. Treatment with drugs (dexamethasone and tetracycline) was initiated 7 days before *C. albicans* inoculation and lasted until the end of the experiments, day 9 post-inoculation, but at a higher dose for dexamethasone and at a lower dose for tetracycline. The infection was done twice at a 24 hours difference. Establishment of *C. albicans* infection was done by oral inoculation with 0.1 ml of saline suspension containing $3.108 \pm 1 \times 10^3$ viable cells of *C. albicans* strain ATCC 10231. Tissue injury was determined by the histological quantification of organs collected at days 3, 6 and respectively 9 of the experiment. Our results have shown that candidiasis affected organs in male rats in the following order: tongue, intestines, liver, spleen and kidney. There was a gradual evolution of the histological lesions severity in the tongue starting from the third day towards the end, but, in reverse, all the other organs affected started having regenerative healing processes at the end of the experiment.

Key words: oral, infection, *Candida albicans*, rats, histo-architectonics.

INTRODUCTION

Although a known issue, the presence of *Candida albicans* infections is still commonly reported and studied in animals and humans by many researchers (Chami N et al., 2004, 2005, Takakura N et al., 2003). Candidiasis remains a current topic, *Candida albicans* being structures able to show a strong and extensive action, expressed with specific morbid conditions (mycoses) whose

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evolution are, often, train / rebellious to treatment. Our approach, related to the development of candidiasis in animal studies was aimed as a preamble of a future study wished to confirm or refute the effectiveness of different regimens applied as part of the allopathic and the phytotherapeutical management of these diseases. The objectives of the present research were to identify with certainty the effects of *Candida albicans* infections, on target organs, occurring in immunosuppressed rats, consecutive oral infection and to evaluate the organs histo-cytological changes, as a basis for assessment of the consequences regarding invasive *Candida albicans* infections.

MATERIALS AND METHODS

The in vivo experiment was performed on young Wistar male rats, about the same weight (average weight of 190 +10 grams). For the experiment, immunosuppressed rats were used to highlight the *Candida albicans* infection progress. The animals were maintained on premises especially for this purpose, with temperature and light controlled (22 °C cycle for 12 hours starting at 8), and had free access to food and water ad libitum.

We have used a model of oral candidiasis in immunosuppressed rats that was reported by Martinez i sar., 2001. The rats were immunosuppressed with dexamethasone (Dex) (Corthametasone, Vetoquinol) and treated with tetracycline 4% soluble powder (Tc) (Laprophan) for one week before the experimental infection, rats were given drinking water with 0.5 mg / liter dexamethasone with tetracycline (0.1%). On the day of infection, the dexamethasone dose was raised at 1 mg / liter, while the tetracycline concentration was reduced to 0.01% and was maintained throughout the experiment. Rats were infected orally, twice, every 24 h (days 0 and 1) with 0.1 ml of saline suspension containing $3.108 \pm 1 \times 10^3$ viable cells of *C. albicans* strain ATCC 10231.

Experimental series were composed of nine groups each consisting of three individuals: - lots IS1 - IS3- male rats, immunosuppressed, euthanized at the same time with the treated groups. The batches of animals which were immunosuppressed but not infected were used as negative control groups. - lots infected with *Candida albicans* known as: ICA1, ICA2 ,ICA3 - established from immunosuppressed rats that were infected on day 0 and 1 of the experiment. Rats were euthanized after 3, 6 and 9 days post-infection and samples were taken for histo-cytological investigations.

The organs harvested for samples were: tongue, respiratory tract, intestine, liver, spleen, kidney and brain. Stain samples were collected and processed through the histological technique HE stain or hematoxylin and eosin stain

and microscopy was performed with the x100 and x400 objectives, and the images were processed with an Olympus CX 41 microscope with image capture and data interpretation software.

RESULTS AND DISCUSSIONS

The main histological alterations were identified in: tongue, intestine, liver, spleen and kidney, respectively. For the trachea, lung and nervous system histological changes were not relevant for our study.

Most conspicuous changes were found in the immunosuppressed rat groups, infected with *Candida albicans*. Histologically, the changes regarding the tongue were recorded only for individuals within the immunosuppressed and infected groups; the tongue of subjects only immunosuppressed being without any histological changes.

We observed the progressive development of histological severity of injuries by the end of the third day, the main changes are: parakeratosis, acanthosis, dystrophy of the cells in the lingual epithelium with the discontinuity of the basement membrane. In the intestine of immunosuppressed rats main lesions were found in the first part of the immunosuppression (third day).

The main histological changes were: destruction of large areas of intestinal villi, discrete edema of the intestinal glands and discrete edema of villi axis, periglandular lymphohistiocytic infiltration. The regenerative capacity of the intestine caused the tissue architecture at the end to recover almost completely, situation which was the same also with the immunosuppressed and infected batches, specifying that the lesions in the first half of the experiment were more serious.

In the liver, in the third and sixth day degenerative processes of hepatocytes with balloon dystrophy were observed accompanied by karyolysis and kariopcnosis phenomena which have affected the liver function and the cellular integrity in immunosuppressed male (to a lesser extent) and especially in those immunosuppressed and infected.

In the samples collected on the ninth day normal liver tissue was found which confirms the regenerative healing, observation justified by the return to a normal state of the cytoplasmic and nuclear structure of hepatocytes and also the function restoration.

These observations, although not backed by laboratory tests, which are consider later, shows that although *Candida albicans* infections causes a major affect on the liver within the first six days, this organ has regenerative capacity, which in our case appeared after the ninth day. At a spleene level in both cases there is a dramatic reduction in the first phase of leucopoiesis areas, a pheno-

menon that affects decisively the leukopoetic function, erythropoiesis areas remaining unchanged with a trend towards the recovery of the lymphoid tissue.

In the case of the infected males, splenic limphonodes are present only in the form of lymphocyte chords, the *Candida albicans* infection blocking in a first phase the development of the expected and accepted fungal diseases, that of the leukocyte reactivity and proliferation, but with the restoration of the lymphoid function and architecture at the end of the experimental period. At kidney level, after three days, in infected and immunosuppressed males, an extensive degenerative phenomena was found, manifested by granular dystrophy of the nephrocytes in the distal and proximal contort tubules, Bowman capsule destruction and loss of the uriniferous spaces. These changes affect seriously, in a first phase, the renal function, the phenomena stepping towards glomerulonephritis, but, with the increase of the exposure time until the ninth day, the new developments having a reversible tendency to restore the function in the renal corpuscles. In the case of the immunosuppressed males, these changes were much reduced in intensity and extension.

CONCLUSIONS

1. Candidiasis affected organs in male rats in the following order: tongue, intestines, liver, spleen and kidney in the experimental oral infection.
2. Oral candidiasis did not affect the lung's, trachea's and the nervous system's histo-architectonics, in the experimental oral infection in male rats.
3. In immunosuppressed and infected males there was a gradual evolution of the histological lesions severity in the tongue starting from the third day towards the end.
4. In the liver, in the third and sixth day after exposure degenerative processes of hepatocytes were observed, the analysis from the ninth day indicating the regenerative healing of the cytoplasmatic structure.
5. At spleen level, in both cases, is found, in the first phase, the reduction of the leucopoiesis areas process that affects the leukopoetic function, erythropoiesis areas remaining unchanged with tendency towards lymphoid tissue recovery.
6. In the kidney, after three days, was found, in the case of the infected and immunosuppressed males, the installation of degenerative phenomena, but, with the increase of the exposure time until the ninth day the new developments having a tendency towards restoring the renal corpuscles function.

LITERATURE

1. Chami N., Chami F., Bennis S., Trouillas J., Remmal A.: Antifungal Treatment with Carvacrol and Eugenol of Oral Candidiasis in Immunosuppressed Rats. *Braz. J. Infect. Dis.*, 8, 217- 226, 2004
2. Chami N., Bennis S., Chami F., Aboussekhra A., Remmal A.: Study of anti-candidal activity of carvacrol and eugenol in vitro and in vivo. *Oral Microbiology and Immunology*, 20,106–111, 2005
3. Martinez A., Ferrer S., Santos I., Jimenez E., Sparrow J., Regardera J., De Las Herras G., and Gargallo-Viola D.:Antifungal activities of two new Azasordarins, GW471552 and GW471558, in experimental models of Oral and Vulvovaginal Candidiasis in Immunosuppressed Rats. *Antimicrob. Agents Chemother.* 45, 3304-3309, 2001
4. Takakura N., Wakabayashi H., Ishibashi H.,Teraguchi S.,Tamura Y.,Yamaguchi H, Abe S.: Oral lactoferrin treatment of experimental oral candidiasis in mice, *Antimicrobial Agents And Chemotherapy*, 47, 8, 2619–2623, 2003

Primljeno: 15.02.2010.

Odobreno: 20.02.2011.