Conference presentation

Evaluation of *Leishmania infantum* 30x biotherapy effects in the prevention and treatment of visceral leishmaniasis: *in vivo* and *in vitro* studies

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Background:

Leishmaniasis is a serious public health problem especially in developing countries [1]. The therapeutic potential of biotherapies against several microorganism has been described *in vitro* [2,3] and *in vivo* studies [4,5,6,7,8,9]. Considering the resistance of leishmaniasis to conventional treatment as well as previous studies with biotherapeutic, we evaluated the effects of *Leishmania infantum* 30x (BioLi30x) biotherapy.

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Aim: evaluate the antileishmanial effects of BioLi30x in *in vivo* and *in vitro* models.

Methodology:

The *in vivo* experiments were performed using BALB/c mice (n=138), divided into 8 groups: G1-healthy, G2-infected with *L. infantum*, G3-BioLi30x pre-treated, G4-BioLi30x pre/post-treated, G5-BioLi30x post-treated, G6-H₂O30x post-treated, G7-*Antimonium crudum* 30x post-treated and G8-Glucantime® post-treated. After 49 days of treatment, the animals were submitted to euthanasia (ethical approval ECUA/UFRJ/066/14). Liver and spleen histological changes were evaluated, and serum samples were aliquoted and storage at -20°C for cytokine assays. The *in vitro* assays were performed using RAW 264.7 macrophages treated with BioLi30x and infected with *L. infantum*. The morphological aspects were evaluated by scanning electron microscopy (SEM), and the nitric oxide (NO) release was quantified in the supernatant of infected macrophages.

Results:

The histological analysis from 4 independent experiments showed livers with normal appearance (G1); periportal chronic hepatitis (G2,G4,G5,G8); discrete (G3,G7), moderate (G4,G5,G6), and severe (G2,G8) vacuolar hydropic degeneration; congestion and neutrophilic inflammation (G2,G4,G5,G6,G8), and possible amastigotes within macrophages (G2-G8). Spleens presented healthy appearance only in G1. All treated animals presented histological alterations, with different lesions severity, which involved spleen pulp hyperplasia with moderate disruption (G2,G8), as well as megakaryocytes and macrophages proliferation (G2- G8). SEM analyses showed BioLi30x treatments induced significant protozoan morphology alterations when compared to H₂O30x. Besides, a 19% increase in the NO release was detected in RAW supernatants, when compared to H₂O30x.

Conclusions:

BioLi30x and *Antimonium crudum* 30x modified the infection animal process, involving several cellular mechanisms as well as different histological damage. The *in vitro* experiments will be repeated in order to confirm these preliminary results.

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Keywords: *Leishmania infantum*, *Antimonium crudum*, visceral leishmaniasis, biotherapeutic, nitric oxide.

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We had full access to all data from this study, and we take complete responsibility for the integrity and accuracy of the data analysis.

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