Real Time PCR Development to Quantify the Parasite Load in Golden Hamsters Infected with *Leishmania infantum*

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Introduction: In Americas, visceral leishmaniaisis (VL), a systemic disease characterized by an intense immunossupression and inflammatory response, is caused by *Leishmania* (*Leishmania*) *infantum*. The use of experimental models is essential to investigate the immunopathogenesis of this disease. The Golden hamster (*Mesocricetus auratus*) model best reproduces the human VL disease. In this context, an accurate and rapid method to quantify the parasite load (PL) in infected hamster is a necessary research tool for monitoring drug's efficacy, vaccine candidates and the evolution of VL. **Objectives:** To standardize and validate a real time PCR assay to quantify with sensitivity and reproducibility the PL in hamsters infected by *L.infantum*. **Material and Methods**: Hamsters were intraperitoneally infected with $2x10^7$ amastigotes of *L.infantum* and treated with anti-*Leishmania* therapy after 45 days post infection. Spleen and blood were removed for DNA extraction right before and after anti-*Leishmania* treatment (120 days post infection). qPCR assays for absolute quantification of PL using SYBR Green® will be based on a standard curve produced from the extracted hamster's tissues DNA spiked with *L. infantum* promastigotes. This curve will be designed using a 10-fold serially diluted sample of *L.infantum* DNA, with 10^6–10^{-1} parasites/reaction tube. To perform quantification, kinetoplast DNA (kDNA) and single copy genes will be used as molecular targets and the most sensitive and specific one will be selected. Also, GAPDH target from *golden* hamster will be included as internal reference. **Results and Discussion**: We´ve obtained DNA from *Leishmania*-infected hamsters. Next, we will standardize optimal primers concentrations, annealing temperature and standard curve efficiency of qPCR. We expect to validate this methodology as a parasitological cure criteria in hamsters infected with *L.infantum* to assess the PL throughout the clinical evolution of the animal. **Conclusion:** Syber Green-based real-time PCR could be an accurate, rapid and cheaper promising method to ease the immunopathogenesis studies of experimental VL.

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Keywords: parasite load, real time PCR and visceral leishmaniasis.