

04. ASYMPTOMATIC LEPTOSPIRAL INFECTION IS ASSOCIATED WITH CANINE CHRONIC KIDNEY DISEASE

Infecção leptospiral assintomática é associada à doença renal canina crônica

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Introduction: There are scarce studies associating asymptomatic leptospiral infection with chronic kidney disease (CKD) in dogs, recent research has suggested that in human being asymptomatic renal colonization is a neglected risk for CKD, especially in endemic areas. **Objective:** Investigate whether the occurrence of CKD may possibly be associated with asymptomatic leptospiral infection in dogs in endemic regions. **Methods:** It was studied 64 dogs, divided into group A and B. Group A (N 16) adult dogs with confirmed diagnosis of CKD and Group B (N 48) healthy dogs control group. The study was conducted on an endemic area for leptospirosis (São Gonçalo/RJ, Brazil). Serology (MAT) and urine PCR were performed in dogs of group A and B. Dogs with CKD were diagnosed based on ultrasonography, hematological (BCC), biochemical and urinalysis tests. For each dog DRC (group A), three healthy dogs of the same age, sex and neighborhood were studied (Group B) and submitted to the same exams. Blood and urine were collected by three collections divided into monthly collections for three months. **Results:** In the serology group A, the dogs that presented exposure, were defined by titles ≥ 100 (12/16, 75%). In group B, 25% were exposed (12/48) and presented titers of 100. For both groups, the reactions observed were directed against serogroups Icterohaemorrhagiae (58.4%) and Canicola (41.6%). In group A, 12/16 animals (75%) were positive, whereas in group B, 10/48 animals (20.8%) presented a positive result, a significant difference ($p = 0.0002$, RR 3.3). As for amplicon sequencing, all five amplicons that were sequenced were confirmed as *Leptospira* sp. All animals in Group

A presented their hematology tests and ultrasound altered, confirming the diagnosis of chronic kidney disease. **Conclusion:** Our results demonstrate that asymptomatic leptospiral infection is associated with canine chronic kidney disease and that differential diagnosis is important for dogs from endemic areas presenting CKD. The early detection of shedders, besides the obvious impact on public health may also help to improve the animal health and avoid the development of CKD. **Ethics Committee approval number:** 709/2015. **Funding:** This study was supported by the Fundação de Amparo à Pesquisa do Estado do Rio de Janeiro (Faperj).

05. AVALIAÇÃO DE MÉTODOS DIAGNÓSTICOS DE INFECÇÃO POR LEPTOSPIRA SPP. EM SUÍNOS ABATIDOS PARA CONSUMO

Evaluation of diagnostic methods of infection by *Leptospira* spp. in pigs slaughtered for consumption

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Introdução: As leptospiroses são zoonoses de importância global, causadas por leptospiras patogênicas que acometem os animais domésticos, silvestres e o homem.

Objetivo: Avaliar três técnicas para diagnóstico de *Leptospira* spp. em suínos abatidos para consumo.

Metodologia: 139 suínos abatidos nas cidades de Teresina-PI e Timon-MA foram utilizados. Obteve-se o soro para realização da soroaglutinação microscópica (SAM). Fragmentos de rim foram fixados em formol tamponado para imunoistoquímica e em meio RPMI com glicerol a 10% para realizar a PCR. **Resultados:** Pela SAM, oito animais foram positivos. A técnica de

imunoistoquímica detectou marcação positiva para antígeno de *Leptospira* spp. em 60 animais. A técnica de PCR revelou sete animais positivos. O diagnóstico de leptospirose é complexo, pois, dos três métodos utilizados, apenas a IMH detectou antígeno onde a sorologia também detectou infecção, mas a recíproca não é verdadeira, e a sorologia e IMH detectaram infecção onde a PCR não detectou. A comparação dos três testes utilizados para o diagnóstico de infecção por leptospirosas revelou que a imunoistoquímica apresentou sensibilidade de 53,6% e 53,1% comparada à sorologia e PCR, respectivamente, e especificidade de 100%. **Conclusão:** Os resultados mostraram que a imunoistoquímica constitui um diagnóstico específico e sensível e pode ser usada para complementar o diagnóstico de leptospirose quando for possível a colheita de amostras de tecido.

06. BASIC MICROBIOLOGY OF LEPTOSPIRA SPP.: A TOOL FOR IMPROVING STUDIES ON LEPTOSPIROSIS

Microbiologia básica de *Leptospira* spp.: uma ferramenta para melhorar estudos sobre leptospirose

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Introduction: Leptospirosis is a zoonosis with a high incidence worldwide, caused by spirochetes belonging to the genus *Leptospira*. The maintenance of the strains and the *in vitro* growth of the microorganism remain time-consuming and difficult to be performed. Demonstrating the need to optimize laboratory culture techniques, towards improving studies in leptospirosis.

Objective: The objective of this work was to evaluate different conditions for the *in vitro* growth of *Leptospira* spp. and the corresponding virulence in an animal model for acute leptospirosis. **Methods:** *L. interrogans* strains L1-130 and RCA and *L. kirschneri* strain UFPel-61H were cultured under different conditions *in vitro* using commercial EMJH culture medium (Difco) and homemade EMJH supplemented with rabbit serum (EMJH++) using different temperatures (28 °C and 37 °C), flasks and inocula (10⁰ to 10⁴) to evaluate the growth

dynamics of the bacteria. Leptospire growth stages of bacterial growth were used to evaluate their impact on virulence in the hamster model for acute infection.

Results: All of the *in vitro* conditions were viable for leptospiral growth, with the exception of the Difco culture medium at 37 °C. The highest bacterial densities (10⁹ leptospire/ml) and the best doubling times were obtained with the Difco medium at 28 °C, especially when associated with orbital agitation. The EMJH++ medium was the most efficient with a low initial inoculum (1 leptospire), especially for *L. interrogans* strains. The *in vivo* experiments demonstrated that growth in Difco medium at 28 °C was more suitable for maintaining the stability and virulence of leptospire across the different bacterial growth stages. **Conclusion:** In conclusion, *in vitro* growth conditions influenced leptospiral virulence, demonstrating the importance of understanding the dynamics of *in vitro* growth of the microorganism. The standardization of leptospire culture techniques will improve the reproducibility of experiments involving pathogenic leptospire. **CEEA UFPel:** 4337-2015. **Funding:** Capes, CNPq.

07. BOVINE LEPTOSPIROSIS: MOST PREVALENT SEROGROUPS IN THE MUNICIPALITY OF NOVO REPARTIMENTO - PARÁ

Leptospirose bovina: sorogrupos mais predominantes no município de Novo Repartimento, Pará

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Introduction: In cattle, leptospirosis is particularly manifested by reproductive disorders, leading to losses in the production of these animals. Once the disease is present in a herd, control becomes difficult, especially by the adaptation of the bacterium to the animal species and can become a reservoir and/or maintenance host, such as serovars of the Sejroe serogroup for cattle. **Objective:**