

Identify the serogroup(s) most prevalent(s) in bovine females in the municipality of Novo Repartimento/PA, using the microscopic agglutination test (MAT).

Methods: Blood samples were collected from 208 bovine females, raised in the municipality of Novo Repartimento/Pará, during slaughter in a slaughterhouse in the Baixo Tocantins/PA region. After the blood serum was obtained, the samples were submitted to microscopic agglutination test (MAT) using 24 serovars of *Leptospira* spp. that represents 18 serogroups. **Results:** From the total of 208 samples, 97 (46.63%) were reagents, titers ranged from 100 to 3.200 for 11 of the 18 serogroup tested. Sejroe was the most prevalent, with frequency of 65.00%; followed by Shermani (10.30%); Canicola (6.18%); Tarassovi (4.12%); Grippotyphosa, Australis and Pomona (3.09%); Autumnalis (2.04%); Celledoni, Icterohaemorrhagiae and Javanica, were the least prevalent, with a frequency of 1.03% each one of them.

Conclusion: Bovine leptospirosis is present in the municipality of Novo Repartimento/PA, with Serogroup Sejroe as the most prevalent. **CEUA:** CEUA/FMVZ/USP N° 5893100816. **Funding:** CNPq (MBH fellowship), Capes (Finance code 001).

08. CERVICO-VAGINAL MUCUS PCR AS AN ALTERNATIVE TOOL TO DETECT UTERINE LEPTOSPIRAL INFECTION

RCP do muco cervico-vaginal como ferramenta alternativa para a detecção de infecção leptospiral uterina

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Introduction: Bovine leptospirosis is characterized by reproductive failure, mainly embryonic death and abortion, leading to economic losses. Nevertheless, the majority of the studies focus on the kidneys, and little is known about genital infection. **Objective:** Investigate the uterine leptospiral infection among slaughtered non-pregnant cows and if its correlation with the infection of cervico-vaginal mucus and urine from the same cows. **Methods:** Samples of cervico-vaginal mucus (CVM), uterine fragments (UF) and urine (n = 34) were

collected from non-pregnant cows at a slaughterhouse. A *LipL32* PCR was performed in all samples in order to detect positivity for pathogenic *Leptospira* in these samples. A correlation between positivity for CVM-UF and the positivity for CVM-urine was assessed by chi-squared analysis and logistic regression. **Results:** It was verified that, among the eight PCRpos CVMs, six were also positive on uterine fragment and one was urine positive. Thus, CVM-UF positivity occurred in 14.3% of the samples, while CVM-urine positivity occurred in 2.3%, a significant difference ($p < 0.05$). Logistic regression indicated that positivity in the uterus is a risk factor for positivity in CVM (OR = 3.4), while positivity in urine is not. **Conclusion:** Although not as sensitive as performing PCR on uterine fragments, CVM may be recommended as a relatively easy to perform, reliable sample that can facilitate the diagnosis of genital leptospirosis by PCR. **CEUA:** Number 863 (CEUA/UFF). **Funding:** CNPq, Faperj.

09. CHRONIC EXPERIMENTAL GENITAL LEPTOSPIROSIS WITH AUTOCHTHONOUS LEPTOSPIRA SANTAROSAI STRAINS OF SEROGROUP SEJROE

Leptospirose genital experimental crônica com estirpes autóctones de *Leptospira santarosai* do sorogrupo Sejroe

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Introduction: Leptospirosis in livestock is usually a chronic infection that leads to reproductive problems. Few studies succeeded on reproducing the chronic genital infection on experimental conditions. **Objective:** To assess the chronic experimental genital leptospirosis with *Leptospira santarosai* strains of serogroup Sejroe.

Methods: Six Santa Inês lamb ewes were inoculated with 1×10^8 leptospores, with three different strains of the serogroup Sejroe: FV237 (Group A), FV52 (Group B) and U81(Group C). Blood samples were collected daily for 28 days, and urine and vaginal fluid were collected at D0, D7, D16 and D22. Laparotomy was performed at D30, D60 and D90. On those days, all samples were collected, in addition to uterus fragment, uterine fluid and follicular aspirate samples. Study was conducted

during 90 days at Unidade de Pesquisa Experimental em Caprinos e Ovinos (UniPECO-UFF). A PCR targeting the *lipL32* gene was performed. Microscopic Agglutination Test was performed using an antigen panel represented by 28 reference strains. Sera displaying at least 50% agglutinating activity at a 1:100 dilution were considered positive. **Results:** All animals showed titers from D7 to D22, and animals inoculated with FV52 and FV237 presented higher titers (≥ 400). All of them, except for one (Group C), were positive on urinary PCR. Four of the six animals presented positivity at least once in vaginal fluid samples. One ewe (Group A) was positive in PCR of uterine wash, and all animals were negative in PCR of follicular aspirate. Regarding to the uterus fragment samples, Group A showed more positive results in PCR. In this group all animals were positive, and one ewe was positive from D30 to D90. Group B presented one positive reaction and Group C none. **Conclusion:** It was confirmed the presence of strains of serogroup Sejroe in genital tract, indicating a chronic infection in this site. **Ethics committee approval number:** 814/2016. **Funding:** This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

10. DESENVOLVIMENTO DE UM ELISA INDIRETO UTILIZANDO PEPTÍDEOS PARA DIAGNÓSTICO DA LEPTOSPIROSE BOVINA

Development of an indirect elisa using peptide for diagnosis of bovine leptospirosis

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Introdução: A leptospirose é uma importante zoonose de ocorrência mundial, que pode reduzir os indicadores da performance da reprodução em bovinos. O teste de aglutinação microscópica (MAT) é considerado padrão-ouro, entretanto, apresenta baixa sensibilidade.

Objetivo: Analisar a eficiência de três peptídeos sintéticos (preditos *in silico* e denominados peptídeo (p)

127, p128 e p129), oriundos de uma proteína hipotética da membrana externa de *L. interrogans*, aplicados em uma plataforma ELISA para a detecção de anticorpos IgG bovinos anti-*Leptospira interrogans*. **Métodos:** A padronização do teste ELISA indireto foi iniciada pela determinação das concentrações ótimas de antígenos, bloqueio, anticorpos primários e anticorpos secundários por meio da metodologia de *checkerboard*. Os três peptídeos foram testados nas concentrações ($\mu\text{g/ml}$) 10, 5, 2,5 e 1,25. O bloqueio foi realizado com albumina sérica bovina (BSA) a 5%. Os anticorpos primários (soros sanguíneos de bovinos, nunca vacinados, negativos ou positivos no MAT) foram testados nas diluições 1:400, 1:800 e 1:1600 em triplicata. O anticorpo secundário (IgG de cabra anti-IgG bovino/HRP) foi testado nas diluições 1:5000; 1:10000 e 1:20000. A revelação foi realizada com tetrametilbenzidina (TMB) e H_2O_2 30%. O bloqueio da reação foi realizado com H_2SO_4 2N. A leitura foi efetuada em 450 nm. O ponto de corte foi calculado pela média das densidades óticas (DO) dos soros negativos acrescida de três desvios padrões. **Resultados:** As condições ótimas da reação foram: peptídeos 5 $\mu\text{g/ml}$; anticorpos primários diluição 1:400 e anticorpo secundário diluição 1:20.000, combinação que melhor diferenciou, respectivamente, as amostras positivas e negativas no MAT. **Conclusão:** Os resultados sugerem que os três peptídeos sintéticos podem ser promissores para a detecção de anticorpos IgG anti-*Leptospira* em soros de bovinos naturalmente infectados. **CEUA:** 006/2017 (Uniube). **Financiamento:** Fundação de Amparo à Pesquisa de Minas Gerais (Fapemig).

11. PROJETO-PESQUISA MOLECULAR E CULTIVO DE LEPTOSPIRA SPP. NA URINA DE BOVINOS ABATIDOS EM FRIGORÍFICO: RESULTADOS PARCIAIS.

Molecular project-research and cultivation of *Leptospira* spp. in urine of bovines slaughtered in slaughterhouse: partial results.

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Introdução: A leptospirose é uma zoonose cujos agentes infecciosos são espécies patogênicas do