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## Brucella canis GroEL recombinant protein as a diagnostic antigen for canine brucellosis

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Canine brucellosis caused by *Brucella canis* is a worldwide distributed zoonosis. Infection often results in abortion, orchitis, Gepididymitis, and discospondylitis. The 2-mercaptoethanol rapid slide agglutination test (2ME-RSAT) is currently the gold standard diagnostic tool for *B. canis*. Although it has been a widely used test, it detects IgG and IgM antibodies and has low sensitivity and specificity. The antigen used in this diagnostic test commonly cross-reacts with other pathogens like *Escherichia coli* O157: H7, *Francisella tularensis, Vibrio cholerae, Salmonella N* group y *Pseudomonas maltophilia*, and its production require a level III biosafety laboratory. As a limiting factor, this test is not commercially available in our country. For this reason, it is necessary to seek additional antigen candidates for the diagnosis of canine brucellosis with a methodology of easy access for use in the veterinary clinic. Our group has demonstrated high levels of the GroEL protein in the serum of animals experimentally infected with B. canis, suggesting its role as a candidate protein for detection in diagnostic tests. For recombinant protein preparation, specific primers were designed to amplify the GroEL gene and later cloning into the pQE60 plasmid. Further protein expression requires the *E. coli* M15 strain that harbors the plasmid pREP4 (*lacI*<sup>q</sup> repressor protein). Protein semiquantification by Western Blot will compare recombinant GroEL with native protein. Purification by FPLC (fast protein liquid chromatography) and assessment as the capture antigen by indirect ELISA will be performed with serum from experimentally infected dogs.

## **Recent Publications:**

- 1. Purvis T J, Krouse D, Miller D, Livengood J, Thirumalapura N R and Tewari D (2017) Detection of *Brucella canis* infection in dogs by blood culture and bacterial identification using matrix assisted laser desorption ionization time of flight mass spectrometry. Journal of Veterinary Diagnostic Investigation 29(4):586-588.
- 2. Gomez G, Adams L G, Rice Ficht A and Ficht T A (2013) Host-Brucella interactions and the *Brucella genome* as tools for subunit antigen discovery and immunization against brucellosis. Frontiers in Cellular and Infection Microbiology 3:17.
- 3. Hollett R B (2006) Canine brucellosis: outbreaks and compliance. Theriogenology 66(3):575-587.
- 4. Lucero N E, Escobar G I, Ayala S M and Jacob N (2005) Diagnosis of human brucellosis caused by *Brucella canis*. Journal of Medical Microbiology 54(5):457-461.

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