TITLE: Bioinformatics identification of antigenic proteins of *Mycoplasma capricolum* subspecies *caprineumoniae*: providing background data for reverse vaccinology

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ABSTRACT:
Contagious Caprine Pleuropneumonia (CCPP), a disease caused by *Mycoplasma capricolum* subspecies *caprineumoniae* (Mccp), affects goats and some wild ruminant species of all ages and both genders. CCPP is characterized by a unilateral sero-fibrinous pleuropneumonia with pleural fluid accumulation that leads to fever, tachypnea, cough, and nasal mucopurulent discharge. This respiratory disease is extremely contagious and frequently fatal for animals from naive herds, where morbidity and mortality rates can be as high as 70-100%. Therefore, CCPP causes severe economic losses in goat production worldwide and is a notifiable disease to the World Organisation for Animal Health (OIE). Despite its great economic impact for producers, CCPP is a highly neglected disease of livestock and little is known about the mechanisms by which Mccp causes disease, in particular its antigens. Thus, the objective of the present study was to identify the main antigenic proteins of Mccp using bioinformatics algorithms. For this purpose, all 703 predicted proteins of Mccp reference strain F38 (NZ_LN515398.1) were analyzed by bioinformatic algorithms regarding their subcellular localization prediction (PSORTb). Proteins predicted as being located at the cytoplasmic membrane or extracellularly were analyzed using the IEDB (Immune Epitope Database) Analysis Resource for the prediction of beta turn, flexibility, antigenicity, hydrophilicity, accessibility and linear epitopes regions. Discontinuous epitopes were predicted using CBTOPE (Prediction of Conformational B-cell epitopes). Thresholds of 1.0 (IEDB) and -0.2 (CBTOPE) were used to avoid false positives. A total of 213/703 (30%) proteins were predicted as membrane-associated. Following the application of antigen prediction algorithms, 136 sequences were identified with linear B-cell epitopes and 180 proteins with conformational epitopes. This difference between the amount of linear and conformational epitopes is expected since most of the B-cell epitopes (~ 90%) are conformational. According to genome annotation, most of these proteins 128/180 (71%) have unknown function or are proteins involved in the transport of molecules. These results support the *in silico* identification and characterization of proteins with possible antigenic epitopes that can aid in the development of new subunit vaccines and immunodiagnostic tests of CCPP, as current control and prevention strategies are still ineffective.

Keywords: Contagious Caprine Pleuropneumonia, *Mycoplasma capricolum* subspecies *caprineumoniae*, goats, antigenic proteins.