

LETTER TO THE EDITOR

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Immunogenicity Assessment of *Brucella melitensis* HSP and TF Proteins by Immunized Rabbit Serum

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Bacteria of the genus *Brucella* are facultative intracellular pathogens which have developed the capacity to survive and multiply in professional and nonprofessional phagocytes. Due to drawbacks of live attenuated vaccines, much attention has been focused on screening *Brucella*-protective antigens as subunit vaccine candidates. In order to screen immunogenic candidate antigens for the development of a *Brucella* subunit vaccine, we cloned, expressed and purified Heat Shock Protein (HSP) and Trigger Factor (TF) from *Brucella melitensis*. These recombinant antigens were then evaluated by serum from a *B. melitensis*-vaccinated rabbit using ELISA and Western blot. Our results showed that the immunized rabbit serum reacted with recombinant HSP and TF in ELISA and Western blot. These results may suggest that *B. melitensis* rTF and rHSP may serve as candidate subunit vaccine components for protection against the infection.

Brucella spp. are Gram-negative and facultative intracellular bacteria which cause brucellosis, a

worldwide zoonotic disease causing abortion in domestic animals and Malta fever in humans.¹ *Brucella melitensis* can invade macrophage-monocyte lineage cells and replicate within the phagosomes by inhibiting phagosome-lysosome fusion.² In intracellular environment of macrophages, bacteria are subjected to hard conditions³ thus, the identification of bacterial proteins essential for intracellular survival in this niche is critical in understanding the protective mechanisms and pathogenesis of the disease. Expression of heat shock proteins in this situation, causes bacteria to adapt not only to thermal but also to various other environmental stresses, and the accumulation of heat-shock proteins (HSPs) is thought to preserve bacterial cellular functions.⁴ The heat shock protein is a small HSP (sHSP) (Accession No. 1197813). sHSPs are molecular chaperones that suppress protein aggregation and protect against cell stress.⁵ Trigger Factor (TF) protein (Accession No. 1196780) is an ATP independent chaperone⁶ and has also been reported to act as a protective antigen against *B. melitensis* infection.⁷ In view of the immunological importance of HSP and TF, we decided to clone, express in *E. coli* and purify the HSP and TF proteins from *B. melitensis* and study the antibody response to this protein in sera from *B. melitensis*-vaccinated rabbit by ELISA and

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