Serological diagnostic potential of recombinant outer membrane proteins (rOMPs) from Brucella melitensis in mouse model using indirect enzyme-linked immunosorbent assay

ABSTRACT

Background: Brucella melitensis is the most important pathogenic species of Brucella spp. which affects goats and sheep and causes caprine and ovine brucellosis, respectively. Serological tests for diagnosis of brucellosis such as Rose Bengal plate test (RBPT) and enzyme-linked immunosorbent assay (ELISA) usually utilize smooth lipopolysaccharides (S-LPS) as a diagnostic antigen which could give false positive serological reactions. Outer membrane proteins (OMP) of B. melitensis have been used as alternative diagnostic antigens rather than S-LPS for differential serological diagnosis of brucellosis, mainly in ELISA with single recombinant OMP (rOMP) as a diagnostic antigen. Nevertheless, the use of single format mainly showed lack of sensitivity against the desired rOMP. Therefore, this study aimed to determine whether a newly developed rOMPs indirect ELISA (rOMPs I-ELISA), based on combination of rOMP25, rOMP28 and rOMP31of B. melitensis, has a potential benefit for use in the serodiagnosis of brucellosis. Methods: In this study, omp25, omp28 and omp31 of B. melitensis were cloned and expressed using prokaryotic pET-32 Ek/LIC system and their respective rOMPs were combined as one coating antigen to develop rOMPs I-ELISA. Three groups of BALB/c mice were used to elicit antibody response. Group 1, infected with B. melitensis strain 0331 field strain; group 2, injected with B. melitensis Rev.1 vaccine strain and group 3, infected with Yersinia enterocolitica O:9. Antibody responses in three groups of mice were investigated using Rose Bengal plate test (RBPT) and rOMPs I-ELISA. Results: The production of rOMP25, rOMP28 and rOMP31 of B. melitensis were achieved and Western immunoblotting analysis demonstrated their reactivity. The RBPT was unable to differentiate the vaccinated mice (group 2) and mice infected with Y. enterocolitica O:9 (group 3) and categorized them wrongly as positive for brucellosis. In contrast, the rOMPs I-ELISA was able to differentiate the mice infected with B. melitensis strain 0331 (group 1) from both of group 2 and group 3, and recorded 100% sensitivity and 100% specificity. Conclusions: The results of this study suggested that rOMPs of B. melitensis has potential diagnostic ability to differentiate the FPSR in serological diagnosis of brucellosis.

Keyword: Brucella melitensis; rOMPs; FPSR; Mice; ELISA; Recombinant protein