10th World Congress on

Veterinary & Animal Science

May 18-19, 2018 Osaka, Japan

Cloning and expression of Eimeria necatrix microneme 5 gene in E. coli

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Micronemes are the smallest secretory organelles of the invasive stages of apicomplexan parasites and contain proteins that are important for parasite motility and host cell invasion. Molecular identification and immunogenic potency of these proteins can be used for developing of recombinant vaccine. In this study, after isolation of *Eimeria necatrix* from Khuzestan province, a pair of primers was designed based on the published nucleotide sequence of micronem 5 gene of *Eimeria necatrix* LZ strain. Partial sequence of a cDNA encoding 758 bp fragment of microneme 5 protein (EnMIC5) was amplified by seminested RT-PCR, cloned and expressed in a maltose binding protein (MBP) containing expression vector (pMAL-c2x) in *E. coli* TG1 strain. The amplified fragment contains an open reading frame of 252 amino acids with high degree of conservation with adhesive plasma pre-kallikrein and seven hydrophilic motifs. The results of SDS-PAGE revealed that the fusion protein with molecular weight about 70 kDa was over-expressed after induction of IPTG. Western-blot results demonstrated that the expressed recombinant protein was reacted with sera of infected chicks with *Eimeria necatrix*, suggesting that this protein should have good immunogenicity and can be used for further studies.

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