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Veterinarni Medicina

Purification of Escherichia coli-expressed HIS-tagged Maedi-Visna p25 core antigen by Ni2+-chelate affinity chromatography

Molinková D.:

Veterinarni Medicina, 46 (2001): 50-54

[fulltext]

In this study, recombinant histidine tagged p25 capsid protein of Maedi-Visna virus was developed. Part of the viral genome coding p25 protein was positioned downstream and in frame with a metal binding domain in pRSET-B vector. Recombinant protein was expressed in E. coli cells and soluble fraction of the protein was subsequently purified by Ni2+-chelate affinity chromatography. Purified protein was then used as antigen in an indirect ELISA test. One hundred fifty ovine serum samples were screened for antibodies to p25 protein of the virus. Immunoblot with whole virus antigen was used as a gold standard. The total number of positive

results in the ELISA was 38 (25.33%). Immunoblot failed to confirm a positive result in 2 (1.33%) of them and these results were therefore considered to be false positive. The number of true positive results in the ELISA was thus 36 (24%). All immunoblot positive samples were also positive by ELISA test. In conclusion, recombinant His-tagged capsid protein showed very high sensitivity and specificity in detecting antibodies to Maedi-Visna virus.

Keywords:

lentivirus; recombinant antigen; ELISA; immunoblo

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