Production and characterization of egg yolk antibody (IgY) against recombinant VP8-S2 antigen


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Abstract

Bovine Rotavirus and Bovine Coronavirus are the most important causes of diarrhea in newborn calves and in some other species such as pigs and sheep. VP8 subunit of rotavirus is the major determinant of the viral infectivity and neutralization. Spike glycoprotein of coronavirus is responsible for induction of neutralizing antibody response. Studies showed that immunoglobulin of egg yolk (IgY) from immunized hens has been identified to be a convenient source for specific antibodies for using in immunotherapy and immunodiagnostic to limit the infections.

In this study, chimeric VP8-S2 gene was designed using by computational techniques. The chimeric VP8-S2 gene was cloned and sub-cloned into pGH and pET32a (+) vectors. Then, recombinant pET32a-VP8-S2 vector was transferred into E. coli BL21 CodonPlus (DE3). The expressed protein was purified by Ni-NTA chromatography column. Hens were immunized with the purified VP8-S2 protein three times. IgY was purified from egg yolks using polyethylene glycol precipitation method. Activity and specificity of anti-VP8-S2 IgY were detected by dot-blotting, Western-blotting and indirect ELISA.

We obtained anti-VP8-S2 IgY by immunizing hens with the recombinant VP8-S2 protein. The anti-VP8-S2 IgY was showed to bind specifically to the chimeric VP8-S2 protein by dot-blotting, Western-blotting analyses and indirect ELISA. The result of this study indicated that such construction can be useful to investigate as candidates for development of detection methods for simultaneous diagnosis of both infections. Specific IgY against the recombinant VP8-S2 could be recommended as a candidate for passive immunization against bovine rotavirus and bovine coronavirus.

Key words: bovine rotavirus, bovine coronavirus, immunization, recombinant protein, IgY

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