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Evaluation of rabbit polyclonal antibody against influenza virus conserved proteins (M2 and HA2)

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Background: Influenza virus is one of the most important factors for sporadic pandemics that usually cause higher morbidity and mortality in the world. The envelope of viruses contains two immunogenic surface glycoproteins, hemagglutinin (HA) and neuraminidase (NA), since these antigenic proteins are variable in antigenic sites so vaccine and serological researches have been focused on influenza conserved antigenic proteins. One of these proteins is hemagglutinin stalk domain (HA2) that is highly conserved in all influenza A virus strains and it plays a major role in the fusion of the virus with the endosomal membrane in host cells during the course of viral infection. Another one of conserved antigens is matrix protein 2 (M2) that is a proton selective ion channel. The immune system is able to produces antibody against M2 and HA2 antigenic region. These antibodies can be used to vaccines and serological methods development.

Methods: In the present study, recombinant HA2 and M2 protein with Freund's adjuvant were injected to rabbits by intramuscularly route. Immunizations were continued for about 9 weeks and function of rabbit antiserums were evaluated using ELISA, RID and western blotting as serological assays. Finally, the IgGs were purified from the antiserum by DEAE-cellulose and antibody functions were evaluated using ELISA.

Results: The ELISA results showed that two antibodies (anti-HA2 and anti M2) titer were raised approximately after first injection and the purified IgGs titer were 1:256000 for anti-HA2 IgG and (1:128000) for anti-M2 IgG using this method. These antibodies had shown positive results in RID assay (sedimentary line) and western blotting.

Conclusion: These findings showed that recombinant HA2 and M2 could stimulate humoral immune response in rabbits and these polyclonal antibodies are proper for serological methods.

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