Full Length Research Paper

Approaches toward the development of DNA vaccine for influenza virus

T. A. F. Bouback and N. A. Redwan*

Department of Biological Sciences, Faculty of Science, King Abdulaziz University, Jeddah, 21589, P.O. Box 80103, Saudi Arabia.

The main goals of this investigation were to prepare a viral DNA vaccine to help stimulate the immune system of poultry and to increase the efficiency of this vaccine. To accomplish this work, a strain of H5N1 circulating in Egypt was confirmed using rapid diagnostic methods and also, reverse transcriptase polymerase chain reaction (RT-PCR) to hemagglutinin and neuraminidase genes. The virus was propagated in MDCK cell line and the viral genes were extracted and reverse transcribed individually. Individual genes were cloned in gene expression vector (PHW2000) and were used as DNA vaccine. The level of maternal antibodies was determined by ELISA to appoint the right time to give the vaccine. The chicks were divided into eight groups and each group was vaccinated by the couple of DNA NP with one of the other genes. The efficiency of coupled DNA vaccine was determined by neutralization assay and compared with the inactivated vaccine. The results showed that the vaccine that had NP with NS had adequate protection for poultry.

Key words: H5N1, virus, vaccine, poultry, DNA.

INTRODUCTION

Immunity to conserved viral components can provide broad protection against different influenza A strains and subtypes. Research immunity has been long studied in animals (Lamb and Krug, 2001) and there is evidence that it may exist in humans (Fouchier et al., 2005; Horimoto and Kawaoka, 2005). Vaccines based on such cross-protection would not require knowing the identity of the strains that would circulate during the coming season, information which is never certain and could avoid hurried manufacturing in response to outbreaks. Inactivated vaccines given intranasal can induce cross-protection (Johnson and Mueller, 2002; Reid et al., 2004), but DNA prime-viral boost regimens offer the advantage of endogenously expressed antigens that may induce a broader range of immune effectors.

DNA vaccination to conserved influenza nucleoprotien NP or NP and matrix proteins (M), has been studied in animal models and protection against both matched and mismatched challenge viruses has been demonstrated (Gamblin et al., 2004; Horimoto and Kawaoka, 2005), albeit with relatively low dose challenges. H5N1 viruses from the 1997 human outbreak in Hong Kong presented a demanding test because of their virulence and rapid kinetics of infection (Matrosovich et al., 2000). Limited protection was achieved by DNA vaccination with NP + M against some lethal H5N1 challenges (Subbarao et al., 1998). To improve the efficacy of this approach, Epstein et al. (2005) explored the ability of a recombinant adenovirus (rADV) vector to enhance potency of vaccination to NP, an antigen with >90% protein sequence homology among influenza A isolates (Buxton et al., 2000) and containing dominant target epitopes (Xu et al., 1999). The ability of the regimen to protect against high virus challenge doses and against challenge with mismatched highly pathogenic H5N1 strains was tested. The DNA prime-rADV boost regimen is more effective than vaccination with NP construct, conferring protection against doses of challenge virus that were lethal to mice vaccinated only with DNA or rADV. In this study, investigation was carried out to find effective system for protection against lethal doses of H5N1 by preparing DNA

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*Corresponding author. E-mail: nezar_redhwan@yahoo.com or Nezarredhwan@yahoo.com.