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M2-based influenza A vaccine: Immunogenicity and protective efficacy of a homologous prime-boost regimen

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Introduction: Influenza A viruses are globally important respiratory pathogens which cause a high degree of morbidity and mortality during annual epidemics. The M2 protein is conserved among all Influenza A viruses and has been considered for developing a universal influenza vaccine which could provide cross protection against different virus strains. Here we report protective efficacy of a DNA vaccine consisting Influenza virus M2 gene boosted by prokaryotic recombinant M2 protein.

Methods: The entire open reading frame of M2 was cloned into pcDNA as well as pET28 vectors and confirmed. The definite eukaryotic expression vector encoding M2 was prepared using Endofree plasmid Mega Kit (Qiagen). Recombinant M2 protein was expressed in *Escherichia coli* purified using Ni-TED columns under denaturing conditions, refolded and desalted by dialysis. Six-week-old BALB/c mice were immunized interdermally with one dose M2 DNA and boosted twice with M2 protein supplemented with Alum. Specific anti-M2 antibodies including IgG sub classes were measured using ELISA method. T-cell immune response was assessed by measuring of different cytokines and MTT. Finally, mice were challenged with one lethal dose (LD90) of PR8 virus.

Results & Conclusion: The results showed that the administration of DNA vaccine followed by purified M2 protein with Alum adjuvant as booster induced high level of anti-M2 antibodies. Also, IgG subclass evaluation showed that both TH1 and TH2 cells have been involved which was in accordance with T cell immune responses. Moreover, these mice exhibited miler illness and fewer deaths upon lethal H1N1 (PR8) challenge. In Conclusion, immunization with a DNA vaccine encoding M2 protein followed by boosting with homologous recombinant protein could be considered as a suitable strategy for prevention influenza disease.

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