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Effective protection by high efficiency bicistronic DNA vaccine against anthrax expressing protective antigen and catalytically inactivated lethal factor

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Limitations of currently available anthrax spore vaccine necessitate the development of an improved vaccine for animals. In the present study lethal factor gene of *Bacillus anthracis* was made catalytically inactive by primer based site directed mutagenesis. To explore whether immunization with plasmid encoding this mutated lethal factor (mLF) and protective antigen (PA) can provide protection against anthrax, a bicistronic DNA vaccine encoding PA and mLF was then made along with mono-cistronic constructs encoding PA/ mLF. The ability of the constructs to express the encoded genes was verified by transfection in MDBK cells followed by indirect immunofluorescence analysis. To investigate the immunogenic potential of the made constructs immunization trials were conducted in mice. After primary immunization with these DNA vaccines the mice were boosted with DNA vaccines, recombinant proteins or formalin inactivated spores (FIS) on 14th and 28th days post vaccination. Subsequently, indirect ELISA, toxin neutralization assays (TNA) and monitoring of cytokines (IL-4, IL-2 and IFN- γ) were done to monitor the immune response. The direct challenge test of immunized mice was done using 1000 LD50 of virulent *B. anthracis* IVRI strain. The results showed that the heterologous prime boost regimen involving priming with bicistronic DNA construct encoding PA and mLF or mono-cistronic DNA construct encoding PA and boosting with recombinant proteins can provide better protection (66.66%) compared to other groups. At the same time immunization with bicistronic DNA construct encoding PA and mLF and boosting with recombinant proteins could provide higher antibody titer, toxin neutralization titer and Th1 and Th2 response ($p < 0.0001$) compared to all other groups illustrating that DNA vaccine encoding PA and mLF conferred a broader spectrum of immune reaction than PA alone. The increase in serum concentration of IL-2, IL-4 and IFN- γ indicated that both humoral and cell mediated immune response were elicited by DNA vaccination. Thus, the results of the present study indicated the feasibility of DNA prime protein boost based immunization strategy based on PA and mLF being developed into nontoxic, effective and stable anti-anthrax vaccine.

Biography

Bincy Joseph has completed her Graduation in Veterinary Sciences (BVSc & AH) from Kerala Agricultural University with KAU merit scholarship. She has completed Post graduation in Veterinary Bacteriology from Indian Veterinary Research Institute with ICAR Junior Research Fellowship and completed Doctorate in Veterinary Bacteriology from Indian veterinary research institute with Senior Research Fellowship. She has published 8 research articles in various reputed journals and participated in more than 10 national and international conferences and presented research papers.

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