EVALUATION OF BACILLE CALMETTE GUÉRIN, MONTANIDE INCOMPLETE SEPPIC AND ALUMINIUM HYDROXIDE AS ADJUVANTS FOR LEISHMANIA VACCINE IN BALB/c MICE

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Leishmaniasis are parasitic diseases caused by protozoan flagellates of the genus \textit{Leishmania}. These parasites infect numerous mammalian species, including humans, and are transmitted through the infective bite of an insect vector, the female \textit{Phlebotomus} sandfly. Leishmaniasis is currently endemic in 88 countries, and is a threat to 350 million people with a worldwide prevalence of 14 million clinical cases and 12 million new cases each year. Drugs for leishmaniasis are generally toxic and their cost is prohibitive. Vector control measures are poorly implemented and inaccessible to many people in developing countries. In leishmaniasis, protection requires leishmanial-specific CD4\textsuperscript{+} T helper-I (Th1) cells. Effective vaccination against leishmaniasis would be cheaper and accessible. Unfortunately, so far there is no vaccine against leishmaniasis for routine use. Immunity to cutaneous leishmaniasis, a chronic skin-disfiguring lesion affecting millions, has been historically achieved by inoculation with live virulent preparations of the parasite. Killed antigens that could be safer as vaccines generally require an adjuvant for the induction of strong Th1 response in murine models. The objective of this study was to assess and compare the immune responses and efficacy of a vaccine containing Bacille calmette guerin (BCG), aluminium hydroxide (alum) and Montanide ISA 720 (MISA 720) as adjuvants combined with killed \textit{Leishmania} (\textit{L.}) \textit{major} vaccine (KLM) in BALB/c mice. Sixty mice were immunized three times at weeks 0, 4 and 6 and two weeks later, either sacrificed for comparative immunogenicity analysis or challenged with virulent \textit{L. major} for efficacy monitoring. Peripheral blood mononuclear cells (PBMC) were stimulated in culture with KLM antigens or concanavalin A and their proliferation quantified. Sera immunoglobulin
gamma (IgG) and in vitro interferon gamma (IFN-γ) production were measured by enzyme linked immunosorbent assay (ELISA) using Leishmania antigen and cross-reactive monoclonal antibody. Lesion development in infected mice footpads were monitored for 8 weeks and parasite loads determined there after. Higher IgG responses were observed in the BCG-KLM and alum-KLM vaccinated mice as compared to the MISAKLM mice. Antigen-specific lymphoproliferative in vitro response showed significantly higher (p<0.01) responses in the MISAKLM as compared to both alum-KLM and BCG-KLM groups. The BCG-KLM group recorded significantly higher IFN-γ production (p<0.001) as compared to both alum-KLM and MISAKLM vaccinated groups. Efficacy evaluations showed significantly reduced lesion sizes in the MISAKLM than in both alum-KLM and BCG-KLM vaccinated groups. There were significantly reduced (p<0.001) parasite loads in both the MISAKLM and BCG-KLM groups as compared to the alum-KLM vaccinated animal group. It was however, noted that, BCG vaccination caused inflammatory reaction that led to highest lesion sizes observed in the mice vaccinated with BCG as compared to both the alum and montanide ISA 720 vaccinated mice groups. The results from this study conclude that Montanide ISA 720 adjuvant is safe and could be used to induce protection against cutaneous leishmaniasis caused by L. major in susceptible BALB/c mice. The study has contributed valuable data to be used in further studies and the development of a potential adjuvant for human Leishmania vaccine.