Quil-A - Chitosan : A novel mucosal adjuvant

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The potential to generate both a local and systemic immune response makes the mucosal immune system an attractive site for immunization. However, mucosal administration of protein and peptide antigens generally results in a poor immune response. Successful mucosal vaccination is therefore largely dependent on the development of effective mucosal adjuvants. In recent years, many adjuvants and vaccine delivery systems have shown an ability to enhance immune responses to mucosally administered antigens. These include bacterially-derived products such as monophosphoryl lipid A, toxins and immunostimulatory DNA sequences.

Cholera toxin consists of a pentameric B oligomer that binds to GM-1 receptors and an enzymatically active A subunit that is responsible for the toxicity of this agent. The toxicity of the CT holotoxin limits its usefulness as an adjuvant; therefore the cholera toxin B-subunit (CTB) which consists only of the non-toxic B-subunit of the cholera enterotoxin is more widely used in candidate mucosal vaccines (Goto et al 2000). Fractions prepared from *Quillaja saponaria* (Quil-A) can also be used as adjuvants. Quillaja saponins are known to increase the effectiveness of both injected and oral vaccines (Sjolander and Cox 1998). In previous studies we have shown that chitosan is able to enhance the effects of other adjuvants when administered intranasally (Bramwell et al 1999). In this study we have examined the effect of vaginal and rectal administration of diphtheria toxoid in the presence of the experimental combination of chitosan plus Quil-A or CTB as a known mucosal adjuvant on the systemic and mucosal immune response.

Three groups of five BALB/c mice were immunized *via* either the rectal or vaginal route (six groups in total) with 20µg of diphtheria toxoid in either 30µl of sterile PBS, 0.2% w/v chitosan glutamate plus 15µg of Quil-A or 10µg of CTB on day 1, day 7 and day 21. Blood samples, vaginal washes and fecal mater were collected on day 14 and 27 and assayed for anti-diphtheria specific antibody by indirect ELISA.

The results showed that CTB produced the highest serum IgG response compared to the group of mice which received free toxoid or the group of mice which received free toxoid with chitosan glutamate plus Quil-A. The group of mice which received diphtheria toxoid with chitosan plus Quil-A also showed enhanced systemic IgG responses compared to the group of mice which received free toxoid. This trend was the same for either rectal or vaginal administration. We are in the process of evaluating the mucosal and cellular responses of these groups, but the preliminary results presented in this study clearly show that chitosan plus Quil-A can act as an adjuvant for rectally and vaginally delivered antigens.

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