

Selection of Polysaccharide Length, Conjugation Procedure and Carrier Protein for Vi Polysaccharide Conjugate Obtaining

J.P. Soubal, Y. Soroa, S. Fernández, S. Darielys, R. Garrido, J. Pedroso, D. García, Y. Valdés, V. Vérez V.

Finlay Institute of Vaccines, Havana, Cuba

Background: There is an estimate of 11.9 million cases of typhoid fever, with 190,000 deaths every year. The development of conjugate vaccines provides an opportunity to better combat this serious disease. This work evaluated the influence of some parameters in physicochemical and immunological characteristics of Vi polysaccharide conjugates and characterized the immune response of a selected conjugate.

Methods: The parameters evaluated were: polysaccharide length (native or fragmented), linker (no linker, hydrazine or ADH) and carrier protein (DT or TT). The conjugates were analyzed by colorimetric methods, SEC-HPLC and H¹-NMR. The immune response was evaluated in BALB/c mice.

Results: All conjugates were suitable physicochemical characteristics, with higher K_D for conjugate obtained from fragmented polysaccharide and for conjugate without linker. All conjugates were more immunogenic than the unconjugated polysaccharide. The avidity of IgG antibodies generated by the fragmented polysaccharide conjugate was greater than the rest of the conjugates immunized. For native Vi ADH conjugated to DT was observed a mixed Th1/Th2 response pattern of isotypes and cytokines. It was shown that the conjugate induces a memory response and that the presence of free polysaccharide in the same amount as the conjugate does not affect the IgG response.

Conclusions: In summary, the study determined the parameters which influence physicochemical and immunological characteristics and demonstrated a thymus-dependent response against the conjugate.