Liposomes containing chemically synthesized FP20 and QS-21_EA to mimic the AS-01b as an effective vaccine adjuvant system

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Abstract: Adjuvants play an important role in the vaccines, as they increase the immune response at a lesser antigen level and as well increases the stability of the antigens involved. MPLA and QS-21 has been widely used as adjuvant in several commercially available vaccines. The immunostimulating action of MPLA is due to its activity as agonist of the Toll-like Receptor 4 (TLR4) whereas, QS-21, a molecule of natural origin, whose mechanism of action is still unclear. The combination of MPLA and QS-21 in the Adjuvant System 01b (AS01b) potentiates the activity of adjuvants and partially decreases the side effects.

In this study, we lipo-formulated two simplified vaccine adjuvants FP-20 and QS-21_EA with an aim to mimic the AS01b. FP20 is a TLR4 agonist and a glycolipid, which is chemically simplified MPLA, whereas QS-21_EA is a simpler QS-21 based saponin variant. Liposomal formulations of the adjuvants were prepared by using DOPC and were extruded using different filters (0.1, 0.4 and 0.8 micron). Two different approaches of adjuvant addition was tested as both the adjuvants are not completely soluble in PBS and have similar behavior as lipids. The liposomal formulations have been standardized to increase the stability and characterized by cryoTEM and DLS. The internalization of the adjuvants were observed on the liposomes as well as inside the liposomes. The TLR4 agonist has been observed integrating into the liposomal bilayer as well as forming micellar structures inside and outside the liposomes due to its lipid nature. Similar result of liposomal encapsulation and internalization has been observed in the QS-21 Echinocystic variant due to its polar and non-polar regions in its structure. The liposomal formulations were purified to separate the liposomes from the free adjuvants by using size exclusion chromatography. As the TLR4 agonist (FP-20) used are forming micellar structures, further experiments can be carried out to improve the use of the TLR4 agonist as an adjuvant and as a delivery system. This work can pave the way for larger applications in the vaccine development.