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Electrospraying Chitosan Particles for Oral Vaccine Delivery

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PURPOSE: To utilize electrospray to prepare chitosan particles in the micrometer size range encapsulating the antigen ovalbumin and the adjuvant, quil-A. Furthermore, the properties of the microparticles were investigated for its potential as an oral vaccine delivery system.

METHODS: Three different types of chitosan were used; 70, 85 and 90, and they were depending on the degree of acetylation of chitosan. Chitosan was dissolved in ethanol and aqueous acetic acid solution (1:1) as the solvent, and the model antigen, ovalbumin was added in either 30, 40 or 50 w/w% in relation to the chitosan concentration, and further 10w/w% quil-A (in relation to the ovalbumin content) was added as the adjuvant. The solutions were electrosprayed either with the distance between the emitter tip and the plate being 10 cm and a flow of 10 $\mu\text{L}/\text{min}$ or with a distance of 15 cm and a flow of 16 $\mu\text{L}/\text{min}$. The morphology of the particles was checked using scanning electron microscopy (SEM). Moreover, the size and zeta potential were measured using dynamic light scattering (DLS), and the encapsulation efficiency and the release of ovalbumin was studied in buffer at pH 6.8. Furthermore, the *in vitro* activity on dendritic cells was studied during a period of 18 h and by using an ELISA kit. The production of IL-6 from the cells was measured from all the produced chitosan formulations.

RESULTS: It was possible to electrospray all the various chitosan's and to encapsulate approximately 100 % of the added ovalbumin. A scanning electron microscope image of an example of some of the particles can be seen in Figure 1. When tested on the dendritic cells, the chitosan with acetylation grade of 70 and with 50 % ovalbumin was found to have the largest production of IL-6 and therefore, the greatest dendritic cell activity compared to the other chitosan formulations. This resulted in that the characterization was continued with this formulation.

When the distance between the emitter tip and the plate was 10 cm and a flow of 10 $\mu\text{L}/\text{min}$, the average size of the chitosan particles with ovalbumin and quil-A was found to be $4.7\pm 0.8 \mu\text{m}$ and the zeta potential was measured to be $45.0\pm 2.4 \text{ mV}$. Approximately 20 % ovalbumin was released from the particles after 2 min, and after 8 h of release in buffer at pH 6.8, $39.6\pm 6.1\%$ of the added ovalbumin was released from the chitosan formulation (Figure 2).

CONCLUSION: The developed chitosan particles with ovalbumin and quil-A show potential to be used for oral vaccine delivery and therefore, the particles will be filled into polymeric drug delivery devices called microcontainers. The microcontainers show the features of being able to protect the particles through the stomach and can provide release in the intestine, and have therefore shown promise as an oral drug delivery system.

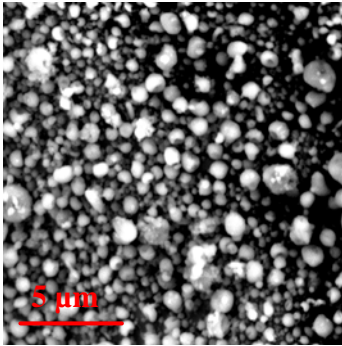


Figure 1: A SEM image of the chitosan microparticles containing ovalbumin and quil-A produced by electrospray

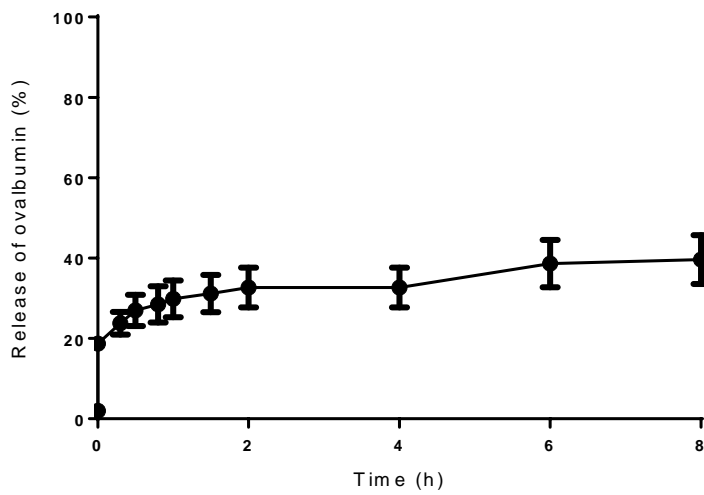


Figure 2: Release of ovalbumin from the chitosan particles in buffer at pH 6.8