**Whey protein nanoparticles produced by electrospraying**

**T Moschakis1, AT Chatzitaki2, G Charitou1, A Lazaridou1, DG Fatouros2,**

*1Department of Food Science and Technology, School of Agriculture, Aristotle University of Thessaloniki, GR-54124 Thessaloniki,* *Greece*

*2Department of Pharmaceutical Technology, School of Pharmacy, Aristotle University of Thessaloniki, GR-54124 Thessaloniki, Greece*

\*Corresponding Author ([tmoschak@agro.auth.gr](mailto:tmoschak@agro.auth.gr))

Electrospray is a process able to produce nanoparticles in different sizes and shapes by controlling the operation conditions1. The goal of the present study was to facilitate the development of nanoparticles by modifying the applied voltage and flow rate during electrospraying and to investigate the impact of different ethanol content on whey protein (WPI) denaturation and consequently nanoparticles morphology.

Aqueous/ethanol solution of whey protein (1.5% w/w) was electrosprayed by controlling the flow rate (0.25-1.0 mL/h), using high-voltage power supply (10-20 kV) and an 18-gauge stainless-steel needle, whereas the operation was set at room temperature and 45% humidity. The distance between the needle tip and the aluminum foil, used as collector, was set to 15 cm. Aqueous WPI solutions without and with ethanol at 10, 30, 50% and 70% w/w, followed by 1 hour heating (70°C), were electrosprayed under the optimized process conditions, as evaluated previously. The formed particles were characterized by means of scanning electron microscopy (SEM), light scattering, and attenuated total reflectance FTIR (ATR-FTIR) spectroscopy.

SEM micrographs confirmed the creation of spherical nanoparticles of *ca*. 400 nm at the lower flow rate (0.25 mL/h), independently of the applied voltage. Increasing the flow rate resulted in size and morphological variations, exhibiting both linear rod-like structures and spherical particles with sizes ranging from nano- to micro-scale range. At a slower flow rate and higher voltage applied uniform nanoparticles were formed at an adequate yield. Considering the cost and time effectiveness of electrospraying with 0.5 mL/h flow rate, in comparison to 0.25 mL/h, the process was opted for 0.5 mL/h and 20 kV for further investigation. Under these conditions, the optimized nanoparticles for uniformity in size and production yield had a size of 524 ± 149 nm. SEM micrographs revealed the crucial impact of ethanol content on protein denaturation since different alcohol concentrations influenced the morphology of the generated particles. The absence of alcohol resulted in aggregation, whereas the ethanol content at concentrations >30% w/w was sufficient to form protein nanoparticles. Ethanol at 50% with and without heat treatment formed spherical nanoparticles with smaller size compared to other conditions. The results suggest that the electrospray method can successfully produce nanoparticles of whey proteins able to be incorporated in food products.

*References:*

1 Coelho, S.C., Estevinho, B.N., Rocha, F. (2021). Encapsulation in food industry with emerging electrohydrodynamic techniques: Electrospinning and electrospraying – A review. Food Chemistry, 339, 127850.