

Supporting Information

Manual versus Microfluidic-assisted Nanoparticle

Manufacture: Impact of Silk Fibroin Stock on Nanoparticle

Characteristics

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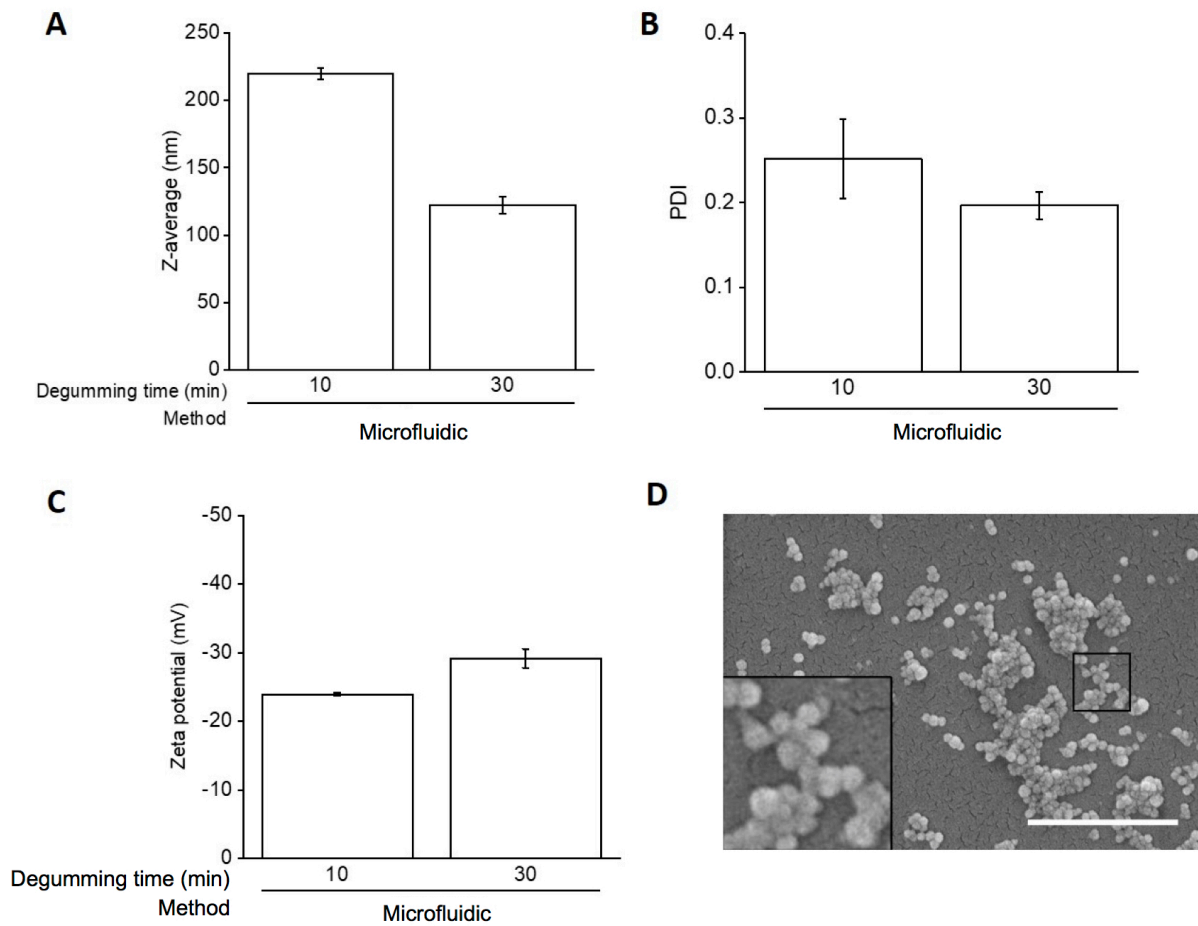


Figure S1. Characterisation of nanoparticles manufactured by the microfluidic method using 10 and 30 minutes degummed silk. Nanoparticles were characterised by DLS in terms of (A) z-average, (B) PDI and (C) zeta potential (mean \pm SD, 10 minutes n=1 (3 technical replicates), 30 minutes n=2). (D) Morphology of nanoparticles from 30 minutes degummed silk was investigated by SEM.

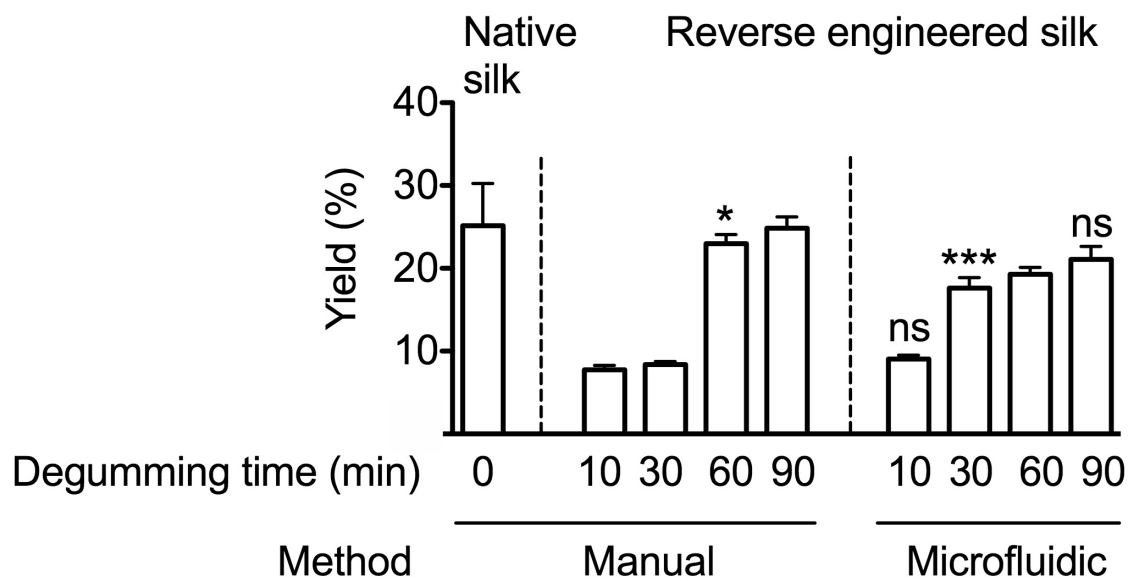


Figure S2. Impact of degumming time and manufacturing method on nanoparticle yields. Statistical analyses were performed by comparing equal degumming times of manual with the microfluidic method using the Student's t-test (samples run as three technical replicates unless stated otherwise. Native silk n=2 ; Manual: 10 minutes n=3; 30 minutes n=2 ; 60 minutes n=3 ; 90 minutes n=3; Microfluidic: 10 minutes n=1 only 2 replicates; 30 minutes n= 2; 30 minutes n=2 ; 60 minutes n=3 ; 90 minutes n=2).

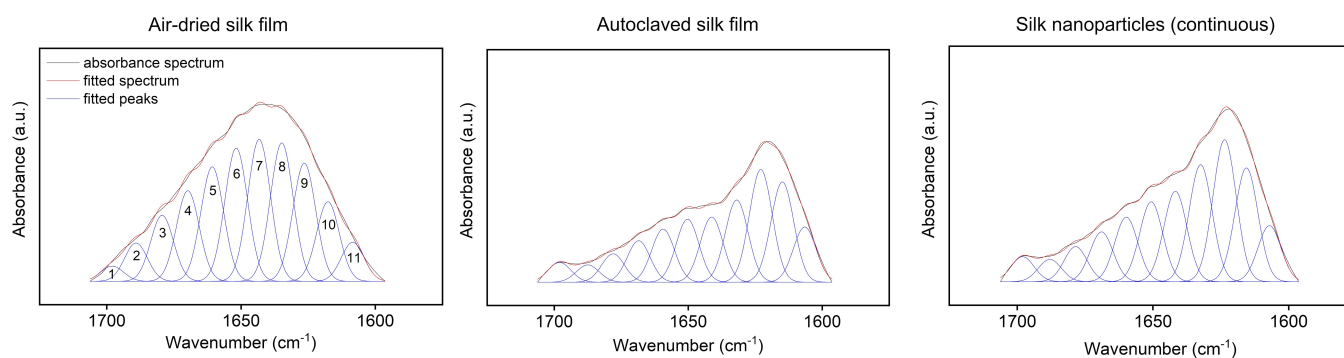


Figure S3. Band fitting to absorbance spectra shown exemplary for 60 minutes degummed silk (A) air dried film, (B) autoclaved film and (C) nanoparticles formulated by the batch method. Peaks are numbered according to Table S1.

Table S1. Assignment of fitted peaks to secondary structure according to Hu et al.⁴⁶

Peak	1	2	3	4	5	6	7	8	9	10	11
Wavenumber (cm ⁻¹)	1697-1703	1686-1696	1671-1685	1663-1670	1656-1662	1647-1655	1638-1646	1628-1637	1622-1627	1616-1621	1605-1616
Secondary structure	Beta sheet	Turns	Turns	Turns	Alpha helix	Random coil	Random coil	Beta sheet	Beta sheet	Beta sheet	Side chain

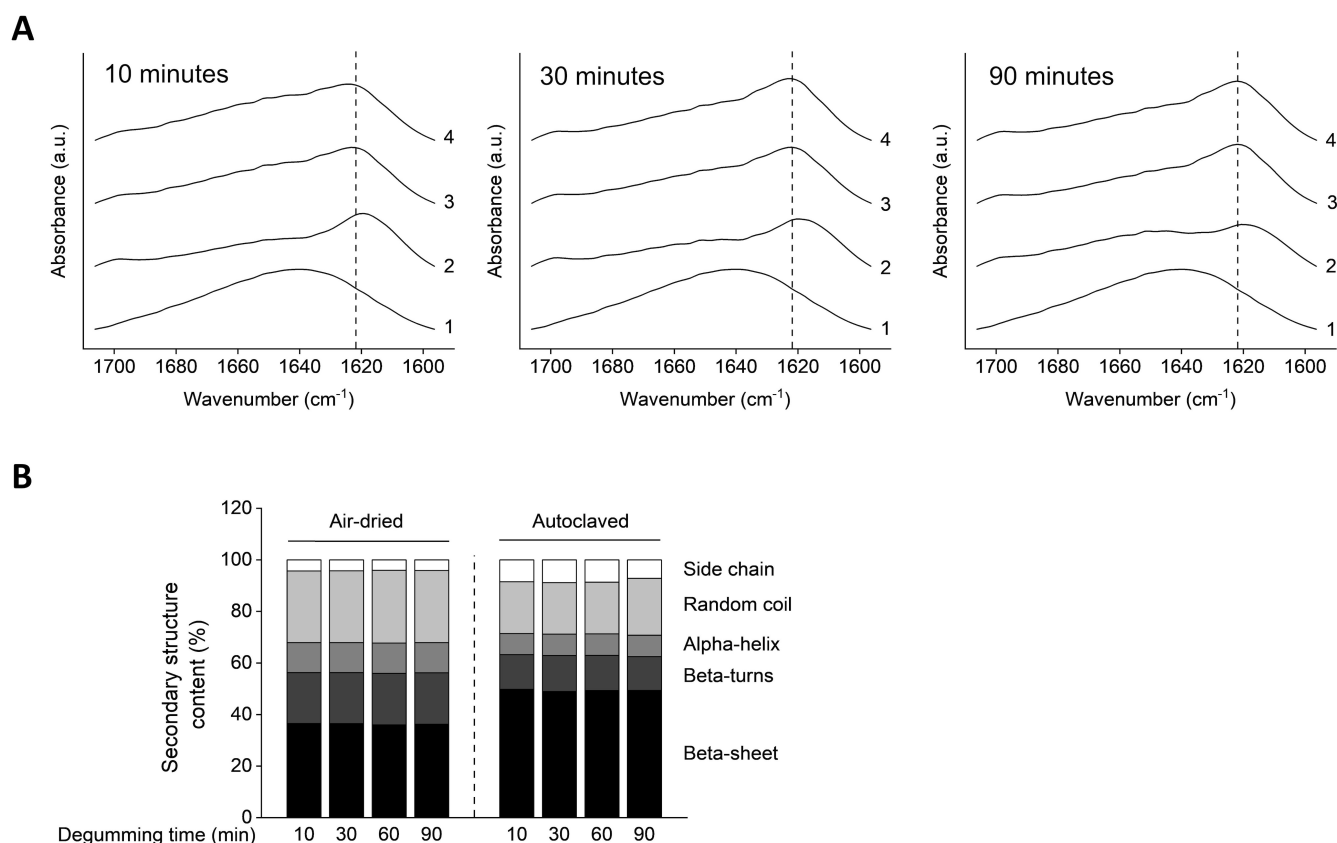


Figure S4. (A) FTIR absorbance spectra in the amide I region of (1) air dried silk film, (2) autoclaved silk film, silk nanoparticles manufactured by (3) batch method and (4) continuous method using 10, 30 or 90 minute degummed silk. The dashed line marks the characteristic absorption band for antiparallel beta-sheet structure between 1622-1627 cm⁻¹. (B) Comparison of secondary structure content of air dried and autoclaved silk films made from 10-, 30-, 60- or 90-minute degummed silk.