

Silver/fibroin/alginate microspheres for wound and ulcer therapy

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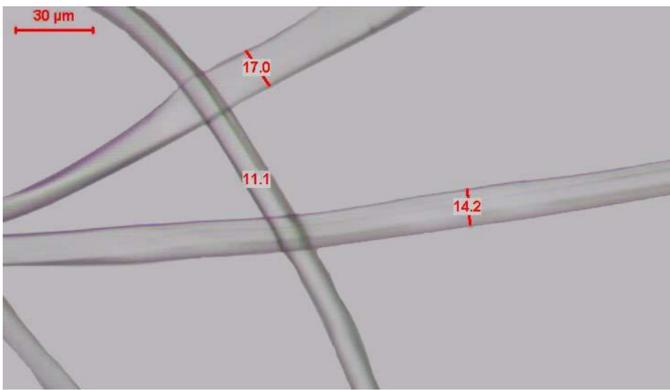
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Silver has been used for centuries in the treatment of wounds and cutaneous ulcers: it is a potent antimicrobial agent, acting by blocking the respiratory enzyme pathways and altering the microbial DNA and cell wall. The aim of this work is to prepare microsphere of alginate and silk fibroin for topical silver spray administration: the stabilization of the silk fibroin β -sheet form was promoted using PEG 1500 as osmolyte.

Silk fibroin solution

Cocoons of *Bombyx mori* were degummed, cut in small pieces and stirred in an aqueous solution of CaCl_2 and ethanol. The solution was dialyzed using cellulose membranes. The final concentration of silk fibroin aqueous solution was 2% w/w.



Coomassie brilliant blue staining was used to check the sericin absence.

Microspheres

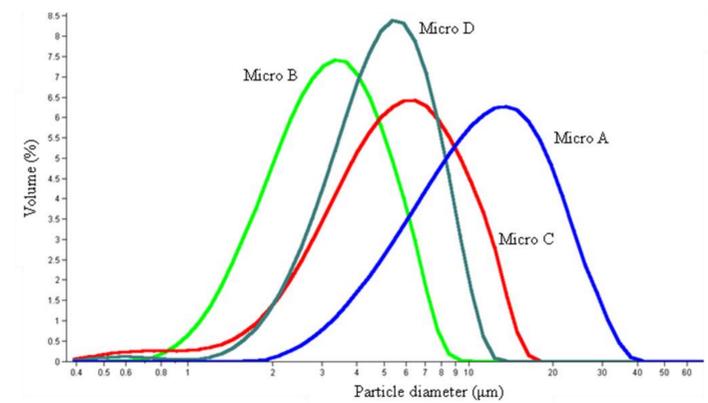
Microspheres were obtained by spray drying (Büchi Mini Spray Dryer). Colloidal silver aqueous solution was electrochemically obtained at the concentration of 80 ppm and diluted in silk fibroin solution, 1:1 volume ratio; polyethylenglycol was added at the concentration of 0.05% w/v (Micro B and D) and 0.1% w/v (Micro C); sodium alginate solution was added at the concentration of 1% w/v (Micro D).

Micro	Alginate	Fibroin	PEG
A	0	100	0
B	0	95	5
C	0	90	10
D	48.75	48.75	2.5

Percent composition of microspheres

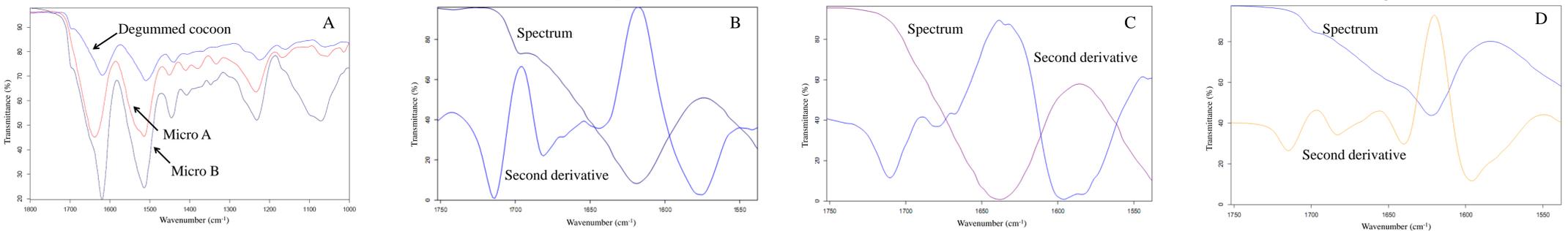
Granulometric analysis

Granulometric analysis of microspheres was performed in water by a laser light scattering granulometer (Beckman Coulter LS230).

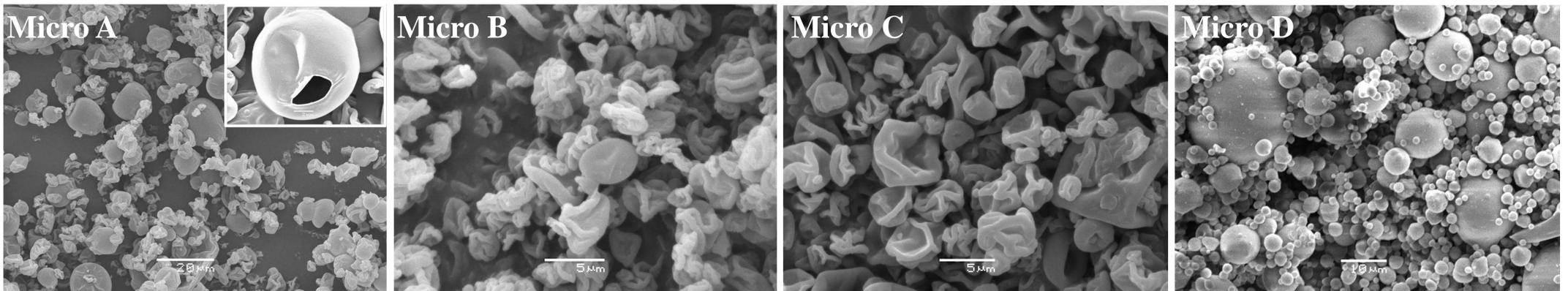


Micro B show a size distribution with volume-weighted diameter lower than the other types of microspheres (Micro A=13.03 μm ; Micro B=3.36 μm ; Micro C=5.99 μm ; Micro D=5.15 μm).

Fourier Transform Infrared Spectroscopy (FTIR). The conformational transition of microspheres containing silk fibroin and silk fibroin/PEG was carried out by means of infrared spectroscopy. The degummed cocoons have absorption bands at 1620cm^{-1} and 1510cm^{-1} for amide I, both attributed to β -sheet conformation, while Micro A at 1640cm^{-1} for amide I and 1515cm^{-1} for amide II, both attributed to random coil; Micro B has a similar behavior to degummed cocoons (A). The second derivative spectra (B, C, D) suggest that degummed cocoons and Micro B have the same behaviour (B and D), whilst the Micro A present the second derivative of random coil conformation (C).



SEM analysis. Micro A, Micro B, and Micro C containing silk fibroin are generally collapsed and vacuolated, while Micro D, containing alginate and fibroin, appears smooth and spherical shaped.



These data suggest that colloidal silver can be loaded in microspheres containing alginate and silk fibroin. Polyethylenglicol may be added in small quantities to guarantee the silk fibroin β -sheet structure and alginate improves the microsphere quality and morphology. Microspheres containing alginate, silk fibroin and colloidal silver are promising devices for the therapy of wounds and cutaneous ulcers by topical spray administration.

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