Stimuli responsive emulsion-based micro/nanospheres for the delivery of lipophilic therapeutics

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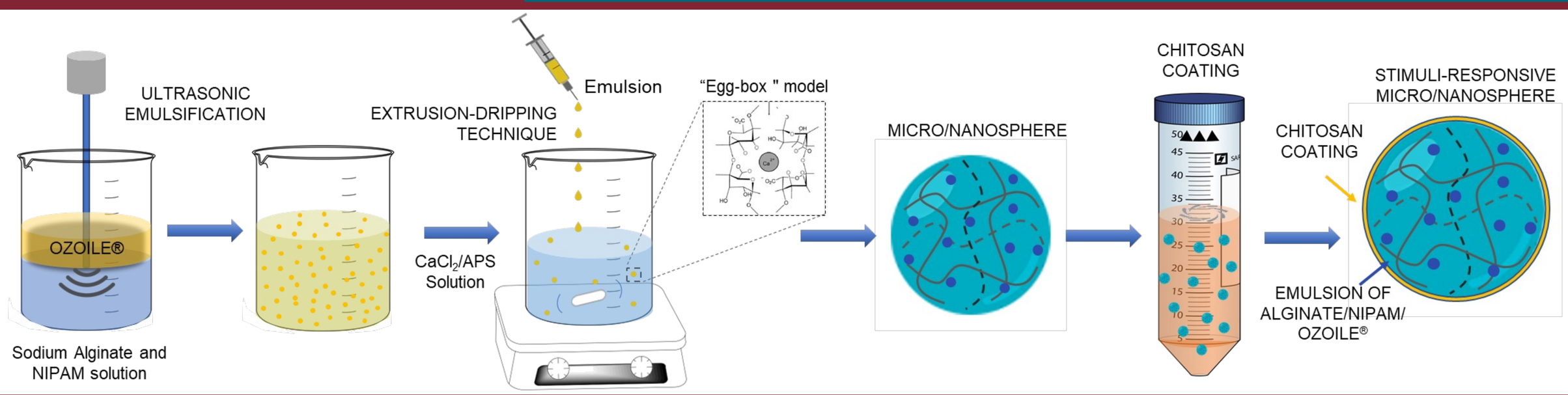
Summary

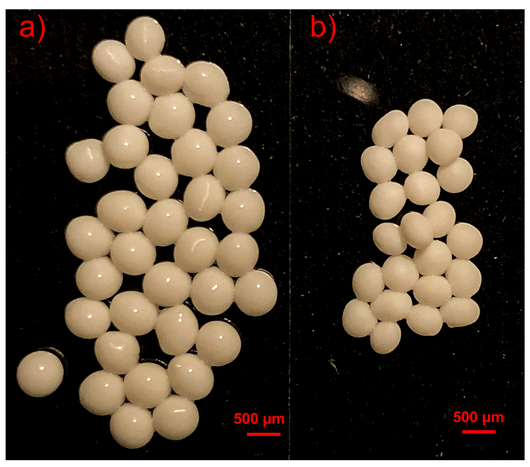
Hydrogel micro/nanospheres are of great interest in the field of drug and gene delivery. They can be applied to overcome the limitations of traditional drug administration in terms of therapeutic efficiency and patient compliance. In this work, advanced multi-responsive hydrogel micro and nano-spheres were developed using an emulsion prepared with sodium alginate and N-isopropyl-acrylamide (NIPAM) with Stable Ozonides (Ozoile) produced in a patented process. Ozoile acts as a biological inducer, regulating the main metabolic pathways and stimulating the endogenous defense system, and promotes tissue regeneration and damage-injury repair through the regulation of gene transcription. Ozoile was dispersed in the alginate solution forming an emulsion by high-intensity ultrasound (HIU) method. The micro/nanospheres were prepared from an Alginate/NIPAM-based emulsion with Ozoile (20 wt%). The emulsification process was optimized by varying the types and concentrations of surfactants and stabilizers. For the fabrication of the spheres, the extrusion-dripping technique followed by ion gelation crosslinking was used. Next, the micro/nano spheres were optimized by a chitosan coating to improve the stability.

Fabrication of Hydrogel Micro/Nanospheres



Chitosan Coating

Hydrated Alginate/PNIPAM/Ozoile microspheres without chitosan coating (a) and with chitosan coating (b)

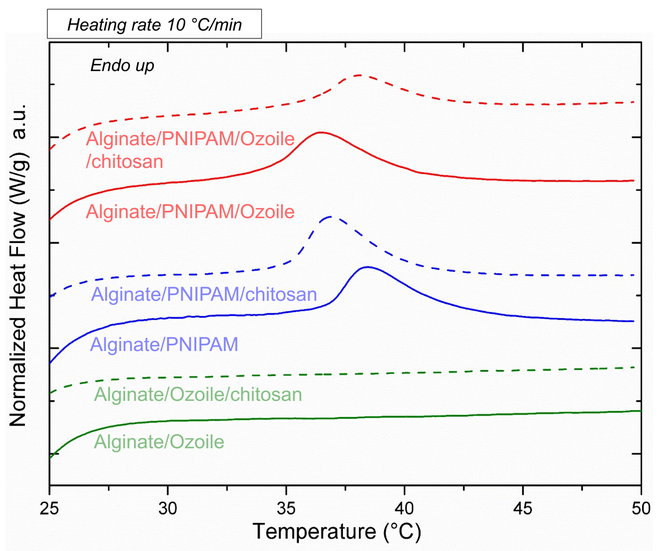


- Chitosan coating obtained upon stirring the microspheres for 30 min at 600 rpm
- NH_3^+ groups of chitosan strongly interact with the alginate carboxyl by ionic bonding
- Coating improves microspheres stability and makes them more compact

Thermal Characterization by DSC

Analysis by differential scanning calorimetry shows endothermic phenomena in the range 35 – 40 °C for the microspheres with PNIPAM due to volume phase transition (VPT) driven by hydrophobic collapse.

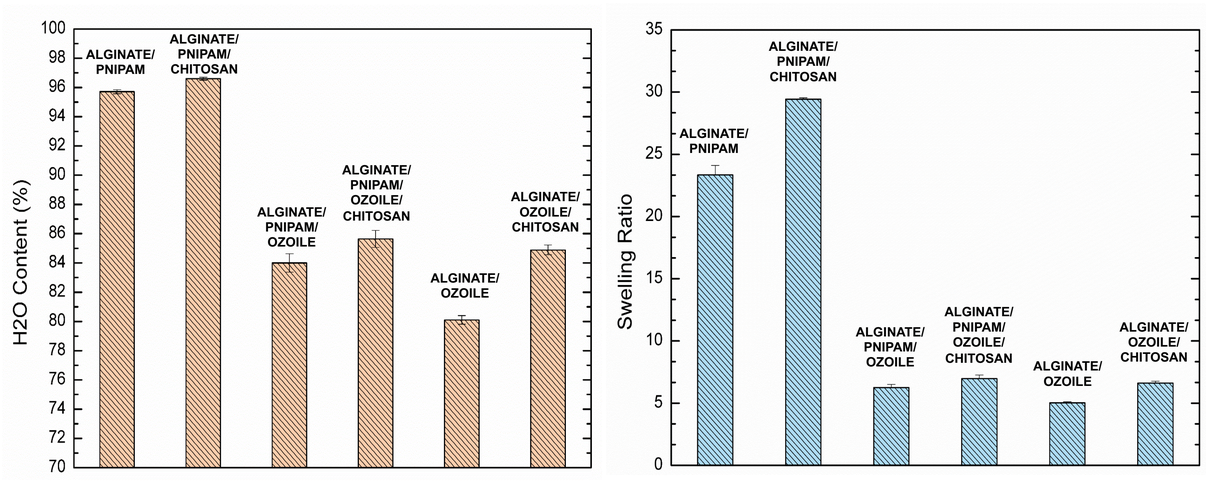
In microspheres with Ozoile, VPT peak is delayed when chitosan coating is applied, indicating stabilizing effect of the coating.

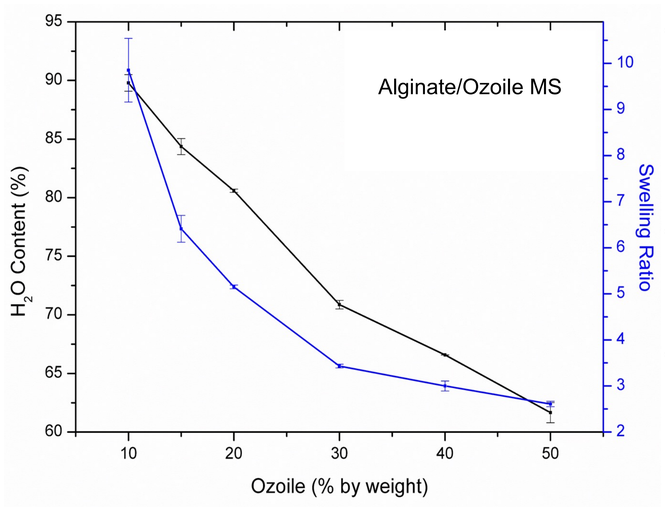


SAMPLE	VPT (°C)	ΔH (J/g)
ALGINATE/PNIPAM/OZOILE	36.1 ± 0.3	0.78 ± 0.03
ALGINATE/PNIPAM/OZOILE/CHITOSAN	37.9 ± 0.2	0.42 ± 0.03
ALGINATE/PNIPAM	38.5 ± 0.2	0.38 ± 0.02
ALGINATE/PNIPAM/CHITOSAN	36.3 ± 0.2	0.19 ± 0.09

DSC analysis shows that addition of NIPAM to alginate sol makes the microspheres responsive to temperature in physiological range.

Swelling Properties of Hydrogel Microspheres



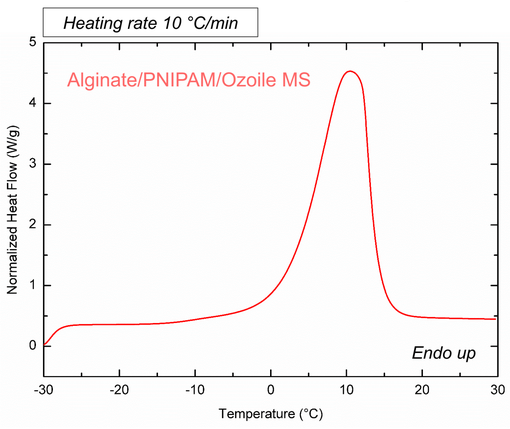


- Unloaded Alginate/PNIPAM microspheres (MS) show higher water content (>90%) with respect to those containing Ozoile (water content 80-85%).
- MS coated with chitosan exhibit higher water content than those without.
- Presence of Ozoile in the alginate matrix induces a decrease in water content and swelling ratio

Water bound fraction (X_{BW}) calculated according to the following equation:

$$X_{BW} = X_{TW} - \frac{Q_{endo}}{Q_f}$$

X_{TW}: total water fraction
Q_{endo}: heat of fusion of freezable water
Q_f: heat of fusion of pure water (333 J/g)



SAMPLE	ΔH (J/g)	X_{BW}
ALGINATE/PNIPAM	272.2 ± 1.3	0.14 ± 0.01
ALGINATE/PNIPAM/CHITOSAN	269.4 ± 1.1	0.16 ± 0.01
ALGINATE/PNIPAM/OZOILE	201.7 ± 0.4	0.23 ± 0.01
ALGINATE/PNIPAM/OZOILE/ CHITOSAN	192.4 ± 1.4	0.28 ± 0.01
ALGINATE/OZOILE	241.43 ± 1.9	0.08 ± 0.01
ALGINATE/OZOILE/CHITOSAN	252.2 ± 1.7	0.09 ± 0.01

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