

Development of nano particles encapsulating bioactive molecules by using microstructure of reversed micelles

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Reversed micelles and water-in-oil microemulsions can disperse nano-size water droplets in a polar solvents. The nano water droplets stably solubilize biomolecules such as proteins, peptides and DNA, and the size is controllable by adjusting water contents. On the other hand, the sol-gel method is a low temperature process widely used to produce optically transparent, microporous glasses by the polycondensation of liquid phase alkoxide precursors. Reversed micellar solution facilitates forming nano-size glasses in the hydrophilic core containing bioactive molecules. The hydrophilic nano sphere is favorable for solubilizing biomolecules and for hydrolysis of the alkoxide precursors. To create a novel nano carrier for bioactive materials, therefore, my strategy is based on the sol-gel encapsulation of the nano-size water droplets solubilizing biomolecules. Transmission electron microscope (TEM) observation shows that nano particles can be produced by the AOT and NP-5 reversed micellar droplets. The monodispersed particles with 10 - 20 nm diameter were obtained. However, in fact, almost all the immobilized protein preparations we obtained are a micro-size aggregate of the nano particles. In addition, the protein encapsulation profiles indicate that proteins solubilized in reversed micelles are completely incorporated into them with the growth of silica particles. The efficiency of protein encapsulation into the nano silica particles depends on the water droplet pH, W_o value ($= [\text{water}]/[\text{surfactant}]$), and the composition of alkoxide precursors. It seems that these factors affected not only the surface condition of cytochrome c but also the sol-gel processing. The encapsulated cytochrome c and subtilisin Carlsberg in nano silica particles retain their catalytic activities. Subtilisin Carlsberg entrapped in nano silica particles shows 40-fold greater transesterification activity than any other immobilized subtilisin Carlsberg. Protein encapsulation in silica gel prevents from denaturing protein by organic solvent. Molecular isolation of proteins in nano silica particles enhances the effective enzyme quantity and the surface area to contact with bulk substrate solution. These effects facilitate the high performance of subtilisin Carlsberg entrapped in nano silica particles for transesterification reaction in organic media. Hereafter we will develop new nanocapsules including biomolecules with high dispersibility and good reactivity.