O7-3	**Supramolecular structures produced from bovine serum albumin and high methoxy pectin as the building blocks of multilayer microcapsules**	

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INTRODUCTION

In view of its ability to protect sensitive ingredients and to control their targeted release, microencapsulation has become an important technique in the food industry. Incorporation of microcapsules into food systems requires the use of food grade ingredients as shell materials. The purpose of this study is to produce microcapsules using supramolecular assemblies consisting of common food ingredients such as bovine serum albumin (BSA) and high methoxyl (HM) pectin. Two supramolecular structures were used in this research, i.e. BSA fibrils and BSA pectin soluble complexes. Fibrillar aggregates of BSA were chosen because previous studies (Sagis, 2008) showed that inclusion of whey protein isolate fibrils in the layers yields microcapsules with high mechanical stability, due to the fact that the fibrils give the shell a structure of a fiber-reinforced nanocomposite. Although the production of multilayer emulsions by charge driven adsorption of polysaccharide onto the surface of protein stabilized oil droplets has been studied extensively, the production of multilayer microcapsules by simultaneous addition of protein and polysaccharide in a form of stable protein-polysaccharide nanoparticles has not been reported before. In this study, microcapsules produced from soluble complexes of BSA-HM pectin and BSA fibrils will be characterized based on size distribution, ζ-potential, morphology and mechanical properties.

MATERIALS AND METHODS

BSA-HM pectin complex solutions were prepared by mixing 0.05% (w/w) BSA and HM pectin solutions at pH 4 in a 200 mM sodium acetate buffer at a weight ratio of 1:2, and were left stirring overnight. BSA fibrils were prepared based on the method developed by Veerman (Veerman, 2003). Microcapsules with different number of layers were produced by alternately adsorbing 0.05% (w/w) complexes of BSA-HM pectin and 0.05% (w/w) BSA fibrils onto oil in water emulsion droplets.

RESULTS AND DISCUSSIONS

Using BSA and a combination of BSA and HM pectin, two different supramolecular structures were produced: spherical complexes and semi-flexible protein fibrils (Figure 1). The hydrodynamic diameters of the spherical complexes are in the range of 300 nm, and the contour length of the semi flexible fibrils are ranging from approximately 100 nm to 500 nm.

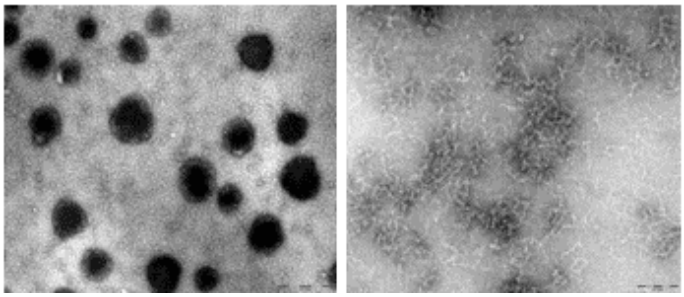

Figure 1 : TEM micrographs of soluble BSA-HM pectin complexes (left) and BSA fibrils (right)

Multilayer microcapsules were obtained through alternate adsorption of BSA/HM pectin complexes and BSA fibrils onto the surface of oil droplets. Microcapsules with an odd number of layers have BSA/HM pectin complexes as the outer layer. Those with an even number of layers have BSA fibrils as the outer layer. Figure 2 shows the size distribution and ζ-potential of the microcapsules after each adsorption cycle.

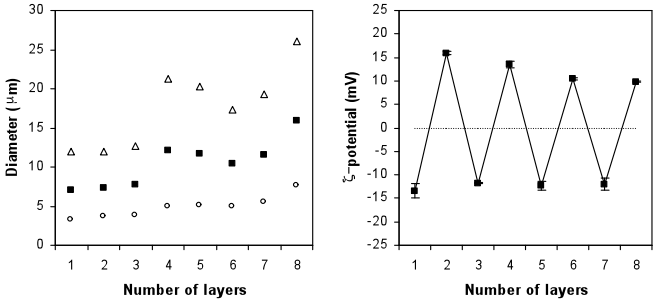

Figure 2 : Size and ζ-potential of multilayer microcapsules. Size at various number of adsorption cycle. (■) $D_{4,3}$ is the volume mean diameter, (○) d(0.1) and (Δ) d(0.9) are the diameters below which 10% and 90% of the microcapsules lie, respectively.

In order to obtain microcapsules with a narrow size distribution, stable oil in water template emulsion with a narrow size distribution is needed. The size distributions of 1% (w/w) oil in water template emulsions stabilized with 0.05% (w/w) BSA/HM pectin complex and pure BSA with concentrations of 0.05% (w/w) and 0.017% (w/w) are given in Figure 3. The 0.05% (w/w) soluble complex gives a narrower size distribution of oil droplets compared to the 0.05% (w/w) of pure BSA. The concentration of BSA in a 0.05% (w/w) soluble complex solution is equal to 0.017% (w/w), and when the same concentration of pure BSA is used to stabilize an oil in water emulsion, a wider size distribution was obtained. The ζ-

potentials of the oil droplets stabilized with the soluble complex are negative, while those stabilized with pure BSA have positive ζ-potentials

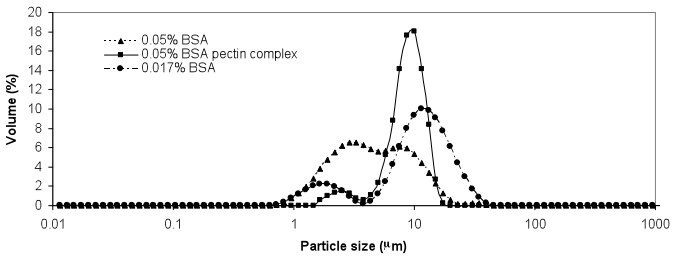

Figure 3 : Size distribution of the oil droplets stabilized using BSA pectin complex and BSA monomers

The efficiency of BSA fibrils as a building block in microcapsules was compared to that of pure monomeric BSA. BSA fibrils and BSA monomers, at a protein concentration of 0.05% (w/w), were adsorbed to the surface of oil droplets previously coated with soluble complex of BSA/HM pectin. Adsorption of 0.05% (w/w) BSA monomers leads to extensive creaming and aggregation, as demonstrated by the large increase in mean particle diameter (Figure 4). In contrast, when 0.05% (w/w) of BSA fibrils was used, the droplets remained stable. More importantly, droplet charge reversal, which is paramount to the formation of multilayer microcapsules (Guzey, 2006), occurs only when BSA fibrils are used. BSA fibrils are more effective than BSA monomers in compensating the negative charge of oil droplets stabilized by the soluble complex of BSA/HM pectin

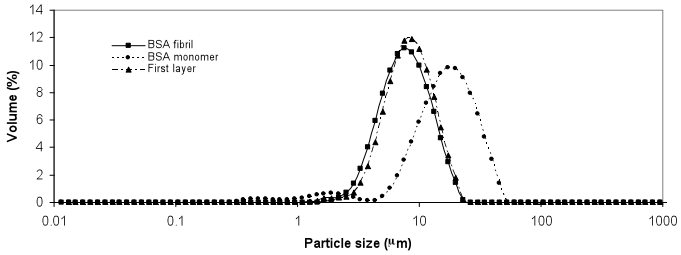

Figure 4 : Size distribution of the oil droplets stabilized using BSA pectin complex, BSA fibrils and BSA monomers.

After removal of the oil template, the shell structure of the hollow multilayer microcapsules made from complexes of BSA-HM pectin and BSA fibrils is visualized by SEM. Figure 5 shows four and seven layer microcapsules. Comparing the surface morphology of microcapsules with the outermost layer of BSA fibrils and BSA pectin complex, the first has a fibrous structure, while the second has a coarse structure. With increasing number of layers, it can be seen that the thickness of the shell increases, leading to stronger microcapsules. This is evident from the fact that microcapsules with lower number of layers tend to shrink and buckle after drying, while microcapsules with higher number of layers have maintained their spherical shape.

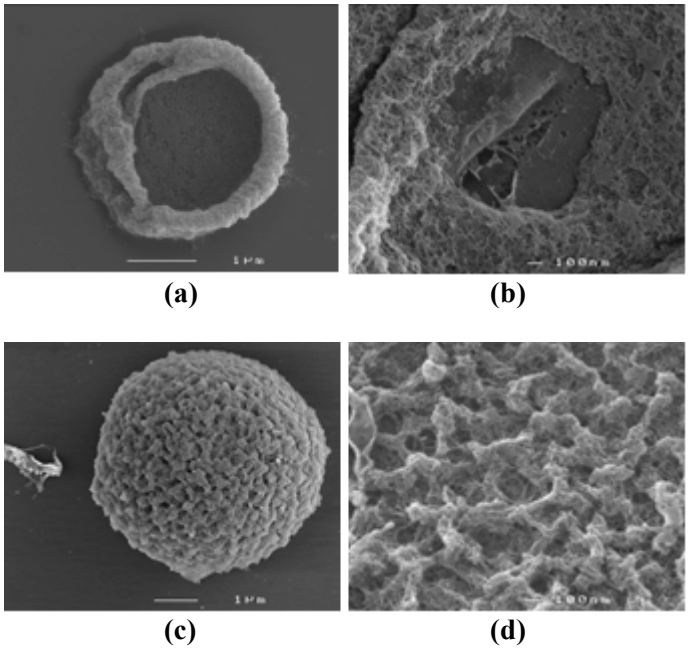

Figure 5 : SEM images of multilayer hollow microcapsules with four (a) and seven (c) layers. The surfaces of the shell of the microcapsules for four (b) and seven (d) layers.

CONCLUSION

Microcapsules with multilayer shells were produced using two supramolecular structures, i.e spherical soluble complexes of BSA-HM pectin and semi flexible BSA fibrils. It is shown that with increasing number of layers, the thickness of the shell increases, leading to stronger microcapsules. Compared to native BSA, the two supramolecular structures are more effective building blocks for creating stable microcapsules. The microcapsules produced by layer-by-layer adsorption of soluble complexes of BSA-HM pectin and BSA fibrils described here can be applied in a food system.

REFERENCES

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