Fabrication of PVA Hydrogel/Chitosan capsules for Bioencapsulation

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INTRODUCTION

The unique properties of poly (vinyl alcohol) **(PVA)**, attract a great deal of attention because of its high degree of swelling in water, desirable mechanical properties and low toxicity (Bray, 1973 & Kobayashi, 1992). It plays a significant role in biomaterial engineering such as artificial cartilage, drug delivery systems, microorganism enwrapping, cell micro-capsulation, anti-thrombin materials, and biomedical sponges due to its biocompatible, biodegradable property as well as mechanical properties (Francios, 2007, Coluccio, 2006, Lio, 1994 & Bajpai, 2006). One of the important properties of PVA is the formation of hydrogel in aqueous system upon standing at room temperature. Its hydroxyl groups get chemically and physically cross-linked with glutarldehyde, epichlorohydrin and borax (Kanaya, 2006). Their water content and elastic property matches that of biological tissue, which offers a unique mechanical properties and friction characteristics so they have been proposed as materials to be used in reconstructive joint surgery for synthetic articular cartilage applications (Swieszkowski, 2006). PVA gels crosslinked with glutaraldhyde, glyoxal or borate have been proposed as drug delivery carriers. In these gels, the drug is able to be released fast or slowly due to the gel's high or low swelling ratio upon immersion in water (Dai, 1999). These excellent biomedical applications of PVA hydrogel encouraged us to use them in LbL (Decher, 1997, Caruso, 1999 & Manna, 2008) assembly as a novel delivery system. But PVA is an uncharged polymer which restricts employing in LbL approach by using electrostatic interaction as a driving force for self-assembly. In this work, we have employed a simple cross-linking reaction between PVA and borax to obtain physically cross-linked gel and induce a negative charge on PVA backbone. Further, charged PVA gel used for self-assembly on colloidal particles in combination with chitosan to obtain stable microcapsules for potential drug delivery application. Rhodamine-B (Rh-B) used as a model drug for encapsulation in microcapsules.

MATERIALS AND METHODS

Chitosan (Mw~2,00,000) and polyvinyl alcohol (Mw~14000) was obtained from Aldrich. Melamine formaldehyde (MF) particles (hydrochloric acid, HCl, soluble) were procured from Microparticles GmbH, Berlin, Germany. HCl, AcOH and calcium chloride were obtained from Qualigens, India. All materials were used without further purification. Ultrapure water (Millipore) with specific resistance around 18 M**Ω**cm was used. All experiments were carried out at room temperature.

Capsule Preparation

Chitosan was dissolved in millipore water at pH 4.0 with a concentration of 1 mg/ml. The positively charged MF particles (0.1 ml of 10 wt % dispersion) were incubated with PVA hydrogel solution and allowed to adsorb for 1 h. The suspension was centrifuged, followed by four washings steps with water in order to remove non-adsorbed hydrogel on MF particles. Then, chitosan solution (1 mg/ml) was added to the MF-particle suspension and allowed to adsorb for 20 min, and followed by three washing steps of water were carried out before depositing the next layer of PVA hydrogel. Alternatively, PVA hydrogel and chitosan steps were repeated to construct multilayers on MF particles. Hollow capsules were obtained by dissolving MF particles in 0.1N HCl solution for 10 min, and then washed three times with water.

RESULT AND DISCUSSION

PVA-Borax Hydrogel

PVA forms chemically crosslinked gels with many organic and inorganic cross-linkers. Particularly gelation of PVA in the presence of boric acid and borax has been studied extensively (Chung, 2004, Seeboth, 1999 & Keita, 1990). Borate anion acts as a bridging agent between two PVA polymer chains. In addition to that intermolecular hydrogen bonding takes place between hydroxyl groups on PVA chains which helps for the gelation process. This mechanism is well established in literature (Kanaya, 2005). Additionally, borate anion has negative charge and we have exploited the negative charge and excellent biomedical properties of hydrogel to grow multilayer thin films on colloidal particles.

Multilayer Coatings on Colloidal Particles & capsule fabrication

The PVA/Chitosan multilayer growth on MF particles was characterized by TEM and CLSM. Figure 1a shows the TEM images of coated MF particles. Thickness of each bilayer corresponds to 11.6 nm. Figure 1a also shows the homogenous coating on MF surface, so the hydrogel characteristic of PVA did not affect uniform coating. This could be a due to different kind of interaction in between hydrogel and chitosan unlike linear PE. CLSM images were recorded for dye (Rh-B) containing particles and it clearly shows a ring like structure as shown in Figure 2b. So, this multilayer membrane is efficient to load hydrophilic molecules.

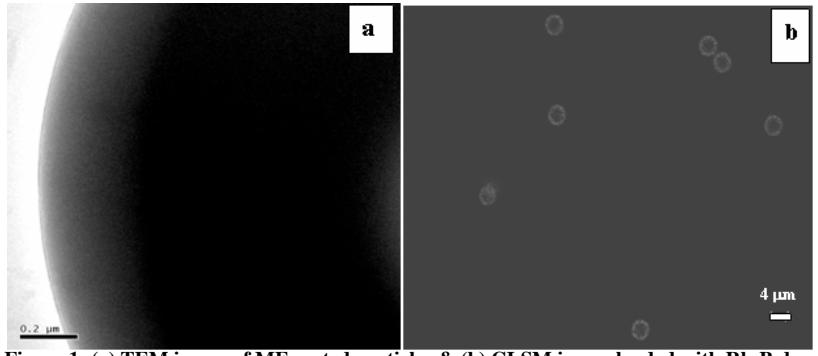

Figure 1: (a) TEM image of MF coated particles & (b) CLSM image loaded with Rh-B dye.

The capsules were prepared by exposing PVA-Borax hydrogel/chitosan multilayer (both 9 layers & 10 layers) coated MF particles to 0.1M HCl solution. The resulting hollow capsules were characterized by TEM as shown in Figure 2. The morphology of capsules is invariably different from the coated particle. The dissolution of MF particle induces the change in the shape of hollow capsules. Average size of hollow microcapsule with hydrogel as outermost layer (9 layers) is 350 nm but shape of the microcapsules remains spherical, where the average size of microcapsules with chitosan as outermost layer is ~1.8 µm. The outermost hydrogel layer of the microcapsules (9 layers) governs change in dimension of the microcapsules upon air drying due to change in water content. (Manna, 2009)

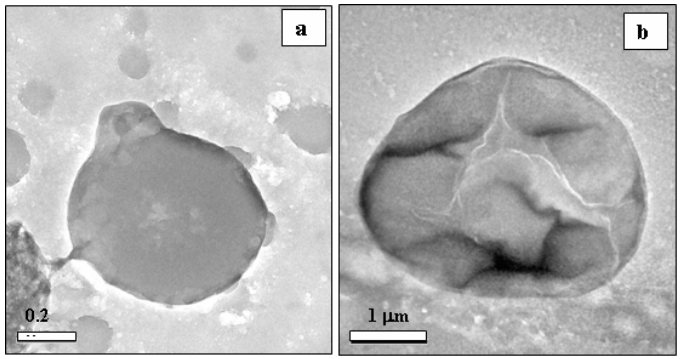

Figure 2: (a) TEM images of hydrogel/chitosan multilayer hollow microcapsule with 9 and 10 layers coating.

Loading of probe molecules in capsules

Hollow capsules have been loaded with hydrophilic Rh-B molecules (model drug) as shown in Figure 3.

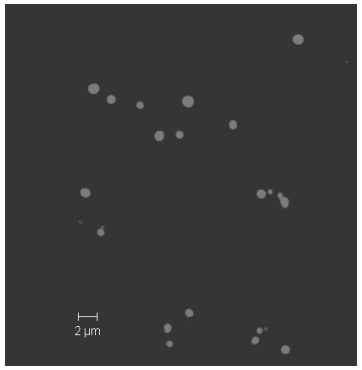

Figure 3: CLSM image of Rh-B loaded PVA-borate/chitosan microcapsules.

The incubation of microcapsules in Rh- B solution (concentration of 50 ppm) for 24 hr results into loading of dye and it alters the size of microcapsules. It could be due to osmotic pressure from concentration gradient of Rh-B molecules between inside & outside of the capsules. But loaded microcapsules remain almost unaltered in shape. These results suggest that these microcapsules can be used for potential drug delivery application.

CONCLUSIONS

In conclusion, the multilayers of neutral PVA were fabricated by inducing negative charge by forming a complex with borax. This results into physically cross-linked PVA gel. LbL approach was employed for multilayer preparation on spherical particles with chitosan as an oppositely charged polyelectrolyte. Removal of core results into hollow microcapsules. The microcapsules of PVA-borax/Chitosan were loaded with model drug Rhodamine-B to establish the drug delivery potential of these microcapsules.

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