Utilization of Polysaccharides Extracted from Tofu Processing Wastewater for Microencapsulation Processes in the Food Industry

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Authors’ contributions
This work was carried out in collaboration among all authors. Author TSS designed the study and operations of the manuscript. Author MKS managed the responsible for preparation of bench reagents and proof reading of the manuscript. Authors AN and MSD performed the typing, proof reading and preparation of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT
The aim of this study was to extract polysaccharides from tofu processing wastewater obtained from tofu factory located at the outcast of Wuxi city, PR China. The extracted polysaccharide was used as wall material for microencapsulation processes in the food industry. This is expected to serve as an environmentally friendly research approach as it converts wastewater into useful polysaccharides that could be used in food industries. This study used vegetable oil as core material, which is used as a test case to ascertain whether a successful encapsulation process can be achieved – an indication of the possibility of the encapsulation of oil soluble minerals and vitamins.

The polysaccharides extracted from tofu processing wastewater were used as wall materials for the encapsulation of vegetable fat load. The basic method applied in the microencapsulation process is indicative of the fact that with the addition of polysaccharides, the interfacial coacervation between...
the protein and the polysaccharide around the oil droplets induces the formation of microcapsules. The Seagull Model Z-Series Photomicrographic equipment was used to obtain Photomicrographs of the various phases in the development of microcapsules. Microencapsulation efficiency, the peroxide value and visual observation on spray-dried microcapsules were studied. Microencapsulated capsules were produced through thermal solidification of the coating to form self-sustaining entities or microcapsules. Photomicrographs were used to clearly visualize the microcapsules. The Microencapsulation efficiency of the microcapsules was studied and results showed that the highest MEE (93.04%) was obtained with a comparatively low vegetable fat load. This shows that there is a relationship between percent fat load and MEE as the higher the vegetable fat load the lower the microencapsulation efficiency. Studies conducted on the Peroxide value of the microcapsules indicated shorter shelf-life at elevated temperatures of 38°C compared to room temperature. Polysaccharides extracted from tofu processing wastewater could be used as wall material for microencapsulation in food industries.

**Keywords:** Tofu wastewater polysaccharides; utilization of wastewater polysaccharides; microencapsulation.

### 1. INTRODUCTION

Microencapsulation is a packaging technology by which small droplets of liquid or solid particles are packed into continuous individual shells. The shells (walls) are designed to protect the encapsulated material from factors which may cause its deterioration; e.g. oxygen, moisture and light. Another important feature of encapsulation is that the walls of the encapsulated materials are designed specifically to enable a controlled release of materials encapsulated under desired conditions [1]. This technology, according to Bakan [2] and Dziejak [3] has made it possible for the production of a wide variety of encapsulated ingredients which are useful in today's food industries. Such ingredients are products of a process that totally envelopes the ingredient in a coating or capsule, thereby conferring many useful and otherwise unusual properties to the original ingredient [4]. Food industries apply microencapsulation for a number of reasons which include:

1. to stabilize the core material;
2. to control the release of the core material (both the rate of release and the commencement of release);
3. or to separate reactive or incompatible components of a formulation.

Under normal circumstances, the microcapsule is expected to offer the food processor a means to protect sensitive food components, prevent or minimize nutritional loss, utilize otherwise sensitive ingredients, incorporate unusual or time-release mechanisms into the formulation, mask or preserve flavors and aromas, and transform liquids into easily handled solid ingredients [5]. Another important application of microencapsulation in the food industry is that it enables the addition of vitamins and minerals to nutritional dry mixes [6]. This is used to fortify a variety of foods including breakfast cereals, dairy products, infant formulas and animal feeds. It also makes it possible for the encapsulation of both fat- and water-soluble vitamins and minerals which provide a lot of advantages for the food processor [7,8]. Encapsulation obviously reduces off-flavors contributed by certain vitamins specifically under extreme temperatures and high moisture conditions [9]. Food items that have benefited from microencapsulation techniques include; natural colors, flavoring agents and spices, leavening agents, sodium chloride, sweeteners, vitamins and minerals, etc.

It has also been noted that one of the most important industrial applications of the interfacial properties of protein-polysaccharide complexes is the formation of microcapsules or micro particles. The formation of micro gel or microencapsulation results from the ability to form a solid film around emulsion droplets containing the product to be encapsulated (microcapsules) and also the possibility of entrapping solvent molecules into the coacervate (micro gel). Schmitt and others [10,11] have investigated the utilization of protein-polysaccharide complexes in microencapsulation and have concluded that the most widespread method of encapsulation is to produce emulsion stabilized with protein. It was therefore with this background that this research was dedicated to the investigation of the possible utilization of polysaccharides extracted from tofu processing wastewater polysaccharides.
wastewater in microencapsulation processes. A successful production of microcapsules using the extracted polysaccharides will serve as a cheap source of obtaining wall materials for encapsulation processes.

2. MATERIALS AND METHODS

Polysaccharide was extracted from tofu processing wastewater (PTW) obtained from the tofu processing factory located at the outcast of Wuxi, Jiangsu Province, PR China [12]. Pure soybean salad oil produced by East Ocean Oils and Grains Industries, Zhang Jiagang, Jiangsu Province, P.R. China. Other materials and chemicals used were either obtained from the chemical store of JiangNan University or from other laboratories. Equipment used were common laboratory equipment. The method used for the encapsulation process was a modified form of methods described by Schmitt [11] and Bakan [2]. According to Schmitt [11] the commonly used method is where an emulsion is produced and stabilized with protein. The dispersed oil droplets contain the desirable oil soluble product. This study only used oil as core material, which is used as a test case to ascertain whether a successful encapsulation process can be achieved – an indication of the possibility of the encapsulation of oil soluble minerals and vitamins.

2.1 Microencapsulation Process

The basic principle applied in the microencapsulation process is that with the addition of polysaccharides, the interfacial coacervation between the protein and the polysaccharide around the oil droplets induces the formation of microcapsules. This principle was used along with the method described by Bakan [2] and Ayoub and co-worker [9], which involved the following steps:

1. Formation of three immiscible chemical phases – a liquid manufacturing vehicle phase, a core material phase, and a coating material phase;
2. Deposition of the liquid polymer coating around the core material. This was accompanied by controlled physical mixing of the coating material (while liquid) and the core material in the manufacturing vehicle;
3. Thermal solidification of the coating to form self-sustaining entities – microcapsules.

Based on the above steps the microencapsulation process was carried out as follows: 200 ml of de-ionized water used as the liquid vehicle was put into a 400 ml beaker, placed in water bath and temperature adjusted to 55ºC The following are the percentage composition of both core and wall materials added to the liquid vehicle in the order in which they are listed at an interval of 15 minutes agitation after each addition. PTW was the main wall material.

The emulsion was agitated for .5 hrs after the addition of the vegetable fat load using a variable speed agitator. Photomicrographs of the various phases in the development of microcapsules were obtained using the Seagull Model Z-Series Photo micrographic equipment produced by Shanghai 3rd Camera Factory, P. R. China. The solidification of the coating was done by spray drying to form self-sustaining entities known as microcapsules.

2.2 Determination of Microencapsulation Efficiency

Microencapsulation efficiency (MEE) was determined using a modified form of the method described by Matsuno and co-worker (14). 2g of sample was put into a 50 ml conical flask with known weight and 30 ml of petroleum ether was added. Using a stirring rod, the mixture was stirred vigorously for 30 min. The resulting solution was then filtered into a beaker using a funnel and a filter paper of known weight. After

<table>
<thead>
<tr>
<th>Test No.</th>
<th>Malto dextrin DE 20</th>
<th>Gum arabic</th>
<th>PTW¹</th>
<th>VFL²</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>25</td>
<td>5</td>
<td>40</td>
<td>30</td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>5</td>
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</tr>
<tr>
<td>3</td>
<td>10</td>
<td>5</td>
<td>40</td>
<td>45</td>
</tr>
</tbody>
</table>

¹PTW = polysaccharides extracted from tofu processing wastewater
²VFL = vegetable fat load

Table 1. Percent composition of core and wall materials
filtration the filter paper was carefully removed and put into a conical flask of a known weight and then put into an oven at 70ºC for 15 min. It was then removed from the oven and put into a desiccator for 15 min and weighed. The following formula was used to calculate MEE:

\[ MEE(\%) = \left( \frac{W_1 - (W_1 + W_2)}{W_0 + VFL} \right) \times 100 \]

Where \( W_1 \) = weight of empty conical flask; \( W_2 \) = weight of filter paper; \( W_3 \) = weight of sample + conical flask (after evaporation); \( W_0 \) = weight of sample.

VFL was calculated as follows:

\[ VFL(\%) = \left( \frac{\text{core}}{\text{core} + \text{wall}} \right) \times 100 \]

2.3 Visual Observation on Spray-Dried Microcapsules

Visual observation was also carried out on the spray-dried microcapsules in an attempt to establish MEE. 5g of sample was evenly spread on a filter paper placed in a petri-dish and kept at room temperature for 72 h and the filter paper was observed for oil droplets.

2.4 Determination of the Rate of Peroxide Value (POV) of Microcapsules

This chemical constant (POV) is a specific characteristic of the oxygen bond as peroxide which can be found, for example, in the primary products of autoxidation. For this reason the peroxide value is one of the most important chemical constants for appraising the degree of deterioration of fats. The encapsulated oil samples were therefore sealed in polyethylene bags and kept at room temperature and at an elevated temperature of 38ºC for analysis of the peroxide value. The peroxide values were determined every other week for twelve weeks.

The initial stage of the determination of POV involved breaking of the wall of the encapsulated material so as to extract the entrapped oil. The method used for breaking the wall was that described by Gejl-Hansen and co-worker [13]. After breaking the wall and extracting the entrapped oil POV was determined using the extracted oil by the method of Gejl-Hansen [13].

3. RESULTS AND DISCUSSION

3.1 Production of Microcapsules

The production of microcapsules using polysaccharides recovered from tofu processing wastewater was successfully achieved as shown in Figs. 1–3.

Fig. 1 shows the gradual establishment of the three-phase system as the first step in the encapsulation process. The photomicrograph shows the droplets to be coated, surrounded by the coating material in liquid form, all dispersed in the vehicle phase [14].
The photomicrograph in Fig. 2 shows the deposition of the liquid coating around the core material. This was achieved by the controlled physical mixing of the coating material, in the liquid state, and the core in the manufacturing vehicle. The deposition of the polymer coating around the core material occurred when the polymer was adsorbed at the interface formed between the core material and the liquid vehicle phase – this sorption phenomenon serves as a prerequisite to effective coating as shown in the photomicrographs.

The solidification of the coating or encapsulation process by spray drying completes the microencapsulation process as shown in the photomicrograph in Fig. 3.

Results obtained indicate that polysaccharides extracted from tofu processing wastewater can be used as a cheap source of coating material to encapsulate a variety of food and non-food materials. Fat-soluble vitamins and other functional food ingredients can be encapsulated for both man and farm animals. Similar findings have been reported by Good and co-workers [15].

3.2 Microencapsulation Efficiency (MEE)

The visual observations made on the spray dried microcapsules obtained from the 3 test samples were positive as there were no visible signs of oil droplets on the filter papers after 72 hrs (Table 2). This was clearly indicative of the fact that the encapsulation was successful to some extent, although the test cannot independently give a reliable confirmation of the efficiency of any encapsulation process.

The MEE of the various test results are shown in Table 2.
Table 2. Microencapsulation efficiency of extracted polysaccharides

<table>
<thead>
<tr>
<th>Test number</th>
<th>Visual observation</th>
<th>Microencapsulation efficiency – MEE (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>NOD*</td>
<td>93.04</td>
</tr>
<tr>
<td>2</td>
<td>NOD*</td>
<td>84.70</td>
</tr>
<tr>
<td>3</td>
<td>NOD*</td>
<td>75.23</td>
</tr>
</tbody>
</table>

*NOD* = No visible oil droplets on the filter papers

Results from the calculation of MEE indicated that the highest MEE (93.04%) was obtained in Test1 with a comparatively low vegetable fat load. This is indicative of the fact that there exists a relationship between percent fat load and MEE as the higher the former the lower the latter. Jyothi and Co-workers [10] and Mutka and co-worker [16,17] also reported similar findings.

3.3 The Rate of Oxidative Deterioration of Encapsulated Oil

Microencapsulation, no matter how successful the process is achieved, can never be enough unless the shelf life or the stability of the encapsulated ingredients is encouraging. If the encapsulated material is unstable then the extent to which it is used is minimized. For this reason the peroxide value which is considered one of the most important chemical constraints in appraising the degree of fat deterioration, and hence the degree of utilization of microcapsules, was determined. The determination was carried out at room and an elevated temperature. Results obtained are shown Fig. 4.

The peroxide value of encapsulated lipid was fairly stable. Gradual increase in peroxide value was observed during the first 4 weeks when a sharp increase occurred between the 4th and 6th weeks.

![Fig. 4. Fat peroxidation in encapsulated sample stored at room temperature over time (weeks)](image1)

![Fig. 5. Fat peroxidation in encapsulated sample stored at 38°C over time (weeks)](image2)
The results in Fig. 5 show that the encapsulated lipids were sensitive to elevated temperature of 38ºC as the POV values obtained over the same period were much higher than those obtained at room temperature. This means that food materials requiring elevated temperatures are not suitable for the encapsulation process as the shelf life of the encapsulated lipid is shorter at storage temperatures of 38ºC which is in accordance with the findings reported by Jyothi and others [11].

4. CONCLUSION

The use of polysaccharides extracted from tofu processing wastewater as wall materials for encapsulation of oil has been successfully demonstrated. This has opened the door for further investigations for the possibilities of encapsulating other essential food ingredients. An encapsulation efficiency of about 90% was achieved. The shelf life for the micro gels produced could be up to 6 months. The waste material recovered could be used as a cheap source of wall material for the encapsulation of fat-soluble minerals and vitamins.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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