ENCAPSULATION OF HORSE RADISH PEROXIDASE INTO POLY(D,L-LACTIDE) BY THE MODIFIED PRECIPITATION METHOD

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Introduction

Injectable poly-D,L-lactide (PDLLA) microspheres containing proteins or peptides as controlled release devices have been widely used for the treatment of human diseases and animal health. Fundamental understanding of the relationship among the size of microspheres, encapsulation efficiency and protein release capacity are essential for the design of microsphere delivery systems [1,2].

The modified precipitation method [3-6] is a method of encapsulating hydrophilic drugs, especially protein and peptide ones, into microspheres. Since the release profiles of proteins dominantly depend on the morphology and nature of the polymer, drug distribution within microspheres and release temperature, the fabrication of microspheres with specific morphology and drug distribution is a challenge for chemical engineers [7]. PDLLA microspheres can protect proteins from chemical and biological inactivation and can ensure their release for long time frames, and at specified time. Finally, the size of the particles can be used to passively target the delivery of protein to physiological specific types of cells, such as professional antigen-presenting cells, or to target specific tissues [8].

Materials and Methods

![Diagram](image1.png)

**Table 1. Results of stereological analysis**

<table>
<thead>
<tr>
<th>Condition</th>
<th>Size (nm)</th>
</tr>
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<tbody>
<tr>
<td>Microsphere</td>
<td>460 ± 400</td>
</tr>
<tr>
<td>Microsphere</td>
<td>1130 ± 920</td>
</tr>
<tr>
<td>Microsphere</td>
<td>530 ± 720</td>
</tr>
<tr>
<td>Microsphere</td>
<td>350 ± 210</td>
</tr>
</tbody>
</table>

![Diagram](image2.png)

**Fig. 2. Results of stereological analysis**

**Fig. 3. Spectrophotometric analysis of PDLLA-HRP powder prepared by precipitation method**

Conclusions

HRP-loaded PDLLA particles were successfully obtained by precipitation method. PDLLA-HRP particles, prepared by modified precipitation method, have perfectly spherical shape, smooth surface and are non-agglomerated. In addition, the optimal particles were obtained with ethanol and 5% PVA. The mean diameter of the particles is 530 nm, and encapsulation efficiency is 48%. The main advantage of this method is that it does not require an increase in temperature and, therefore, may be useful when the heat-sensitive drugs, like proteins, are used.

**References**


**Fig. 4. DSC diagrams of PDLLA powder and PDLLA-HRP powder**

**Fig. 5. XRD diagrams of PDLLA, HRP and PDLLA-HRP powder**