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Microsphere Synthesis

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Protocol status: In development
We are still developing and optimizing this protocol

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Abstract

Microspheres are micron- sized plastic beads that have been used in drug delivery systems, typically with poly-lactide:glycolide (PLGA). The PLGA beads are able to encapsulate small molecules and they will degrade over time, releasing the drug. Synthesis of microspheres involves Water-in-Oil synthesis; PLGA or PLA is dissolved in Dicloromethane (DCM), creating the organic phase, and it is added drop-wise to an emulsifier (PVA), the aqueous phase, that is being mixed at a high rate of speed. The consistency of the beads and the size of the beads are dependent on mixing speed. This protocol aims to create PLA microspheres in order to create 'Mock organoids' utilizing the Water-in-Oil' synthesis methodology.

Materials

Dichloromethane (DCM),4% Polyvinyl Alcohol, PLA (preferably with color), Stir bar, Stir mantle, 10 mL Glass Scintillating vial or 50 mL Glass Pyrex bottle, 250 mL Glass Beaker, Secondary Containment, Glass Seriological (optional), Automated pipette aid (optional), Cell Strainer (100 microns), Water, Chemical Waste Beaker, 100 mL or 250 mL erlenmeyer flask, 250 mL Pyrex bottle, 0.22 micron filter that can attach to a pyrex bottle, aluminium foil, parafilm.

Before start

Collect all materials, and read SDS.

Dissolving PLA

1 Collect PLA and record the weight. Aim for 0.5-1 gram. 10m

2 Depending on the amount weighted, calculate the amount of DCM needed. The final concentration of PLA in DCM is 50 mg/mL 1m

3 In either a glass scintillation vial or a glass pyrex bottle, add the PLA to the DCM. DCM dissolves plastic, do not allow it to come in contact with the plastic lid of the scintillation vial or the pyrex bottle. Use aluminium foil and parafilm to make a makeshift cap. 4m

4 Wait till the PLA is fully dissolved 1h

Water-In-Oil emulsion

5 Create 50 mL of a 0.5% solution of PVA in water. 10m

6 Add the 0.5% PVA solution to a 250 mL Glass beaker. 1m

7 Place beaker onto a stirring mantle and add a stir bar to the bottom of the beaker. 1m

8 Set the speed to 50% -100% of the max speed; a vortex must be present. 5m

9 Add the PLA solution dropwise into the PVA. Allow the solution to stir for 5- 20 minutes. Continue to next step. 10m

Microsphere collection 18m

10 Place cell strainer on top of 100 mL or 250 mL erlenmeyer flask. 1m

11 Decant the PVA solution containing the microspheres over the 100 micron cell strainer. 1m

12 Wash the microspheres with excess water. Collect microspheres in a 50 mL conical tube. 5m

13 Add a 0.22 micron filter to a pyrex bottle, and decant the once filtered solution of PVA. 1m

14 Label the containers with chemical waste contents. 10m

Protocol references

Damptey, R., Torres, S., Cummings, L. *et al.* Synthesis and characterization of polylactic acid microspheres via emulsion-based processing. *MRS Advances* **8**, 982–987 (2023). https://doi.org/10.1557/s43580-023-00634-x