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Spectroscopic Evaluation of PEGDM Hydrogels for Osteogenic Progression Monitoring

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Abstract: To mitigate bone implant failure, PEGDM hydrogels were evaluated for their suitability for osteogenesis with the aid of Raman spectroscopy. Ability to detect hydroxyapatite as a spectral marker in hydrogels was also evaluated via Raman. © 2023 The Author(s)

1. Introduction

Bone damage affects over 100 million individuals within Europe, totaling €37.5 billion for osteoporotic bone damage alone [1]. Traditional treatment approaches involve autologous bone grafting to support osteogenesis. However, owing to limitations in autologous bone grafting engineered bone implants are becoming a popular regenerative medicine alternative. High failure rates (2-62%) of these implants [2] create a crucial need to improve newly formed bone quality and reduce healing times.

Histology, the traditional approach to assessing success of implantation, is destructive, requires staining and data acquisition takes place 4-6 weeks following implantation [3]. However, an approach that allows for longitudinal monitoring of mineral deposition would enable scaffold optimisation prior to implantation.

Thus, a hydrogel-based bone-on-chip (BOC) device coupled to a non-ionising optical spectroscopic technique such as Raman (Figure 1), allows for non-invasive, long-term monitoring of osteogenic progression in bone implants, understand bone formation mechanisms on hydrogel scaffolds and evaluate hydrogel scaffold quality.

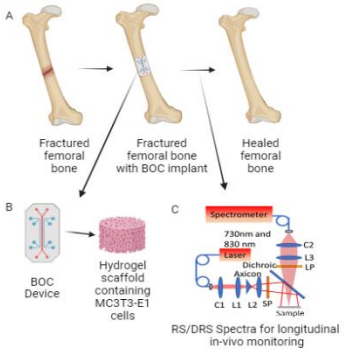


Figure 1: (A) Schematic representation of fracture healing with the aid of a BOC device. (B) BOC device containing MC3T3-E1 cells in a hydrogel scaffold. (C) In-vitro longitudinal data acquisition for osteogenic progression monitoring using optical spectroscopy.

2. Materials and Methods

Owing to its ability to promote osteogenesis [4], the suitability of poly(ethyleneglycol) dimethacrylate (PEGDM) hydrogels as a BOC substrate was evaluated with the aid of Raman spectroscopy.

PEGDM hydrogels of concentrations 10%, 15%, 20% and 30% were prepared in phosphate buffered saline (PBS) by curing under a UV light source at 400 nm for 15 minutes with the aid of a photoinitiator, Irgacure 2959.

The curing process was optimised with respect to Irgacure concentration and UV intensity to create stable hydrogels. Raman spectra were acquired at 730 nm, 785 nm and 830 nm to visualise the entire range of wavenumbers. The mechanical strength of PEGDM hydrogels were evaluated with the aid of an Instron 5565 mechanical testing system. Agarose hydrogels were used as a control substrate for PEGDM.

The feasibility of Raman spectroscopy for osteogenesis monitoring was determined by hydroxyapatite (HA) detection, which was incorporated into varying concentrations of PEGDM and agarose hydrogels. HA is an inorganic mineral found in bone, which is expected to be present in increasing quantities on scaffolds as bone growth occurs, enabling it to act as a spectral marker for longitudinal monitoring.

3. Results and Discussion

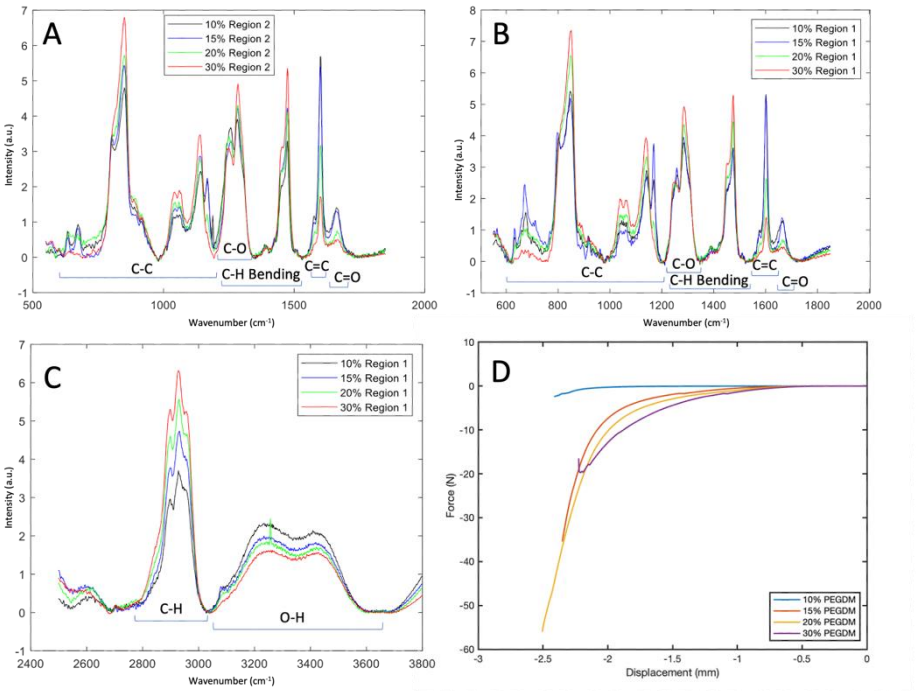


Figure 2: (A-C) Raman spectra of 10%-30% PEGDM hydrogels acquired at (A) 785 nm, (B) 830 nm and (C) 730 nm. (D) Compression curves of 10%-30% PEGDM hydrogels.

As indicated by the Raman spectra in Figure 2 (A-C), higher PEGDM concentrations corresponds to a higher degree of polymerisation, leading to increased spectral intensities of bonds associated with the PEGDM polymer (C-C, C-O, C-H), while bonds that undergo photopolymerisation (C=C, C=O) decrease in intensity. A lowering of the OH peak intensity at 730 nm (Figure 2C) could also be observed as the polymer concentration was increased, indicating a higher degree of crosslinking and a higher polymer density. Pure PEGDM and Irgacure 2959 powders and solutions indicated Raman spectra different to cured PEGDM hydrogels, owing to polymerisation and the presence of water in solution phase. Agarose hydrogels showed similar variations in their Raman spectra, however, it should be noted that agarose gelation doesn't undergo UV-assisted curing. It was also observed that PEGDM hydrogels increased in rigidity and became prone to breakage with increasing polymer concentration. This is indicated by the reducing area under the curve in the compression curve (Figure 2D), denoting a decrease in work required to break the hydrogel samples as concentration increased. HA detection by Raman spectroscopy was observed, where HA related spectral intensities increased as the amount of HA incorporated into the hydrogel increased.

4. Conclusion

Increased PEGDM polymer concentrations indicated an increase in the degree of polymerisation and in the rigidity of the hydrogels. Raman spectroscopy was observed to be a suitable means for longitudinal osteogenesis monitoring via the detection of HA present in hydrogel samples. As next steps, fabrication of the BOC device and cell culture studies using the mouse MC3T3-E1 cell line on the PEGDM substrate will be conducted.

5. References

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