WHEY PROTEIN ISOLATE-ARAGONITE COMPOSITES FOR BONE TISSUE ENGINEERING

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Introduction

Hydrogels, or highly hydrated three-dimensional polymer networks can be improved for applications in bone regeneration by mineralization to create hydrogel-inorganic composites. In this study, whey protein isolate (WPI) hydrogels were mineralized by incorporation of preformed aragonite particles. WPI is a by-product from the production of cheese and Greek yoghurt. Hence, its usage is advantageous for environmental and financial reasons. Also, previously it has been demonstrated that WPI in solution promotes proliferation and osteogenic differentiation of cells [1]. WPI hydrogels can be formed by heat sterilization, e.g. autoclaving. Aragonite is a polymorph of calcium carbonate ($CaCO_3$). It has successfully been used to promote bone regeneration [2].

Materials and Methods

Hydrogel-$CaCO_3$ composites were produced by the heat-induced gelation of 40% WPI solution, with 0, 100, 200 or 300 mg/ml aragonite particles added (denoted as WPI, WPI/100$CaCO_3$, WPI/200$CaCO_3$, WPI/300$CaCO_3$). 1 ml composites were formed in 2 ml Eppendorf tubes. Composite properties were investigated by swelling studies, degradation (BCA assay), morphology (SEM), structure (FTIR, Raman spectroscopy), mechanical properties (compressive modulus), particle distribution (Micro-CT imaging) and cytocompatibility (cell metabolic activity and alkaline phosphatase activity (ALP)) using MG63 osteoblast-like cells, after autoclaving.

Results and Discussion

Particles had a positive impact on mechanical properties. The highest compression modulus was observed in WPI/300$CaCO_3$ hydrogels *c.a* 3.15 MPa (FIG. 1). SEM and Micro-CT analyses suggested that aragonite particles were uniformly distributed within hydrogels (FIG. 2). MG63 metabolic activity and ALP activity were also highest for WPI/300$CaCO_3$ hydrogels, suggesting positive effect of aragonite incorporation on MG63 cell survival and early osteogenic differentiation, respectively.

Conclusions

Physicochemical, mechanical and cytocompatibility studies indicated that WPI/300$CaCO_3$ were most suitable for cell growth and possibly bone tissue engineering applications.

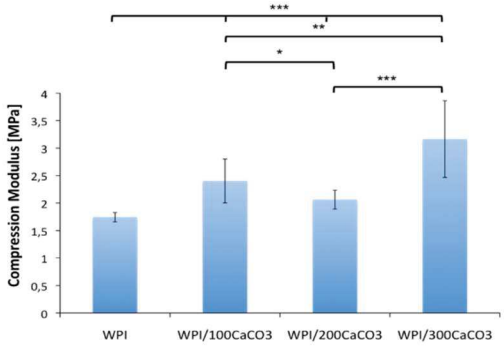

FIG. 1. Compressive modulus of composites

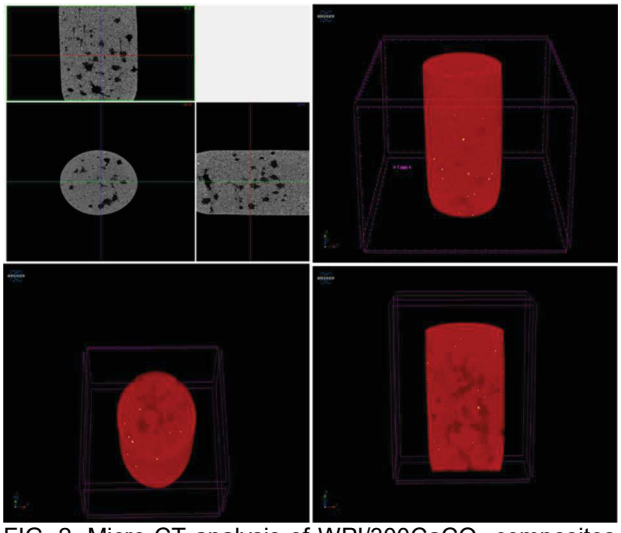

FIG. 2. Micro-CT analysis of WPI/300$CaCO_3$ composites (diameter 8 mm). Top left and bottom right: cross-sections Red: hydrogel. Yellow: $CaCO_3$

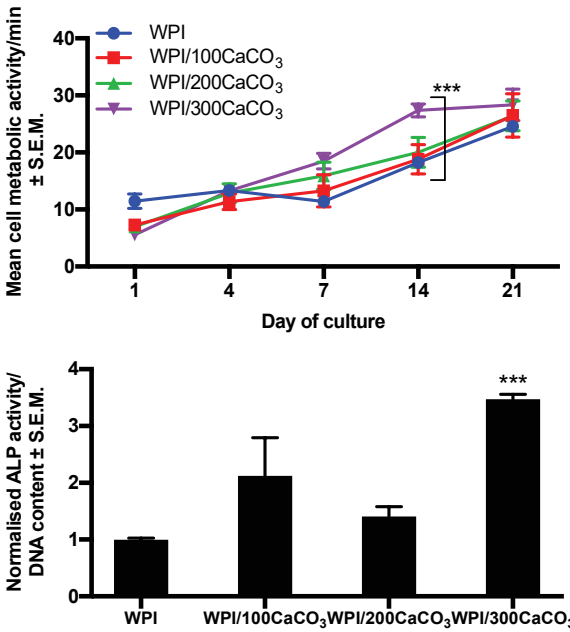

FIG. 3. Metabolic activity (top) and ALP activity (bottom) of MG63 cells on composites after 21 d.

Acknowledgment

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References

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