

WHEY PROTEIN ISOLATE COATINGS FOR BIOMATERIALS

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[ENGINEERING OF BIOMATERIALS 153 (2019) 48]

Introduction

Whey protein isolate (WPI) is a by-product of the dairy industry. Its main component β -lactoglobulin is able to assemble into fibrils with functional properties that can serve as a new coating material for biomaterial surfaces. The aim was to develop sterile fibrillar coatings on glass substrates and assess the effect of the coatings on growth, morphology and differentiation of human bone marrow stromal cells (hBMSC). Three sample groups were compared: (i) uncoated glass and glass coated with fibrils formed at (ii) pH 2 and (iii) pH 3.5.

Materials and Methods

For the fibril formation a 2.5 wt% WPI (BiPro, Davisco Foods International Inc., USA) solution was incubated at 90°C for 5 h under stirring conditions at a low pH (2, 3.5). The fibrillar suspension was placed on glass coverslips of diameter 1 cm to allow adsorption of fibrils to the surface. After rinsing and drying, glass substrates coated with fibrils were autoclaved at 121°C for 15 minutes. Coating morphology was assessed using scanning electron microscopy (SEM). hBMSC (primary osteoblast-like cells) were seeded at a density of 5,555 hBMSC/cm² in DMEM with 10% heat-inactivated fetal calf serum with antibiotics (BM), from day 4 in BM with 10 mM β -glycerophosphate and 300 μ M ascorbate (OM/D); medium was changed twice per week. Metabolic activity was assessed using the MTS assay on day 2 & 4, cell morphology by immunofluorescence staining 2 h after seeding, differentiation by activity of tissue non-specific alkaline phosphatase (TNAP) on day 11.

Results and Discussion

SEM confirmed the presence of fibrillary coatings after autoclaving (FIG. 1). Cell adhesion was observed on all sample groups, but spreading and staining intensity were superior on coated samples (FIG. 2). TNAP activity was observed on all sample groups (FIG. 3).

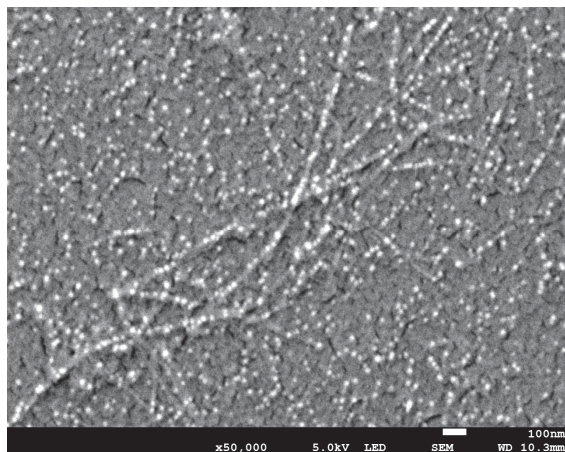


FIG. 1. SEM image of WPI fibrillary coating formed at pH 2 after autoclaving. Scale bar = 100 nm.

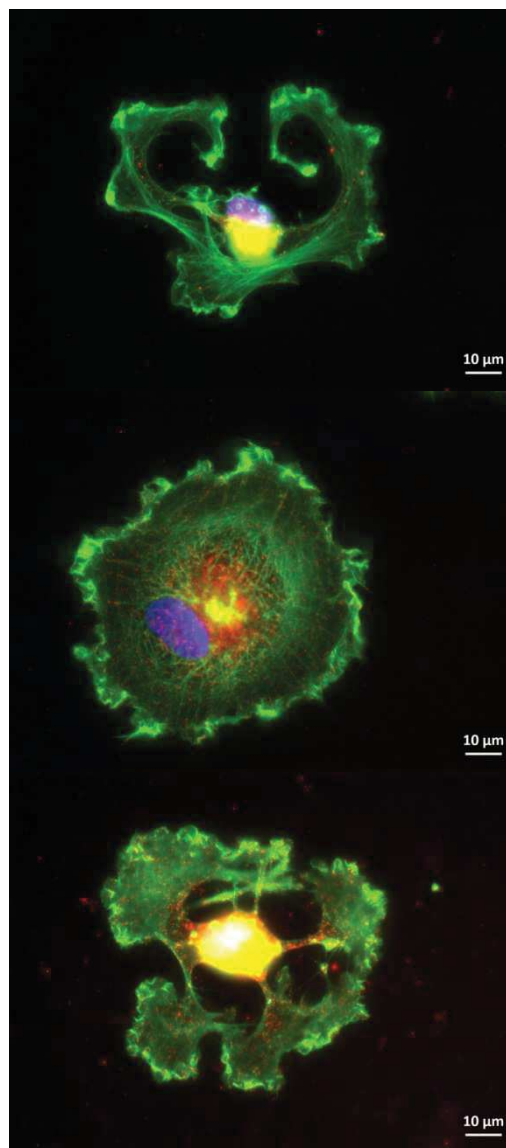


FIG. 2. Morphology of hBMSC on glass (top), fibrils formed at pH 2 (middle) and pH 3.5 (bottom), 2 h after seeding. Scale bar = 10 μ m.

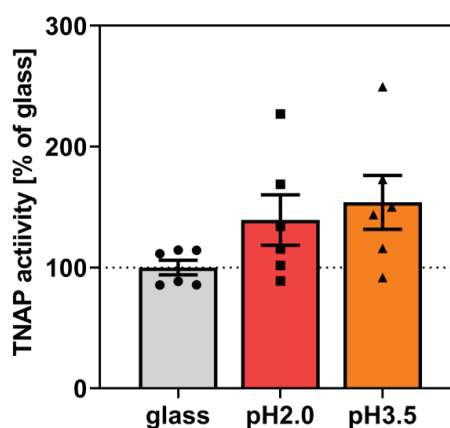


FIG. 3. TNAP activity in hBMSC cultured for 11 days in differentiation medium, n=3.

Conclusions

Coatings withstand autoclaving and support adhesion and differentiation of hBMSC. This paves the way for enhancement of the fibrils with further biomolecules.

Acknowledgment

N8 Agrifood grant "Food2Bone" (T.E.L.D.), German Research Foundation (DFG), SPP 1934 DiSPBiotech (project number 273937032) (J.K.K.).