

Valorization of Sea Urchin Wastes: Characterization of Marine Collagen Peptides

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Abstract

The purpose of the recently launched CIRCULAR and BRITeS projects is the recycling of wastes from the sea urchin food industry and its valorization in diversified products, including innovative collagen-based biomaterials for applications in regenerative medicine. In previous works, we have developed and characterized prototypes of biodegradable medical devices (skin substitute) made of this eco-friendly marine collagen, analyzing their microstructure, mechanical performances and *in vitro* cytocompatibility. In the current work, we want to further characterize them in terms of the amino acid (aa) composition of the starting material (collagen) and the *in vitro* degradation rates in both physiological (PBS) and enzymatic (collagenase) conditions. We also evaluated the antioxidant activity and *in vitro* cytotoxicity of the biomaterial degradation products. Indeed, the *in vivo* physiological degradation of collagen-based biomaterials leads to the formation of collagenous peptides, which can have anti-inflammatory and antioxidant effects and could in turn become "bioactive molecules" useful in the regenerating tissue microenvironment. Analysis of the amino acid profile of sea urchin-derived collagen confirmed that it is composed of glycine, hydroxyproline and proline and its overall aa composition is similar to that of human collagen, with some small exceptions for alanine, arginine, methionine and glutamic acid. The degradation test showed that, in enzymatic condition, the biomaterial is degraded by 66% after 48 hours and completely degraded after 10 days. In PBS, as expected, the degradation rate was slower than in collagenase, with a reduction of 54% after 10 days. A commercially available bovine collagen membrane (Integra) was used as control. In collagenase, Integra is degraded by less than 10% after 48 hours and not yet completely degraded after 3 weeks, while in PBS it remains intact even after 10 days. The faster degradation rate of the sea urchin biomaterial, due to a lack of cross-linking procedures, may promote a better incorporation *in vivo* conditions. Preliminary analysis of the antioxidant activity of collagen peptides suggests that these may have a potential role as a radical scavenger

and thus anti-inflammatory biomolecules. Finally, the *in vitro* tests on human fibroblasts showed that at short time (24h) high concentrations (100 µg/mL) of collagen peptides increase vitality (and, indirectly, proliferation) of human dermal fibroblasts. Overall, this work has strengthened the potential usefulness of our innovative and environmentally friendly biomaterial in tissue regeneration. Further *in vivo* studies will provide information on the actual regeneration efficacy of this new biomaterial.

Keywords: collagen peptides; sea urchin; waste valorization; circular economy

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