A novel collagen skin-like scaffold improves skin regeneration in sheep

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Skin wound healing is a multi-phase dynamic process fundamental for the restoration of skin integrity. Skin wounds are a common occurrence in Veterinary Medicine and an inadequate wound care management might lead to a poor prognosis, with a consequent impact on the health of the animal and the economic sector. Conventional treatments (e.g. autografts) have many limitations and do not provide a proper wound healing. Tissue engineering techniques could provide a solution by the production of scaffolds able to support skin regeneration, in function and structure, and promote wound closure. In this work, we describe the application of a novel designed collagen-based skin-like scaffold (CBSS) [1], produced with collagen extracted from sea urchin food wastes, for the treatment of experimental second intention healing wounds in the sheep (approved by the Italian Ministry of Health n°51/2015-PR, in accordance with the Body for the Protection of Animals (OPBA)). After 7, 14, 21 and 42 days from wound creation, clinical observations were performed along with skin biopsies collected for histopathology (H&E and IHC for Ki67 and α-smooth muscle actin (αSMA)) and molecular analysis (RT-PCR for collagen type I and III, VEGF and hair-Keratin (hKER)) for untreated (control) and treated (CBSS) wounds. CBSS-treated wounds showed a higher inflammatory cell infiltration than control wounds at 7 days; nonetheless, at 42 days treated wounds showed no inflammation while it was still present in control wounds. The CBSS application led to a higher deposition of granulation tissue (GT) at day 7 while in control wounds it was still low. In treated wounds, the amount of GT started to diminish by day 14 while increasing in control wounds. These results were reflected by the gene expression analysis of the immature (type III) and mature (type I) form of collagen: while in the control wounds type III expression was increasing, reaching its peak at 21 days, in CBSS-treated wounds the up-regulated one was the mature type I. This scenario resulted into an appropriate maturation (also supported by the induced gene expression of VEGF) of the GT in treated wounds at 42 days while the higher amount of collagen type III led to dermal fibrosis in control wounds, also characterized by αSMA immunopositivity. Moreover, the application of the CBSS led to a higher re-epithelialization rate at 14 and 21 days in comparison to the control; concomitantly, a higher immunopositivity for Ki67 (marker of proliferation) was observed in the basal layer of the newly formed epidermis. Histologically, in treated wounds along with the earlier presence of a neoepidermis at day 14, it was also observed the appearance of skin adnexa with the concomitant gene expression of hKER, absent in control wounds until day 42. In conclusion, the application of a collagenous biomaterial anticipated the inflammatory phase, promoted the maturation and remodelling of the GT into a mature and well-organized dermis along with skin adnexa,

and accelerated the re-epithelialization process. Overall, these preliminary findings suggest that this marine collagen scaffold possesses regenerative properties, worthy of further investigations, might be a useful tool for the treatment of second intention healing skin wounds in Veterinary patients.

[1] Ferrario et al. From Food Waste to Innovative Biomaterial: Sea Urchin-Derived Collagen for Applications in Skin Regenerative Medicine, Marine Drugs, Aug 6;18(8):414, 2020.