1 Abstract

Within the last decades the classical wound treatment has fundamentally been revolutionized. Functions of wound dressings should optimally include a targeted stimulation of particular biological processes. As the direct application of growth factors had only moderate success, the modulation of their activity has been suggested as a promising target. The objective of this study was to investigate the effects of glycosaminoglycans (GAGs) as growth factor modulating compounds on keratinocyte functions. The predominant GAG in the extracellular matrix (ECM) of the skin is chondroitin/dermatan sulfate (CS/DS). CS/DS are highly-sulfated linear polysaccharides composed of defined, repeating disaccharide units. Distinct sulfation patterns are known to specifically modulate the function of growth factors like the fibroblast growths factor 7 (FGF7). The clinical consequences of a disrupted CS/DS pattern became obvious in patients suffering from connective tissue disorders summarized as Ehlers-Danlos syndrome (EDS). A mouse model, lacking the proteoglycan decorin (*Dcn*$^{-/-}$) with altered GAG fine structure, presented a skin and wound healing phenotype similar to the human EDS. The results indicated that CS/DS were able -even in the absence of exogenous FGF7- to induce or inhibit the proliferation of keratinocytes. In this context the microheterogeneity of the GAGs correlated with their biological function. While *Dcn*$^{-/-}$ CS/DS with reduced 2-O-sulfation and altered IdoA blocks was able to trigger proliferation of keratinocytes in higher concentrations, CS/DS from wild-type mice blocked proliferation. DS from porcine mucosa also promoted proliferation, unlike to shark cartilage 6-sulfated CS (CS6S). Although wild-type and *Dcn*$^{-/-}$ CS/DS had a different impact on keratinocyte proliferation, both GAGs activated extracellular regulated kinase-1, 2 to a similar extent. In order to explain this contradiction, the influence on the differentiation of keratinocytes was studied. 3D fibroblasts embedded in their own matrix were used to establish an *in vivo* like co-culture system, which enabled the multilayered growth of keratinocytes. Indeed, CS/DS also differentially affected the differentiation of keratinocytes. *Dcn*$^{-/-}$ CS/DS treated co-cultures pointed out a decreased involucrin expression indicating a delayed differentiation of keratinocytes. However, wild-type CS/DS and DS treatment did not influence differentiation. In conclusion, GAGs with distinct fine structure properties had different impact on keratinocyte proliferation and differentiation.

Water-soluble plant arabinoxylans from *Plantago ovata* seed husks (P1) enhanced keratinocyte proliferation at similar dosages as CS/DS. The analysis of the monosaccharide linkage pattern revealed highly branched xylose nodal points within the complex heteropolysaccharide. P1 was clathrin-independently internalized into keratinocytes and accumulated in the lysosomes for at least 24 h.

In summary, polysaccharides can improve the keratinocyte driven reepithelialization and therefore their application is suggested as a potential wound healing therapeutic.