application for the assessment of physiological and pathological states of connective tissues. J Chromatogr B. 2002;781:21-38.). The resulting disaccharide mixtures can be analyzed by PAGE (Hampson IN, Gallagher JT. Separation of radiolabeled glycosaminoglycan oligosaccharides by polyacrylamide-gel electrophoresis Biochem J. 1984;221:697-705.), SAX-HPLC (Skidmore M AA, Yates E and Turnbull JE. Labelling heparan sulfate saccharides with chromophore, fluorescence and mass tag for HPLC and MS separations. Methods in Molecular biology. 2009;534:157-69.), or highly sensitive capillary electrophoresis (CE) (Lamari F, Militsopoulou M, Gioldassi X, Karamanos NK. Capillary electrophoresis: a superior miniaturized tool for analysis of the mono-, di-, and oligosaccharide constituents of glycan moieties in proteoglycans. Fresenius J Anal Chem. 2001;371:157-67.; Karamanos NK, Vanky P, Tzanakakis GN, Hjerpe A. High performance capillary electrophoresis method to characterize heparin and heparan sulfate disaccharides. Electrophoresis. 1996;17:391-5.; Sudhalter J, Folkman J, Svahn CM, Bergendal K, Damore PA. Importance of size, sulfation, and anticoagulant activity in the potentiation of acidic fibroblast growth-factor by heparin J Biol Chem. 1989;264:6892-7.; Militsopoulou M, Lamari FN, Hjerpe A, Karamanos NK. Determination of twelve heparin- and heparan sulfate-derived disaccharides as 2-aminoacridone derivatives by capillary zone electrophoresis using ultraviolet and laser-induced fluorescence detection. Electrophoresis. 2002;23:1104-9) by comparison to known disaccharides standards.

**Summary**

In one aspect, the present invention refers to an isolated or substantially purified heparan sulphate HS6, wherein the heparan sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1), wherein following digestion with heparin lyases I, II and III and then subjecting the resulting disaccharide fragments to HPLC analysis the heparan sulphate HS6 has a disaccharide composition comprising:

| Disaccharide | Normalised weight percentage |
|---|---|
| ΔUA,2S-GlcNS,6S | 12.9 ±3.0 |
| ΔUA,2S-GlcNS | 6.8 ±2.0 |
| ΔUA-GlcNS,6S | 11.7 ±3.0 |
| ΔUA-GlcNS | 24.5 ±3.0 |
| ΔUA,2S-GlcNAc | 0.5 ±0.5 |
| ΔUA-GlcNAc,6S | 12.1 ±3.0 |
| ΔUA-GlcNAc | 31.6 ±3.0 |

In another aspect, the present invention refers to a composition comprising isolated or substantially purified heparan sulphate HS6 according to the present invention and a growth factor, preferably PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B. In still another aspect, the present invention refers to the use of isolated or substantially purified heparan sulphate HS6 in the manufacture of a medicament for the treatment of a disease, condition or injury to tissue, wherein the heparan sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1), and wherein the method involves the repair and/or regeneration of skin tissue. In one aspect, the present invention refers to a biocompatible implant or prosthesis comprising a biomaterial and isolated or substantially purified heparan sulphate HS6 according to the present invention. In one aspect, the present invention refers to the use of isolated or substantially purified heparan sulphate HS6 in the manufacture of a medicament for use in a therapeutic method of repair and/or

regeneration of skin, wherein the heparin sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1). In one aspect, the present invention refers to a method of stabilising PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B, the method comprising contacting PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B *in vitro* with isolated or substantially purified heparan sulphate HS6 according to the present invention. In one other aspect, the present invention refers to a method of preserving biological material, the method comprising contacting the biological material *in vitro* with isolated or substantially purified heparan sulphate HS6 according to the present invention, wherein the biological material is cellular material, tissue, blood, a blood-derived product, cells, or stem cells. In one other aspect, the present invention refers to a preparation comprising biological material and a preservative, wherein the preservative comprises isolated or substantially purified heparan sulphate HS6 according to the present invention, wherein the biological material is cellular material, tissue, blood, a blood-derived product, cells, or stem cells. In one aspect, the present invention refers to a method of isolating, processing, culturing or expanding stem cells, the method comprising contacting the stem cells with isolated or substantially purified heparan sulphate HS6 according to the present invention.

The present disclosure concerns a heparan sulphate species and heparan sulphate preparations comprising or consisting of the heparan sulphate species. The heparan sulphate species is called HS6 (or HS6+). HS6 refers to a novel class of structurally and functionally related isolated heparan sulphate.

HS6 has been found to bind PDGF-B and/or PDGF-BB. HS6 has also been found to enhance the proliferation of human dermal fibroblasts *in vitro*, enhance migration of human dermal fibroblasts in an *in vitro* model of wound healing, and enhance wound healing *in vivo*. HS6 has also been found to increase/enhance binding of PDGF-BB to PDGFRβ.

In one example of the present disclosure a heparan sulphate HS6 is provided. HS6 may be provided in isolated form or in substantially purified form. This may comprise providing a composition in which the heparan sulphate component is at least 80% HS6, more preferably one of at least 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%.

**Claims:**

1. Isolated or substantially purified heparan sulphate HS6, wherein the heparan sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1), wherein following digestion with heparin lyases I, II and III and then subjecting the resulting disaccharide fragments to HPLC analysis the heparan sulphate HS6 has a disaccharide composition comprising:

| Disaccharide | Normalised weight percentage |
|---|---|
| ΔUA,2S-GlcNS,6S | 12.9 ±3.0 |
| ΔUA,2S-GlcNS | 6.8 ±2.0 |
| ΔUA-GlcNS,6S | 11.7 ±3.0 |
| ΔUA-GlcNS | 24.5 ±3.0 |
| ΔUA,2S-GlcNAc | 0.5 ±0.5 |
| ΔUA-GlcNAc,6S | 12.1 ±3.0 |
| ΔUA-GlcNAc | 31.6 ±3.0 |

2. Isolated or substantially purified heparan sulphate HS6 according to claim 1, obtained by a method comprising:

(i) providing a solid support having polypeptide molecules adhered to the support, wherein the polypeptide comprises a heparin-binding domain having the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK;

(ii) contacting the solid support with a mixture comprising glycosaminoglycan, preferably a heparan sulphate preparation, such that polypeptide-glycosaminoglycan complexes are allowed to form;

(iii) partitioning polypeptide-glycosaminoglycan complexes from the remainder of the mixture;

(iv) dissociating glycosaminoglycans, preferably heparan sulphate species, from the polypeptide-glycosaminoglycan complexes;

(v) collecting the dissociated glycosaminoglycans, preferably one or more heparan sulphate species.

3. Isolated or substantially purified heparan sulphate HS6 according to claim 2 wherein the mixture comprising glycosaminoglycans is a heparan sulphate preparation obtained from porcine mucosa ($HS^{PM}$).

4. A composition comprising isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3 and a growth factor, preferably PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B.

5. Use of isolated or substantially purified heparan sulphate HS6 in the manufacture of a medicament for the treatment of a disease, condition or injury to tissue, wherein the heparan sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1), and wherein the method involves the repair and/or regeneration of skin tissue.

6. A biocompatible implant or prosthesis comprising a biomaterial and isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3.

7. Use of isolated or substantially purified heparan sulphate HS6 in the manufacture of a medicament for use in a therapeutic method of repair and/or regeneration of skin, wherein the heparan sulphate HS6 is capable of binding a peptide or polypeptide having, or consisting of, the amino acid sequence RAKTPQTRVTIRTVRVRRPPKGKHRKFKHTHDK (SEQ ID NO:1).

8. The use of claim 7 wherein the therapeutic method involves treatment of a skin wound or scar, optionally selected from skin burns, ulcers, excisional wounds, cut, stab or puncture wounds.

9. The use of claim 7 wherein the therapeutic method involves skin graft healing, skin reconstruction, or skin plastic surgery.

10. The use of any one of claims 7 to 9 wherein the isolated or substantially purified heparan sulphate HS6 is formulated for topical or transdermal administration.

11. The use of any one of claims 7 to 10 wherein the isolated or substantially purified heparan sulphate HS6 is formulated as a gel, spray, paste, ointment, cream, lotion, salve, oil, aqueous solution, suspension, dispersion, patch, adhesive plaster, bandage, dressing, depot, or reservoir.

12. The use of any one of claims 7 to 11 wherein the method further comprises administration of a growth factor, preferably PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B.

13. The use of any one of claims 7 to 12, wherein the isolated or substantially purified heparan sulphate HS6 is formulated as a combined preparation with a growth factor, preferably PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B.

14. A method of stabilising PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B, the method comprising contacting PDGF-B, PDGF-BB or a heterodimer comprising PDGF-B *in vitro* with isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3.

15. A method of preserving biological material, the method comprising contacting the biological material *in vitro* with isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3, wherein the biological material is cellular material, tissue, blood, a blood-derived product, cells, or stem cells.

16. A preparation comprising biological material and a preservative, wherein the preservative comprises isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3, wherein the biological material is cellular material, tissue, blood, a blood-derived product, cells, or stem cells.

17. A method of isolating, processing, culturing or expanding stem cells, the method comprising contacting the stem cells with isolated or substantially purified heparan sulphate HS6 according to any one of claims 1 to 3.