CLAIMS

1. A glycosaminoglycan comprising 78-99% N-sulfated (NS) disaccharide group and having a weight average molecular weight appropriate to form a final heparin product of 15,000-19,000 Da, the percentage of heparin chains with a molecular weight of greater than 24,000 Da being not more than 20% of the total, and the ratio of chains between molecular weights of 8,000 to 16,000 Da to chains with molecular weights of 16,000 to 24,000 Da being not less than 1.0.

2. The glycosaminoglycan of claim 1 comprising 81-97% of N-sulfated (NS) disaccharide group.

3. The glycosaminoglycan of claim 1 comprising 83-95% of N-sulfated (NS) disaccharide group.

4. The glycosaminoglycan of claim 1 comprising 85-93% of N-sulfated (NS) disaccharide group.

5. A glycosaminoglycan comprising 44-80% of N-sulfated, 2-sulfated (NS2S) disaccharide group and 13-39% of NS group and having a weight average molecular weight appropriate to form a final heparin product of 15,000-19,000 Da, the percentage of heparin chains with a molecular weight of greater than 24,000 Da being not more than 20% of the total, and the ratio of chains between molecular weights of 8,000 to 16,000 Da to chains with molecular weights of 16,000 to 24,000 Da being not less than 1.0.

6. The glycosaminoglycan of claim 5 comprising 44-80% of NS2S group.

7. The glycosaminoglycan of claim 5, comprising 50-78% of NS2S group.

8. The glycosaminoglycan of claim 5, comprising 55-77% of NS2S group.

9. The glycosaminoglycan of claim 5, comprising 60-76% of NS2S group.

10. The glycosaminoglycan of claim 5, comprising 14-35% of NS group.

11. The glycosaminoglycan of claim 5, comprising 15-30% of NS group.

12. The glycosaminoglycan of claim 5, comprising 16-26% of NS group.

13. A glycosaminoglycan comprising 36-70% of N-sulfated, 2-sulfated, 6-sulfated (NS2S6S) disaccharide group, 6-32% of N-sulfated, 6-sulfated (NS6S) disaccharide group, 0-27% of NS2S group and 1-22% of NS group and having a weight average molecular weight properties appropriate to form a final heparin product of 15,000-19,000 Da, the percentage of heparin chains with a molecular weight of greater than 24,000 Da being not more than 20% of the total, and the ratio of chains between molecular weights of 8,000 to 16,000 Da to chains with molecular weights of 16,000 to 24,000 Da being not less than 1.0.

14. The glycosaminoglycan of claim 13 comprising 40-67% of NS2S6S disaccharide group and 6-26% of NS6S disaccharide group

15. The glycosaminoglycan of claim 13 comprising 43-64% of NS2S6S disaccharide group and 6-22% of NS6S disaccharide group.

16. The glycosaminoglycan of claim 13 comprising 12-27% of NS2S group and 1-17% of NS group.

17. The glycosaminoglycan of any one of claims 13 to 16, which does not contain 3S disaccharide groups.

18. A method for producing a biosynthetic heparin, comprising;

a. obtaining a glycosaminoglycan comprising 36-70% of NS2S6S disaccharide group, 6-32% of NS6S disaccharide group, 0-27% of NS2S group and 1-22% of NS group;

b. treating the glycosaminoglycan with an enzyme, which is 3-O-sulfotransferase isoform 1 (3OST-1), in the presence of a sulfate donor.

to produce a biosynthetic heparin batch.

19. The method of claim 18, wherein in step (b) sufficient glucosamine residues are converted to 3-O-sulfoglucosamine residues so that the produced batch has anti-Xa and anti-IIa activities of no less than 180 units/mg and an anti-Xa/anti-IIa ratio of 0.85-1.15.

20. The method of claim 18, wherein the anti-Xa/anti-IIa ratio is 0.9-1.1.

21. A method of making a second glycosaminoglycan intermediate comprising 44 to 80% of NS2S group and 13 to 39% of NS group; the method comprising:

a. converting an amount of N-acetyl glucosamine residues in heparosan to produce a first glycosaminoglycan comprising 78-99% of N-sulfated (NS) disaccharide group, wherein the amount of the converted N-acetyl glucosamine residues corresponds to the amount of the NS group in the first glycosaminoglycan intermediate; and

b. treating the first glycosaminoglycan intermediate with an enzyme, which is C5-epimerase (C5-epi) and 2-O-sulfotransferase (2OST), in the presence of a sulfate donor

to produce the second glycosaminoglycan intermediate.

22. The method of claim 21, wherein said converting comprises reacting the heparosan with a base and a sulfonating reagent.

23. The method of claim 21, wherein said converting comprises reacting the heparosan with N-deacetylase, N-sulfotransferase (NDST).

24. A method of making a third glycosaminoglycan intermediate comprising 36-70% of NS2S6S disaccharide group, 6-32% of NS6S disaccharide group, 0-27% of NS2S group and 1-22% of NS group:

the method comprising

treating a second glycosaminoglycan intermediate comprising 44-80% of NS2S group and 13-39% of NS group with an enzyme, which is 6-O-sulfotransferase isoforms 1 and/or 3 (6OST-1/3), in the presence of a sulfate donor to convert the second glycosaminoglycan intermediate to the third glycosaminoglycan intermediate.

25. The method of any one of claims 18-24, wherein the sulfate donor comprises PAPS.

26. The method of claim 25, wherein PAPS is in a solution.

27. The method of any one of claims 18-24, wherein the sulfate donor comprises PAP and PNPS.

28. The method of any one claims 18-24, wherein said treatment is performed in the presence of a recycling system comprising PAPS, PNPS and a catalyst.

29. The method of claim 28, wherein the catalyst is aryl sulfotransferase IV (AST-IV).

30. The method of any one of claims 18-29, wherein the enzyme is in solution.

31. The method of any one of claims 18-29, wherein the enzyme is immobilized.

32. The method of any one of claims 18-24, wherein said treating is performed within an engineered microbial strain containing the genes encoding for NDST, C5-Epi, 2OST, 3OST-1, one or both of 6OST-1 or 6OST-3, and a source of PAPS.