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**Differential Scanning Fluorimetry measurement of protein stability changes upon
binding to glycosaminoglycans: a rapid screening test for binding specificity**

Figure S-1 Sequence data for tested FGF-s

Figure S-2 Sulfation pattern of the major repeating disaccharides of the heparin
derivatives

A)

FGF-1 (aFGF)

UniProt Accession: P05230 (isoform 1)

MAEGEITTTALTEKFNLPNGYKKPKLLYCSNGGHFLRLPDGTVDGTRDRSDQHIQLQLSAESVGEVYIKSTETGQYLAMDDTGLLYGSQTPNE
ECLFLERLEENHYNTYISKKHAENWFVGLKKNKGSCKRGPRTHYGQKAILFLPLPVSSD

Residues: 1-155

Expressed / assayed protein:

**MFNLPPGNYKKPKLLYCSNGGHFLRLPDGTVDGTRDRSDQHIQLQLSAESVGEVYIKSTETGQYLAMDDTGLLYGSQTPNEECLFLERL
EENHYNTYISKKHAENWFVGLKKNKGSCKRGPRTHYGQKAILFLPLPVSSD**

Residues: 1 (Met), 16-155

B)

FGF-2 (bFGF)

UniProt Accession: P09038-2 (isoform 3)

MAAGSITTLPALPEDGGSGAFPPGHFKDPKRLYCKNGGFLLRIHPDGRVDGVREKSDPHIKLQLQAEERGVSISIKVCANRYLAMKEDGRLLASK
CVTDECFERLESNNYNTYRSRKYTSWYVALKRTGQYKLGSKTGPGQKAILFLPMSAKS

Residues: 1-155

Expressed / assayed protein:

**MAAGSITTLPALPEDGGSGAFPPGHFKDPKRLYCKNGGFLLRIHPDGRVDGVREKSDPHIKLQLQAEERGVSISIKVCANRYLAMKEDGR
LLASKCVTDECFERLESNNYNTYRSRKYTSWYVALKRTGQYKLGSKTGPGQKAILFLPMSAKS**

Residues: 1-155

C)

FGF-18 (zFGF5)

UniProt Accession: O76093

MYSAPSACTCLCLHFLLLCFQVQVLVAEENVDFRIHVENQTRARDDVSRKQLRLYQLYSRTSGKHIQVLGRRISARGEDGDKYAQLLVETDTFGS
QVRIKGKETEFYLCMNRKGKLVGKPDGTSKECVFIEKVLENNYTALMSAKYSGWYVGFTKKGRPRKGPKTRENQQDVHFMKRYPKGQPELQKP
FKYTTVTKRSRIRPHTPA

Residues: 1-207

Expressed protein:

**MKHHHHHHHPMSDYDIPTTENLYFQGAMEENVDFRIHVENQTRARDDVSRKQLRLYQLYSRTSGKHIQVLGRRISARGEDGDKYAQLLVET
DTFGSQVRIKGKETEFYLCMNRKGKLVGKPDGTSKECVFIEKVLENNYTALMSAKYSGWYVGFTKKGRPRKGPKTRENQQDVHFMKRY
PKGQPELQKPFKYTTVTKRSRIRPHTPA**

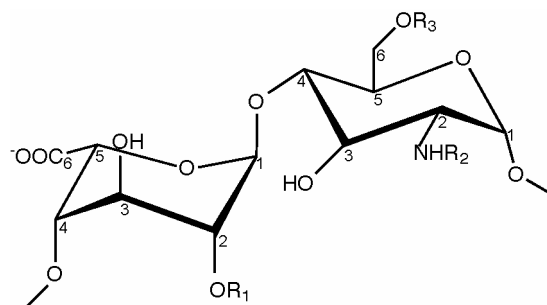
Residues: 1-27 (tag), 28-207

Assayed protein:

**GAMEENVDFRIHVENQTRARDDVSRKQLRLYQLYSRTSGKHIQVLGRRISARGEDGDKYAQLLVETDTFGSQVRIKGKETEFYLCMNRK
GKLVGKPDGTSKECVFIEKVLENNYTALMSAKYSGWYVGFTKKGRPRKGPKTRENQQDVHFMKRYPKGQPELQKPFKYTTVTKRSRRI
RPTHPA**

Residues: 1-3 (tag), 28-207

Fig. S-1. Sequence data for tested FGF-s. A) FGF-1 was expressed and assayed without the signal sequence (1-15 amino acids). B) FGF-2 was expressed and assayed as a full-length protein. C) FGF-18 was expressed without its signal sequence (1-27 amino acids), but with a N-terminal histidine tag, which was subjected to enzymatic cleavage prior to the analysis. The sequences corresponding to the final proteins were compared with appropriate UniProt records and are highlighted bold, where there is an overlap between UniProt and expressed / assayed protein sequence.



Name	Predominant structure	R1	R2	R3
Heparin	$I_{2S} A_{NS}^{6S}$	SO_3^-	SO_3^-	SO_3^-
NAc	$I_{2S} A_{NAc}^{6S}$	SO_3^-	NAc	SO_3^-
2OH	$I_{2OH} A_{NS}^{6S}$	OH	SO_3^-	SO_3^-
6OH	$I_{2S} A_{NS}^{6OH}$	SO_3^-	SO_3^-	OH
2OH NAc	$I_{2OH} A_{NAc}^{6S}$	OH	NAc	SO_3^-
6OH NAc	$I_{2S} A_{NAc}^{6OH}$	SO_3^-	NAc	OH
2OH 6OH	$I_{2OH} A_{NS}^{6OH}$	OH	SO_3^-	OH
2OH 6OH NAc	$I_{2OH} A_{NAc}^{6OH}$	OH	NAc	OH
Persulfated Heparin	$I_{2S}^{3S} A_{NS}^{6S\ 3S}$	SO_3^-	SO_3^-	SO_3^-

Fig. S-2. Sulfation pattern of the major repeating disaccharides of the heparin derivatives. Top panel illustrates the repeating disaccharide unit, where position C-2 of the iduronate (I) can be either O – sulfated or not modified (R1), position C-2 of the glucosamine (A) can be N – sulfated or acetylated (R2), and position C-6 of the glucosamine can be O – sulfated or not modified (R3).