SUPPLEMENTAL DATA

Table S1. pI values of WT FGF-1 and mutant proteins[1].[a]

Protein	pI
WT FGF-1	7.93
Ala66Cys	7.89
Cys16Ser	7.98
Cys117Ala	7.98
Pro134Ala	7.93
Pro134Val	7.93
C16S/A66C/C117A	7.98
C16S/A66C/C117A/P134A	7.98
C16S/A66C/C117A/P134V	7.98

[a]Includes 6x His tag

Figure S1

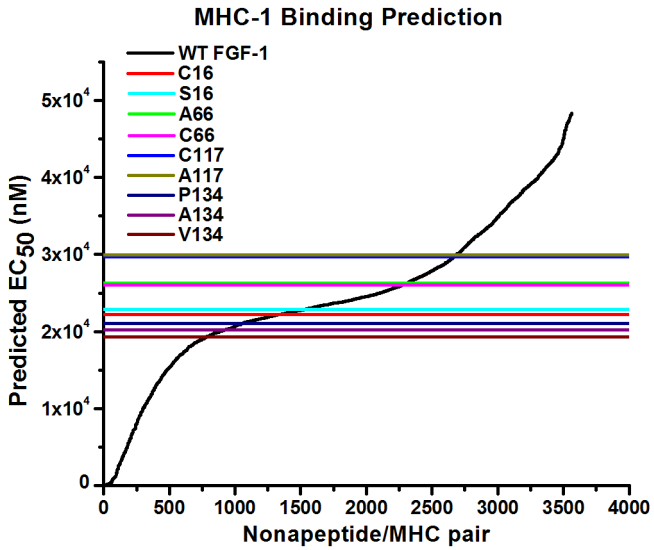

Figure S1. **Predicted MHC-1 binding affinity for nonapeptide epitopes in WT FGF-1 and mutant proteins**. The mutations associated with design of the Cys-free form of FGF-1 have minimal predicted effect as nonapeptide epitopes upon MHC-1 binding affinities [2] compared to the associated WT FGF-1 epitopes.

Figure S2

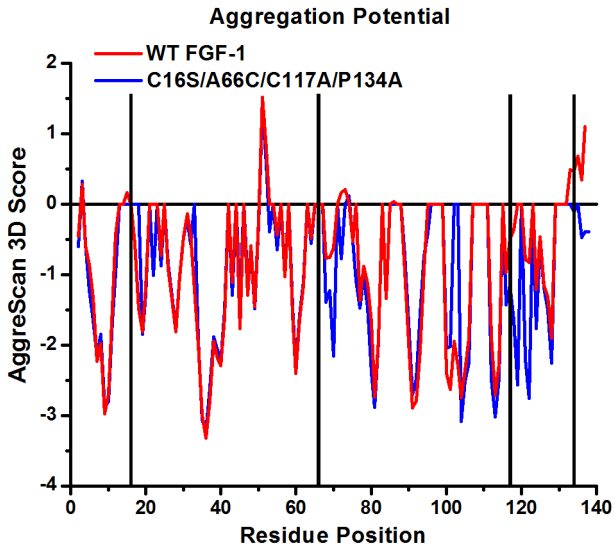

Figure S2. **Predicted aggregation potential, by residue position, for WT FGF-1 and C16S/A66C/C117A/P134A mutant protein**. The predicted aggregation potential was calculated from the X-ray structures of WT FGF-1 (PDB accession 1JQZ) and the C16S/A66C/C117A/P134A mutant protein (PDB accession 4YOL) using the AggreScan 3D server [3]. A greater score indicates increased aggregation potential. The sites of mutation are indicated by the black bars.

Figure S3

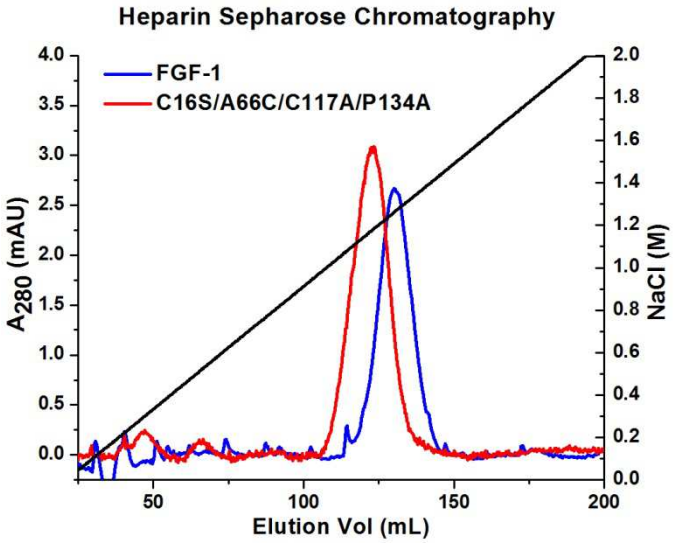

Figure S3. **Heparin Sepharose elution profile of WT FGF-1 and mutant C16S/A66C/C117A/P134A**. The WT FGF-1 elution peak from heparin Sepharose analytical chromatography occurs at 1.26 M NaCl; whereas, the C16S/A66C/C117A/P134A mutant elution peak occurs at 1.18 M NaCl.

Figure S4

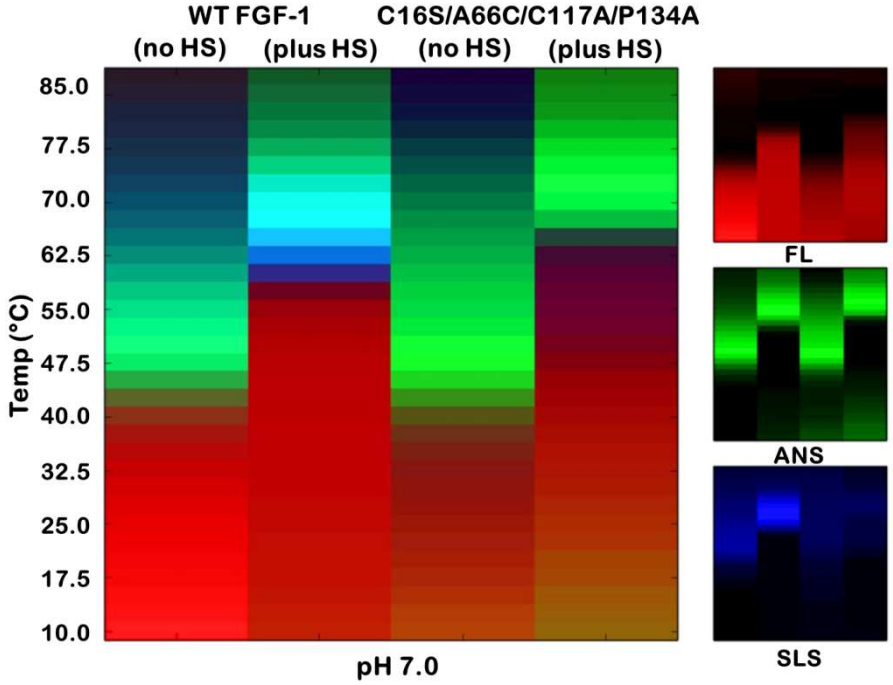

Figure S4. **EPD of WT FGF-1 and mutant C16S/A66C/C117A/P134A, with and without heparin sulfate, at pH 7.0**. The EPD for WT FGF-1 and mutant C16S/A66C/C117A/P134A, both in the presence and absence of 3-fold mass heparin sulfate, at pH 7.0. The EPD data indicate a similar magnitude of thermal stabilization in response to heparin sulfate complexation. However, the SLS data indicate a reduced potential for thermally-induced aggregation with the C16S/A66C/C117A/P134A mutant.

REFERENCES

1. Gasteiger E, Hoogland C, Gattiker A, Duvaud S, Wilkins MR, Appel RD, Bairoch A. 2005. Protein Identification and Analysis Tools on the ExPASy Server;. In Walker JM, editor The Proteomics Protocols Handbook, ed.: Humana Press.

2. Kim JJ, Iyer V, Joshi SB, Volkin DB, Middaugh CR 2012. Improved data visualization techniques for analyzing macromolecule structural changes. Protein Science 21:1540-1553.

3. Zambrano R, Jamroz M, Szczasiuk A, Pujols J, Kmiecik S, Ventura S 2015. AGGRESCAN3D (A3D): server for prediction of aggregation properties of protein structures. Nucleic acids research 43:W306-W313.