Supporting information:

Design and functional studies of xylene-based cyclic mimetics of SOCS1 protein

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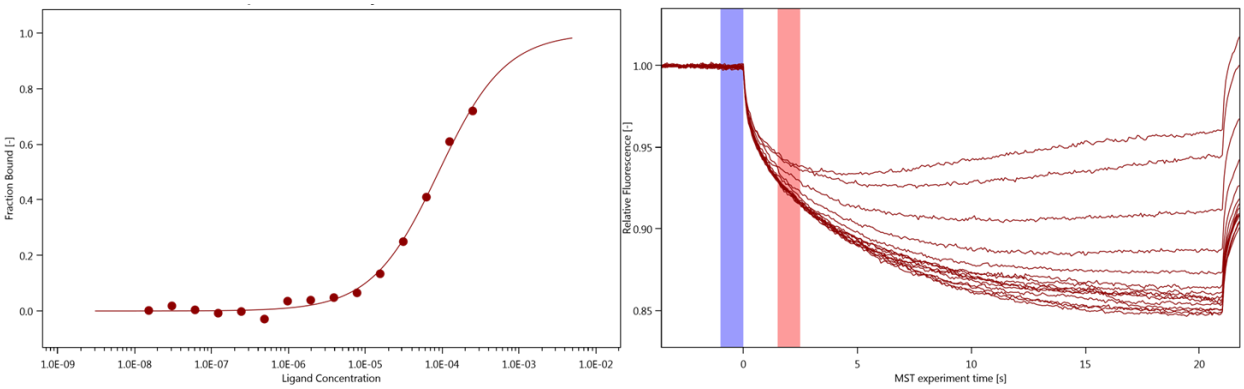

Figure S1. MST assay of curcumin toward JAK2: (right) thermophoretic traces of MST for the binding to JAK2. (left) binding isotherms for MST signals versus curcumin concentrations (250 - 0.0153 μM) 50 mM Tris-HCl, 150 mM NaCl, 0.05 %, Triton X-100, 1 mM dithiothreitol (DTT), 10 % glycerol, at pH 7.5, 15% (v/v) TFE.

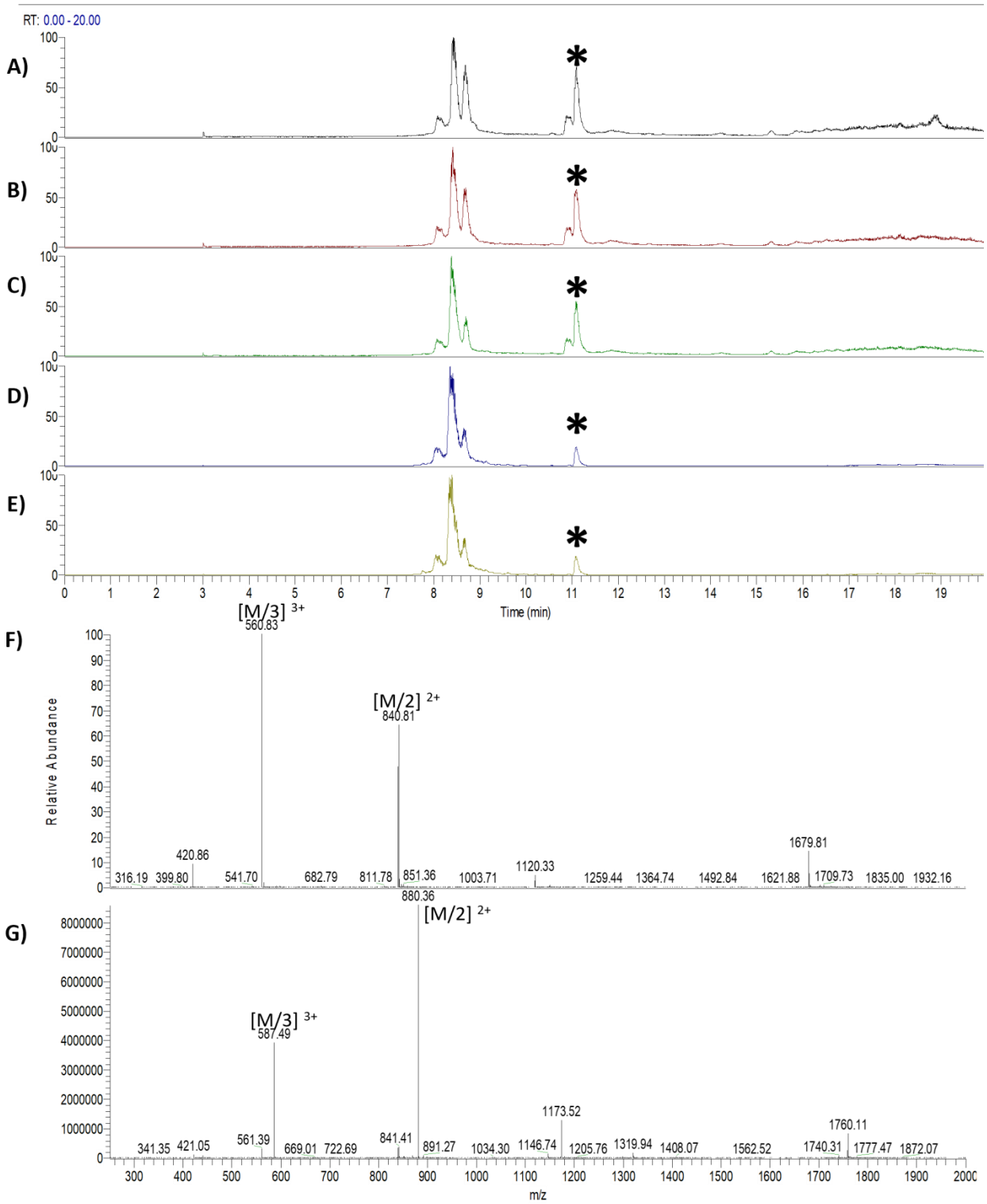

Figure S2. LC-MS of the inhibition of phosphorylation. The peak at retention time (RT): 8.43 min belongs to Srctide and the peak at RT: 8.69 min corresponds to p-Srctide. (**A**) Spectrum in the absence of thio-monocycle PS5(Nal1) with 6.4×10^{-3} eq of JAK2, 64 eq of ATP and 1 eq of Srctide. (**B**) Spectrum in the presence of 4×10^{-2} eq of thio-monocycle PS5(Nal1), 6.4×10^{-3} eq of JAK2, 64 eq of

ATP and 1 eq of Srctide. (**C**) Spectrum in the presence of $4x10^{-1}$ eq of thio-monocycle PS5(Nal1), $6.4x10^{-3}$ eq of JAK2, 64 eq of ATP and 1 eq of Srctide. (**D**) Spectrum in the presence of 1 eq of thio-monocycle PS5(Nal1), $6,4x10^{-3}$ eq of JAK2, 64 eq of ATP and 1 eq of Srctide. (**E**) Spectrum in the presence of 2 eq of thio-monocycle PS5(Nal1), $6,4x10^{-3}$ eq of JAK2, 64 eq of ATP and 1 eq of Srctide. ESI-MS spectrum of (**F**) Srctide and (**G**) p- Srctide. ***=** peak of JAK2 protein.

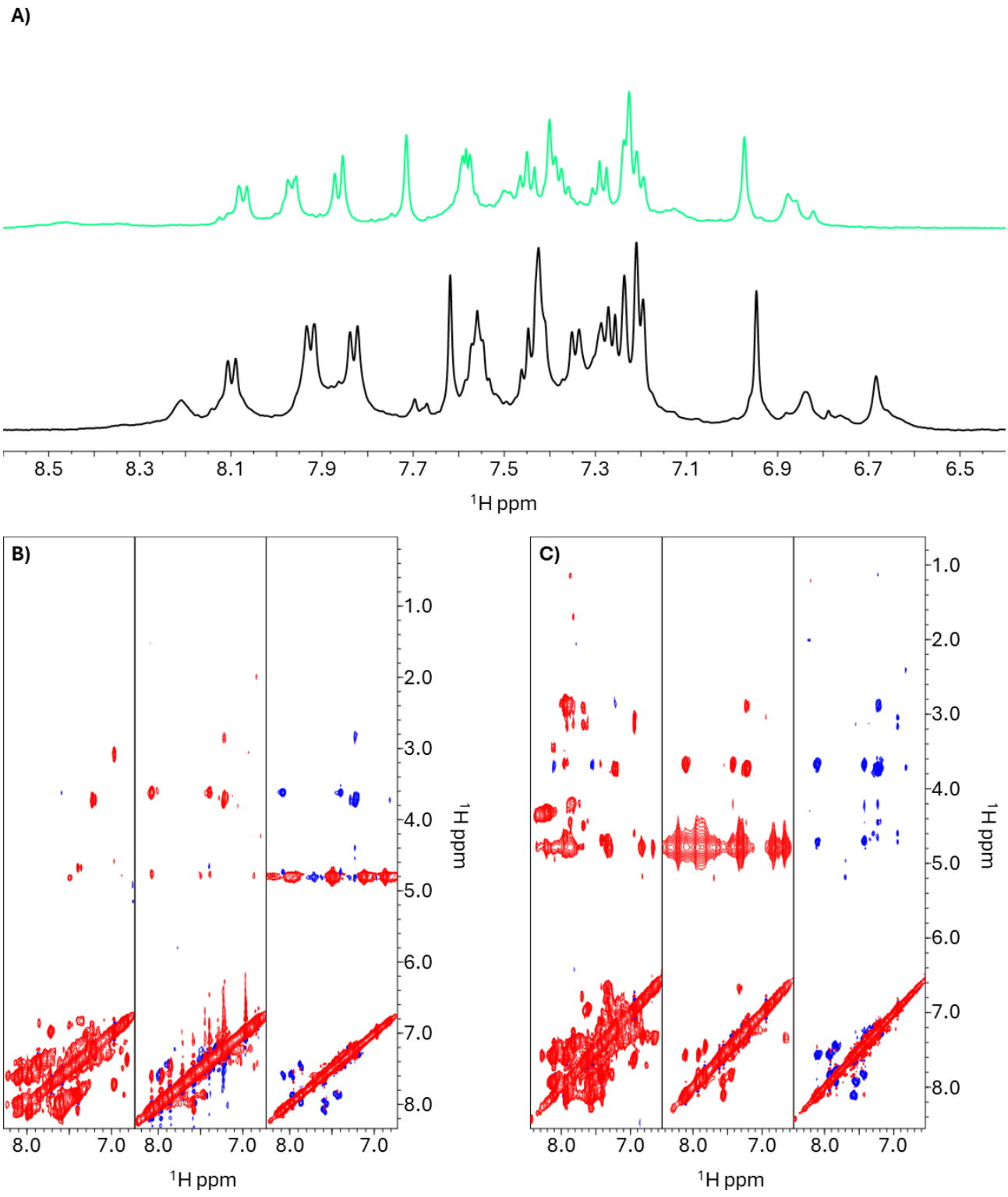

Figure S3. (**A**) Comparison of 1D [^{1}H] spectra of thio-monocycle PS5(Nal1) (329 µM concentration) in 10 mM NaP pH 7.39 (green) and in a mixture TFE/NaP (50/50 v/v) (black) pH 7.51. (**B, C**) Comparison of 2D [^{1}H, ^{1}H] TOCSY (left panels), NOESY 300 (middle panels) and ROESY (right panels) spectra of thio-monocycle PS5(Nal1) acquired in 10 mM NaP (**B**) and in NaP/TFE (50/50

v/v) (**C**). Peaks of opposite sign with respect to the diagonal are colored blue. Spectral regions containing correlations from H_N and aromatic protons are shown.

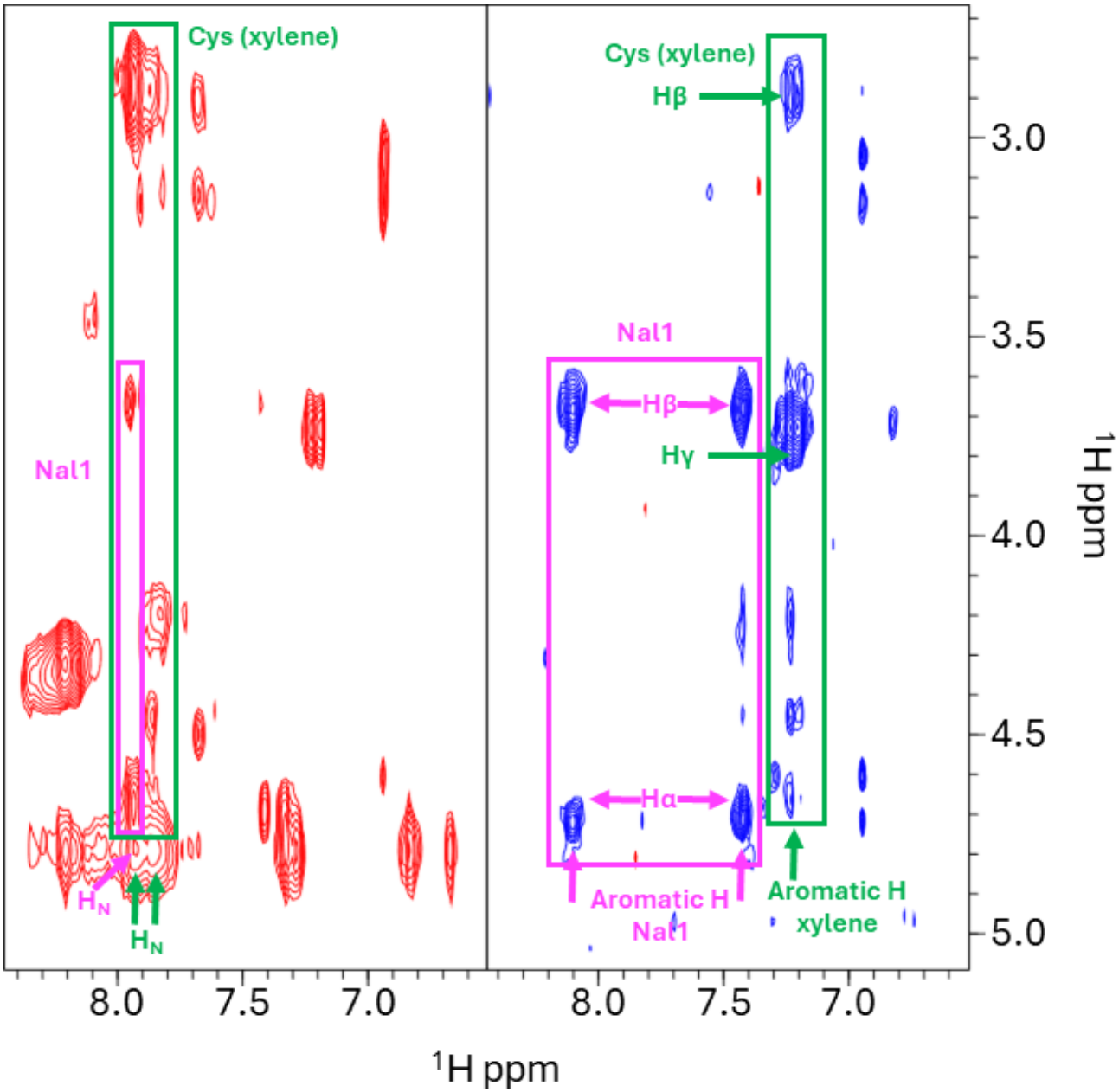

Figure S4. 2D [^{1}H, ^{1}H] TOCSY (left panel) and ROESY (right panel) spectra of thio-monocycle PS5(Nal1) in NaP/TFE (50/50 v/v): spin systems from the two cysteines linked by xylene are enclosed in green rectangles, those from Nal1 are highlighted by the magenta box. Through-space correlations between $_S$CH2 β and γ protons of the two cysteines (Cys1 and 9) and xylene aromatic protons are evident in the ROESY spectrum (right panel). Intra-residue correlations involving the backbone H_N of Cys (1, 9), Hα and Hβ protons can be instead seen in the TOCSY spectrum (left panel).

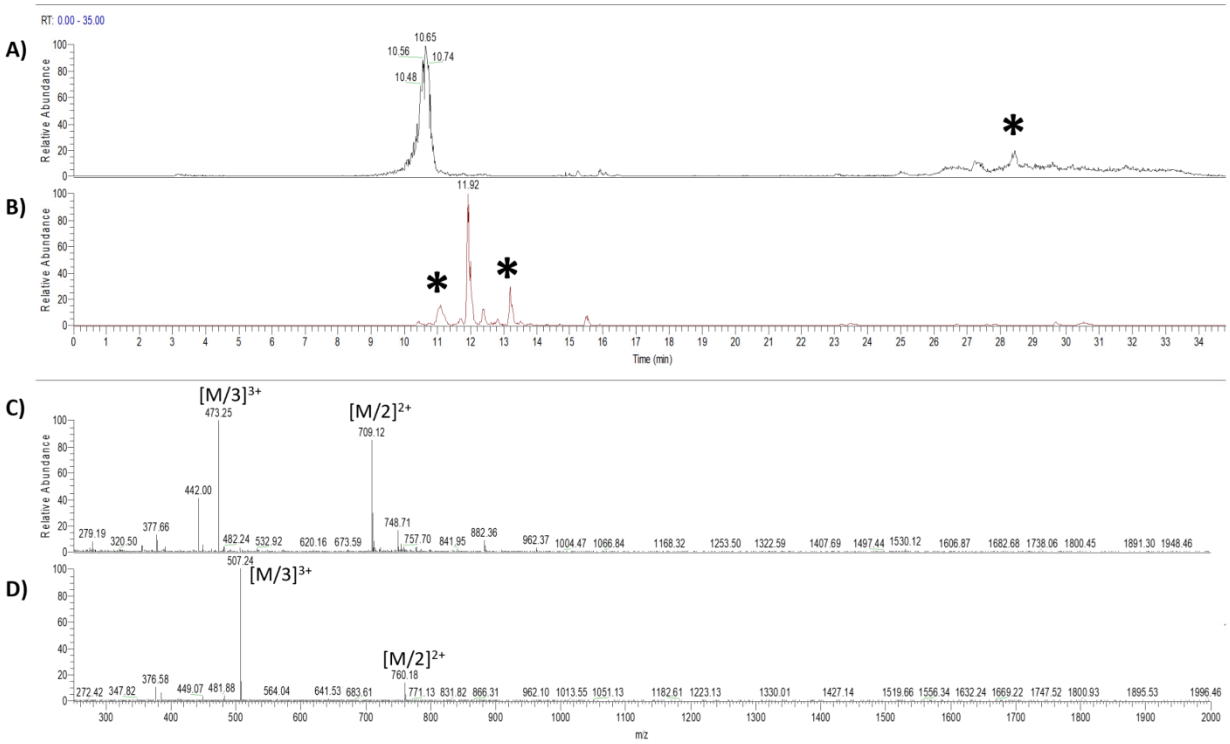

Figure S5. LC-MS profiles of (**A**) linear (**B**) thio-monocycle PS5(Nal1). MS spectra of the main peaks in linear (**C**) thio-monocycle PS5(Nal1) (**D**) chromatograms. * = impurity.

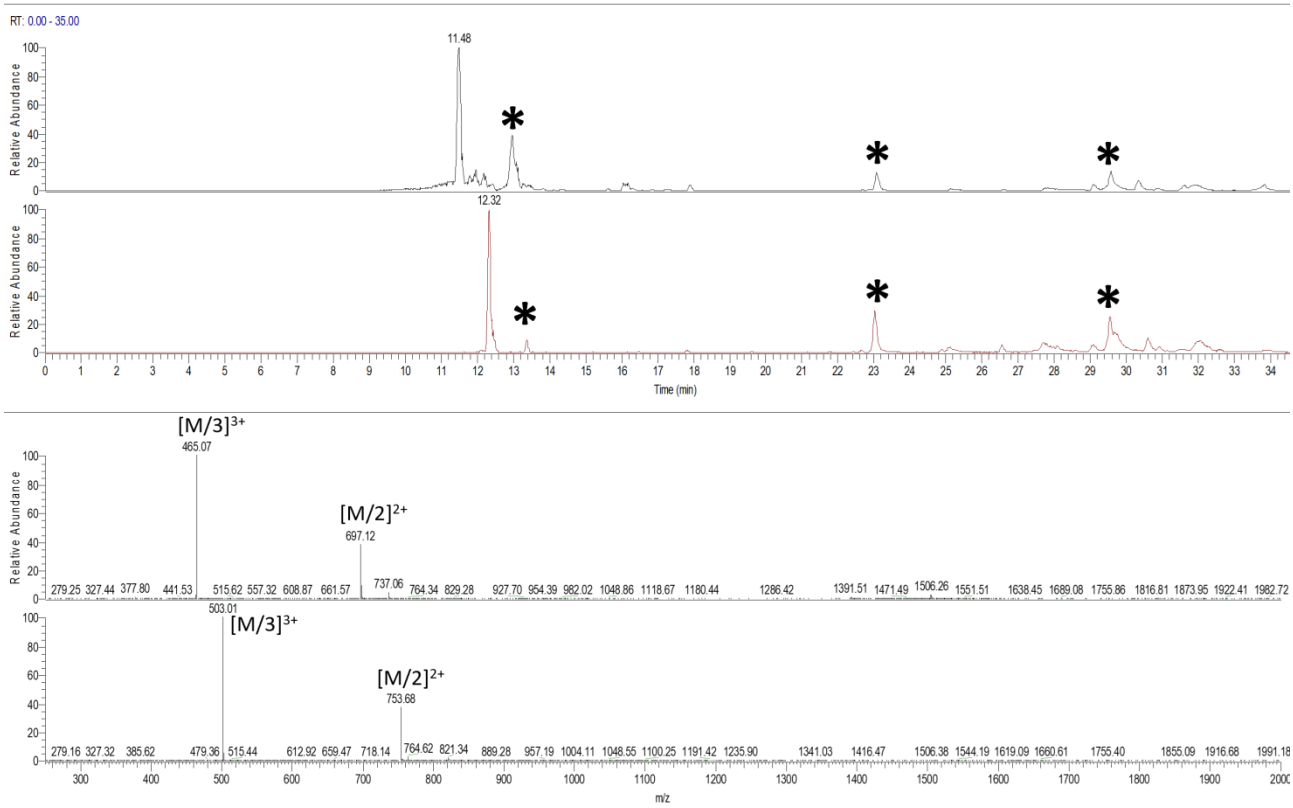

Figure S6. LC-MS profiles of (**A**) linear (**B**) thio-bicycle PS5(Nal1). MS spectra of the main peaks in linear (**C**) thio-bicycle PS5(Nal1) (**D**) chromatograms. * = impurity.

Table S1. Name and sequence of SOCS1 peptidomimetics. The substitution Phe[58]/Nal1 is reported in magenta, lactam or xylene linkages, in blue.

Protein	Name	Sequence
SOCS1	**PS5**	**D[1]TC(Acm)RQ[5]TFRK[9]H**
	***ic*PS5(Nal1)**	**D[1]TC(Acm)RQ[5]TNalRK[9]H**
	thio-monocycle PS5(Nal1)	**C[1]TC(Acm)RQ[5]TNalRC[9]H**
	thio-bicycle PS5(Nal1)	**C[1]TC(Acm)RC[5]TNalRC[9]H**

Table S2. Deconvolution of CD spectra of thio-PS5(Nal1) analogues.

peptide	TFE	helix	beta	turn	others
thio-monocycle PS5(Nal1)	0%	0.0	24.4	15.9	59.7
	50%	5.4	33.0	13.1	48.5
thio-bicycle PS5(Nal1)	0%	0.0	36.9	15.2	47.9
	50%	5.3	34.8	11.8	48.2

Table S3. Proton chemical shift assignments (ppm) of thio-monocycle PS5(Nal1) in NaP/TFE (50/50 v/v).

Residue	H_N	Hα	Hβ	Hγ	Others
1, 9 C	7.94 7.86	4.66 4.45	2.83-2.92 2.86-2.93	3.74-3.67	xylene: 7.23
2, 6 T		4.18 4.33	4.27 4.46	1.14 1.28	
3 C(Acm)				4.31	Hζ 8.21 Q1 CH_3 2.02
4, 8 R		4.11 4.20	1.77-1.84 1.74-1.83	1.63 1.49	Hδ 3.14
5 Q		4.22	1.97-2.07	2.37	Hε 7.33-6.67
7 Nal1	7.93	4.67	3.62-3.67		Aromatic protons: 8.10 7.83 7.61-7.55 7.93
10 H		4.60	3.04-3.18		