

Supplementary Figure 2.

Mass spectrometry analysis to confirm the identity of native Cystatin and recombinant FhStf-1, 2 and 3

A) Mascot Results for Native Cystatin

Match to: **FhStf-1** Score: **408**

Nominal mass (M_r): **11290**; Calculated pI value: **6.95**

NCBI BLAST search of [FhStf-1](#) against nr

Unformatted [sequence string](#) for pasting into other applications

Fixed modifications: Carbamidomethyl (C)

Variable modifications: Oxidation (M)

Cleavage by Trypsin: cuts C-term side of KR unless next residue is P

Sequence Coverage: **15%**

Matched peptides shown in **Bold Red**

1 M MCGGCTQTR **APSAEEKTEL EIVLREQLES** HIGHKPHIIE VVEICTQVVA
51 GLNYFAKVKI GNDNYIHARI **FQELPCNGGK** KKVHSLKDK SATDALQYF

Start - End	Observed	Mr(expt)	Mr(calc)	Delta	Miss	Sequence	
11 - 25	562.2813	1683.8221	1683.9043	-0.0822	1	R.APSAEKTELEIVLR.E	(Ions score 43)
11 - 25	562.2928	1683.8565	1683.9043	-0.0477	1	R.APSAEKTELEIVLR.E	(Ions score 41)
11 - 25	562.2928	1683.8565	1683.9043	-0.0477	1	R.APSAEKTELEIVLR.E	(Ions score 30)
11 - 25	842.9437	1683.8728	1683.9043	-0.0314	1	R.APSAEKTELEIVLR.E	(Ions score 107)
11 - 25	842.9446	1683.8745	1683.9043	-0.0297	1	R.APSAEKTELEIVLR.E	(Ions score 18)
11 - 25	842.9483	1683.8820	1683.9043	-0.0223	1	R.APSAEKTELEIVLR.E	(Ions score 42)
11 - 25	842.9501	1683.8857	1683.9043	-0.0186	1	R.APSAEKTELEIVLR.E	(Ions score 94)
11 - 25	842.9518	1683.8890	1683.9043	-0.0152	1	R.APSAEKTELEIVLR.E	(Ions score 108)
11 - 25	562.3042	1683.8908	1683.9043	-0.0135	1	R.APSAEKTELEIVLR.E	(Ions score 29)

B) Mascot results for rFhStf-1

Match to: **FhStf-1** Score: **1628**

Nominal mass (M_r): **11290**; Calculated pI value: **6.95**

NCBI BLAST search of [FhStf-1](#) against nr

Unformatted [sequence string](#) for pasting into other applications

Fixed modifications: Carbamidomethyl (C)

Variable modifications: Oxidation (M)

Cleavage by Trypsin: cuts C-term side of KR unless next residue is P

Sequence Coverage: **38%**

Matched peptides shown in **Bold Red**

1 M MCGGCTQTR **APSAEEKTEL EIVLREQLES** HIGHKPHIIE VVEICTQVVA
51 GLNYFAKVKI GNDNYIHARI **FQELPCNGGK** KKVHSLKDK SATDALQYF

Start - End	Observed	Mr(expt)	Mr(calc)	Delta	Miss	Sequence	
11 - 25	562.2698	1683.7877	1683.9043	-0.1166	1	R.APSAEKTELEIVLR.E	(Ions score 27)
11 - 25	562.2813	1683.8221	1683.9043	-0.0822	1	R.APSAEKTELEIVLR.E	(Ions score 77)

11 - 25	562.2813	1683.8221	1683.9043	-0.0822	1	R.APSAEKTELEIVLR.E	(Ions score 87)
11 - 25	562.2813	1683.8221	1683.9043	-0.0822	1	R.APSAEKTELEIVLR.E	(Ions score 87)
11 - 25	562.2813	1683.8221	1683.9043	-0.0822	1	R.APSAEKTELEIVLR.E	(Ions score 83)
11 - 25	842.9332	1683.8518	1683.9043	-0.0524	1	R.APSAEKTELEIVLR.E	(Ions score 140)
11 - 25	562.2928	1683.8565	1683.9043	-0.0477	1	R.APSAEKTELEIVLR.E	(Ions score 53)
11 - 25	562.2928	1683.8565	1683.9043	-0.0477	1	R.APSAEKTELEIVLR.E	(Ions score 41)
11 - 25	842.9488	1683.8831	1683.9043	-0.0212	1	R.APSAEKTELEIVLR.E	(Ions score 104)
11 - 25	842.9492	1683.8839	1683.9043	-0.0204	1	R.APSAEKTELEIVLR.E	(Ions score 112)
11 - 25	842.9495	1683.8844	1683.9043	-0.0198	1	R.APSAEKTELEIVLR.E	(Ions score 118)
11 - 25	842.9500	1683.8855	1683.9043	-0.0188	1	R.APSAEKTELEIVLR.E	(Ions score 31)
11 - 25	842.9501	1683.8857	1683.9043	-0.0186	1	R.APSAEKTELEIVLR.E	(Ions score 122)
11 - 25	842.9507	1683.8869	1683.9043	-0.0173	1	R.APSAEKTELEIVLR.E	(Ions score 135)
11 - 25	562.3042	1683.8908	1683.9043	-0.0135	1	R.APSAEKTELEIVLR.E	(Ions score 65)
11 - 25	562.3042	1683.8908	1683.9043	-0.0135	1	R.APSAEKTELEIVLR.E	(Ions score 64)
11 - 25	562.3042	1683.8908	1683.9043	-0.0135	1	R.APSAEKTELEIVLR.E	(Ions score 57)
11 - 25	842.9528	1683.8910	1683.9043	-0.0132	1	R.APSAEKTELEIVLR.E	(Ions score 101)
11 - 25	842.9538	1683.8930	1683.9043	-0.0113	1	R.APSAEKTELEIVLR.E	(Ions score 97)
58 - 69	700.3379	1398.6613	1398.7368	-0.0754	1	K.VKIGNDNYIHAR.I	(Ions score 89)
58 - 69	467.2442	1398.7107	1398.7368	-0.0261	1	K.VKIGNDNYIHAR.I	(Ions score 53)
58 - 69	700.3664	1398.7182	1398.7368	-0.0186	1	K.VKIGNDNYIHAR.I	(Ions score 63)
60 - 69	586.7635	1171.5125	1171.5734	-0.0609	0	K.IGNDNYIHAR.I	(Ions score 85)
70 - 80	631.7655	1261.5165	1261.6125	-0.0960	0	R.IFQELPCNGGK.K	(Ions score 36)

C) Mascot results for rFhStf-2

Match to: **FhStf-2** Score: **142**

Nominal mass (M_r): **12805**; Calculated pI value: **5.74**

NCBI BLAST search of [FhStf-2](#) against nr

Unformatted [sequence string](#) for pasting into other applications

Fixed modifications: Carbamidomethyl (C)

Variable modifications: Oxidation (M)

Cleavage by Trypsin: cuts C-term side of KR unless next residue is P

Sequence Coverage: **17%**

Matched peptides shown in **Bold Red**

1 MLRILLGICI LHFMSCDVFG EMLVGGYTEP RSVTPEERSV **FQPMILSKLL**
51 TAGSVESSECE LELLQVSTQV VAGINYKFKV SGGATCPGCW EVVVFVPLYS
101 SK**SATSVGTP** TRVSCT

Start - End	Observed	Mr(expt)	Mr(calc)	Delta	Miss	Sequence	
39 - 48	575.3022	1148.5898	1148.6264	-0.0365	0	R.SVFQPMILSK.L	(Ions score 54)
39 - 48	583.2663	1164.5179	1164.6213	-0.1033	0	R.SVFQPMILSK.L	Oxidation (M) (Ions score 44)
39 - 48	1165.5376	1164.5303	1164.6213	-0.0910	0	R.SVFQPMILSK.L	Oxidation (M) (Ions score 49)
103 - 112	488.7540	975.4934	975.4985	-0.0051	0	K.SATSVGTPTR.V	(Ions score 34)

A) Mascot results for rFhStf-3

Match to: **FhStf-3** Score: **355**

Nominal mass (M_r): **13052**; Calculated pI value: **5.04**

NCBI BLAST search of [FhStf-3](#) against nr

Unformatted [sequence string](#) for pasting into other applications

Fixed modifications: Carbamidomethyl (C)

Variable modifications: Oxidation (M)

Cleavage by Trypsin: cuts C-term side of KR unless next residue is P

Sequence Coverage: 31%

Matched peptides shown in **Bold Red**

1 MKLITFLICL ICVSGCRSTE **KRLCGGYDEP** KAVQQDDIDK FESVVTENLI
51 **RSGSLTHDAY** **LTFLTVSKQI** **VAGINYK**FLV KSSGGQCFEV LLFEPLPYTE
101 LPPSVTSFPT VECPS

Start - End	Observed	Mr(expt)	Mr(calc)	Delta	Miss	Sequence	
22 - 31	597.7598	1193.5050	1193.5499	-0.0448	1	K.RLCGGYDEPK.A	(Ions score 26)
52 - 68	920.4377	1838.8608	1838.9414	-0.0806	0	R.SGSLTHDAYLTFLTVSK.Q	(Ions score 117)
69 - 77	503.2497	1004.4849	1004.5655	-0.0806	0	K.QIVAGINYK.F	(Ions score 60)
69 - 77	503.2880	1004.5614	1004.5655	-0.0041	0	K.QIVAGINYK.F	(Ions score 58)
69 - 77	503.2892	1004.5638	1004.5655	-0.0016	0	K.QIVAGINYK.F	(Ions score 54)
69 - 77	503.2903	1004.5661	1004.5655	0.0006	0	K.QIVAGINYK.F	(Ions score 65)
69 - 77	503.2917	1004.5688	1004.5655	0.0034	0	K.QIVAGINYK.F	(Ions score 24)
69 - 77	503.2947	1004.5748	1004.5655	0.0093	0	K.QIVAGINYK.F	(Ions score 42)