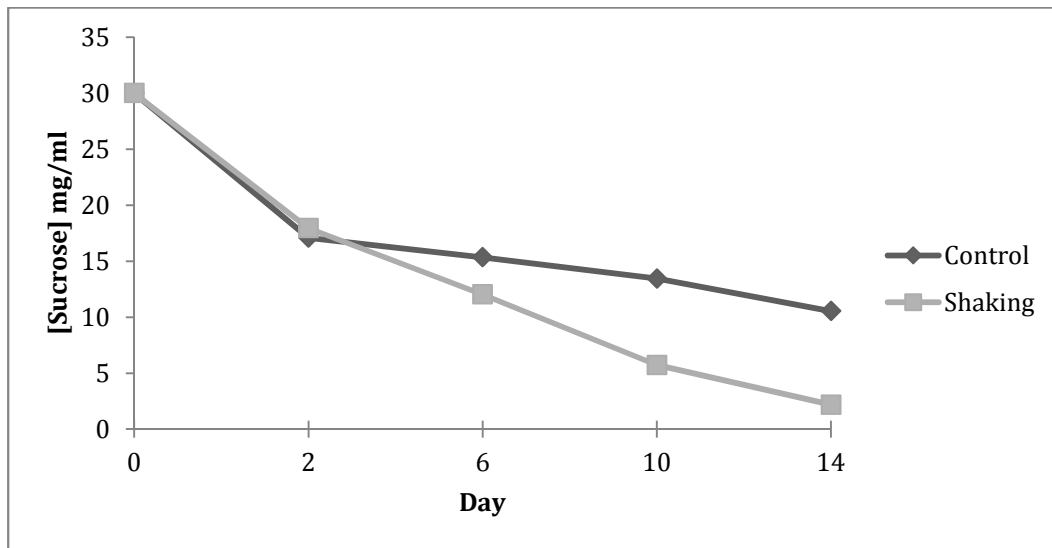


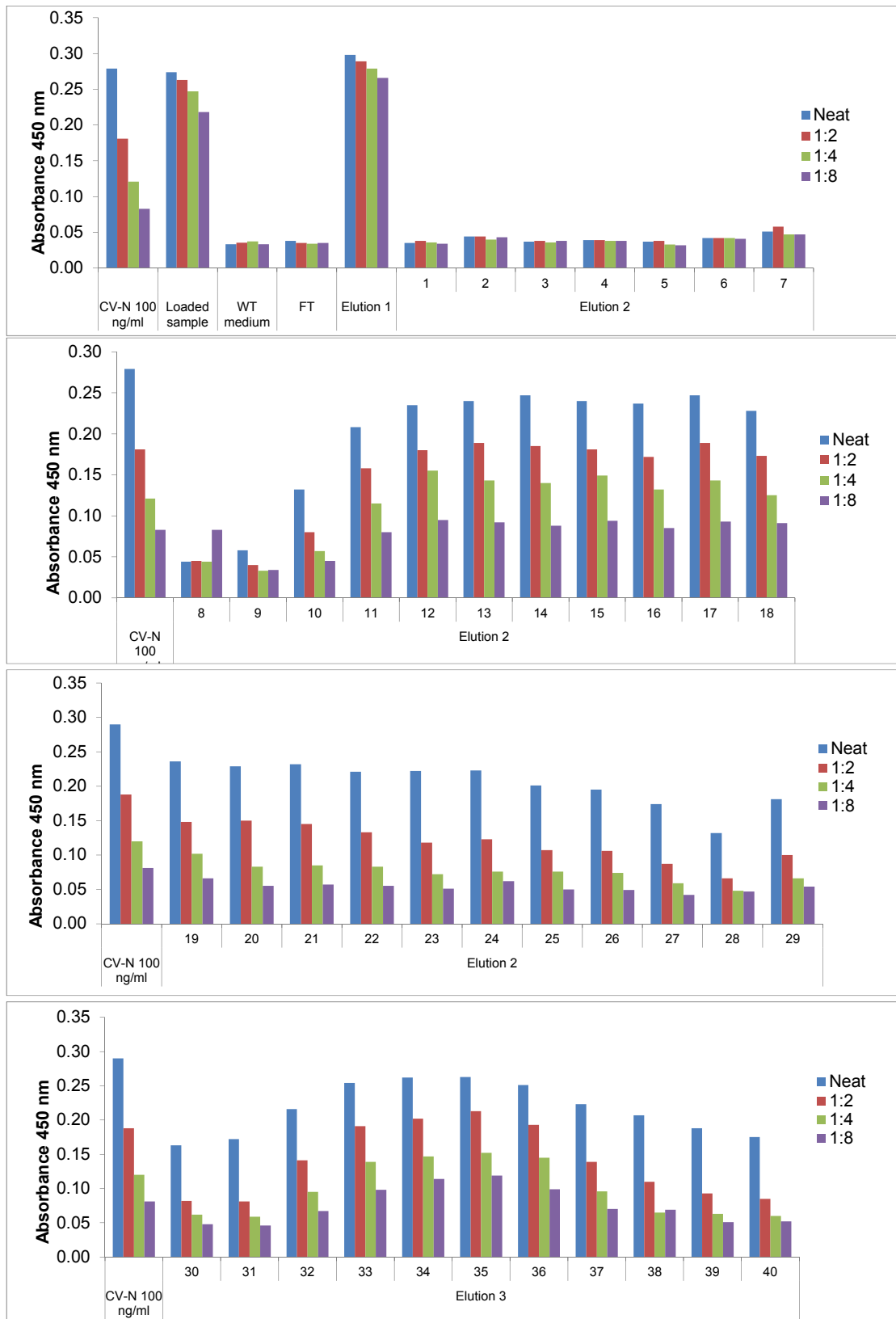
Supplementary figures



Supplementary figure 1. Sucrose depletion in hydroponic medium between weeks 7 and 9 for the stationary control and the shaking group. Values are for pooled samples from 3 plants.

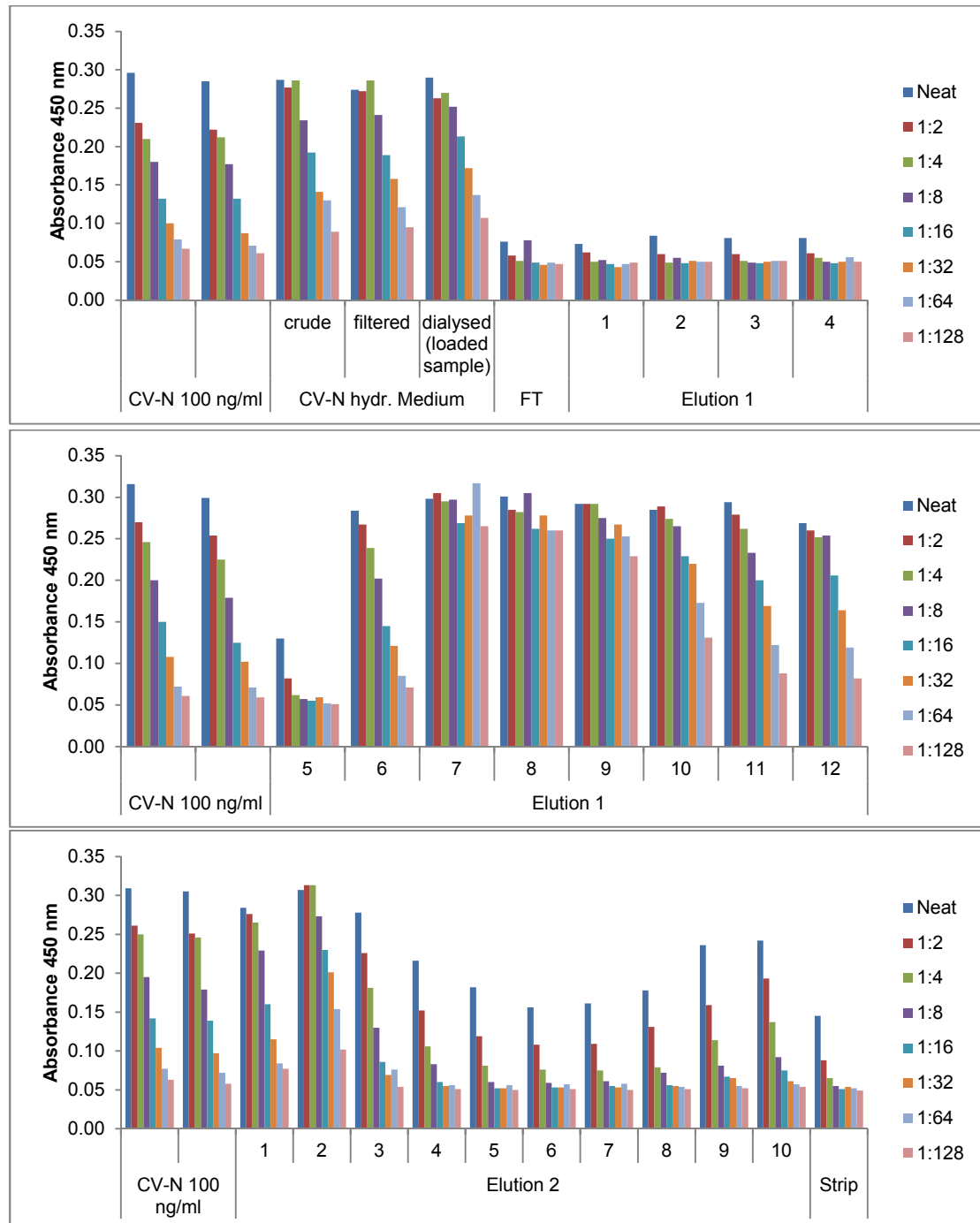
Methodology

Sucrose concentration was measured with a Sucrose Assay Kit (Sigma-Aldrich, Poole, UK - product code SCA-20). The assay was performed according to the manufacturer's instructions.



Supplementary figure 2. gp120 binding ELISA with eluted samples from first IEX run. 50 ml of dialyzed medium were loaded onto a 1 ml HiTrap Q HP column. After washing, elution was performed in three steps: 1 – 0 to 200 mM NaCl in buffer A (10 ml); 2 – gradient from 200mM to 1M NaCl in buffer A (30 ml, 1 ml fractions); 3 – 1M NaCl (10 ml, 1 ml fractions). Loaded samples and Flow-

through samples were diluted 10 times and elution fraction 100 times prior to loading (neat).



Supplementary figure 3. gp120 binding ELISA with eluted samples from second IEX run. 50 ml of dialysed medium were loaded onto a 1 ml HiTrap Q HP column. After washing, elution was performed in three steps: 1 – gradient from 0 to 200 mM NaCl in buffer A (12 ml, 1 ml fractions); 2 – gradient from 200mM to 1M NaCl in buffer A (10 ml, 1 ml fractions); Strip – 1M NaCl (10 ml). Hydroponic medium samples and FT were diluted 10 times and elution fractions were diluted 100 times prior to loading (neat).

Supplementary Table 1. Proteins identified in 'CV-N with NAA in MS' sample. Recombinant protein is highlighted in red, hits highlighted in blue indicate proteins differing from supplementary table 2.

Hit number	Database access number	Protein description	Mascot score	Mass	emPAI
1	gi 148299083	subtilisin-like protease [Nicotiana tabacum]	1211	81137	1.67
2	gi 108752035 gb ABG02914.1	cyanovirin-N [synthetic construct]	750	10923	27.27
3	gi 116314	Endochitinase A precursor (CHN-A)[Nicotiana tabacum]	629	35134	1.76
4	gi 64976605	peroxidase [Nicotiana tabacum]	593	38740	2.65
5	gi 131015	Pathogenesis-related protein R major form precursor (Thaumatococcus-like protein E22)[Nicotiana tabacum]	590	24650	2.65
6	gi 63002585	peroxidase [Nicotiana tabacum]	571	35563	1.48
7	gi 63253080	peroxidase [Nicotiana tabacum]	562	38505	2.35
8	gi 129837	Lignin-forming anionic peroxidase precursor (TOPA)[Nicotiana tabacum]	544	34652	2.83
9	gi 24430421	41 kD chloroplast nucleoid DNA binding protein (CND41) [Nicotiana sylvestris]	528	53600	0.83
10	gi 2541876	CND41, chloroplast nucleoid DNA binding protein [Nicotiana tabacum]	507	53413	0.96
11	gi 62362434	nectarin IV [Nicotiana langsdorffii x Nicotiana sanderae]	504	46948	0.85
12	gi 467689	chitinase/lysozyme [Nicotiana tabacum]	483	41993	1.35
13	gi 83281191	putative glycerophosphoryl diester phosphodiesterase family protein [Nicotiana tabacum]	442	81954	0.3
14	gi 505267	chitinase, class V [Nicotiana tabacum]	414	41754	0.68
15	gi 119011	Glucan endo-1,3-beta-glucosidase, basic vacuolar isoform GLB precursor ((1->3)-beta-glucan endohydrolase) ((1->3)-beta-glucanase) (Beta-1,3-endoglucanase, basic) (Glucanase GLB)[Nicotiana tabacum]	391	40417	1.22
16	gi 37359706	LEXYL1 [Solanum lycopersicum]	385	83111	0.42
17	gi 116342	Acidic endochitinase Q precursor (Pathogenesis-related protein Q) (PR-Q)[Nicotiana tabacum]	345	27615	0.92
18	gi 5360263	NtPRp27 [Nicotiana tabacum]	337	27361	0.92
19	gi 1167854	PR protein; osmotin [Nicotiana tabacum]	335	26744	1.22
20	gi 131017	Pathogenesis-related protein R minor form precursor (PR-R) (PROB12) (Thaumatococcus-like protein E2)[Nicotiana tabacum]	327	24535	1.06
21	gi 3790355	chitinase 134 [Nicotiana tabacum]	319	29184	0.84
22	gi 5381253	peroxidase [Nicotiana tabacum]	299	34348	0.52
23	gi 37359708	LEXYL2 [Solanum lycopersicum]	299	68869	0.3
24	gi 7547630	pathogen- and wound-inducible antifungal protein CBP20 precursor [Nicotiana tabacum]	285	22153	0.91
25	gi 116332	Acidic endochitinase precursor[Nicotiana tabacum]	279	31263	0.98
26	gi 3913932	Proteinase inhibitor type-2 precursor (Proteinase inhibitor type II)	258	20970	0.96
27	gi 1255951	PS60 [Nicotiana tabacum]	251	60107	0.2
28	gi 19771	acidic chitinase PR-P [Nicotiana tabacum]	241	27512	0.68
29	gi 8919178	alpha-glucosidase [Solanum tuberosum]	236	104620	0.19
30	gi 267124	Thioredoxin H-type 1 (Trx-H1) [Nicotiana tabacum]	229	13947	1.1
31	gi 100284	superoxide dismutase (EC 1.15.1.1) (Fe) - curled-leaved tobacco	215	25434	0.52
32	gi 27803890	NtPRp27-like protein [Solanum tuberosum]	211	25115	0.33
33	gi 130840	Pathogenesis-related protein PR-4B precursor [Nicotiana tabacum]	210	16225	0.53
34	gi 6538778	biotic cell death-associated protein [Nicotiana glutinosa]	201	25142	0.53
35	gi 48375044	putative mitochondrial malate dehydrogenase [Nicotiana tabacum]	201	22024	0.9
36	gi 19962	pathogenesis-related protein 4A [Nicotiana tabacum]	200	16195	1.36
37	gi 90992878	phylloplanin [Nicotiana tabacum]	199	11302	1.47
38	gi 157830095	Chain A, Pathogenesis-Related Protein 5d From Nicotiana Tabacum	197	22745	0.86
39	gi 21388550	putative mitochondrial NAD-dependent malate dehydrogenase [Solanum tuberosum]	192	36144	0.49
40	gi 5725356	alpha-D-xylosidase [Tropaeolum majus]	190	104870	0.15
41	gi 83283995	fructose-bisphosphate aldolase-like protein [Solanum tuberosum]	187	38594	0.31
42	gi 30526289	29 kDa chitinase-like thermal hysteresis protein [Solanum dulcamara]	175	28827	0.64
43	gi 77999255	unknown [Solanum tuberosum]	169	38413	0.32
44	gi 19944	unnamed protein product [Nicotiana tabacum]	165	18575	1.13
45	gi 2497757	Non-specific lipid-transfer protein 1 precursor (LTP 1) (Pathogenesis-related protein 14) (PR-14) [Nicotiana tabacum]	164	11516	0.81
46	gi 2213867	fructose-biphosphate aldolase [Mesembryanthemum crystallinum]	163	38134	0.33
47	gi 130846	Basic form of pathogenesis-related protein 1 precursor (PRP 1) [Nicotiana tabacum]	159	19716	0.71
48	gi 32482806	putative xyloglucanase inhibitor [Solanum tuberosum]	159	46779	0.14

49	gi 74355968	alpha-L-arabinofuranosidase [Raphanus sativus]	156	83723	0.14
50	gi 786337	osmotin I=group 5 basic pathogenesis-related protein (N-terminal) [Nicotiana tabacum=tobacco, Samsun NN, Peptide Partial, 25 aa]	155	2696	21.45
51	gi 130827	Pathogenesis-related protein 1B precursor (PR-1B) [Nicotiana tabacum]	145	18488	1.31
52	gi 8439545	methionine synthase [Solanum tuberosum]	144	84613	0.14
53	gi 46250719	pathogenesis-related protein [Nicotiana tabacum]	144	27523	0.47
54	gi 182410392	PR1 protein [Solanum lycopersicum]	131	20110	0.42
55	gi 547733	Proteinase inhibitor I-B precursor (PI-IB) (Inhibitor of microbial serine proteinases major isoform)[Nicotiana tabacum]	131	11908	0.78
56	gi 24638017	Malate dehydrogenase, mitochondrial precursor Fragaria x ananassa	128	35600	0.22
57	gi 157331729	unnamed protein product [Vitis vinifera]	125	60362	0.2
58	gi 115486089	Os11g0592000 [Oryza sativa (japonica cultivar-group)]	124	16246	0.54
59	gi 498926	glucan endo-1,3-beta-D-glucosidase [Solanum lycopersicum]	121	37839	0.21
60	gi 544202	Glucan endo-1,3-beta-glucosidase, acidic isoform PR-Q~ precursor ((1->3)-beta-glucan endohydrolase) ((1->3)-beta-glucanase) (Beta-1,3-endoglucanase) (PR-35)[Nicotiana tabacum]	116	36972	0.21
61	gi 9759007	pectin methylesterase-like protein [Arabidopsis thaliana]	114	63463	0.12
62	gi 547743	Trypsin inhibitor 1 precursor (Trypsin inhibitor I) (NSTI-I)Nicotiana sylvestris	114	10458	0.92
63	gi 130828	Pathogenesis-related protein 1C precursor (PR-1C)Nicotiana sylvestris [Nicotiana tabacum]	113	18571	0.76
64	gi 7939623	putative beta-galactosidase [Lycopersicon esculentum]	110	93184	0.12
65	gi 7672732	LRR receptor-like protein kinase [Nicotiana tabacum]	109	101724	0.07
66	gi 58416137	serpin [Cucumis sativus]	108	42576	0.09
67	gi 146762261	beta-xylosidase/alpha-L-arabinosidase [Medicago sativa subsp. x varia]	108	83674	0.09
68	gi 130826	Pathogenesis-related protein 1A precursor (PR-1A)[Nicotiana tabacum]	106	18562	0.76
69	gi 586099	Thioredoxin H-type 2 (Trx-H2)[Nicotiana tabacum]	104	13039	0.62
70	gi 18403247	receptor protein kinase-related [Arabidopsis thaliana]	103	27846	0.29
71	gi 116337	Basic endochitinase precursor[Nicotiana tabacum]	102	32373	0.25
72	gi 83778371	expansin-like protein precursor [Solanum tuberosum]	99	28340	0.46
73	gi 688429	tumor-related protein [Nicotiana glauca x Nicotiana langsdorffii]	96	13798	1.12
74	gi 37951323	BYJ15 [Nicotiana tabacum]	95	16921	0.51
75	gi 75153341	NAD(P)H:quinone oxidoreductase (NAD(P)H:QR)	94	21302	0.39
76	gi 157355185	unnamed protein product [Vitis vinifera]	92	39303	0.1
77	gi 82568706	Serine carboxypeptidase [Prunus mume]	88	55936	0.07
78	gi 75755983	TO76-23 [Taraxacum officinale]	87	12020	0.33
79	gi 157328520	unnamed protein product [Vitis vinifera]	84	33094	0.11
80	gi 40288318	putative 6 repeat proteinase inhibitor [Nicotiana attenuata]	83	46464	0.08
81	gi 113531030	properoxidase [Picea abies]	81	32859	0.24
82	gi 15219493	peroxidase, putative [Arabidopsis thaliana]	80	37717	0.1
83	gi 15234910	CYTC-2 (CYTOCHROME C-2); electron carrier [Arabidopsis thaliana]	74	12231	0.33
84	gi 121663827	class IV chitinase [Nicotiana tabacum]	76	29885	0.38
85	gi 119012	Glucan endo-1,3-beta-glucosidase, acidic isoform G19 precursor ((1->3)-beta-glucan endohydrolase) ((1->3)-beta-glucanase) (Beta-1,3-endoglucanase) (PR-2B) (PR-36)	74	37699	0.1
86	gi 123965222	germin-like protein [Capsicum chinense]	68	21032	0.18
87	gi 147820458	hypothetical protein [Vitis vinifera]	65	64811	0.06
88	gi 16903357	endo-beta-1,4-glucanase precursor [Nicotiana tabacum]	62	34512	0.14
89	gi 18447763	putative serine carboxypeptidase precursor [Gossypium hirsutum]	57	56558	0.14
90	gi 62909955	peroxidase [Pisum sativum]	54	34257	0.11
91	gi 15231303	ATEXLA1 (ARABIDOPSIS THALIANA EXPANSIN-LIKE A1) [Arabidopsis thaliana]	54	28683	0.13
92	gi 146216710	leucine-rich repeat protein [Nicotiana tabacum]	52	21382	0.18
93	gi 83584404	EIN3-binding F-box protein 2 [Lycopersicon esculentum]	52	71154	0.05
94	gi 417371	Non-specific lipid-transfer protein 2 precursor (LTP 2)[Nicotiana tabacum]	50	11407	0.35
95	gi 55294668	putative ML domain protein [Nicotiana glauca]	49	20139	0.19
96	gi 125573356	hypothetical protein Os_004696 [Oryza sativa (japonica cultivar-group)]	48	43722	0.08
97	gi 157341025	unnamed protein product [Vitis vinifera]	48	43416	0.09

Supplementary Table 2. Proteins identified in 'CV-N without NAA in MS' sample. Recombinant protein is highlighted in red.

Hit number	Database access number	Protein description	Mascot score	Mass	emPAI
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1	gi 63002585	peroxidase [Nicotiana tabacum]	885	35563	4.01
2	gi 75315324	Peroxidase N1 precursor (Peroxidase B2) (Peroxidase B3)	769	35709	3.49
3	gi 116314	Endochitinase A precursor (CHN-A)	691	35134	2.39
4	gi 63253080	peroxidase [Nicotiana tabacum]	531	38505	1.78
5	gi 64976605	peroxidase [Nicotiana tabacum]	521	38740	1.77
6	gi 62362434	nectarin IV [Nicotiana langsdorffii x Nicotiana sanderae]	498	46948	0.85
7	gi 467689	chitinase/lysozyme [Nicotiana tabacum]	498	41993	0.98
8	gi 226121	glucan endoglucosidase [Nicotiana tabacum]	468	39137	1.28
9	gi 129837	Lignin-forming anionic peroxidase precursor (TOPA) [Nicotiana tabacum]	446	34652	1.28
10	gi 131015	Pathogenesis-related protein R major form precursor (Thaumatococcus-like protein E22) [Nicotiana tabacum]	445	24650	1.74
11	gi 108752035 gb ABG02914.1	cyanovirin-N [synthetic construct]	422	10923	7.87
12	gi 148299083	subtilisin-like protease [Nicotiana tabacum]	415	81137	0.37
13	gi 505267	chitinase, class V [Nicotiana tabacum]	401	41754	0.68
14	gi 19970	prb-1b [Nicotiana tabacum]	357	20107	1.86
15	gi 182410392	PR1 protein [Solanum lycopersicum]	334	20110	1.86
16	gi 116332	Acidic endochitinase precursor [Nicotiana tabacum]	332	31263	0.77
17	gi 19859	glucan endo-1,3-beta-glucosidase [Nicotiana tabacum]	316	37835	0.46
18	gi 131017	Pathogenesis-related protein R minor form precursor (PR-R) (PROB12) (Thaumatococcus-like protein E2) [Nicotiana tabacum]	300	24535	1.06
19	gi 116342	Acidic endochitinase Q precursor (Pathogenesis-related protein Q) (PR-Q)	287	27615	0.9
20	gi 130846	Basic form of pathogenesis-related protein 1 precursor (PRP 1) [Nicotiana tabacum]	282	19716	1.04
21	gi 19783	osmotin [Nicotiana tabacum]	266	27110	0.69
22	gi 157355185	unnamed protein product [Vitis vinifera]	215	39303	0.44
23	gi 7547630	pathogen- and wound-inducible antifungal protein CBP20 precursor [Nicotiana tabacum]	207	22153	0.62
24	gi 3913932	Proteinase inhibitor type-2 precursor (Proteinase inhibitor type II) [Nicotiana tabacum]	207	20970	0.96
25	gi 19771	acidic chitinase PR-P [Nicotiana tabacum]	203	27512	0.47
26	gi 2497757	Non-specific lipid-transfer protein 1 precursor (LTP 1) (Pathogenesis-related protein 14) (PR-14) [Nicotiana tabacum]	194	11516	0.34
27	gi 119012	Glucan endo-1,3-beta-glucosidase, acidic isoform G19 precursor ((1->3)-beta-glucan endohydrolase) ((1->3)-beta-glucanase) (Beta-1,3-endoglucanase) (PR-2B) (PR-36) [Nicotiana tabacum]	182	37699	0.46
28	gi 83281191	putative glycerophosphoryl diester phosphodiesterase family protein [Nicotiana tabacum]	179	81954	0.14
29	gi 130840	Pathogenesis-related protein PR-4B precursor [Nicotiana tabacum]	178	16225	1.49
30	gi 30526289	29 kDa chitinase-like thermal hysteresis protein [Solanum dulcamara]	175	28827	0.45
31	gi 6538778	biotic cell death-associated protein [Nicotiana glutinosa]	174	25142	0.53
32	gi 37359708	LEXYL2 [Solanum lycopersicum]	173	68869	0.17
33	gi 218308	peroxidase [Nicotiana tabacum]	173	34504	0.51
34	gi 19962	pathogenesis-related protein 4A [Nicotiana tabacum]	172	16195	1.36
35	gi 547743	Trypsin inhibitor 1 precursor (Trypsin inhibitor I) (NSTI-I)	165	10458	1.65
36	gi 1255951	PS60 [Nicotiana tabacum]	162	60107	0.2
37	gi 5381253	peroxidase [Nicotiana tabacum]	161	34348	0.23
38	gi 32482806	putative xyloglucanase inhibitor [Solanum tuberosum]	146	46779	0.17
39	gi 21495	chitinase; endochitinase [Solanum tuberosum]	145	28777	0.28
40	gi 11528439	chitinase [Fragaria x ananassa]	144	28307	0.29
41	gi 2541876	CND41, chloroplast nucleoid DNA binding protein [Nicotiana tabacum]	142	53413	0.22
42	gi 547732	Proteinase inhibitor I-A precursor (PI-IA) (Inhibitor of microbial serine proteinases minor isoform)	132	11872	0.78
43	gi 37359706	LEXYL1 [Solanum lycopersicum]	126	83111	0.09
44	gi 21668053	non-specific lipid transfer protein [Nicotiana tabacum]	123	11723	0.14
45	gi 113531030	properoxidase [Picea abies]	117	32859	0.24
46	gi 547733	Proteinase inhibitor I-B precursor (PI-IB) (Inhibitor of microbial serine proteinases major isoform)	111	11908	0.78
47	gi 90992878	phyloplanin [Nicotiana tabacum]	108	11302	0.83
48	gi 157355449	unnamed protein product [Vitis vinifera]	107	34368	0.23
49	gi 146216710	leucine-rich repeat protein [Nicotiana tabacum]	105	21382	0.39
50	gi 76446068	trypsin proteinase inhibitor precursor [Nicotiana corymbosa]	101	16198	0.54
51	gi 119507463	pectin methylesterase 2 [Pyrus communis]	99	62606	0.12
52	gi 218304	PR1a protein precursor [Nicotiana tabacum]	97	18578	0.46
53	gi 18403247	receptor protein kinase-related [Arabidopsis thaliana]	97	27846	0.29
54	gi 115486089	Os11g0592000 [Oryza sativa (japonica cultivar-group)]	97	16246	0.54
55	gi 417371	Non-specific lipid-transfer protein 2 precursor (LTP 2)	88	11407	
56	gi 688429	tumor-related protein [Nicotiana glauca x Nicotiana langsdorffii]	87	13798	0.65
57	gi 267124	Thioredoxin H-type 1 (Trx-H1)	87	13947	0.28

58	gi 19948	PR1b preprotein [<i>Nicotiana tabacum</i>]	86	16693	0.52
59	gi 169555	anionic peroxidase	82	37324	0.2
60	gi 85813772	xylan 1,4-beta-xylosidase [<i>Populus alba</i> x <i>Populus tremula</i>]	79	81424	0.05
61	gi 7939581	thionin like protein [<i>Nicotiana tabacum</i>]	76	8571	0.48
62	gi 10798758	P-rich protein NtElG-C29 [<i>Nicotiana tabacum</i>]	75	13376	0.29
63	gi 32351452	class III peroxidase [<i>Gossypium hirsutum</i>]	75	35399	
64	gi 172046673	Unknown protein 1	63	1031	7.22
65	gi 15231303	ATEXLA1 (ARABIDOPSIS THALIANA EXPANSIN-LIKE A1) [<i>Arabidopsis thaliana</i>]	60	28683	0.13
66	gi 536789	PR-1c protein [<i>Nicotiana tabacum</i>]	58	17717	0.21
67	gi 82568706	Serine carboxypeptidase [<i>Prunus mume</i>]	57	55936	0.07
68	gi 498926	glucan endo-1,3-beta-D-glucosidase [<i>Solanum lycopersicum</i>]	56	37839	0.1
69	gi 15219493	peroxidase, putative [<i>Arabidopsis thaliana</i>]	56	37717	0.1
70	gi 62732994	Ribosome inactivating protein, putative [<i>Oryza sativa</i> (japonica cultivar-group)]	54	46294	0.08
71	gi 25044849	peroxidase [<i>Ananas comosus</i>]	50	35591	0.11
72	gi 6716760	methionine synthase [<i>Coffea arabica</i>]	49	24430	0.16
73	gi 115489060	Os12g0560400 [<i>Oryza sativa</i> (japonica cultivar-group)]	48	11736	0.34
74	gi 15218665	peroxidase, putative [<i>Arabidopsis thaliana</i>]	47	34310	0.11