

Supplementary Materials: The Leaf Color Regulation Mechanism in Chimera *Hosta* “Gold Standard” Leaves Revealed from Proteomic Analysis

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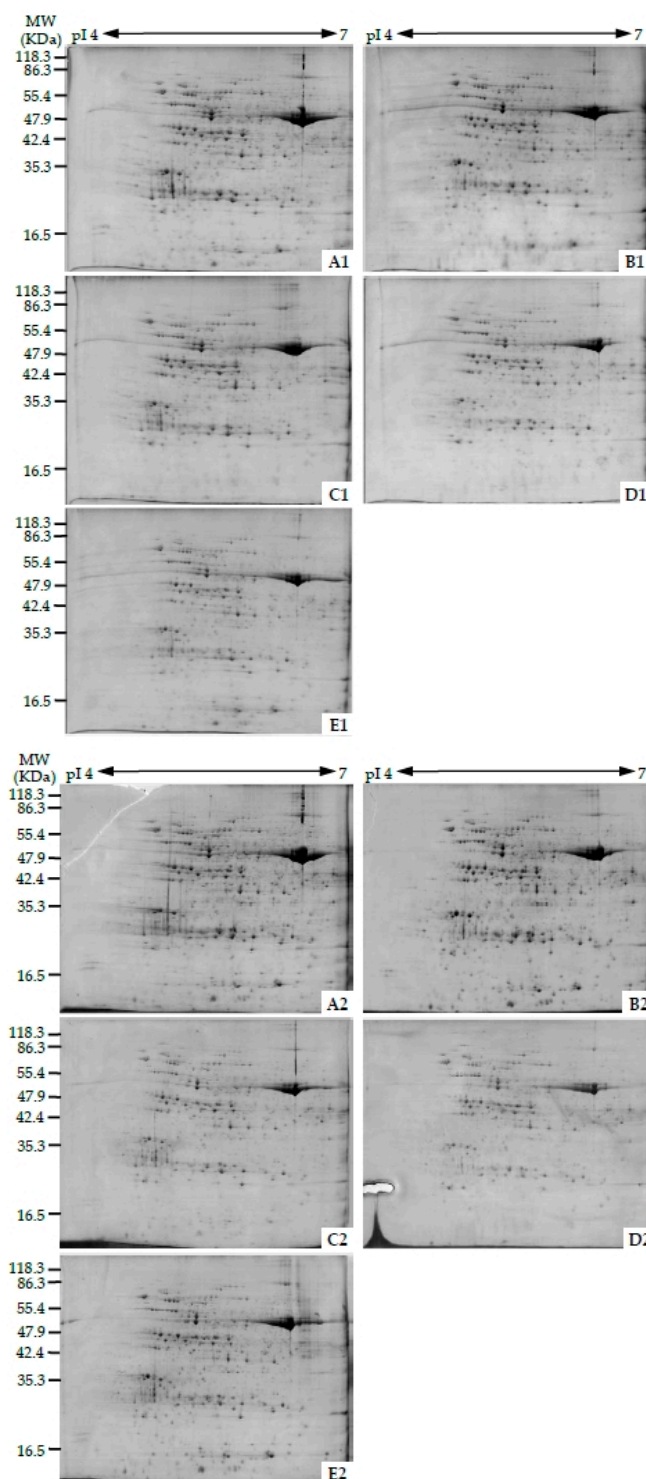


Figure S1. Cont.

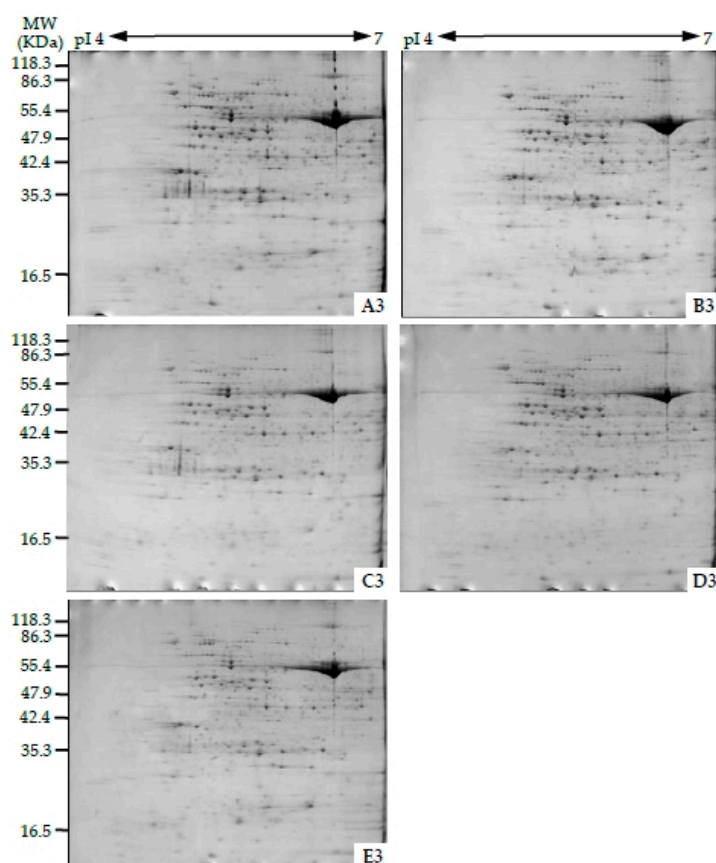


Figure S1. Three biological replicates of 2-DE gels of proteins extracted from various leaf regions in different development stages of *Hosta* “Gold Standard”. (A1–A3) young leaf margin (YLM); (B1–B3) young leaf center (YLC); (C1–C3) mature leaf margin (MLM); (D1–D3) mature leaf center (MLC); (E1–E3) leaf under excess nitrogen fertilization (LENF). Proteins were separated on 24 cm IPG strips (pH 4–7 linear gradient) using isoelectric focusing (IEF) in the first dimension, followed by 12.5% SES-PAGE gels in the second dimension. The 2-DE gel was stained with Coomassie Brilliant Blue. Molecular weight (Mw) in kilodaltons (kDa) and pI of proteins are indicated on the left and top of the gel, respectively.

gi 71361904 spot 795 GS1c	MALLTDLNLDLTDSTEKI IAEYIWIWGGSGMDLRKARTLPGPVTDP SKLPKWN YDGSSTGQAPGEDSEVILYPQAI FKD	80
gi 71361904 spot 806 GS1c	MALLTDLNLDLTDSTEKI IAEYIWIWGGSGMDLRKARTLPGPVTDP SKLPKWN YDGSSTGQAPGEDSEVILYPQAI FKD	80
consensus	malltdlnldltdstekiaeyiwiggsgmdlrskartlpgpvt dpsklpkwnydgsgtgqapgedsevilypqai fkd	
gi 71361904 spot 795 GS1c	PFRKGNNILVMDCYTPAGVPIPTNKRYNAV KIFSNPDVAKEEPWY GIEQ EYTL LQKDINWPLGWPVGGFPGPGQGPYYCS	160
gi 71361904 spot 806 GS1c	PFRKGNNILVMDCYTPAGVPIPTNKRYNAV KIFSNPDVAKEEPWY GIEQ EYTL LQKDINWPLGWPVGGFPGPGQGPYYCS	160
consensus	pfrkgnnilvmcdcytpagvp iptnkrynavkifsnpdvakeepwygieqeytllqkdinwplgwpvggfp gpgqgpyy cs	
gi 71361904 spot 795 GS1c	IGADKSFGRDIVDSHYKACLFAGVNI SINGINGEVM PGQW E FQVGPTVGISAGDQVWVARYLLERITEIAGVVVTFDPKPIP	240
gi 71361904 spot 806 GS1c	IGADKSFGRDIVDSHYKACLFAGVNI SINGINGEVM PGQW E FQVGPTVGISAGDQVWVARYLLERITEIAGVVVTFDPKPIP	240
consensus	igadksfgrdivdshy k a c l f a g v n i s i n g i n g e v m p g q w e f q v g p t v g i s a g d q v w v a r y l l e r i t e i a g v v t f d p k p i p	
gi 71361904 spot 795 GS1c	GDWNGAGAHTNYSTESMRKDG GFKVIVDAVEK LK LKHKEHIAAYGEGNERRLTGKHETADINTFSWGVANRGASVRVGRE	320
gi 71361904 spot 806 GS1c	GDWNGAGAHTNYSTESMRKDG GFKVIVDAVEK LK LKHKEHIAAYGEGNERRLTGKHETADINTFSWGVANRGASVRVGRE	320
consensus	gdwn g a g a h t n y s t e s m r k d g g f k v i v d a v e k l k l k h k e h i a a y g e g n e r r l t g k h e t a d i n t f s w g v a n r g a s v r v g r e	
gi 71361904 spot 795 GS1c	TEQNGKGYFEDRRPASNM DPYVVTSMIAETTILWK	355
gi 71361904 spot 806 GS1c	TEQNGKGYFEDRRPASNM DPYVVTSMIAETTILWK	355
consensus	teqngkgy fedrrpasnm dpyvvt smiaett ilwk	A
gi 194396261 spot 338 TK	MAS S S L T L S Q A I L S R S V P R H G S A S S S O L S P S S I T F S G L K S N P N I T T S R R R T P S S A A A A V V R S P A T R A S A A T E T I E K T E	80
gi 224063766 spot 346 TK	MAS S S L T L S Q A I L A R A V S H N A I D N L R D S R L S L V S L P A F S G L K S T I C T A T R A T T T S R R R R V S S Q R V R A A V E T L D A T E	80
consensus	mas s s l t l s q a l r v s s s o l s p s s i t f s g l k s n p n i t t s r r r v s s r a a t e	
gi 194396261 spot 338 TK	T L V L K S V N T I R F L A I D A V E K A N S G H P G L P M G C A P M G H I L Y D E V M R Y N P K N P Y W F N R D R F V L S A G H G C M L Q Y A L L H L A G Y	160
gi 224063766 spot 346 TK	T S L V E K S V N T I R F L A I D A V E K A N S G H P G L P M G C A P M G H I L Y D E V M R Y N P K N P Y W F N R D R F V L S A G H G C M L Q Y A L L H L A G Y	160
consensus	t l v k s v n t i r f l a i d a v e k a n s g h p g l p m g c a p m g h i l y d e v m r y n p k n p y w f n r d r f v l s a g h g c m l q y a l l h l a g y	
gi 194396261 spot 338 TK	D A V R E E D L K S F R Q W G S R T P G H P E N F E T P G V E V T T G P L G Q G I A N A V G L A L V E K H L A A R N K P D A E I V D H Y T Y V I L G D G C Q M	240
gi 224063766 spot 346 TK	T S V K E E D L K S F R Q W G S R T P G H P E N F E T P G V E V T T G P L G Q G I A N A V G L A L V E K H L A A R N K P D N E I V D H Y T Y V I L G D G C Q M	240
consensus	d v e e d l k s f r q w g s t p g h p e n f e t p g v e v t t g p l g q g i a n a v g l a l e k h l a a r n k p d a e i v d h y t y v i l g d g c q m	
gi 194396261 spot 338 TK	E G I S Q E A C S L A C H W G L G K L I A F Y D D N H I S I D G D T E I A F T E D V C A R F E L G W H V I W K N G N T G Y D E I R A A I K E A K V I T D K P	320
gi 224063766 spot 346 TK	E G I S N E A C S L A A H W G L G K L I A F Y D D N H I S I D G D T E I A F T E D V K R F E L G W H V I W K N G N T G Y D E I R A A I K E A K S V R D K P	320
consensus	egis q e a c s l a h w g l g k l i a f y d d n h i s i d g d t e i a f t e d v r f e l g w h v i w k n g n t g y d e i r a a i k e a k v d k p	
gi 194396261 spot 338 TK	T I K V T T T I G G S P N K A N S Y S V H G S A L G A K E V D A T R N L G W P Y E P F H V P E D V K H W S R H V T E G A A E A G W N T K F A E Y E K K	400
gi 224063766 spot 346 TK	T I K V T T T I G G S P N K A N S Y S V H G S A L G A K E V D A T R N L G W P Y E P F H V P E D V K H W S R H T P A G A A F A E A W N K F A E Y E K K	400
consensus	t i k v t t t i g g s p n k a n s y s v h g s a l g a k e v a t r n l g w p y e p f h v p e d v k h w s r h g a a e a w n k f a e y e k k	
gi 194396261 spot 338 TK	Y E E E A A E L K S I T T G E L P A G W E K A L P T Y T P E S P A D A T R N L S Q Q N L N A L V K V L P G L G G S A D L A S S N M T L K M F G D F Q K T P	480
gi 224063766 spot 346 TK	Y S E E A A E L K S I A N G E L P A G W E K A L P T Y T P E S P A D A T R N L S Q Q N L N A L V K V L P G L G G S A D L A S S N M T L K M F G D F Q K T P	480
consensus	y e e a a e l k s i g e l p a g w e k a l p t y t p e s p a d a t r n l s q q n l n a l v k v l p g l g g s a d l a s s n m t l k m f g d f q k t p	
gi 194396261 spot 338 TK	E E R N V R F G V R E H G M G A I C N G I A L H S P G L I P Y C A T F F V F T D Y M R A M R I S A L S E A G V I Y V M T H D S I G L G E D G P T H Q P I E H L	560
gi 224063766 spot 346 TK	E E R N V R F G V R E H G M G A I C N G I A L H C P G L I P Y C A T F F V F T D Y M R A M R I S A L S E A G V I Y V M T H D S I G L G E D G P T H Q P I E H L	560
consensus	e e r n r f g v r e h g m g a i c n g i a l h p g l i p y c a t f f v f t d y m r a m r i s a l s e a g v i y v m t h d s i g l g e d g p t h q p i e h l	
gi 194396261 spot 338 TK	A S F R A M P N I L M R P A D G N E T A G A Y K V A V I K M K T P S I L A L S R Q K L P O L A G S S I E C A K G G Y I L S D N S S C N K P D V I L I G T G S	640
gi 224063766 spot 346 TK	A S F R A M P N I L M R P A D G N E T A G A Y K V A V I N K R P S I L A L S R Q K L P O L A G T S I E C V K G G Y I L S D N S S C N K P D V I L I G T G S	640
consensus	a s f r a m p n i l m r p a d g n e t a g a y k v a v i k p s i l a l s r q k l p o l g s i e c k g g y i s d n s s n k p d v i l i g t g s	
gi 194396261 spot 338 TK	E L E I A V K A A E L R K E G K A V R V V S F V W E L F E Q S A D Y K E S V L E S S V T A R V S I E A G S T F G W E K Y V G S K G K A I G I D R F G A S A	720
gi 224063766 spot 346 TK	E L E I A K A A E L R K E G K A V R V V S F V S N E L F E Q S D A Y K E S V L E A A V E A R V S I E A G S T F G W E K I V G K G K A I G I D R F G A S A	720
consensus	e l e i a k a a e l r k e g k a v r v v s f v w e l f e q s a d y k e s v l e s s v t a r v s i e a g s t f g w e k y v g s k g k a i g i d r f g a s a	
gi 194396261 spot 338 TK	P A G K I Y K E Y G I T E A V V A A A K V	743
gi 224063766 spot 346 TK	P A G K I Y K E Y G I T E A V T A A A K E V	743
consensus	p a g k i y k e y g i t e a v a a a k v	B

Figure S2. Cont.

gi 242090109 spot 459 RBP- αMASANAI STASLISPLSQ GRARRARNGRSQRTFVVRBAKDIATFDOK..SRAAL	51
gi 223948025 spot 472 RBP- βMASTFGATSTVGLMAA.PTGKNVRLQRRANFRVKAAKELVFNKDGSAIKKL	50
gi 225442531 spot 476 RBP- β	MASSFATMSSSIGYFTSSSSHTMDKKFLGSSRRLESFASISANSFEGRKQSMVLQKRCSFPKVRAMAKELVFNKDGSAIKKL	80
consensus	v ak f l	
gi 242090109 spot 459 RBP- α	CAGVEKLANAVGVTLGPRGRNVVLD.EYGSPPVNDGVITARA TELYDP ENAGALIREVASKTND SAGD GTITIASVLA	130
gi 223948025 spot 472 RBP- β	CTGVNKLADLVGVTLGPKGRNVVLESKYGSPKIVNDGVTVAREVELDDPVENIGARLVROAPAKTND LAGD GTITIVVLA	130
gi 225442531 spot 476 RBP- β	CTGVNKLADLVGVTLGPKGRNVVLESKYGSPKIVNDGVTVAREVELDDPVENIGARLVROAPAKTND LAGD GTITIVVLA	160
consensus	q gv kla vgvtlgp grnvvl ygspp vndgvt ar el dp en ga l r a ktnd agdgttt vla	
gi 242090109 spot 459 RBP- α	REITIKLCLLSVTSGANPVSLKKGIDKTVQGLICELENKARPKGGGDIKAVASISAGNDEFTGSMIAEATDKVGPDPVLS	210
gi 223948025 spot 472 RBP- β	QGLIABGVKVVAAGANPVQITRGIETAKAVBELRKLKSKVEEDS.ELADVAAVSAGNNYIEIGNMIAEAMSVKGRKGVVT	209
gi 225442531 spot 476 RBP- β	QGLITEGVKVVAAGANPVQITRGIETAKAVBELRKLKSKVEEDS.ELADVAAVSAGNNYIEIGNMIAEAMSVKGRKGVVT	239
consensus	i g v ganpv gi t l el v va sagn g miaea vg gv	
gi 242090109 spot 459 RBP- α	TESSSEFTTVEVBEGMEDRGIYSQFVTNLEKSI VEENAKVLT TDCKISSIKEIL PILEKTTQLRAEL IIAEDITG	290
gi 223948025 spot 472 RBP- β	TEEGKSENFLVVEGMQDRGIYSFVFTDSEKMAVE ENCKLLV PKITNARDLIN VEDAIRGAE PIIIIAEDIEQ	289
gi 225442531 spot 476 RBP- β	TEEGKSAENNLVVEGMQDRGIYSFVFTDSEKMAVE ENCKLLV PKITNARDLIN VEDAIRGAE PIIIIAEDIEQ	319
consensus	e s e v egm drgyisp fvt ek e en k l d ki le p iiaedi	
gi 242090109 spot 459 RBP- α	EALATLVVNKLRGILNVAARKAPSGFERKKA VL ODIAITGA EF LAKDLGLLVEN TEAQLGTARKVT HQTITTTIADA	370
gi 223948025 spot 472 RBP- β	EALATLVVNKLRGSLKTAARKAPSGFERKKT QYLD DAITGTATVIRDEVGLSLDKADK SVLGTAAK VLTKESTTIVGDS	369
gi 225442531 spot 476 RBP- β	EALATLVVNKLRGALKTAARKAPSGFERKKS QYLD DAITGTATVIREEVGLSLDKAE EV LGHAAKVLTKDITTTIVGDS	399
consensus	ealatlvvnklrg l aa kap fger l diai tg gl a lg a kv tt d	
gi 242090109 spot 459 RBP- α	ASKDEIQARVA LKKELAE TSVYDTEKLA ERIAKL AGGVAVIRVGAATETEL EDRQLR IEDAKNATEAAIEEGIVPGGG	450
gi 223948025 spot 472 RBP- β	STQEEVTKRVACIKNLIEAAEQYEKEKLN ERIAKL AGGVAVICVGAATETEL KEKKLR VEDA NATKA AVEEGIVVGGG	449
gi 225442531 spot 476 RBP- β	STQEA VNKR VACIRNLVEAADQYEKEKLN ERIAKL SGGVAVICVGAATETEL KEKKLR VEDA NATKA AVEEGIVVGGG	479
consensus	rvaq y ekl eriakl ggavai vga tetel lr eda nat aa eegiv ggg	
gi 242090109 spot 459 RBP- α	TAYVHLSTVPSIK EKTE EDPRLGADTIQKALVA SLIA NAGVGE EV VVEKIKDS.VWEVGYNAMTDTYENIT EAGV	529
gi 223948025 spot 472 RBP- β	CTLLRLAAKVDAIK TE LENDEQKVGA ITVRRAL SYPLKLI ARNAGV NGSVVTEKVLSDNPKYGYNA PTQ QVEDIMPA GI	529
gi 225442531 spot 476 RBP- β	CTLLRLAAKVDAIK TE LENDEQKVGA ITVRRAL SYPLKLI ARNAGV NGSVVIEKVLSSDNPKYGYNA PTQ QVEDIMPA GI	559
consensus	l v ik ga i al p lia nagv g vv ek gyna t ye l ag	
gi 242090109 spot 459 RBP- α	IDP AKVRC ALONAASVAGMVL LTQAL VVEKPK KFQ .VAESEGS LAV	577
gi 223948025 spot 472 RBP- β	IDP TKVRC OLEEHAASVAK TFL ASD VVV VDIKEE EAAP VANEMD NSGYG	578
gi 225442531 spot 476 RBP- β	IDP AKVRC OLEEHAASVART FL ASDA VVV VDIKEE EP IPAGNEMD NSGYG	608
consensus	idp kv rc l aasva lt vv p p s	

C

gi 255646270 spot 832 PRK	MAACTVYSTQSLSTNCSISTPSKTQLSFFHQKQVFWRNSSKKGSCSSRRSYVITCAAGDSQ TV VIGLAADSGCGKSTFMR	80
gi 1885326 spot 840 PRKAGDSQ TV VIGLAADSGCGKSTFMR	24
consensus	agdsqt viglaadsgcgkstfmr	
gi 255646270 spot 832 PRK	RLTSVFGGAAEPPKGGNPDSENTLISD TTT VICLDDYHSLDRTGRKEKGV TALDE BRANDFDLMYEQVKA KDG VAVEKPIY	160
gi 1885326 spot 840 PRK	RLTSVFGGAAEPPKGGNPDSENTLISD TTT VICLDDYHSLDRTGRKEKGV TALDE BRANDFDLMYEQVKA KDG SKVCKPIY	104
consensus	rltsvfggaaepkgnpdsentlisdtttvi clddyhslrdrtgrkekgv taldp andfdlmyeqvka kdg v kpiy	
gi 255646270 spot 832 PRK	NHVTGLLDPELIKPPKILVIEGLHPMD SRVR LLDFSIYLDISNEVKFAWKIQ RDMA ERGHSL ESIKASIE ARKPD FE	240
gi 1885326 spot 840 PRK	NHVTGLLDPELIKPPKILVIEGLHPMD SRVR LLDFSIYLDISNEVKFAWKIQ RDMA ERGHSL ESIKASIE ARKPD FE	184
consensus	nhvtgll d pelikppkilvieglhpm dsrvr lldfs iyl disnevkfawki qrdmaerghslesikasiearkpdfe	
gi 255646270 spot 832 PRK	AYIDPQKQYADAVIEVLPTQLIPDDNEGKILRVRLIQ RGV KYFSPVYLFDDGSTISWIPCGRKL TC SYPGIKFFYGPET	320
gi 1885326 spot 840 PRK	AYIDPQKQYADAVIEVLPTQLIPDDNEGKILRVRLIQ RGV KYFSPVYLFDDGSTISWIPCGRKL TC SYPGIKFFYGPET	264
consensus	ayidpqkyadavievlptqlipddnegkilrvrliq rgv kyf spvylfd gstiswipcgrkl tcsypgikffygpet	
gi 255646270 spot 832 PRK	YKGNEV LV VEMDGGQFDR LD ELIYVESHL SNLS TKFYGEV TQ QMLKHADFP GS NNGTG GF FQ TI VGLKIR DL VEQLTAS KAE	400
gi 1885326 spot 840 PRK	YKGNEV LV VEMDGGQFDR LD ELIYVESHL SNLS TKFYGEV TQ QMLKHADFP GS NNGTG GF FQ TI VGLKIR DL VEQLTAS KAE	344
consensus	y gnev lv emdggqfdrldeliyveshlsnls kfygevtq qmlkhadfp gsnngtg fgtivglkirdl eq as ae	
gi 255646270 spot 832 PRK	TPV AKKA	407
gi 1885326 spot 840 PRK	TPV AKKA	351
consensus	tpv a	

D

Figure S2. Cont.

gi 225449497 spot 355 HSC70-1	MACKGE.....GPAIGIDLGTITSCVGVVQHDRVEITANDQGNRTTPSVAFDTT.ERLIGDAAKNQVAMNPNNTVFDA	73
gi 225449497 spot 361 HSC70-1	MACKGE.....GPAIGIDLGTITSCVGVVQHDRVEITANDQGNRTTPSVAFDTT.ERLIGDAAKNQVAMNPNNTVFDA	73
gi 225573627 spot 358 HSC70-2	MACKGE.....GPAIGIDLGTITSCVGVVQHDRVEITANDQGNRTTPSVAFDTT.ERLIGDAAKNQVAMNPNNTVFDA	73
gi 225434984 spot 367 HSC70-2	MACKGE.....GPAIGIDLGTITSCVGVVQHDRVEITANDQGNRTTPSVAFDTT.ERLIGDAAKNQVAMNPNNTVFDA	73
gi 147805297 spot 332 cpHSP70	MVRKASFHVVNEKVVIGDLGTTISAAAMEGGKPTITVNAEGORTTPSVVAYTKNGDMVGCIAARCAVNVNPNNTFESV	80
consensus	m k g d l g t t s v i n g r t t p s v a t l g a k q n p n t f	
gi 225449497 spot 355 HSC70-1	KRLIGRRFSDASVQSDIKHWSFKVVPFGDKPMITVYKGEDKQFAAEETISSMVLTKMRETAAYLGSTVKNNAVVTVPAY	153
gi 225449497 spot 361 HSC70-1	KRLIGRRFSDASVQSDIKHWSFKVVPFGDKPMITVYKGEDKQFAAEETISSMVLTKMRETAAYLGSTVKNNAVVTVPAY	153
gi 225573627 spot 358 HSC70-2	KRLIGRRFSDASVQSDIKHWSFKVVPFGDKPMITVYKGEDKQFAAEETISSMVLTKMRETAAYLGSTVKNNAVVTVPAY	153
gi 225434984 spot 367 HSC70-2	KRLIGRRFSDSSVQSDIKLWPEKVIIPGPGDKPMIVVYKGEKQFAAEETISSMVLTKMRETAAYLGSTVKNNAVVTVPAY	153
gi 147805297 spot 332 cpHSP70	KRLIGRKMD..EVGEEAKQVSKVWRDENG..VKLECPVLAKFAAEETISQVLRKLADDKSKFNDKVKTKNAVVTVPAY	156
consensus	kr igr v k kv k faaeetis vl k a l v avvtvpay	
gi 225449497 spot 355 HSC70-1	FNDSQROATKDAGVIAGLNVMRIINEPTAAATAYGLDKKATSVGKKNVLI FDLGGGTDFVSLITIEEGHFEVKATAGDTH	233
gi 225449497 spot 361 HSC70-1	FNDSQROATKDAGVIAGLNVMRIINEPTAAATAYGLDKKATSVGKKNVLI FDLGGGTDFVSLITIEEGHFEVKATAGDTH	233
gi 225573627 spot 358 HSC70-2	FNDSQROATKDAGVIAGLNVMRIINEPTAAATAYGLDKKATSVGKKNVLI FDLGGGTDFVSLITIEEGHFEVKATAGDTH	233
gi 225434984 spot 367 HSC70-2	FNDSQROATKDAGVIAGLNVMRIINEPTAAATAYGLDKKATSVGKKNVLI FDLGGGTDFVSLITIEEGHFEVKATAGDTH	233
gi 147805297 spot 332 cpHSP70	FNDSQRTATKDAGRIAGLEVLRINEPTAAATAYGFEKKNN...ETILVFDLGGGTDFVSLIEVGDGFEVFLSLISGDTH	232
consensus	fndsqr atkdag iagl v riineptaa ayg kk l fdlgggtfdvs l g fev t g dth	
gi 225449497 spot 355 HSC70-1	LGGEDFDNRMVNHVQCFKKRKHKKITGNARSLRRIRTSOBRARKLSSTAQTITIEI...DSLYEC..IDFYSTITRARFE	309
gi 225449497 spot 361 HSC70-1	LGGEDFDNRMVNHVQCFKKRKHKKITGNARSLRRIRTSOBRARKLSSTAQTITIEI...DSLYEC..IDFYSTITRARFE	309
gi 225573627 spot 358 HSC70-2	LGGEDFDNRMVNHVQCFKKRKHKKITGNARSLRRIRTSOBRARKLSSTAQTITIEI...DSLYEC..IDFYSTITRARFE	309
gi 225434984 spot 367 HSC70-2	LGGEDFDNRMVNHVQCFKKRKHKKITGNARSLRRIRTSOBRARKLSSTAQTITIEI...DSLYEC..IDFYSTITRARFE	309
gi 147805297 spot 332 cpHSP70	LGGEDFDKRIIDWLAQNFKRDEGILLKDKQATCRITETAFAKIEISSLTQTNISLPFITATSECPKHITETITRAKFE	312
consensus	lgg dfd r v q fkr d l rl e ak lss qt i eg t tra fe	
gi 225449497 spot 355 HSC70-1	ELNMDLFRKCMPEVEKCLRDAKMDKSTIHDVVLVGGSTRIPKVOQLQDFFNGKELCKSINPDEAVAGAAVQAAILSGE	389
gi 225449497 spot 361 HSC70-1	ELNMDLFRKCMPEVEKCLRDAKMDKSTIHDVVLVGGSTRIPKVOQLQDFFNGKELCKSINPDEAVAGAAVQAAILSGE	389
gi 225573627 spot 358 HSC70-2	ELNMDLFRKCMPEVEKCLRDAKMDKSTIHDVVLVGGSTRIPKVOQLQDFFNGKELCKSINPDEAVAGAAVQAAILSGE	389
gi 225434984 spot 367 HSC70-2	ELNMDLFRKCMPEVEKCLRDAKMDKSTIHDVVLVGGSTRIPKVOQLQDFFNGKELCKSINPDEAVAGAAVQAAILSGE	389
gi 147805297 spot 332 cpHSP70	ELCSDLDLRLRTPVETALRDALSFKDLDBVVLVGGSTRIPKVOQLVRKMT..GKEPNVTNPNDEVALGAAVQAAGVLAC	390
consensus	el dl pve lrdak v lvggstrip vq l gke npde va gaavqa l g	
gi 225449497 spot 355 HSC70-1	GNEKVQDLLLLDVTPLSLGLETAGGVMIVLIIPRNTTILPTKKEQVFSTYSDNCPGVLIQVYGERTRTRDNNLLCKFELSG	469
gi 225449497 spot 361 HSC70-1	GNEKVQDLLLLDVTPLSLGLETAGGVMIVLIIPRNTTILPTKKEQVFSTYSDNCPGVLIQVYGERTRTRDNNLLCKFELSG	469
gi 225573627 spot 358 HSC70-2	GNEKVQDLLLLDVTPLSLGLETAGGVMIVLIIPRNTTILPTKKEQVFSTYSDNCPGVLIQVYGERTRTRDNNLLCKFELSG	469
gi 225434984 spot 367 HSC70-2	GNEKVQDLLLLDVTPLSLGLETAGGVMIVLIIPRNTTILPTKKEQVFSTYSDNCPGVLIQVYGERTRTRDNNLLCKFELSG	469
gi 147805297 spot 332 cpHSP70	...DVSNIIVLLDVTPLSLGLETILGGVMITKIIIPRNTTILPTKSEVVFSTAAAGCTISVEINVLQGEREFVRDNKSLGSEFLDC	467
consensus	v lldvtplslglet ggvmv iprntt pt k vfst d q v i v ger rdn lg f l g	
gi 225449497 spot 355 HSC70-1	IPPAPRGVPQITVCFDIDANGILNVSAEDKTTGCKNKITITNDKGRLSKEETIENMVQEAKEYKSEDEBHHKKVKAANKALE	549
gi 225449497 spot 361 HSC70-1	IPPAPRGVPQITVCFDIDANGILNVSAEDKTTGCKNKITITNDKGRLSKEETIENMVQEAKEYKSEDEBHHKKVKAANKALE	549
gi 225573627 spot 358 HSC70-2	IPPAPRGVPQITVCFDIDANGILNVSAEDKTTGCKNKITITNDKGRLSKEETIENMVQEAKEYKSEDEBHHKKVKAANKALE	549
gi 225434984 spot 367 HSC70-2	IPPAPRGVPQITVCFDIDANGILNVSAEDKTTGCKNKITITNDKGRLSKEETIENMVQEAKEYKSEDEBHHKKVKAANKALE	549
gi 147805297 spot 332 cpHSP70	IPPAPRGVPQIEVCFDIDANGILSVTAVDKGTGKKQDITITIG.ASTIPNDEVDRMVKEAEKFAKEKFKRDAIDTKNQAD	546
consensus	ippaprgvpqi v fdidangil v a dk tg k itit l e mv eaek ed e kn	
gi 225449497 spot 355 HSC70-1	NYAYNMRNTVKDEKISAKLEPPADKKKKIEDAVEQAIQWLDSNQLAAEDEFEDKMKELIESICNPIIAKMYOCAGGPDACAGA	629
gi 225449497 spot 361 HSC70-1	NYAYNMRNTVKDEKISAKLEPPADKKKKIEDAVEQAIQWLDSNQLAAEDEFEDKMKELIESICNPIIAKMYOCAGGPDACAGA	629
gi 225573627 spot 358 HSC70-2	NYAYNMRNTVKDEKISAKLEPPADKKKKIEDAVEQAIQWLDSNQLAAEDEFEDKMKELIESICNPIIAKMYOCAG.GEMGA.G	627
gi 225434984 spot 367 HSC70-2	NYAYNMRNTIKDEKIGAKLTIPADKKKKIEDAIDQAIQWLDSNQLAAEDEFEDKMKELIESICNPIIAKMYOCAG.PDMGG.P	627
gi 147805297 spot 332 cpHSP70	SVVYQTEKQLK...ELGDKVPAVKKVKEAKLKKELKDAISSG..STTQTKPAAALNQEVMOISQSLYSQACAPGACAGA	622
consensus	y k k k k e s d m l i y ag g	
gi 225449497 spot 355 HSC70-1	MDDDGPSAGGSAGGPKIEEVD	650
gi 225449497 spot 361 HSC70-1	MDDDGPSAGGSAGGPKIEEVD	650
gi 225573627 spot 358 HSC70-2	MEDDAF..AGGSAGGPKIEEVD	647
gi 225434984 spot 367 HSC70-2	MEDDVFPASGSAGGPKIEEVD	648
gi 147805297 spot 332 cpHSP70	GAAGSTDSKPDGDVIDADFTES	644
consensus	g	

Figure S2. Cont.

gi 9294618 spot 170 PreP1	MSLTSSSTAALRVPSRNLRRRISSPSVAGRLLRLRRGLRIPSAAVRSVNGQFSRLSVRAVATQAPAPLYPDVGQDEAEKLGFE	80
gi 9294618 spot 179 PreP1	MSLTSSSTAALRVPSRNLRRRISSPSVAGRLLRLRRGLRIPSAAVRSVNGQFSRLSVRAVATQAPAPLYPDVGQDEAEKLGFE	80
consensus	msltssstaalrvpsrnlrrrissp svagrllrlrrglripsaavrsvngqfsrlsvravatqapaplypdvggqdeaeaklgfe	
gi 9294618 spot 170 PreP1	KVSEEFISECKSKAILFKHKKTGCEVMSVSNEDENKVFVGVFRTPPKDDSTGIPHILEHSVLGSRKYPVKKEPFVELLKGS	160
gi 9294618 spot 179 PreP1	KVSEEFISECKSKAILFKHKKTGCEVMSVSNEDENKVFVGVFRTPPKDDSTGIPHILEHSVLGSRKYPVKKEPFVELLKGS	160
consensus	kvseefiseckskailfkhhktgcevmsvsnedenkfvfgvfrtppkdstgiphilehsvlgsrkypvkepfvellkgs	
gi 9294618 spot 170 PreP1	LHTFLNAFTYPDRTCYPVASTNTKDFYNLVVDVLDVAVFFPKCVDDAHTFQOEGWHYELNDPSEDISYKGVVFNEMKGVYS	240
gi 9294618 spot 179 PreP1	LHTFLNAFTYPDRTCYPVASTNTKDFYNLVVDVLDVAVFFPKCVDDAHTFQOEGWHYELNDPSEDISYKGVVFNEMKGVYS	240
consensus	lhtflnaftypdrtcypvastntkdfynlvdvyldavffpkcvddahtfqogegwhyelndpsedis ykgvvfnemkgvys	
gi 9294618 spot 170 PreP1	QPDNILGRIAQQALSPENTYGVDSGGDPKDI PNLTFFEEFKEFHRQYYHPSNARIWIFYGDDDPVHRLVRLSEYLDMEFASP	320
gi 9294618 spot 179 PreP1	QPDNILGRIAQQALSPENTYGVDSGGDPKDI PNLTFFEEFKEFHRQYYHPSNARIWIFYGDDDPVHRLVRLSEYLDMEFASP	320
consensus	qpdnilgriaqqalspentygvdsggdpkdipnltf eefkefhrqyyhpsnariwfygdddpvhrlvrlseyldmfeasp	
gi 9294618 spot 170 PreP1	SPNSSKIKFQKLFSEPVRLVEKYPAGRDGDLKKHMLCVNWLLSEKPLDLQTQLALGFLDHLMLGTPASPLRKILLESGL	400
gi 9294618 spot 179 PreP1	SPNSSKIKFQKLFSEPVRLVEKYPAGRDGDLKKHMLCVNWLLSEKPLDLQTQLALGFLDHLMLGTPASPLRKILLESGL	400
consensus	spnsskikfqlfsepvrlveky pagrdgdlkkhmlcvnwllsekpdlqltqlalglf dhlmlgt pasplr killesgl	
gi 9294618 spot 170 PreP1	GEALVSSGLSDELLQPFQIGLKVSEENVQKVEELIMDTLKKLAEEGFDNDAVEASMTNIEFSLRENNTGSPFRGLSLM	480
gi 9294618 spot 179 PreP1	GEALVSSGLSDELLQPFQIGLKVSEENVQKVEELIMDTLKKLAEEGFDNDAVEASMTNIEFSLRENNTGSPFRGLSLM	480
consensus	gealvssglsdellqpqfiglkgvseenvqkveelimdtlkklaeegfndaveasmntiefslrenntgspfrglslm	
gi 9294618 spot 170 PreP1	LQSISKWIYDMDPFEPLKYTEPLKALKTRIAEEGSKAVFSPLIEKLILNNSHRVTIEMQPDPEKATQEEVEEKNILEKVK	560
gi 9294618 spot 179 PreP1	LQSISKWIYDMDPFEPLKYTEPLKALKTRIAEEGSKAVFSPLIEKLILNNSHRVTIEMQPDPEKATQEEVEEKNILEKVK	560
consensus	lqsiskwiymdpfeplkyteplkalktriaeeegskavf splieklilnns hrvtiemqpdpek atqeeveeknilekvk	
gi 9294618 spot 170 PreP1	AAMTEEDLAELARATEELKLKQETPDPEALRCVPSNLNGDIPKEPTYVPTEVGDINGVKVLRHDLFTNDIITYEVVFDI	640
gi 9294618 spot 179 PreP1	AAMTEEDLAELARATEELKLKQETPDPEALRCVPSNLNGDIPKEPTYVPTEVGDINGVKVLRHDLFTNDIITYEVVFDI	640
consensus	aamteedlaelarateelklkqetpdpealrcvpsnlng dipkeptyvp tevgdingvkv lrhdlftndii ytevvfdi	
gi 9294618 spot 170 PreP1	GSCLKHELLPLVPLFCQSLLEMGTKDLTFVQLNQLIGRKTGGISVYPLTSSVRGKDEPCSKIIVRGKSMAGRADDLFNLMN	720
gi 9294618 spot 179 PreP1	GSCLKHELLPLVPLFCQSLLEMGTKDLTFVQLNQLIGRKTGGISVYPLTSSVRGKDEPCSKIIVRGKSMAGRADDLFNLMN	720
consensus	gslkhellplvplfcqsllemgtkdltfvqlnqligrktggisv ypl tssvrgkdep cskii vrgksmagraddlfnlmn	
gi 9294618 spot 170 PreP1	CLLQEVQFTDQQRFKQFVSQSRARMENLRGSGHGIAAARMADAMLNIAAGWMSEQMGGLSYLEFLHLEKKVDEDWEGISS	800
gi 9294618 spot 179 PreP1	CLLQEVQFTDQQRFKQFVSQSRARMENLRGSGHGIAAARMADAMLNIAAGWMSEQMGGLSYLEFLHLEKKVDEDWEGISS	800
consensus	cllqevqftdqqr fkgfvsqsrarmenlrsgshgiaaarmadaml niaagwmseqm ggl syleflhlekkvdedwegiss	
gi 9294618 spot 170 PreP1	SLEEIRRSLLARNGCIVNMTADGKSLTNVEKSAKFLDLLPENPSGGLVTWDGRLPLRNEAIVIPTQVNVYVKGAGNIYST	880
gi 9294618 spot 179 PreP1	SLEEIRRSLLARNGCIVNMTADGKSLTNVEKSAKFLDLLPENPSGGLVTWDGRLPLRNEAIVIPTQVNVYVKGAGNIYST	880
consensus	sleeirrsllarngcivnmtadgksltnveksakfldll penpsgg lvtwdgrlplrneaiviptqvnvyvkgagniyst	
gi 9294618 spot 170 PreP1	GYELDGSAYVISKHISNTWLWDRVRVSGGAYGGFCDFDSHSGVFSYLSYRDPNLLKTLTDIYDGTGDFLRGLDQVDETITK	960
gi 9294618 spot 179 PreP1	GYELDGSAYVISKHISNTWLWDRVRVSGGAYGGFCDFDSHSGVFSYLSYRDPNLLKTLTDIYDGTGDFLRGLDQVDETITK	960
consensus	gyeldgsayviskhisntwldr vrvsggayggfcdf dshsgv fsysyrdpnllktldi ydgtgd flrgldv dgetitk	
gi 9294618 spot 170 PreP1	AIIGTIGDVDSYQLPDAKGYSSLLRHLLGVTDEERQKRKEIILTTSLKDFKDFQAQIDVVRDKGVAVAVASAEDIDAANN	1040
gi 9294618 spot 179 PreP1	AIIGTIGDVDSYQLPDAKGYSSLLRHLLGVTDEERQKRKEIILTTSLKDFKDFQAQIDVVRDKGVAVAVASAEDIDAANN	1040
consensus	aiigtigdvdsyqlpdakgyssllrhllgvtdeerqrkreei lttslkdfkdfaqaidvvr dkgvavavasaedidaann	
gi 9294618 spot 170 PreP1	ERSNFFFEVKA	1051
gi 9294618 spot 179 PreP1	ERSNFFFEVKA	1051
consensus	ersnffevkka	

F

Figure S2. Multiple alignment of the amino acid sequences from proteoforms by using DNAMAN software (Lynnon Corporation, QC, Canada). (A) glutamine synthetase isoform GS1c (GS1), with 100% identities; (B) transketolase (TK), with 84.41% identities; (C) RuBisCO large subunit-binding protein subunit (RBP), with 72.68% identities; (D) phosphoribulokinase (PRK), with 80.64% identities; (E) heat shock protein 70 (Hsp70), with 86.09% identities; (F) zinc metalloprotease (insulinase family), homologous to presequence protease 1 (PreP1), with 100% identities. The gi number, spot number, and protein name of the presented proteins are shown on the left.

gi 71361902 GS1b	1	-----HALLT	
gi 71361904 GS1c	1	-----HALLT	
gi 71361900 GS1a	1	-----HALLT	
gi 40317420 GS-e1	1	-----HSPLA	
gi 40317422 GS-e2	1	-----HSPLA	
gi 40317416 GS-r1	1	-----HASLA	
gi 40317418 GS-r2	1	-----HASLA	
gi 71362638 GS2b	1	MAQAUPAHQCQUGURSAUPARQAGRVUGURATARATSGFKULALGPETTGUIRQH	
gi 71362640 GS2c	1	MAQAUPAHQCQUGURSAUPARQAGRVUGURATARATSGFKULALGPETTGUIRQH	
gi 71362455 GS2a	1	MAQAUPAHQCQUGURSAUPARQAGRVUGURATARATSGFKULALGPETTGUIRQH	
consensus	1	MAQAUPAHQCQUGURSAUPARQAGRVUGURATARATSGFKULALGPETTGUIRQH	
gi 71361902 GS1b	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 71361904 GS1c	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 71361900 GS1a	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 40317422 GS-e2	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 40317416 GS-r1	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 40317418 GS-r2	6	DLNLDLTOSTEKI IAEYI WGGSGHDLRSKARTLPGPUDPSKLPKUNYDGSSTGQAPG	
gi 71362638 GS2b	61	QLLDHDTTPTDKI IAEYI WGGSGIDLRSKARTISKPUEDPSKLPKUNYDGSSTGQAPG	
gi 71362640 GS2c	61	QLLDHDTTPTDKI IAEYI WGGSGIDLRSKARTISKPUEDPSKLPKUNYDGSSTGQAPG	
gi 71362455 GS2a	61	QLLDHDTTPTDKI IAEYI WGGSGIDLRSKARTISKPUEDPSKLPKUNYDGSSTGQAPG	
consensus	61	QLLDHDTTPTDKI IAEYI WGGSGIDLRSKARTISKPUEDPSKLPKUNYDGSSTGQAPG	

Cytosolic GS1

Plastidic GS2

Cytosolic GS1

Plastidic GS2

Figure S3. Cont.

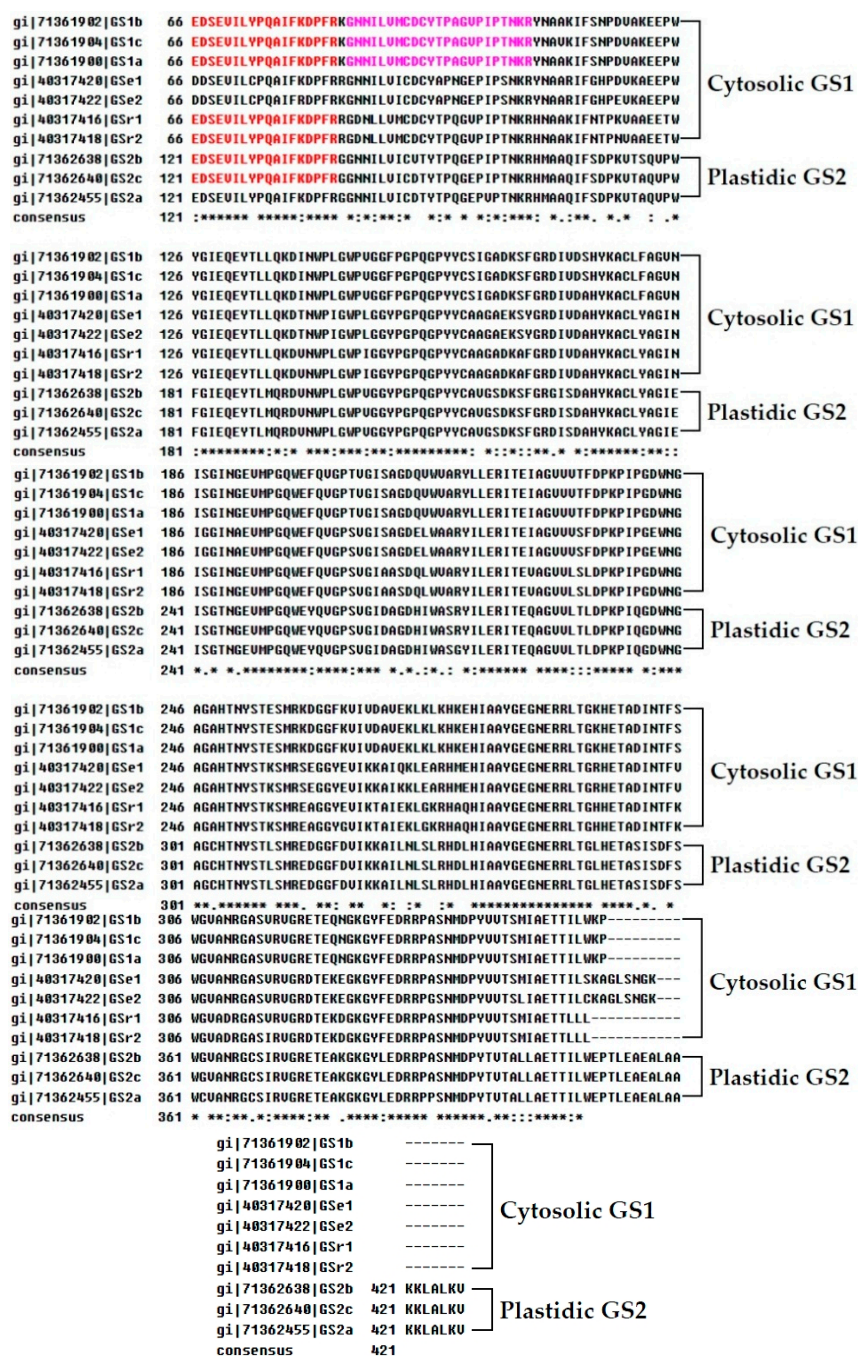


Figure S3. The alignment analysis between the MS-identified peptides from two proteoforms of GS1 (spots 795 and 806) and the amino acid sequences of the ten glutamine synthetase (GS) isozymes in wheat (*Triticum aestivum* L.). The amino acid sequences from wheat used for the multiple sequence alignment were from NCBI protein database (<http://www.ncbi.nlm.nih.gov/protein/>) and submitted by Bernard *et al.* [36]. The accession numbers and names of the GS isozymes were listed on the left, and the subfamilies were shown on the right of the amino acid sequences. The asterisk indicates completely conserved residues; colon indicates highly conserved residues; dot indicates a weak conservative region. The MS-identified peptides from spots 795, 806 and both were highlighted in red, blue, and purple fonts, respectively. The results showed that the MS-identified peptides were well aligned with cytosolic GS1, but not plastidic GS2. The detailed information of the identified peptides can be found in Table S1.

Figure S4. The alignment analysis between the MS-identified peptides of phosphoglycerate kinase (PGK) from *Ricinus communis* and the amino acid sequences of the eight putative protein products encoded from four Pgk genes in *Oryza sativa* sp. Japonica. The amino acid sequences from *O. sativa* sp. Japonica used for the multiple sequence alignment were from GABI primary database (<https://www.gabipd.org>) by using the TIGR ID provided by Joshi *et al.* [37]. The detailed accession numbers and names of the PGK isozymes were listed on the left, and the subcellular locations were shown on the right of the amino acid sequences. The asterisk indicates completely conserved residues; colon indicates highly conserved residues; dot indicates a weak conservative region; the short line indicates the vacancies residues of optimal sequence alignment. The identified peptides of PGK identified from *Ricinus communis* were highlighted in red fonts. The results showed that the unique matched peptides could only be found in the three members of OsPGK3, which were predicted to be localized in chloroplast. The detailed information of the identified peptides can be found in Table S1.

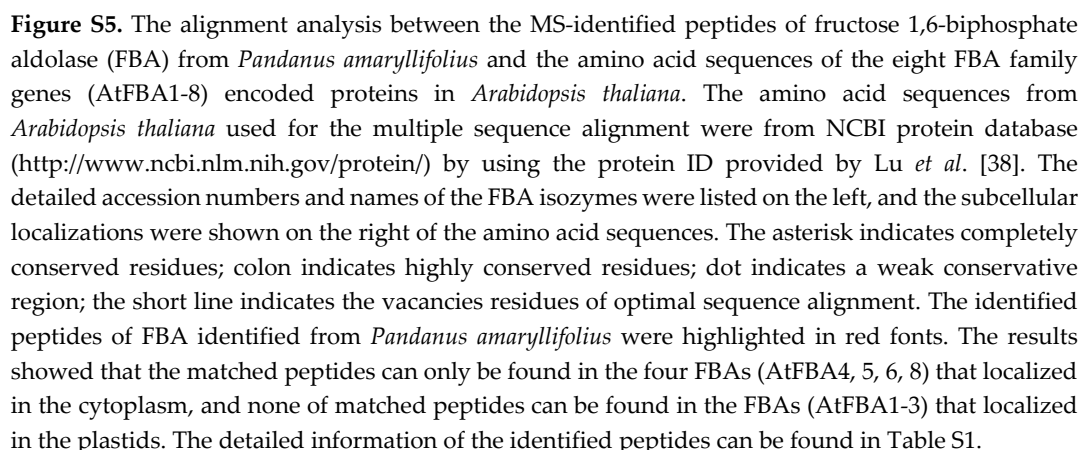


Figure S6. Cont.

g1 75313889 cpHsc70-1	283	GDGUFVULSTSGDTHLGGDFDKRVUUDLAAEFKKDEGIDLLKDKQALQRLTEAAEKAKI	Plastid stroma
g1 75314568 cpHsc70-2	283	GDGUFVULSTSGDTHLGGDFDKRVUUDLAAEFKKDEGIDLLKDKQALQRLTEAAEKAKI	
g1 75299484 mtHsc70-1	256	SSGUFVUKATGDFGLGGDFDNLLEVLUNEFKRSNDHIDTKMDLALQRLREAAEKAKI	Mitochondrion matrix
g1 75311081 mtHsc70-2	261	SSGUFVUKATGDFGLGGDFDNLLEVLUNEFKTEGIDLANDRLALQRLREAAEKAKI	
g1 18206379 B1P-1	244	DNGUFVULSTNGDTHLGGDFDHRUMEFYIKLKKKHQKIDSKDNKALGLRRECEAKR	Endoplasmic reticulum lumen
g1 12643245 B1P-2	244	DNGUFVULSTNGDTHLGGDFDHRUMEFYIKLKKKHQKIDSKDNKALGLRRECEAKR	
g1 75329119 B1P-3	258	DNGUFVULSTSGDTHLGGDFDHRUMYFIKLUKKKYNKIDSKDNKALGLRRECEAKR	Cytoplasm
g1 15241840 Hsc70-1	219	EEGIFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	
g1 12644165 Hsc70-2	219	EEGIFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	Unknown
g1 18206367 Hsc70-3	219	EEGIFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	
g1 75308864 Hsp70t-1	219	EEGIFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	Cytoplasm
g1 75311168 Hsp70t-2	219	EEGIFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	
g1 75313114 Hsp70b	218	EEGUFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	Unknown
g1 75313546 Hsp70t-2	218	EEGUFVUKATAGDTHLGGDFDHRUMHNFVDFEKKNNKIDSGNPRALRLATACERAKR	
consensus	381	AGGUSQIKALAG-SPIGGEDILQNTIRHAPPNEEASG-----LLRVAQDAQTH	
		* : : : * : : : * : : : * : : : * : : : * : : : *	
g1 75313889 cpHsc70-1	343	ELSSLTQTNMSLPFITATADGPKHITETLIRAKFEELCSDLLDRVTPUENSLDAKL SF	Plastid stroma
g1 75314568 cpHsc70-2	343	ELSSLTQTNMSLPFITATADGPKHITETLIRAKFEELCSDLLDRVTPUENSLDAKL SF	
g1 75299484 mtHsc70-1	316	ELSSLTQTTEINLPFITADASGAKHLNITLRSKFEGUGKLIERTSPQCMLKDAQUTI	Mitochondrion matrix
g1 75311081 mtHsc70-2	321	ELSSLTQTTEINLPFITADASGAKHLNITLRSKFETLUNHILERTDPCKMLKDAQUTI	
g1 18206379 B1P-1	304	ALSSQHQURVEIESLFDGUD----FSEPLTRARFEELNIDLFRKTHPUKKAANDAGLQK	Endoplasmic reticulum lumen
g1 12643245 B1P-2	304	ALSSQHQURVEIESLFDGUD----FSEPLTRARFEELNIDLFRKTHPUKKAANDAGLQK	
g1 75329119 B1P-3	318	SLSNHQURVEIESLFDGUD----FSEPLTRARFEELNIDLFRKTHPUKKAANDAGLQK	Cytoplasm
g1 15241840 Hsc70-1	279	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	
g1 12644165 Hsc70-2	279	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	Unknown
g1 18206367 Hsc70-3	279	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	
g1 75308864 Hsp70t-1	279	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	Cytoplasm
g1 75311168 Hsp70t-2	279	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	
g1 75313114 Hsp70b	278	TLSSTAQTITIEIDSLVEGID----FVSTITRAREFEELNIDLFRKTHPUKKAANDAGLQK	Unknown
g1 75313546 Hsp70t-2	296	RLTDQENQIEVDLGNCKNIS-----KULDLRFEFVUKFEECEERLUVCQLDARVUC	
consensus	361	* : : : * : : : * : : : * : : : * : : : * : : : *	
g1 75313889 cpHsc70-1	403	KDIDEUILUGGSTRIPAUQGLURKUT-GKEPNUTUNPDEUVALGAUQAQULAG-----D	Plastid stroma
g1 75314568 cpHsc70-2	403	KDIDEUILUGGSTRIPAUQGLURKUT-GKEPNUTUNPDEUVALGAUQAQULAG-----D	
g1 75299484 mtHsc70-1	376	KEUDEULLUGGSTRIPKUVQIEUSEIF-GKSPCKUNPDEAVAGAAIIGGGLILG-----D	Mitochondrion matrix
g1 75311081 mtHsc70-2	381	KEUDEULLUGGSTRIPKUVQIEUSEIF-GKSPCKUNPDEAVAGAAIIGGGLILG-----D	
g1 18206379 B1P-1	360	SQIDEIULUGGSTRIPKUVQILLKDFFEKPEPKNGUNPDEAVAGAAUQGGILSGEGG-DE	Endoplasmic reticulum lumen
g1 12643245 B1P-2	360	SQIDEIULUGGSTRIPKUVQILLKDFFEKPEPKNGUNPDEAVAGAAUQGGILSGEGG-DE	
g1 75329119 B1P-3	374	SQIDEIULUGGSTRIPKUVQILLKDFFDGKEPKNGUNPDEAVAGAAUQGGILSGEGG-EE	Cytoplasm
g1 15241840 Hsc70-1	335	STUHDVULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	
g1 12644165 Hsc70-2	335	STUHEIULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	Unknown
g1 18206367 Hsc70-3	335	NSIDVULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	
g1 75308864 Hsp70t-1	335	SMUHDVULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	Cytoplasm
g1 75311168 Hsp70t-2	335	SSUHDVULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	
g1 75313114 Hsp70b	334	SSUHDVULUGGSTRIPKUVQILLDFEFNGKELCKSNPDEAVAGAAUQAAILSGEEN-EK	Unknown
g1 75313546 Hsp70t-2	351	GIDIDLIMUGGCVYIPKURTITKWKCKDEYKUNPDEAVAGAAUQAAILSGEEN-EK	
consensus	421	: : : : : * : : : : * : : : : * : : : : * : : : : * : : : : *	
g1 75313889 cpHsc70-1	457	USDVULLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Plastid stroma
g1 75314568 cpHsc70-2	457	USDVULLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 75299484 mtHsc70-1	430	UKDILLLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Mitochondrion matrix
g1 75311081 mtHsc70-2	435	UKELLILLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 18206379 B1P-1	419	TKDILLLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Endoplasmic reticulum lumen
g1 12643245 B1P-2	419	TKDILLLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 75329119 B1P-3	433	TQMLLLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Cytoplasm
g1 15241840 Hsc70-1	394	UQDILLLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 12644165 Hsc70-2	394	UQDILLLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Unknown
g1 18206367 Hsc70-3	394	UQDILLLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 75308864 Hsp70t-1	394	UQDILLLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Cytoplasm
g1 75311168 Hsp70t-2	394	UQDILLLDVTPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
g1 75313114 Hsp70b	393	UQDILLLDVUPLSLGLTETAGGUTHIIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	Unknown
g1 75313546 Hsp70t-2	411	SLDLITIQATPLAUCURANGHIFIPRNTTIPKTSKFEVSTAGDGQTSVFINVLQGER	
consensus	481	: : : : : * : : : : * : : : : * : : : : * : : : : * : : : : *	
g1 75313889 cpHsc70-1	517	EFURDNKSLGSFRLLGIPPAFRGUPQIEUTCFIDANGILSUSADKGTGKKQDITITGA-	Plastid stroma
g1 75314568 cpHsc70-2	517	EFURDNKSLGSFRLLGIPPAFRGUPQIEUTCFIDANGILSUSADKGTGKKQDITITGA-	
g1 75299484 mtHsc70-1	490	EMADNKLGFEDLUGIPPAFRGUPQIEUTCFIDANGITTSADKGTGKKQDITITGA-	Mitochondrion matrix
g1 75311081 mtHsc70-2	495	EMADNKLGFEDLUGIPPAFRGUPQIEUTCFIDANGITTSADKGTGKKQDITITGA-	
g1 18206379 B1P-1	479	SLTKDCLLKGFDLNGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	Endoplasmic reticulum lumen
g1 12643245 B1P-2	479	SLTKDCLLKGFDLNGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	
g1 75329119 B1P-3	493	SLTKDCLLKGFDLNGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	Cytoplasm
g1 15241840 Hsc70-1	454	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	
g1 12644165 Hsc70-2	454	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	Unknown
g1 18206367 Hsc70-3	454	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	
g1 75308864 Hsp70t-1	454	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	Cytoplasm
g1 75311168 Hsp70t-2	454	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	
g1 75313114 Hsp70b	453	ARTDNNLKGFEELSGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	Unknown
g1 75313546 Hsp70t-2	471	ETUEANHLGVYKFLUGIPPAFRGUPQIEUTCFIDANGILNUSADKGTGKKQDITITGA-	
consensus	541	: : : : : * : : : : * : : : : * : : : : * : : : : * : : : : *	

Figure S6. Cont.

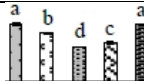
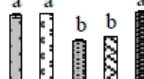
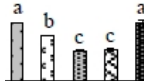
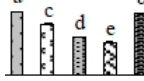
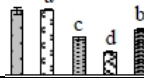
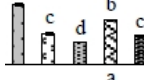
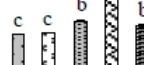
gi 75313889 cpHsc70-1	576	STLPKDEVDQMUQEAERFAKDDKEKRDADITKNDADSUVVOTEKQLKEL---GEKIPGEV	Plastid stroma
gi 75311568 cpHsc70-2	576	STLPKDEVDTMUQEAERFAKDDKEKRDADITKNDADSUVVOTEKQLKEL---GEKIPGPV	
gi 75299484 mtHsc70-1	549	GGLSDDIEINRMUKAEELHAQDKQEKQLIDLNSADTTIYSVEKSLSEY---REKIPAEI	Mitochondrion matrix
gi 75311081 mtHsc70-2	554	GGLSDDIQKMUREAELHAQDKKERKELIDTKNTADTTIYSIEKSLGEY---REKIPSEI	
gi 18206379 B1P-1	539	GRLSQEEIDRMUKAEFEFAEDKKKUEKIDARNALETYVYNNKQVNDKDLADKLEGE	Endoplasmic reticulum lumen
gi 12643245 B1P-2	539	GRLSQEEIDRMUKAEFEFAEDKKKUEKIDARNALETYVYNNKQVNDKDLADKLEGE	
gi 75329110 B1P-3	553	GRLTFFETFEETREAEFEFAEDKINKEKIDARNKLETVYVNNKSTUADKEKLAKKISDED	Cytoplasm
gi 15241849 Hsc70-1	514	GRLSKDEIEKMUQEAKEYKSEDEEHKKKUEAKNLENVAVMNTIIDE-KIGEKLPAD	
gi 12644165 Hsc70-2	514	GRLSKDEIEKMUQEAKEYKSEDEEHKKKUEAKNLENVAVMNTIIRDE-KIGEKLPAD	Cytoplasm
gi 18206367 Hsc70-3	514	GRLSKDEIEKMUQEAKEYKSEDEEHKKKUDAKNLENVAVMNTIIRDE-KIGEKLAGD	
gi 75308864 Hsp70t-1	514	GRLSKDDIEKMUQEAKEYKSEDEEHKKKUEAKNLENVAVMNTIIRDE---MGEKLPAD	Unknown
gi 75311168 Hsp70	514	GRLSKEEIEKMUQEAKEYKAEDEEHKKKUEAKNLENVAVMNTIKDE-KIASKLDAAD	Cytoplasm
gi 75313114 Hsp70b	513	GRLSKEEIEKMUQDAEKYKAEDEQKKKUEAKNLENVAVMNTIKDE-KLAQKLTDQ	
gi 75313546 Hsp70t-2	531	RHPTUDDGHGCAQALNKKYGATLDLITLQAKH-----	Unknown
consensus	601	. : : . : *	
gi 75313889 cpHsc70-1	633	KEKVEAKLQELKDKIGSGSTQETKDAMALNQEVNQIQSLYHQPGAGGPGAGSPGGEG	Plastid stroma
gi 75311568 cpHsc70-2	633	KEKVEAKLQELKEKIASGSTQETKDTHMALNQEVNQIQSLYHQPGAGGPGAGSPGGEG	
gi 75299484 mtHsc70-1	606	ASEIETAUSDRTAHAGEDUEDIKAKUEAANKAUSKIGEHHS-----KCSG	Mitochondrion matrix
gi 75311081 mtHsc70-2	611	AKIEEDAVADLRSSAGDQLNETKAKTEAANKAUSKIGEHHS-----KCSG	
gi 18206379 B1P-1	599	KEKIEAATKEALEMDEHQNSEK-EEYDEKLKEUEAVCNPIITAVYQR-----SG	Endoplasmic reticulum lumen
gi 12643245 B1P-2	599	KEKIEAATKEALEMDEHQNSEK-EEYDEKLKEUEAVCNPIITAVYQR-----SG	
gi 75329110 B1P-3	613	KEKMEGULKEALEMLEENHNAEK-EDYDEKLKEUELUCDPUKSUYEK-----TE	Cytoplasm
gi 15241849 Hsc70-1	573	KKKIEDSIEQA IQMLEGNQLAEA-DEFEDEKMELESICNPITAKMWQG-AGGAGGPGAS	
gi 12644165 Hsc70-2	573	KKKVEDSIEFA IQMLDGNQLAEA-DEFEDEKMELESICNPITAKMWQG-AGGAGGPGAS	Unknown
gi 18206367 Hsc70-3	573	KKKIEDSIEFA IEMLANQLAEC-DEFEDEKMELESICNPITAKMWQG-AGGAGGPGAG	
gi 75308864 Hsp70t-1	571	KKKFEDSIEEV IQMLDDNQLAEA-DEFEHKKMELESUMSTIITAKMWQG-----	Unknown
gi 75311168 Hsp70	573	KKKIEDAIDQA IEMLDGNQLAEA-DEFEDEKMELESICNPITAKMWQG-AGPDHGG--AG	
gi 75313114 Hsp70b	572	KQKIEKAIDET IEWIEGNQLAEV-DEFEYKLEKEGICNPITISKMWQG-----GAAG	Cytoplasm
gi 75313546 Hsp70t-2	-----	-----	
consensus	661		
gi 75313889 cpHsc70-1	693	ASSGD-SSSSKGGD-GDDUIDADFTDSQ----	Plastid stroma
gi 75311568 cpHsc70-2	691	SSSSDTSSSAKGGDGGDUIDADFTDSH----	
gi 75299484 mtHsc70-1	652	SSGSD-GSSGEGTSGTEQTPAEFEFEASGSRK	Mitochondrion matrix
gi 75311081 mtHsc70-2	657	GGAP-GGSGEG--GSDQAPAEVEYUUK----	
gi 18206379 B1P-1	648	GAPGGAGGESSFEEDESDEL-----	Endoplasmic reticulum lumen
gi 12643245 B1P-2	648	GAPG-AGGESSFEEDESDEL-----	
gi 75329110 B1P-3	662	-----GENEDDGDGDD-HDEL-----	Cytoplasm
gi 15241849 Hsc70-1	631	GMD-DDAPPASGGAG-PKIEEVD-----	
gi 12644165 Hsc70-2	632	GMDDEAPPASGGAG-PKIEEVD-----	Cytoplasm
gi 18206367 Hsc70-3	629	GMD-EDUPPSAGGAG-PKIEEVD-----	
gi 75308864 Hsp70t-1	-----	-----	Unknown
gi 75311168 Hsp70	629	GMDDDTPAGGSGGAG-PKIEEVD-----	Cytoplasm
gi 75313114 Hsp70b	624	GMPTDGFSSSGAAGGPKIEEVD-----	
gi 75313546 Hsp70t-2	-----	-----	Unknown
consensus	721		

Figure S6. The alignment analysis between the MS-identified peptides from five proteoforms of heat shock protein 70 (Hsp70) (spots 355, 361, 358, 367, and 332) and the amino acid sequences of the fourteen Hsp70 gene family encoded proteins in Arabidopsis. The amino acid sequences from Arabidopsis used for the multiple sequence alignment were from NCBI protein database (<http://www.ncbi.nlm.nih.gov/protein/>) by using the protein name provided by Sung *et al.* [39]. The detailed accession numbers and names of the HSP70 proteins were listed on the left, and the subcellular locations were shown on the right of the amino acid sequences. The asterisk indicates completely conserved residues; colon indicates highly conserved residues; dot indicates a weak conservative region; the short line indicates the vacancies residues of optimal sequence alignment. The identified peptides from spots 355, 361, 358, 367, and 332 were highlighted with red, blue, purple, yellow, and green underlines, respectively. The results showed that the unique identified peptides from all the spots 355, 361, 358, and 367 could be only matched to the Hsp70s localized in cytoplasm. In contrast, the unique identified peptides from the spot 332 could be only matched to the Hsp70s localized in chloroplast. The detailed information of the identified peptides can be found in Table S1.

Table S3. Proteoforms identified in *Hosta* “Gold standard”.

Protein Name ^a	Identity ^b	Spot No. ^c	SCL ^d	Plant Species ^e	gi Number ^f	Exp. Mw (Da)/pI ^g	Thr. Mw (Da)/pI ^h	Sco ⁱ	Cov (%) ^j	QM ^k	V% ± SE / YLM			
											MLM	LENF	YLC	MLC
glutamine synthetase isoform GS1c (GS1)	100%	795	Cyt	<i>Triticum aestivum</i>	71361904	39,676/5.58	39,445/5.41	89	15	2				
		806	Cyt	<i>T. aestivum</i>	71361904	39,505/5.75	39,445/5.41	168	15	3				
transketolase (TK)	84.41%	338	Chl	<i>Nicotiana tabacum</i>	194396261	78,879/5.95	80,451/6.16	141	17	10				
		346	Chl	<i>Populus trichocarpa</i>	224063766	79,837/5.87	81,122/5.97	337	13	9				
RuBisCO large subunit-binding protein subunit (RBP)	72.68%	476	Chl	<i>Vitis vinifera</i>	225442531	63,140/5.18	65,255/5.6	474	17	8				
		459	Chl	<i>Sorghum bicolor</i>	242090109	66,151/4.84	60,914/5.07	233	31	13				
		472	Chl	<i>Zea mays</i>	223948025	64,795/5.24	61,969/5.42	524	30	11				
phosphoribulokinase (PRK)	80.64%	840	Chl	<i>Pisum sativum</i>	1885326	38,189/4.99	39,230/5.41	113	13	3				
		832	Chl	<i>Glycine max</i>	255646270	38,858/4.85	45,757/5.89	277	38	11				

Table S1. Cont.

Protein Name ^a	Identity ^b	Spot No. ^c	SCL ^d	Plant Species ^e	gi Number ^f	Exp. Mw (Da)/pI ^g	Thr. Mw (Da)/pI ^h	Sco ⁱ	Cov (%) ^j	QM ^k	V% ± SE ^l YLM				
											MLM	LENF	YLC	MLC	
heat shock protein 70 kDa (Hsp70)	86.09%	355	Cyt	<i>V. vinifera</i>	225449497	77,129/5.15	71,525/5.17	1050	61	26					
		361	Cyt	<i>V. vinifera</i>	225449497	77,663/5.10	71,525/5.17	959	58	25					
		358	Cyt	<i>Ricinus communis</i>	255573627	78,743/5.06	71,276/5.14	977	53	23					
		367	Cyt	<i>V. vinifera</i>	225434984	76,864/5.20	71,589/5.17	847	57	25					
		332	Chl	<i>V. vinifera</i>	147805297	80,390/4.75	69,198/5.11	722	36	16					
zinc metalloprotease (insulinase family), homologous to presequence protease 1 (PreP1) *	100%	170	Chl/Mit	<i>Arabidopsis thaliana</i>	9294618	113,711/4.99	118,265/5.32	346	13	11					
		179	Chl/Mit	<i>A. thaliana</i>	9294618	113,515/5.07	118,265/5.32	157	11	9					

^a: The name and functional categories of the proteins identified by MALDI TOF-TOF MS. Protein names marked with an asterisk (*) have been edited by us depending on BLAST against NCBI non-redundant protein database. The detailed information of the NCBI BLAST can be found in Table S2; ^b: The identities of the multiple alignment of the amino acid sequences from different proteoforms. ^c: Assigned spot number as indicated in Figure 5; ^d: Protein subcellular localization predicted by softwares (YLoc, LocTree3, Plant-mPloc, ngLOC, and TargetP). SCL, subcellular location; Chl, chloroplast; Cyt, cytoplasm; Mit, mitochondria; ^e: The plant species that the peptides matched from. ^f: Database accession numbers from NCBI; ^{g,h}: Experimental (f) and theoretical (g) molecular weight (Da) and pI of identified proteins. Experimental values were calculated using Image Master 2D Platinum Software. Theoretical values were retrieved from the protein database; ⁱ: The Mascot score obtained after searching against the NCBI database; ^j: The amino acid sequence coverage for the identified proteins; ^k: The number of unique peptides identified for each protein; ^l: The mean values of protein spot volumes relative to total volume of all the spots. Five samples (from left to right: YLM, YLC, MLM, MLC, LENF) of various leaf regions in different development stages of *Hosta* “Gold Standard” were performed. YLM, young leaf margin; YLC, young leaf center; MLM, mature leaf margin; MLC, mature leaf center; LENF, leaf under excess nitrogen fertilization. The different small letters on the columns indicate significant differences ($p < 0.05$) among the five samples as determined by one-way ANOVA. Error bars indicate \pm standard error (SE).