Supplemental Data. Noël et al. (2007). Interaction between SGT1 and cytosolic/nuclear HSC70 chaperones regulates *Arabidopsis* immune responses.



Supplemental Figure 1 online. Functionality tests of Strepll-tagged SGT1 proteins in transgenic Arabidopsis described in Figure 1. Plants 1/2, 3/4 and 6/7 indicate independent transgenic lines. Line 5 is derived from a cross between a Ler/Ws-0 hybrid *sgt1b*-3/*sgt1b*-3 *sgt1a*-1/SGT1a and plant 4. Background refers to the SGT1 alleles: wt, wild type SGT1a and SGT1b; a, homozygous *sgt1a*-1; b, homozygous *sgt1b*-3; ab, double *sgt1a*-1 *sgt1b*-3 homozygotes; nd, not determined.

(A) Root growth inhibition on 0,075 μ M 2-4D. *, statistically significant differences for values compared with Ler sgt1b-3 mutant as determined by student's *t*-test (p<0,001).

(B) Root growth was measured three days after transfer on 10 μ M methyl jasmonate. n>10 for each data point. Errors bars indicate standard errors. *, statistically significant differences for values compared with wild type Ler as determined by student's *t*-test (p<0,005).

(C) *RPP5* gene-mediated resistance assays were performed using *H. parasitica* avirulent isolate Noco2 on two-week-old *Arabidopsis* seedlings. Spores were counted 6 dpi. One representative experiment from three is shown. Each data point represents the average of triplicate measurements and errors bars indicate standard errors. *, statistically significant differences for values compared with wild type Ler as determined by student's *t*-test (p<0,05).

/	A		m/z	(charge	e)															
	HSC70-1	T10	615,8	(2)	F	S	D	S	s	v	Q	S	D	Μ	к					
	HSC70-2	T10	556,2	(2)	F	S	D	A	S	v	Q	S	D	R						
	HSC70-3	T10	613,8	(2)	F	т	D	S	ទ	v	Q	S	D	Ι	ĸ					
	HSC70-4	T10	555,2	(2)	Y	S	D	Ρ	s	v	0	А	D	ĸ	ន					
	HSC70-5	T11	635,0	(3)	F	s	D	Ρ	S	v	õ	s	D	Ι	L	H	W	Р	F	K
	В		m/z	(charge	e)				-			Ĩ								
	HSC70-1	T16	653,8	(2)	Е	Ι	Α	Е	A	Y	L	G	v	т	Ι	к				
	HSC70-2	T15	639,8	(2)	Е	Ι	Α	Е	A	F	L	G	т	т.	V	к				
	HSC70-3	T16	654,8	(2)	Е	Ι	А	Е	Α	Y	г	G.	т	т	Ι	к				
	HSC70-4	T16	630,8	(2)	Ε	Ι	Α	Е	Α	F	г	G	ន	\mathbf{P}	v	к				
	HSC70-5	T16	496,2	(2)	Е	V	Α	Е	Α	F	L	G	R							

Supplemental Figure 2 online. Mass fingerprinting of two peptides discriminating between cytosolic HSC70 isoforms

The polymorphic tryptic peptides (T) corresponding to T10 (A) and T16 (B) of HSC70-1 were used to identify specifically different cytosolic HSC70 isoforms. Corresponding peptides of HSC70-2 to -5 were aligned and polymorphic amino acids indicated by black shading. Predicted mass/charge values and charge are indicated.



Supplemental Figure 3 online. Interaction studies between SGT1 and RAR1 or HSP90-1 using the Clontech Matchmaker[™] GAL4 yeast two-hybrid system. Different dilutions were plated on SD medium lacking Leucine (L), Tryptophan (W), Histidine (H) or Adenin (A) : -LW, bait/prey plasmid selection; -LWH, weak interaction; -LWHA, strong interaction. Picture was taken after 3 days. HSP90-1 ATPase domain (residue 1-207); SGT1b^{eta3} (residue 1-322). AD, GAL4 activation domain; BD, GAL4 DNA binding domain.

(A) Interaction studies with RAR1, HSP90-1, SGT1b and HSC70-1.

(B) Interaction studies with SGT1b and HSP90-1.



Supplemental Figure 4 online. Individual loss of *Arabidopsis HSC70* isoforms 1, 2 or 3 does not affect plant immune responses

(A) Schematic representation of the intron (chevron) -exon (grey bar) structure of the five *Arabidopsis* cytosolic *HSC70* genes. Arrows indicate translation starts and asterixes the stop codons. Triangles indicate the sites of T-DNA insertions in accession Col-0. Dashed lines mark the regions chosen for RT-PCR analysis of gene expression. Expression results of homozygous T-DNA lines are shown: (+) wild-type; (+/-) reduced; (-) not detectable.

Arabidopsis T-DNA insertion lines were inoculated with avirulent *H. parasitica* isolates Cala2 (B) or Emwa1 (C) and infection measured by trypan blue staining at 6 dpi. Ler and Ws-0 plants are susceptible to Cala2 and Emwa, respectively. HR, hypersensitive response; TN, trailing necrosis.

(D) Bacterial growth of virulent *Pst* DC3000 after vacuum infiltration of different lines, as indicated. The *eds1-2* (Ler) and *pad4-1* (Col) mutations compromise basal resistance. Each data point was analysed in triplicate and errors bars indicate standard errors. **, statistically significant differences for values compared with wild type Col-0 as determined by student's *t*-test (p<0,005).



Supplemental Figure 5 online. *SGT1b* loss-of-function and *HSC70-1* over expression have additive effects on *R* gene-mediated resistance to *H. parasitica*.

Two-week-old *Arabidopsis* seedlings were inoculated with avirulent *H. parasitica* isolates Emwa1. Spores were counted 6 dpi. Ws-0 plants are susceptible to Emwa1.The *pad4-1* (Col) mutation compromises *R* gene-mediated resistance. Each data point is the average of two independent experiments with duplicate measurements. Errors bars indicate standard errors. * and **, statistically significant differences for values compared with wild type Col-0 as determined by student's *t*-test (p<0,05 and p<0,005 respectively). ° and °°, statistically significant differences for values compared with line *eta3* 8-7 as determined by student's *t*-test (p<0,05 and p<0,005 respectively).

SUPPLEMENTAL TABLE

Pentide	m/z	Charge	HSC70 isoform									
replice	111/ Z	Charge	HSC70-1	HSC70-2	HSC70-3	HSC70-4	HSC70-5					
А	559,2	3	T24	T23	T24	T24	T26					
В	608,3 ^b	3	T19		T19							
С	615,8 ^b	2	T10									
D	639,8	2	T25	T24	T25	T25						
E	648,4 ^b	2			T39							
F	657,3	2	Т39									
G	653,8 ^b	2	T16									
Н	654,8 ^b	2			T16							
I^a	679,8 ^b	2	T71	T70	T71	T71						
J	737,4	2	T4	T4	T4	T4						
Κ	824,4	2	T6	T6								
L	840,5	2	T17	T16	T17	T17	T18					
Μ	962,5 ^b	3	T53-54		Т53-54							

Supplemental Table 1 online. Tryptic fragments observed by mass spectrometry of 70-kDa proteins co-purified with over expressed SGT1b-Strep.

^a The corresponding peptide containing an oxidized methionine was also observed

^b Confirmed by MS-MS