

## **SELF-INCOMPATIBILITY IN BRASSICACEAE: IDENTIFICATION AND CHARACTERIZATION OF SRK-LIKE SEQUENCES LINKED TO THE S-LOCUS IN THE TRIBE BISCUTELLEAE**

Jean-Baptiste Leducq\*†, Célia C Gosset§†, Rita Gries†, Kevin Calin†, Éric Schmitt†, Vincent Castric† & Xavier Vekeman†.

\*- *Institut de Biologie Intégrative et des Systèmes, Département de Biologie, PROTEO, Pavillon Charles-Eugène-Marchand, 1030 avenue de la Médecine - Université Laval - Québec (QC) G1V 0A6, Canada*

§- *Institut des Sciences de l'Évolution UMR 5554, CNRS, Université Montpellier 2, Place Eugène Bataillon, C.C. 065 - 34095 Montpellier cedex 05, France*

†- *Laboratoire Génétique et Evolution des Populations Végétales, CNRS UMR 8198, Université Lille1, F-59655 Villeneuve d'Ascq cedex, France*

Corresponding author: Jean-Baptiste Leducq ([jean-baptiste.leducq.1@ulaval.ca](mailto:jean-baptiste.leducq.1@ulaval.ca))

**DOI: 10.1534/g3.114.010843**

**Table S1** Accession numbers of SRK-related sequences for Figure 1, from *Arabidopsis halleri* (Aha), *A. lyrata* (Aly), *A. thaliana* (Ath), *Brassica oleraceae* (Bol), *B. rapa* (Bra) and *Capsella grandiflora* (Cgr).

<i>Sequence</i>	<i>Accession number</i>	<i>Sequence</i>	<i>Accession number</i>
<i>AhaSRK03</i>	EU075126	<i>BolSRK06</i>	M76647.1
<i>AhaSRK04</i>	DQ520278	<i>BolSRK07</i>	AB070624.1
<i>AhaSRK11</i>	EU075132	<i>BolSRK08</i>	AB054708.1
<i>AhaSRK12</i>	EU075133	<i>BolSRK11</i>	AB054709.1
<i>AhaSRK13</i>	EU075134	<i>BolSRK12</i>	AB180901.1
<i>AhaSRK15</i>	EU075136	<i>BolSRK13</i>	AB024420.2
<i>AhaSRK20</i>	EU075141	<i>BolSRK15</i>	AB180903.1
<i>AhaSRK22</i>	EU075143	<i>BolSRK16</i>	AB054710.1
<i>Aly327420</i>	XM_002872670.1	<i>BolSRK18</i>	AB032473.1
<i>AlyARK3_946052</i>	XM_002867806.1	<i>BolSRK23</i>	AB013720.1
<i>AlySRK03</i>	AF328992	<i>BolSRK24</i>	AB054712.1
<i>AlySRK04</i>	AF328994	<i>BolSRK25</i>	AB054713.1
<i>AlySRK05</i>	AF328990	<i>BolSRK29</i>	Z30211.1
<i>AlySRK06</i>	DQ520281	<i>BolSRK33</i>	AB054714.1
<i>AlySRK08</i>	AY186766	<i>BolSRK35</i>	AB054715.1
<i>AlySRK09</i>	DQ520282	<i>BolSRK36</i>	AB054716.1
<i>AlySRK10</i>	AY186767	<i>BolSRK38</i>	AB054717.1
<i>AlySRK12</i>	AY186769	<i>BolSRK39</i>	AB054718.1
<i>AlySRK13</i>	AF328993	<i>BolSRK45</i>	AB054719.1
<i>AlySRK14</i>	AY186770	<i>BolSRK50</i>	AB054720.1
<i>AlySRK15</i>	AY186771	<i>BolSRK51</i>	AB054721.1
<i>AlySRK16</i>	DQ520283	<i>BolSRK57</i>	AB054722.1
<i>AlySRK18</i>	DQ520284	<i>BolSRK58</i>	AB054723.1
<i>AlySRK19</i>	AF328998	<i>BolSRK60</i>	AB032474.1
<i>AlySRK20</i>	AF328995	<i>BolSRK62</i>	AB054724.1
<i>AlySRK22</i>	DQ520285	<i>BolSRK64</i>	AB054725.1
<i>AlySRK23</i>	AF328997	<i>BraARK03_013522</i>	AB041621.1
<i>AlySRK25</i>	DQ520286	<i>BraSRK46</i>	AB013718.2
<i>AlySRK27</i>	EU878017	<i>BraSRK54</i>	AB219162.1
<i>AlySRK29</i>	AY186776	<i>BraSRK60</i>	AB097116.1
<i>AlySRK30</i>	EU878018	<i>CgrSRK01</i>	DQ530637.1
<i>AlySRK33</i>	EU878019	<i>CgrSRK02</i>	DQ530638.1
<i>AlySRK35</i>	EU878021	<i>CgrSRK03</i>	DQ530639.1
<i>AlySRK36</i>	DQ520288	<i>CgrSRK04</i>	DQ530640.1
<i>AlySRK37</i>	DQ520289	<i>CgrSRK05</i>	DQ530641.1
<i>AlySRK38</i>	EU165341	<i>CgrSRK06</i>	DQ530642.1
<i>AlySRK39</i>	EU878022	<i>CgrSRK07</i>	EF530735.1
<i>AlySRK43</i>	EU878024	<i>CgrSRK09</i>	FJ649955.1
<i>AlySRK44</i>	EU878025	<i>CgrSRK11</i>	FJ649956.1
<i>AthSRKC</i>	KC207414.1	<i>CgrSRK14</i>	FJ649958.1
<i>BolSRK01</i>	AB054706.1	<i>CgrSRK18</i>	FJ649953.1
<i>BolSRK02</i>	AJ306588.1	<i>CgrSRK24</i>	FJ649962.1
<i>BolSRK03</i>	X79432.1	<i>CgrSRK37</i>	FJ649959.1
<i>BolSRK05</i>	Y18259.1	<i>CgrSRK38</i>	FJ649960.1

**Table S2 List and sequences of primers targeting candidate sequences of the pollen-recognition extracellular domain of the *SRK* gene in *Biscutella neustriaca*.** T<sub>m</sub> indicates the optimized hybridization temperature during PCR reaction. When the number of PCR cycles used is different from 35 cycles, it is indicated after T<sub>m</sub> in parenthesis. Generalist primers amplify several sequences belonging to the same sequences class.

Class		F-specific primer		R-specific primer	T <sub>m</sub> (°C)	Sequence(s) amplified
<i>Bne327420L</i>	X-F001	GTACCGGAGATGCGGAAGT	X-R540	GAGCTCTCCGGACCAAATTA	60	1 and 2
<i>BneSRKA</i>	A01-F142	AGGTGGAATTCGACGTTAAA	A01-R558	GTAATTACTGATATCCAATA	52	1
	A02-F277	TTTACGAAGAGGAATCCGGT	SLGR		50	2
	A03-F074	TTAAACTCACCAGCCCAGAAT	A03-R421	TATCCACAGTTGCCGCCCTTG	62	3
	A04-F068	ACACTTTTCAACTCACCAAGCGA	SLGR		59	4 and 5
	A06-F238	GACATGGACACTTCACCGAT	A06-R514	CAGACCCATTATTTTCGGACA	62	6
	A-F020	GGGAACACATGGTTTCCAAC	SLGR		50	
	A-F300	GTGGGAAAGGACAAATGGAA	SLGR		60	Generalist A
	A-F294	GSWTGAGTGGGAAAKGRCAA	A-R461	CYGCACATYTTTCTCACA	60	
	A-F330	TGTGAGGAAGACACGGTTGA	SLGR		50	Generalist A and C01
<i>BneSRKB</i>	B01-F079	ATGACCAACCAAACCATACAG	SLGR	ATCTGACATAAAGATCTTGACC	52	1
	B-F075	CTTCATGACCAACCAAAGCA	B-R495	CGCATTTCGCAAATCCTCTAC	60	3
	B04-F159	ATATGGATGGAGACAGATGT	B04-R371	GTAAGCCATCTCCCCTGTA	60	4
	B05-F112	GTGAGTTTACGGGTCTTTTCG	B05-R405	TGTAGTATCCGGCAACTTAA	60	5
	B06-F276	GTTTGATCCTAGAAATCCACAT	B06-R463	CTTTAAGGCACCTCTTCTCA	60	6
	B09-F147	GATTCCGATATCAGATGACT	B-R564	WRCATAATCCGAATATCAA	50	9
	B10-F238	GACATGAACACGTCACCGGT	B10-R452	TTCTTACATTCTTTCGTATCA	52	10
	B11-F153	GACATCATATGAATGGAGCCCG	B-R539	AKCAYTYCYKTCCAAATAAC	52	11
	B12-F110	CTGTGAGTTTTACGGGCTTT	B12-R408	TGTAGTGTCCGGCAGCTTTACC	63	12
	B17-F115	ACTTTCTGGGGTATTTAAATA	B-R388	TTATCTTCTCARCCSGTA	58 (40c)	17
	B-F055	GATGARATCGCTTACACGTT	B-R564	WRCATAATCCGAATATCAA	54	
	B-F064	GCTTACACGTTCYTMATGACC	B-R388	TTATCTTCTCARCCSGTA	50	Generalist B
			B-R539	AKCAYTYCYKTCCAAATAAC	60	
<i>BneSRKC</i>	C-F001	GCTGCAGAGGTGACAAGTTTT	C-R173	GGATGGGTTGTCTGATTGG	56	1

**Table S3 Result of test of deviation from Mendelian expectations in collection F1 (See Table 3 for genotypes).** For each cohort size  $n$ , 100,000 cohorts were simulated according to two scenarios: (1) assuming that both parents were heterozygous at the  $S$ -locus and (2) assuming that one parent was homozygous and one parent heterozygous. For each cohort, the following information is shown: number of seedlings (cohort size), name of F0 parental individuals, absolute expected frequency of each genotype in the cohort and difference (deviation) between observed and expected frequencies under each scenario.

Seedling collection	$n^a$	Parents <sup>b</sup>		Expected genotype frequency in F1 (SD) <sup>c</sup>		Deviation from expected genotype frequency (associated $p$ -value) <sup>d</sup>					
						(1) He <sup>e</sup>			(2) Ho <sup>f</sup>		
						1	2	3	4	1	2
F1o	7	3	6	1.75 (1.15)	3.50 (1.32)	-2.25* (0.01)	-1.25 (0.07)	-	-	-0.5 (0.23)	0.5 (0.50)
	13	3	7	3.25 (1.56)	6.50 (1.8)	-3.75* (0.01)	-2.75* (0.03)	-	-	-0.5 (0.30)	0.5 (0.50)
	5	8	4	1.25 (0.97)	2.50 (1.12)	-0.75 (0.11)	0.25 (0.37)	-0.75 (0.10)	-	-	-
	7	8	6	1.75 (1.15)	3.50 (1.33)	0.75 (0.45)	-1.25 (0.07)	-1.25 (0.07)	-	-	-
	21	9	4	5.25 (1.98)	10.50 (2.29)	1.25 (0.37)	4.25* (0.02)	-4.75* (0.01)	-0.75 (0.26)	-	-
	7	9	6	1.75 (1.14)	3.50 (1.32)	-1.25 (0.07)	-0.25 (0.24)	-0.25 (0.24)	-	-	-
	6	10	4	1.50 (1.06)	3.00 (1.22)	0.5 (0.47)	-2.5* (0.01)	0.5 (0.47)	-	-	-
	16	10	6	4.00 (1.73)	8.00 (2.00)	2 (0.20)	-2 (0.08)	-2 (0.08)	2 (0.20)	-	-
F1c	14	4	20	3.50 (1.62)	7.00 (1.87)	0.5 (0.48)	1.5 (0.28)	-0.5 (0.26)	-1.5 (0.11)	-	-
	30	12	8	7.50 (2.37)	15.00 (2.74)	0.5 (0.49)	1.5 (0.35)	1.5 (0.35)	-3.5 (0.05)	-	-
	30	17	18	7.50 (2.37)	15.00 (2.73)	-5.5* (0.01)	-9.5*** (0.00)	-	-	2 (0.29)	-2 (0.18)
	50	18	1	12.50 (3.06)	25.00 (3.54)	1.5 (0.38)	-2.5 (0.16)	3.5 (0.16)	-2.5 (0.16)	-	-
	14	21	8	3.50 (1.62)	7.00 (1.87)	-0.5 (0.26)	2.5 (0.10)	-1.5 (0.11)	-0.5 (0.26)	-	-

a cohort size

b F0 collection

c expected genotype frequency in F1 collection following symbols are associated with significance of  $p$ -values: "\*\*\*\*":  $p \leq 0.001$ ; "\*\*";  $p \leq 0.05$ .

d expected frequency : see table 3

e assuming that both parents are heterozygous at the  $S$ -locus

f assuming that parent 1 is homozygous and that parent 2 is heterozygous at the  $S$ -locus - Not applied for cohorts with more than two genotypes

**Table S4** Accession numbers of SRK-L sequences found in *Biscutella neustriaca*.

Sequence	Accession number
<i>BneSRKL_A01a</i>	KF905296
<i>BneSRKL_A01b</i>	KF905297
<i>BneSRKL_A02</i>	KF905298
<i>BneSRKL_A03</i>	KF905299
<i>BneSRKL_A04</i>	KF905300
<i>BneSRKL_A05</i>	KF905301
<i>BneSRKL_A06</i>	KF905302
<i>BneSRKL_A07</i>	KF905303
<i>BneSRKL_B01</i>	KF905304
<i>BneSRKL_B03</i>	KF905305
<i>BneSRKL_B04</i>	KF905306
<i>BneSRKL_B05</i>	KF905307
<i>BneSRKL_B06</i>	KF905308
<i>BneSRKL_B09</i>	KF905309
<i>BneSRKL_B10</i>	KF905310
<i>BneSRKL_B11</i>	KF905311
<i>BneSRKL_B12</i>	KF905312
<i>BneSRKL_B13</i>	KF905313
<i>BneSRKL_B17</i>	KF905314
<i>BneSRKL_B18</i>	KF905315
<i>BneSRKL_C01</i>	KF905295
<i>BneARK3L_1</i>	KF905320
<i>BneARK3L_2</i>	KF905321
<i>BneARK3L_3</i>	KF905322
<i>BneARK3L_4</i>	KF905323
<i>BneARK3L_5</i>	KF905324
<i>Bne327420L_1</i>	KF905316
<i>Bne327420L_2</i>	KF905317
<i>Bne327420L_3</i>	KF905318
<i>Bne327420L_4</i>	KF905319

S01			Pollen donors										Controls		S-shared vs. Controls		Expressed in stigma ?	
			F0			F1												
S-haplotypes		Plants	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01	S01
1	2		1	2	1	2	1	2	2	3	4	1						
F0	S01	S02	1	0/5	1/10	0/5	/	/	/	/	/	/	/	59/65		yes		
	S01	S11	2	9/10**	0/10	0/10*	/	/	/	/	/	/	153/185		yes*			
	S01	S01	1	4/5**	0/10*	0/5	0/10*	3/4**	3/4**	9/10**	0/5*	15/23**	5/5**	55/90		yes*		
Pollen receptors (stigma)	S01	S07	2	/	/	0/9	/	/	/	/	/	/	4/5		yes			
	S01	S09	1	/	/	0/5	/	/	/	/	/	/	5/5		yes			
	S01	S04	3	/	/	1/10*	/	/	3/10**	/	/	/	3/5		yes*			
	F1	S01	S08	2	/	/	0/9	/	/	/	/	/	/	NA	NA	yes		
		S01	S10	3	/	/	0/5	/	/	/	/	/	/	1/5		yes		
		S01	S03	4	/	/	0/20	/	/	/	/	/	/	13/15		yes		
		S01	S12	1	/	/	0/5	/	/	/	/	/	/	4/5		yes		
Controls			59/65	136/164	58/85	8/10	5/5	5/5	NA	3/5	2/14	3/4						
S-shared vs. Controls																		
Expressed in pollen ?			no**	yes	yes	yes	no**	no**	no**	yes	no**	no**						

**Figure S1 Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S01 (B10-B11).** For each tested cross-pollination between individuals having at least S01 in common (S-shared crosses), pollen donor is shown in line and pollen receptor (stigma) in column. The number of plants having the same genotype used for cross-pollinations is shown next to genotypes. Slash bars indicate that no cross-pollination was realized for the given combination. For realized cross-pollinations, the number of successful pollinations over the number of realized pollinations is indicated. Black cells indicate that most (>70%) of cross-pollinations were unsuccessful. Controls indicate cross-pollinations realized between the tested genotype and all other plants sharing no common S-haplotype with the tested genotype (in grey when the low proportion of successful pollination could result from low pollen efficiency/receptivity). Histograms indicate the proportion of successful cross-pollinations for S-shared crosses totalized by line or column (black; crosses between identical genotypes were not counted) and controls (grey). We considered that S01 was expressed in either pollen (columns) or stigma (lines) when most of combinations gave more than 70% of unsuccessful pollinations. In complex cases, asterisks indicate what combinations of crosses were considered for each diagnostic of expression. Question marks (?) indicate that results were too ambiguous or insufficient to conclude (for instance in cases of recessive haplotypes for which no homozygous was available to test for haplotype expression). In some cases, different individuals with the same genotype gave different results and then the two possible conclusions were indicated. No positive control could be obtained for plants of genotype (S01, S08).

**S02**

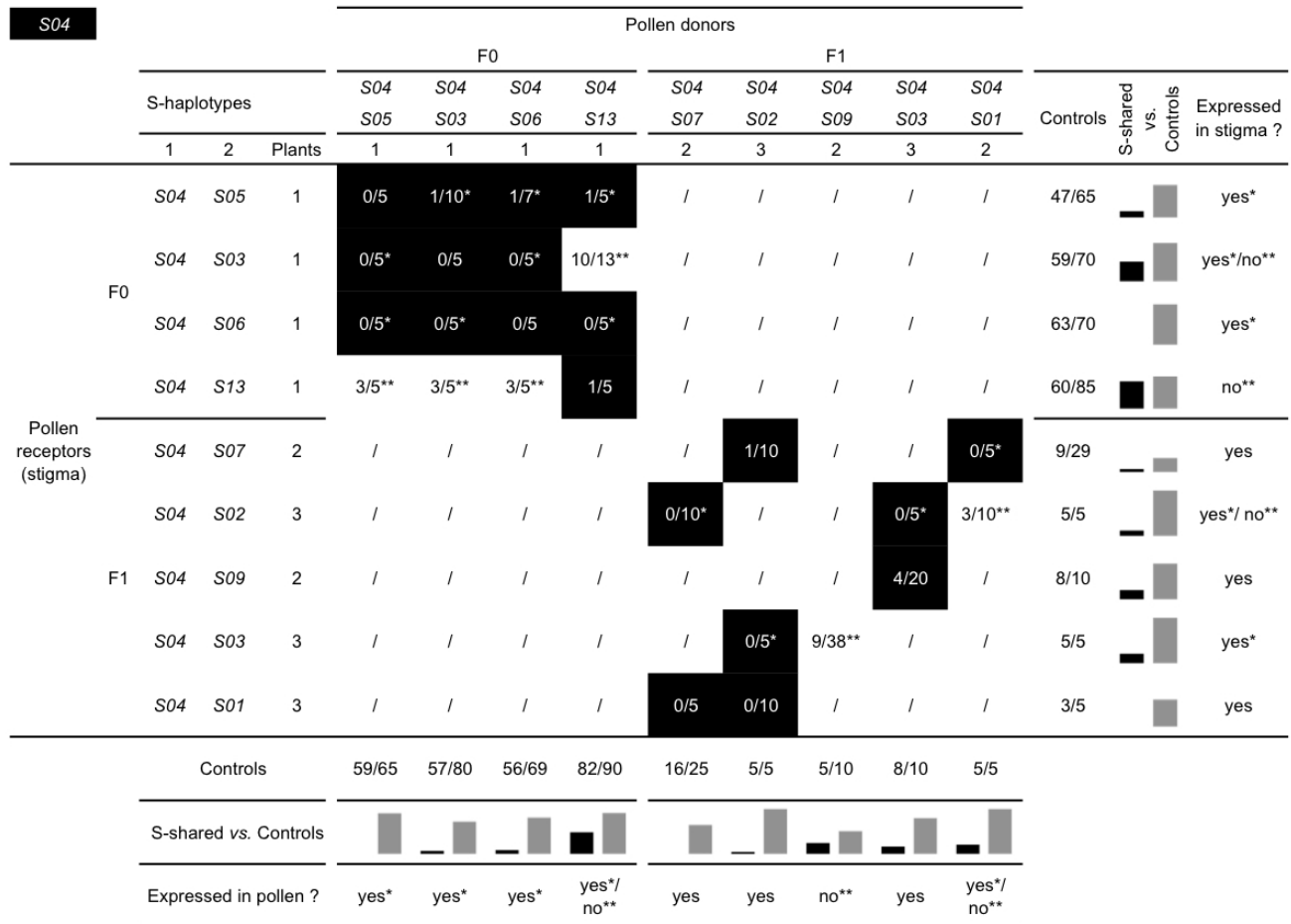
			Pollen donors														
			F0				F1										
S-haplotypes		1	S02	S02	S02	S02	S02	S02	S02	S02	S02	S02	S02	S02	Controls	S-shared vs. Controls	Expressed in stigma ?
1	2	Plants	1	1	1	1	S09	S04	S08	S10	S03	S01	S12				
F0	S02	S07	1	0/5	0/5	0/5	0/5	0/5	0/5	/	/	/	/	/	72/75		yes
	S02	S03	1	0/5	0/5	0/5	0/5	/	/	/	/	/	/	/	51/60		yes
	S02	S11	1	0/5	0/5	0/5	0/5	/	/	/	/	/	/	/	55/65		yes
	S02	S01	1	0/5	0/5	0/5	0/5	/	/	/	/	/	/	/	59/65		yes
Pollen receptors (stigma)	S02	S09	3	0/5	/	/	/	/	/	/	0/60	/	/	7/14		yes	
	S02	S04	3	0/5	/	/	/	/	/	/	0/20	1/44	/	5/5		yes	
	S02	S08	2	/	/	/	/	/	/	0/20	/	/	/	5/5		yes	
F1	S02	S10	2	/	/	/	/	/	0/20	/	/	/	/	23/30		yes	
	S02	S03	5	/	/	/	/	0/48	0/19	/	/	/	1/55	11/15		yes	
	S02	S01	4	/	/	/	/	/	1/39	/	/	/	/	8/15		yes	
	S02	S12	4	/	/	/	/	/	/	/	0/78	/	/	7/10		yes	
Controls			54/73	57/60	56/70	59/65	11/15	5/5	5/5	15/18	5/10	8/19	10/10				
S-shared vs. Controls																	
Expressed in pollen ?			yes	yes	yes	yes	yes	yes	yes	yes	yes	yes	yes				

**Figure S2** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S02 (A01-A03). See Figure S1 for legend details.

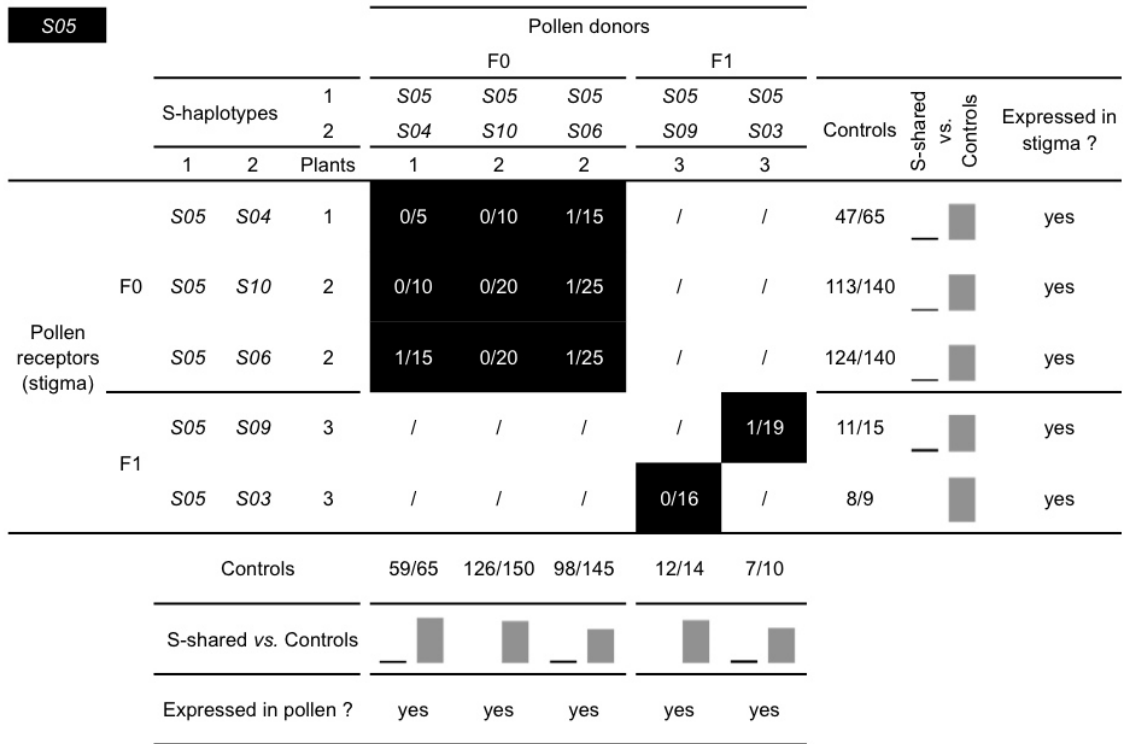
S03			Pollen donors												Controls			S-shared vs. Controls	Expressed in stigma ?
			F0					F1											
S-haplotypes		1	S03	S03	S03	S03	S03	S03	S03	S03	S03	S03	S03	S03	S03	Controls	S-shared vs. Controls	Expressed in stigma ?	
1	2	Plants	S02	S09	S04	S08	S12	S07	S02	S05	S04	S01	S13						
F0	S03	S02	1	0/5	0/5*	0/5*	0/5*	0/5*	/	/	/	/	/	/	51/60		yes		
	S03	S09	1	0/5*	0/5	0/5*	1/5*	0/5*	0/5	/	/	/	/	/	57/78		yes		
	S03	S04	1	1/5**	0/5*	0/5	1/10*	0/5*	0/4	/	/	/	/	/	59/70		yes		
	S03	S08	1	5/5**	2/5**	1/5*	0/5	4/5**	/	/	/	/	/	/	55/76		yes*/no**		
	S03	S12	1	0/5*	0/5*	0/5*	0/5*	0/5	0/5	/	/	/	/	/	60/75		yes		
Pollen receptors (stigma)	S03	S07	2	/	0/5	0/4	/	/	/	0/5	/	/	/	/	20/22		yes		
	S03	S02	4	/	/	/	/	/	0/5	/	/	/	0/35	/	8/10		yes		
F1	S03	S05	3	/	/	/	/	/	/	/	0/15	/	0/19	8/9		yes			
	S03	S04	3	/	/	/	/	/	/	0/15	/	/	0/5	5/5		yes			
	S03	S01	4	/	/	/	/	/	4/33	/	/	/	/	13/15		yes			
	S03	S13	2	/	/	/	/	/	/	1/20	0/5	/	/	9/9		yes			
Controls			57/60	67/85	57/80	59/85	65/70	19/25	4/5	7/10	8/10	2/14	8/10						
S-shared vs. Controls																			
Expressed in pollen ?			yes*	yes*	yes*	yes*	yes*	yes	yes	yes	yes	yes	yes	yes					

**Figure S3** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S03 (B06). See Figure S1 for legend details.





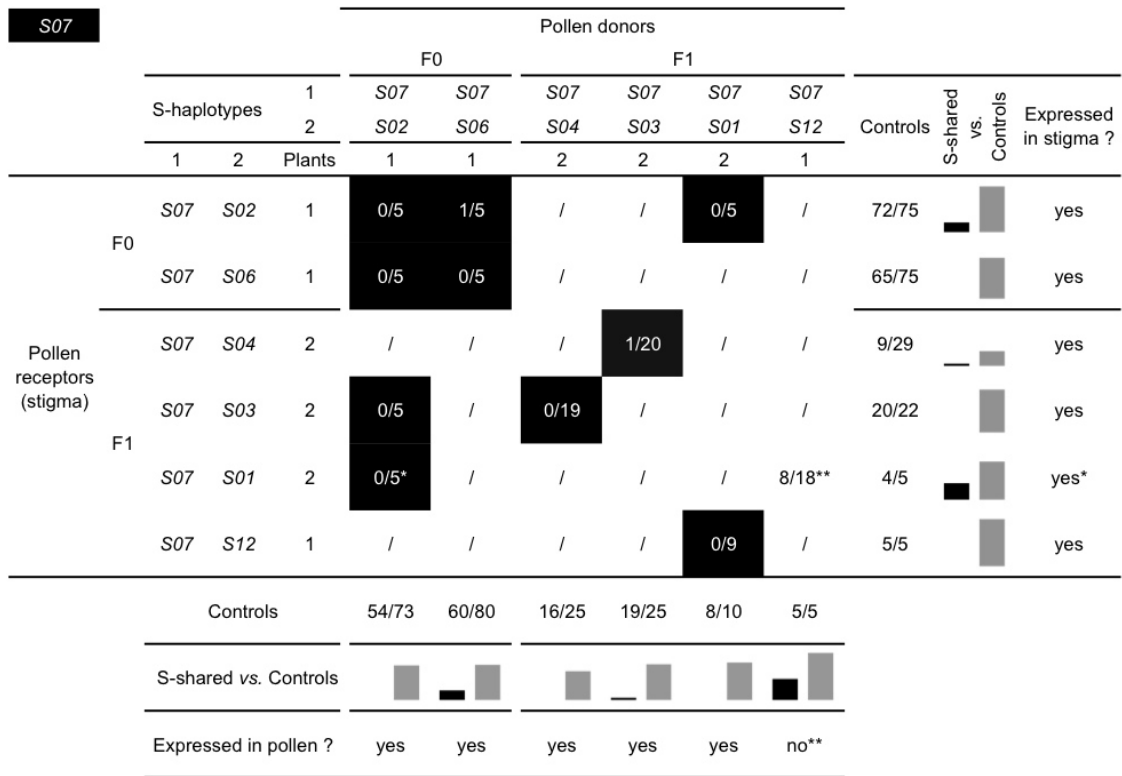
**Figure S4** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S04 (B01-B13). See Figure S1 for legend details.



**Figure S5** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S05 (A05). See Figure S1 for legend details.

S06			Pollen donors				Controls	S-shared vs. Controls	Expressed in stigma ?	
			F0							
S-haplotypes		1	S06	S06	S06	S06	Controls	S-shared vs. Controls	Expressed in stigma ?	
1	2	Plants	1	2	1	1				
Pollen receptors (stigma)	S06	S07	1	0/5	7/10**	5/5**	0/5*	65/75		yes*
	S06	S05	2	10/10**	1/25	9/10**	0/10*	124/140		yes*
	S06	S04	1	5/5**	8/15**	0/5	0/5*	63/70		yes*
	S06	S06	1	3/5**	5/10**	0/5*	0/5	37/75		?
Controls			60/80	98/145	56/69	37/48				
S-shared vs. Controls										
Expressed in pollen ?			no**	no**	yes*/no**	yes*				

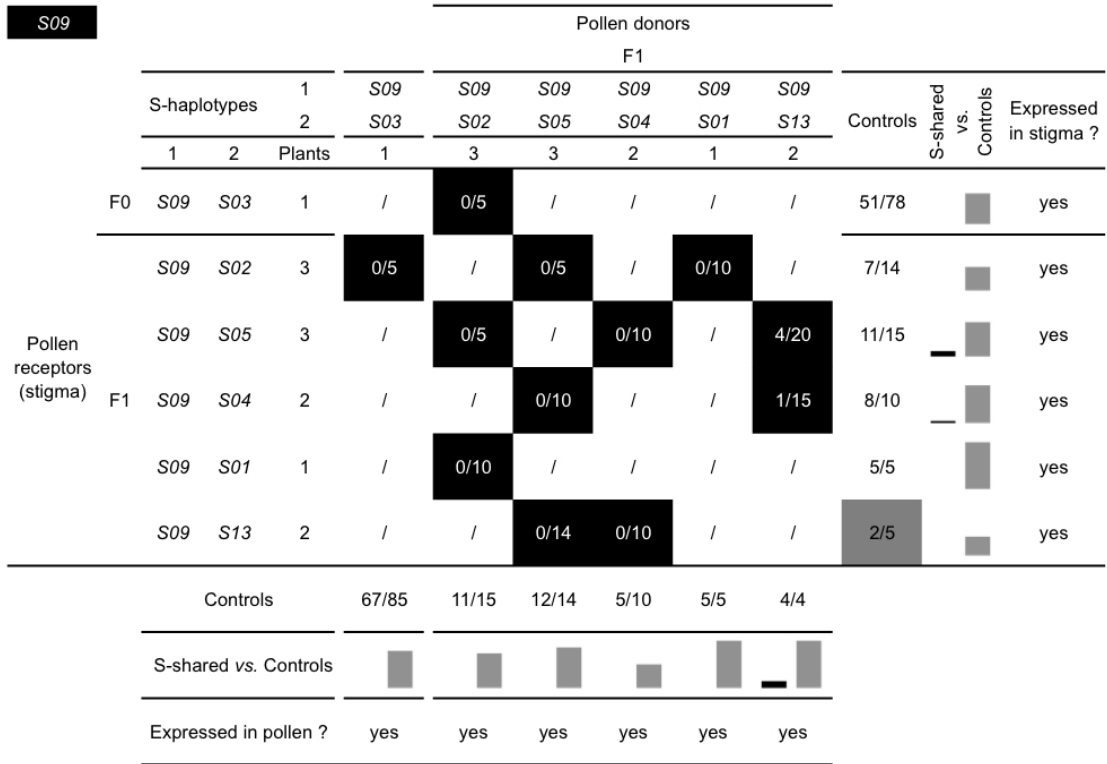
**Figure S6** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S06 (B04-C01). See Figure S1 for legend details.



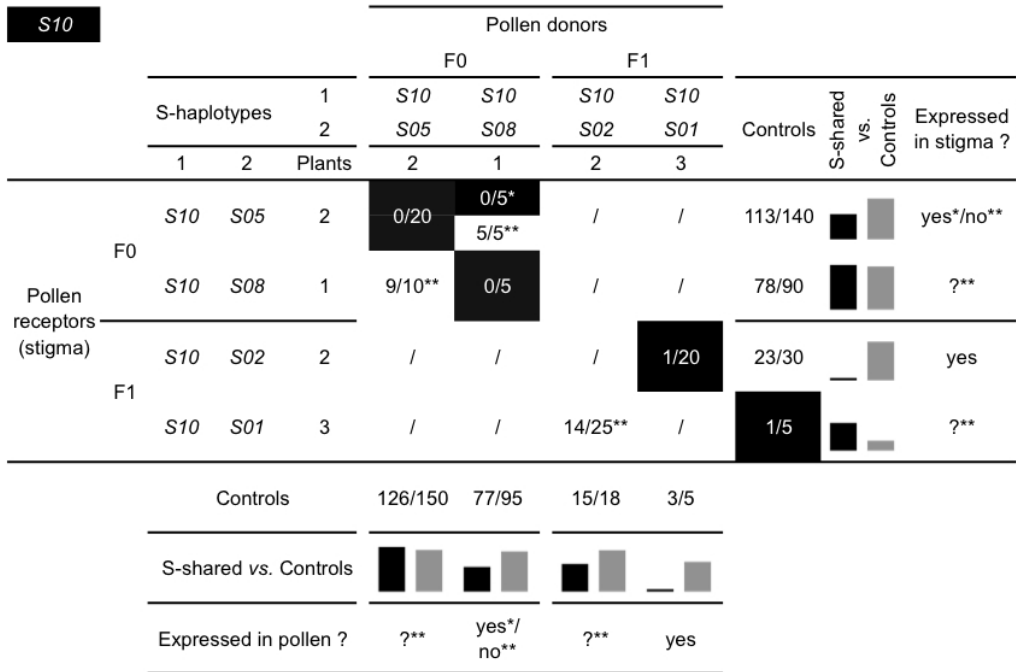
**Figure S7** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S07 (A01-A02). See Figure S1 for legend details.

S-haplotypes			Pollen donors				Controls	S-shared vs. Controls	Expressed in stigma ?	
			F0		F1					
1	2	Plants	1	1	2	2				
Pollen receptors (stigma)	F0	S08 S10	1	0/5	0/5	/	/	78/90		yes
		S08 S03	1	0/5	0/5	/	/	55/76		yes
	F1	S08 S02	2	/	/	/	0/19	5/5		yes
		S08 S01	2	/	/	1/15	/	/		yes
Controls			77/95	59/85	5/5	/				
S-shared vs. Controls							/			
Expressed in pollen ?			yes	yes	yes	yes				

**Figure S8** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype *S08* (*B03*). See Figure S1 for legend details. No positive control could be obtained for plants of genotype (*S01,S08*).



**Figure S9** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S09 (A06-A07). See Figure S1 for legend details.









**Figure S10** Summary of cross-pollinations realized for individuals from collection F0 and F1 having *S*-haplotype *S10* (*B05*). See Figure S1 for legend details.

S11			Pollen donors				Controls	S-shared vs. Controls	Expressed in stigma ?
			F0						
S-haplotypes		1	S11	S11					
		2	S02	S01					
		Plants	1	2					
Pollen receptors (stigma)	S11	S02	1	0/5	4/5*	55/65		?	
	S11	S01	2	5/5**	0/5	82/110		?	
Controls			56/70	68/85					
S-shared vs. Controls									
Expressed in pollen ?			?	?					





**Figure S11** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S11 (B09-B12). See Figure S1 for legend details.



			Pollen donors			Controls	S-shared vs. Controls	Expressed in stigma ?
			F1					
S-haplotypes	1		<i>S12</i>	<i>S12</i>	<i>S12</i>			
	2		<i>S07</i>	<i>S02</i>	<i>S01</i>			
Plants			1	4	1			
Pollen receptors (stigma)		<i>S12</i>	<i>S07</i>	1	5/5	NA	NA	?
	F1	<i>S12</i>	<i>S02</i>	4	5/5	7/10		yes*
		<i>S12</i>	<i>S01</i>	1	5/5	4/5	 	?
Controls			NA	10/10	3/4			
S-shared vs. Controls			NA	 				
Expressed in pollen ?			?	?	yes*			

**Figure S12** Summary of cross-pollinations realized for individuals from collection F0 and F1 having *S*-haplotype *S12* (*B17*-*B18*). See Figure S1 for legend details.

Haplotype *S12* was not typed in collection F1 because of no available specific primers for *B17* or *B18*. The presence of *S12* in collection F1 was thus deduced from the segregation pattern in offspring of individual 9 of (genotype *S03-S12*; see Figure 3 and Table 3). In collection F1, we thus considered that *S12* was present when *S03* (*B06*) was absent.

S13			Pollen donors					
			F1				Controls	
S-haplotypes		1	S13	S13	Controls	S-shared vs. Controls	Expressed in stigma ?	
1	2	Plants	S09	S03				
		2	2					
Pollen receptors F1 (stigma)	S13	S09	2	/	0/20	2/5		yes
	S13	S03	2	0/20	/	9/9		yes
Controls			4/4	8/10				
S-shared vs. Controls								
Expressed in pollen ?			yes	yes				

**Figure S13** Summary of cross-pollinations realized for individuals from collection F0 and F1 having S-haplotype S13 (A04).

<b>S02 vs. S07</b>				Pollen donors												
				F0					F1							
S-haplotypes		1	<b>S02</b>	<b>S07</b>	<b>S07</b>	<b>S07</b>	<b>S07</b>	<b>S07</b>	<b>S02</b>	<b>S02</b>	<b>S02</b>	<b>S02</b>	<b>S02</b>	<b>S02</b>	<b>S02</b>	
		2	<b>S07</b>	S04**	S03*	S01	S12***	S03*	S01	S09	S04**	S08	S10	S12		
1	2	Plants	1	2	2	2	1	5	4	3	3	2	2	4		
F0	<b>S02</b>	<b>S07</b>	1	/	/	/	0/5	/	/	/	0/5	0/5	/	/	/	
	<b>S07</b>	S04**	2	/	/	1/20	/	/	1/5	8/19	/	1/10**	/	/	/	
	<b>S07</b>	S03*	2	0/5	0/19	/	/	/	0/5*	/	/	5/5	/	7/8	5/5	
	<b>S07</b>	S01	2	0/5	/	/	/	8/18***	4/5	/	/	/	/	/	/	
	<b>S07</b>	S12	1	/	/	/	0/9	/	/	/	/	/	/	/	5/5	
Pollen receptors (stigma)	F1	<b>S02</b>	S03*	5	/	3/5	0/5*	8/10	/	/	/	0/48	0/19	/	/	1/55
		<b>S02</b>	S01	4	/	8/15	/	/	/	/	/	/	1/39	/	/	/
		<b>S02</b>	S09	3	0/5	/	/	/	/	0/60	/	/	/	/	/	/
		<b>S02</b>	S04**	3	0/5	0/10**	5/5	/	/	0/20	1/44	/	/	/	/	/
		<b>S02</b>	S08	2	/	/	/	/	/	/	/	/	/	/	0/20	/
		<b>S02</b>	S10	2	/	/	8/10	/	/	/	/	/	0/20	/	/	/
		<b>S02</b>	S12	4	/	/	2/5	/	5/5	0/78	/	/	/	/	/	/

**Figure S14** Cross-pollinations between plants having haplotype S02 and plants having haplotype S07 are mostly successful, indicating that S02 and S07 are distinct functional S-haplotypes, while sharing sequence A01 in common. See Figure S1 for legend details.