

1 S1 Table. Summary of Markers, gene, location and enzymes used in PCR-RFLP (Restriction Fragment Length Polymorphism) (Su et al., 2010)

Marker	Gene	Location	Multiplex PCR primers	nPCR primers	nPCR (bp)	Restriction enzymes	Incubation time and temperature	Reference
SAG1	SAG1	chromosome VIII	F**:GTTCTAACCACGCACCCTGAG R***:AAGAGTGGGAGGCTCTGTGA	F: CAATGTGCACCTGTAGGAAGC R:GTGGTTCTCCGTCCGGTGTGAG	390	Sau96I + HaeII	37°C 1h. 2,5% gel	Grigg et al., 2001b ; Khan et al., 2005a
5'SAG2	SAG2	chromosome VIII	It is not necessary. The DNA fragment for 5'SAG2 is covered by the external alt.SAG2 primers.	F:GAAATGTTTCAGGTTGCTGC R:GCAAGAGCGAACTTGAACAC	242	MboI	37°C 1h. 2,5% gel	Howe et al., 1997; Khan et al., 2005a
3'SAG2	SAG2	chromosome VIII	F:TCTGTTCTCCGAAGTACTCC R:TCAAAGCGTGCATTATCGC	F:ATTCTCATGCCTCCGCTTC R:AACGTTTCACGAAGGCACAC	222	HhaI	37°C 1h. 2,5% gel	Lehmann et al., 2006 Khan et al., 2005a
alt.SAG2	SAG2	chromosome VIII	F:GGAACGCGAACAATGAGTTT R:GCACTGTTGTCCAGGGTTTT	F:ACCCATCTGCGAAGAAAACG R:ATTTCCAGCAGCGGAGCAC	546	HinfI +TaqI	37°C 30min, 65°C 30 min. 2,5% gel	Grigg et al., 2001b; Khan et al., 2005a
SAG3	SAG3	chromosome XII	F:CAACTCTCACCATTCCACCC R:GCGCGTTGTTAGACAAGACA	F:TCTTGTCGGGTGTTCACTCA R:CACAAGGAGACCGAGAAGGA	225	NciI	37°C 1h. 2,5% gel	Khan et al., 2005a
BTUB	BTUB	chromosome IX	F:TCCAAAATGAGAGAAATCGT R:AAATTGAAATGACGGAAGAA	F:GAGGTCATCTCGGACGAACA R:TTGTAGGAACACCCGGACGC	411	BsiEI +TaqI	60°C 1h. 2,5% gel	Khan et al., 2005a
GRA6	GRA6	chromosome X	F:ATTTGTGTTTCCGAGCAGGT R:GCACCTTCGCTTGTGGTT	F:TTTCCGAGCAGGTGACCT R:TCGCCGAAGAGTTGACATAG	344	MseI	37°C 1h. 2,5% gel	Khan et al., 2005a
C22-8	C22-8	chromosome Ib	F:TGATGCATCCATGCGTTTAT R:CCTCCACTTCTTCGGTCTCA	F:TCTCTCTACGTGGACGCC R:AGGTGCTTGGATATTCGC	521	BsmAI + MboII	37°C 30min, 55°C 30 min. 2,5% gel	Khan et al., 2005a
C29-2	C29-2	chromosome III	F:ACCCACTGAGCGAAAAGAAA R:AGGGTCTCTTGCGCATAACAT	F:AGTTCTGCAGAGTGTCCG R:TGTCTAGAAAAGAGGCCG	446	HpyCH4IV + RsaI	37°C 1h. 2% gel	Khan et al., 2005a
L358	L358	chromosome o V	F:TCTCTCGACTTCGCCTCTTC R:GCAATTTCTCGAAGACAGG	F:AGGAGGCGTAGCGCAAGT R:CCCTCTGGCTGCAGTGCT	418	HaeIII + NlaIII	37°C 1h. 2,5% gel	Khan et al., 2005 a,

PK1	PK1	chromosome VI	F:GAAAGCTGTCCACCCTGAAA R:AGAAAGCTCCGTGCAGTGAT	F:CGCAAAGGGAGACAATCAGT R:TCATCGCTGAATCTCATTGC	903	AvaI + RsaI	37°C 1h. 2,5% gel	Khan et al., 2005 a,
APICO	Apico	Apicoplast chromosome	F:ATGGTTTTAACCTAGATTGTGG R:ACGGAATTAATGAGATTGAA	F:GCAAATTCCTGAATTCTCAGTT R:GGGATTCGAACCCTTGATA	640	AflIII + DdeI	37°C 1h. 2,5% gel	Dubey et al; 2007

2 * CutSmart Buffer® was used for all reactions; **F: forward *primer*; ***R: reverse *primer*

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