

	10	20	30	40	50	60	70	80	90	100
BnaA09g45590D
At1g12620	TGAAATTCGT	TCGGTGTGCG	GATCTAGTTG	TGGGTGAAAG	GGCGAA---G	AAGAATC---	---ATCAAGT	CA--CTGAAG	ATGAAGATGG	GAGTGGAGTT
At1g12300	TTTGTTTTAT	TGTGTGTGAA	AGAGGTGTAA	TGAGCAGACA	TTGGTGTCTG	TGGAAGCCGA	ACAAACGAAT	CAGCACAAAA	CCGAAGACGT	GACTCCAATT
Clustal Consensus	** *	** **		* *	*	*	*	* * * *	** *	* * *

	110	120	130	140	150	160	170	180	190	200
BnaA09g45590D
At1g12620	GTTCTTCTTC	TTCTTTCTAG	TTCTTTATTT	TTCCAGATTT	GAAACAAAAA	CAGACAATGT	TGTGTCGGAG	ATTGGTACGT	GAGCCTCGTA	TCCCTCTGAG
At1g12300	TTTTTTTTTT	TAAATCAGAG	CTTTCAAATT	CTTAAC--TT	GAGTCATGCG	GGGATTG-AT	TCAGACACGT	CTTCTCGAGA	CAGGTACTCT	AAGAAGTGCT
Clustal Consensus	* *	**	* **	* *	*	**	*	*	*	**

	210	220	230	240	250	260	270	280	290	300
BnaA09g45590D
At1g12620	TCTTGGAGGT	ACTCTTACAG	TTACAACCTCA	TCATTTGCTC	TCTCGGAAGA	TGTC-----T	TGTTACAAAG	AGAGATTGCG	AAGTGGGATC	GTCGATATTA
At1g12300	TTATCTTTGT	CTTGCTATGG	ACGAGTCTTT	TCTAGTGTCA	GTGATGGGAA	AGGGAAAGTC	TCCTATAGAG	AGAGATTGAG	AAGTGGGATT	GTGATATCA
Clustal Consensus	*	* *	**	** *	*		* * * *	***** *	***** *	* * * * *

	310	320	330	340	350	360	370	380	390	400
BnaA09g45590D
At1g12620	AGAAAGATGA	AGCTGTTGCT	CTGTTTCAA	CTATGATTGT	GTCTCGTCCT	CTTCCT---	-----	-----	-----	-----
At1g12300	AGGAAGATGA	TGCTGTTGAT	CTGTTTCAAG	AAATGACTAG	GTCTCGTCCT	CGTCCTCGTC	TTATTGATTT	CAGTAGATTG	TTTAGTGTG	TTGCCAGAAC
Clustal Consensus	** *****	*** ** *	***** *	***** *	***** *	***** *	***** *	***** *	***** *	***** *

	410	420	430	440	450	460	470	480	490	500
BnaA09g45590D
At1g12620	-----	-----	-----	-ACGCAAATG	GAAATGAATG	GGATTGCGCA	TGACCTCTAC	ACGCTGAGTA	TTGTCATCAA	CTGCTTCTGC
At1g12300	AAAGCAATAT	GATCTAGTGT	TAGATCTCTG	CAAGCAAATG	GAATTAAAGG	GAATTGCGCA	TAACCTCTAC	ACTCTGAGTA	TTATGATTAA	TTGCTGTTGT
Clustal Consensus				* *****	*** * * *	* * * * *	* * * * *	* * * * *	* * * * *	*** **

	510	520	530	540	550	560	570	580	590	600
BnaA09g45590D
At1g12620	CGTCGCCGGA	AATC-----	-----	-----	-----	-----	-----	--GTCACCTT	CTCAACTTTG	ATTAACGGTT
At1g12300	CGATGTCGTA	AATCTCTTTT	GGCTTTTTCT	GCTATGGGGA	AGATCATTA	ACTTGGGTAT	GAGCCTGATA	CAGTCACATT	TTCAACTTTG	ATAAACGGAT
Clustal Consensus	** * * *	*****						***** *	***** *	***** *

	610	620	630	640	650	660	670	680	690	700
BnaA09g45590D
At1g12620	TATGTCTCAA	GGGTCGAGTC	TCTGAAGCTG	TGGAGTTAGT	TGATCGTATG	GTGGAAATGA	AGGTTATTCC	AAATCTCATC	ATACTTAACA	CTCTTGTCAA
At1g12300	TATGTCTCGA	GGGTAGAGTT	TCCGAAGCTC	TGGAATTAGT	TGATCGAATG	GTTGAAATGG	GACATAAACC	CACTCTCATA	ACGCTTAACG	CTCTGGTCAA
Clustal Consensus	***** *	**** *	** *****	**** *****	***** *	** *****	** *	***** *	***** *	***** *

	710	720	730	740	750	760	770	780	790	800
BnaA09g45590D
At1g12620	TGGACTTTGT	CTCCAAGATA	GAGTGTCCGA	AGCAGTGGCT	TTGATAGATC	GAATGGTGGC	TAATGGATGT	CAACCCGATC	AGTTTACCTA	TGGTCCGATT
At1g12300	TGGACTTTGT	CTCAACGGTA	AAGTCTCTGA	TGCTGTGCTT	TTGATTGATC	GAATGGTTGA	GAATGGCTTT	CAACCCAATG	AAGTAACTTA	TGGACCGGTT
Clustal Consensus	***** *	*** ** *	** ** *	** ** *	***** *	***** *	*** ** *	***** *	* ** *	** ** *

	810	820	830	840	850	860	870	880	890	900
BnaA09g45590D
At1g12620	TTGAACAGAA	TGTGCAA---	-----	-----	-----	-----ATG	GAAGATCGAA	AGGTCAAGCC	-----TC	AAAGTA-----
At1g12300	TTAAAGTAA	TGTGTAAGTC	CGGCCAAACT	GCCCTTGCCA	TGGAGTTGCT	CAGAAAGATG	GAAGAAAGAA	AGATCAAGCT	CGATGCAGTC	AAATACAGTA
Clustal Consensus	** ** *	** **** *	CGGCCAAACT	GCCCTTGCCA	TGGAGTTGCT	CAGAAAGATG	GAAGAAAGAA	ATATCAAGCT	CGATGCAGTC	AAATACAGTA

	910	920	930	940	950	960	970	980	990	1000
BnaA09g45590D
At1g12620	TCATCATTGA	TGGTCTGTGC	AAAGACGGGA	GCCTCGACAA	TGCATTCAAC	CTTTTCAATG	AAATGGAAAT	CAAAGGGTTC	AAAGCTGATA	TTATTATCTA
At1g12300	TCATCATTGA	TGGTCTTTGC	AAACACGGGA	GCCTCGACAA	TGCATTCAAC	CTTTTCAATG	AAATGGAAAT	GAAAGGGATC	ACAACAAATA	TTATTACCTA
Clustal Consensus										

	1010	1020	1030	1040	1050	1060	1070	1080	1090	1100
BnaA09g45590D
At1g12620	CACCACTCTC	ATTAGAGGCT	TTTGTTACGC	TGGTAGATGG	GATGATGGTG	CCAAGTTGCT	GAGGGATATG	ATCAAAAGGA	AAATCACACC	AGACGTGGTC
At1g12300	CAACATTCTC	ATTGGCGGCT	TTTGTAATGC	TGGTAGATGG	GATGATGGTG	CCAAGTTATT	GAGGGATATG	ATCAAGAGGA	AAATCAACCC	CAACGTTGTC
Clustal Consensus										

	1110	1120	1130	1140	1150	1160	1170	1180	1190	1200
BnaA09g45590D
At1g12620	-----TG	CGTTGATTGA	TAGTTTTGTG	AAAGAAGGAA	AGCTTACTGA	GGCTAAAGAG	TTGTACAATG	AGATGATCGT	AAGAGGCTTA	GATCCTAATA
At1g12300	GCTTTCAGCG	CGTTAATAGA	TTGTTTTGTG	AAAGAGGGAA	AGCTTCGAGA	GGCTGAAGAA	CTGCACAAGG	AGATGATACA	ACGAGGCATA	TCTCCTGATA
Clustal Consensus		* ** ** *	* *****	***** *	***** *	***** *	** *****	***** *	***** *	***** *

	1210	1220	1230	1240	1250	1260	1270	1280	1290	1300
BnaA09g45590D
At1g12620	CCATTACATA	TGGTTCTCTG	ATACATGGGC	TGTGCATGGA	GAATCGCTTA	GATGAGGCCA	ACCAGATGTT	GGATCTGATG	GTTAGCAAGG	GATGCGATCC
At1g12300	CTGTTACGTA	CACTTCTTTG	ATAGATGGGT	TTTGCAAGGA	GAACCAGCTA	GATAAGGCCA	ACCACATGCT	GGATCTGATG	GTTAGCAAAG	GATGCGGTCC
Clustal Consensus	* * * * *	*****	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *

	1310	1320	1330	1340	1350	1360	1370	1380	1390	1400
BnaA09g45590D
At1g12620	TGATATTTGG	ACGTATAATA	TCCTTATAAA	CGGGTATTGT	AAGGCTAAAC	TGGTTGATGA	AGGTATGAGA	CTTTTCCGCA	AAATGTCTCT	GAGAGGAGTG
At1g12300	TAATATCAGG	ACGTTTAATA	TCCTCATAAA	TGGATATTGT	AAGGCTAATC	TGATTGACGA	TGGTTTGAA	CTCTTCCGCA	AAATGTCTTT	GAGAGGAGTG
Clustal Consensus	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *	* * * * *

	1410	1420	1430	1440	1450	1460	1470	1480	1490	1500
BnaA09g45590D
At1g12620	GTTGCAGATA	CAGTCACTTA	TAGCAGTCTC	ATTCAAGGGT	TTTGTCAATC	AGGAAAACCT	AAAGTCGCCA	AAGAACTCTT	CCAAGAGATG	GTCTCTCAAG
At1g12300	GTTGCAGATA	CGGTTACTTA	TAACACTCTT	ATCCAAGGAT	TTTGTGAATT	GGGAAAACCT	GAAGTTGCCA	AAGAACTCTT	CCAAGAGATG	GTTTCTCGTC
Clustal Consensus	*****	* * * * *	* * * * *	* * * * *	* * * * *	*****	* * * * *	*****	*****	* * * * *

	1510	1520	1530	1540	1550	1560	1570	1580	1590	1600
BnaA09g45590D
At1g12620	GTGTTCATCC	TAATATTGTA	ACCTACAAAA	TTTTGATGGA	TGGA-----	-----	-----	-----	-----	-----
At1g12300	GTGTTCATCC	TAATATTGTA	ACCTACAAAA	TTTTGCTCGA	TGGGTTGTGT	GACAATGGGG	AACCAGAAAA	AGCGCTGGAA	ATATTTGAAA	AAATAGAGAA
Clustal Consensus	**** *	* * * * *	*****	*****	* * *	*****	*****	*****	*****	*****

	1610	1620	1630	1640	1650	1660	1670	1680	1690	1700
BnaA09g45590D
At1g12620	-----	-----	-----	-----	-----	-----	-----GTC	GATGAAGCTT	GGGATCTATT	TTGTAGCCTC
At1g12300	GAGTAAGATG	GAGCTTGATA	TTGGTATCTA	TAACATCATC	ATTCATGGGA	TGTGCAATGC	TAGTAAGGTG	GATGATGCTT	GGGATTTATT	CTGTAGCCTC
Clustal Consensus	GAGTAAGATG	GAGCTTGATA	TTGGTATCTA	TAACATCATC	ATTCACGGGA	TGTGCAATGC	TAGTAAGGTG	GATGATGCTT	GGGATTTATT	CTGTAGCCTC

	1710	1720	1730	1740	1750	1760	1770	1780	1790	1800
BnaA09g45590D
At1g12620	CCTCTCAAAG	AAGTGAAGCC	TGATGTCAAA	ACATACACCA	TAATGATTGG	AGGACTATGT	AAGAAAGGCT	CGATGTCTAA	AGCGGGCCTG	TTATTTAAGA
At1g12300	CCTCTCAAAG	GAGTGAAGCC	CGATGTCAAA	ACATACAATA	TAATGATTGG	GGGACTGTGT	AAGAAAGGCT	CACTGTCTGA	AGCAGACCTG	TTGTTTAGAA
Clustal Consensus	CCCTCAAAG	GAGTGAAGCC	CGGTGTCAAA	ACATACAATA	TAATGATTGG	GGGACTGTGT	AAGAAAGGCC	CATTGTCTGA	AGCGGAACCTG	TTGTTTAGAA

	1810	1820	1830	1840	1850	1860	1870	1880	1890	1900
BnaA09g45590D
At1g12620	AAATGGGAGA	GGATGGGATT	GCACCAGATG	ATTGTACATG	GAACACACTA	ATCCGAGCTC	ATCTCCGAGG	TGGTGACTTA	GCAACATCAG	CTAAACTTAT
At1g12300	AAATGGAAGA	GGATGGGCAT	TCGCCAAATG	GTTGTACATA	CAACATACTC	ATCAGGGCAC	ATCTTGGAGA	AGGTGATGCA	ACCAAATCAG	CTAAACTTAT
Clustal Consensus	*****	***	*****	*	*	***	***	***	*****	*****
	1910	1920	1930	1940	1950	1960	1970	1980	1990	2000
BnaA09g45590D
At1g12620	AGAAGAAATG	AAGAGTTGTG	GGTTCTCTGC	AGATGCTTCC	ACGATAAAGA	TGTTTATGGA	TATGTTATCG	GATGGTAGAA	TGAAGAAAAG	CTTTCTGGAT
At1g12300	TGAAGAAATA	AAGAGGTGCG	GGTTCTCAGT	TGATGCTTCC	ACGGTAAAGA	TGGTTGTCGA	TATGTTATCG	GATGGTAGAT	TGAAGAAAAG	CTTTTGGAT
Clustal Consensus	*****	*	*****	*	*****	*	***	*	*****	*****
	2010	2020	2030	2040	2050	2060	2070	2080	2090	2100
BnaA09g45590D
At1g12620	TTGCTTTCTT	AG-----	-----	-----	-----	-----	-----	-----	-----	-----
At1g12300	ATGCTTTCTT	AGAATTTTGG	TGTTTACATC	ATATTACCGT	CTCAGAGCAT	TGAAAGACTA	AGTTGGAGTT	TCATATGTGA	CAACATTGAT	GTACACAACA
Clustal Consensus	*****	***	*****	*****	*****	*****	*****	*****	*****	*****
	2110	2120	2130	2140	2150	2160	2170	2180	2190	2200
BnaA09g45590D
At1g12620	TTTTTCTTAA	GCAGATCGAC	CACCAAGATT	TATAACCATT	CTCCGGGC-A	TGAAAGCAAG	CTCATAATTT	CCTTATCACT	CTGTTC--AT	CTGTCTACAT
At1g12300	AACCTGAAAA	ACCTTCAGAC	TCTTGGAAC	GGAAGTGTCT	CTTAAGGTTG	CGTTGTTTAG	CAGAAAAC	TGTCAGGTGT	AAGATTGGAG	ATGGAAAGAC
Clustal Consensus	*****	*****	*****	*****	*****	*****	*****	*****	*****	*****
	2210	2220	2230	2240	2250	2260	2270	2280	2290	2300
BnaA09g45590D
At1g12620	TGCT--TTTC	AGATTGTACT	TGCTAAATTC	TG-----	CACAAATATG	TATAG-GTAA	ATGATATAAT	TTGTAGCTTA	AACACACCAT	CGCTATCAAA
At1g12300	TGCTAGTTTC	TGGTT-TGAT	AACTGGACTC	CTTTTGGTCC	CCTAATCAAG	TTCATTGGAA	CAGATGGCCC	CTGTAGTTGA	GAGCCCCTTT	GAATGCTAAG
Clustal Consensus	*****	*****	*****	*****	*****	*****	*****	*****	*****	*****

Figure S1. Sequence alignment of *BnaRFL11* with *Arabidopsis* homologs genes. Gene sequence alignment from *B. napus* (*BnaA09g45590D*) and *A. thaliana* (*AT1G12300*, *AT1G12620*). The base differences are highlighted in grey boxes. Star show the similarities between each copy and portion without star show polymorphism.

10 20 30 40 50 60 70 80 90 100

BnaA09g45590D	YKERLRSGIV	DIKKDEAVAL	FQTMIVSRPL	PTQMEMNGIA	HDLYTLSIVI	NCFCRRRKL-	-----	-----VT	FSTLINGLCL	KGRVSEAVEL	
AT1G12300	YRERLRSGLV	DIKADDAIDL	FRDMIHSRPL	PKQMELKGIA	HNLYTLSIMI	NCFCRCRKLC	LAFSAMGKII	KLGYEPNTIT	FSTLINGLCL	EGRVSEALEL	
AT1G12620	YRERLRSGIV	DIKEDDAVDL	FQEMTRSRRP	PKQMELKGIA	HNLYTLSIMI	NCCRCRKLK	LAFSAMGKII	KLGYEPDVT	FSTLINGLCL	EGRVSEALEL	
Clustal Consensus	*:*****:	*** *: *: *	*: * ***	*.***:***	*:*****:	** ** **		: *	*****	:*****:	**

	110	120	130	140	150	160	170	180	190	200

BnaA09g45590D	VDRMVEMKVI	PNLIILNTLV	NGLCLQDRVS	EVALIDRMV	ANGCQPDQFT	YGPILNRMCK	WKIERSSLKY	ALIDSFVKEG	KLTEAKELYN	EMIVRGDLPN
AT1G12300	VDRMVEMGHK	PDLITINTLV	NGLCLSGKEA	EAMLLIDKMV	EYGCQPNVAVT	YGPVLNVNCK	RNIKLDAVKY	VLIDSFVKEG	KLREAELHK	EMIHRIAPD
AT1G12620	VDRMVEMGHK	PTLITLNALV	NGLCLNGKVS	DAVLLIDRMV	ETGFQPNVET	YGPVLKVMCK	RKIKLDAVKY	ALIDCFVKEG	KLREAELHK	EMIQRGISPD
Clustal Consensus	*****	* ** *: **	*****.: :	:*: ***: **	* *: . *	***: *: **	:*: .: **	.***.*****	** *: *: *: *	*** *: *: *

	210	220	230	240	250	260	270	280	290	300

BnaA09g45590D	TITYGSLIHG	LCMENRLDEA	NQMLDLMVSK	GCDPDIWTYN	ILINGYCKAK	LVDEGMRLFR	KMSLRGVVAD	TVTYSSLIQG	FCQSGKLKVA	KELFQEMVSQ
AT1G12300	TITYTSLIDG	FCKENHLDKA	NQMVDLMVSK	GCDPNIRTFN	ILINGYCKAN	RIDDGLELFR	KMSLRGVVAD	TVTYNTLIQG	FCELGKLNVA	KELFQEMVSR
AT1G12620	TVTYTSLIDG	FCKENQLDKA	NHMLDLMVSK	GCGPNIRTFN	ILINGYCKAN	LIDDGLELFR	KMSLRGVVAD	TVTYNTLIQG	FCELGKLEVA	KELFQEMVSR
Clustal Consensus	*: ** ***. *	: * *: *: *	*: *: *****	**.: * *: *	*****:	: *: *: .***	*****	****.: ****	** *: *: *: *	*****:

	310	320	330	340	350	360	370	380	390	400

BnaA09g45590D	GVHPNIVTYK	ILMDG--VDE	AWDLFCSLPL	KEVKPDVKTY	TIMIGGLCKK	GMSKAGLLF	KKMGEDGIAP	DDCTWNTLIR	AHLRGGLAT	SAKLIEEMKS
AT1G12300	KVPPNIVTYK	ILLDGLKVDD	AWDLFCSLPL	KGVKPGVKTY	NIMIGGLCKK	GPLSEAEELLF	RKMEEDGHAP	DGWTYNILIR	AHLGDGDATK	SVKLIEELKR
AT1G12620	RVRPDIVSYK	ILLDGLKVDD	AWDLFCSLPL	KGVKPDVKTY	NIMIGGLCKK	GSLSEADLLF	RKMEEDGHSP	NGCTYNILIR	AHLGEGDATK	SAKLIEEIKR
Clustal Consensus	* *: *: *	** *: *	*****	* **.: ****	.*****	*.: *: * **	: ** *** : *	.. *: * ***	*** ** :	*.*****:

	410	420	430

BnaA09g45590D	CGFSADASTI	KIVMDMLSDG	RMKKSFLDLL S
AT1G12300	CGFSVDASTI	KMVIDMLSDG	RLKKSFLDML S
AT1G12620	CGFSVDASTV	KMVVDMLSDG	RLKKSFLDML S
Clustal Consensus	***.***:	* *: *****	*.*****: *

Figure S2. Protein sequence alignment from *B. napus* (*BnaA09g45590D*) and *A. thaliana* (*AT1G12300*, *AT1G12620*). The base differences are highlighted in grey boxes. Star show the similarities between each copy and portion without star show polymorphism.

***BnaRFL11* targets**

	10	20	30	40	50	60	70	80	90	100	
BnaA09g45590D	AAATCATCGA	TGAAATTCGT	TCGGTGTGCG	GATCTAGTTG	TGGGTGAAAG	GGCGAAGAAG	AATCATCAAG	TCACTGAAGA	TGAAGATGGG	AGTGGAGTTG	
	110	120	130	140	150	160	170	180	<u>S1</u>	190	200
BnaA09g45590D	TTCTTCTTCT	TCTTCTAGGT	TCTTTATTTT	TCCAGATTTG	AAACAAAAAC	AGACAATGTT	GTGTCGGAGA	TTGGTACGTG	<u>AGCCTCGTAT</u>	<u>CCCTCTGAGT</u>	
	210	220	230	240	250	260	270	280	290	300	
BnaA09g45590D	<u>CTTGG</u> AGGTA	CTCTTACAGT	TACAAC TCAT	CATT TGCTCT	CTCGGAAGAT	GTCTTGTTAC	AAAGAGAGAT	TGCGAAGTGG	GATCGTCGAT	ATTAAGAAAG	
	310	320	330	340	350	360	370	380	390	400	
BnaA09g45590D	ATGAAGCTGT	TGCTCTGTTT	CAAAC TATGA	TTGTGTCTCG	TCCTCTTCCT	ACGCAAATGG	AAATGAATGG	GATTGCGCAT	GACCTCTACA	CGCTGAGTAT	
	410	420	430	440	450	460	470	480	490	500	
BnaA09g45590D	TGTCATCAAC	TGCTTCTGCC	GTGCGCCGAA	ACTCGTCACT	TTCTCAACTT	TGATTAACGG	TTTATGTCTC	AAGGGTCGAG	TCTCTGAAGC	TGTGGAGTTA	
	510	520	530	540	550	560	570	580	590	600	
BnaA09g45590D	GTTGATCGTA	TGGTGGAAAT	GAAGGTTATT	CCAAATCTCA	TCATACTTAA	CACTCTTGTC	AATGGACTTT	GTCTCCAAGA	TAGAGTGTCC	GAAGCAGTGG	
	610	620	630	640	650	660	670	680	<u>S1</u>	690	700
BnaA09g45590D	CTTTGATAGA	TCGAATGGTG	GCTAATGGAT	GTCAACCCGA	TCAGTTTACC	TATGGTCCGA	TTTTGAACAG	<u>AATGTGCAAA</u>	<u>TGGAAGATCG</u>	<u>AAAGG</u> TC AAG	
	710	720	730	740	750	760	770	780	790	800	
BnaA09g45590D	CCTCAAGTAT	GCGTTGATTG	ATAGTTTTGT	GAAAGAAGGA	AAGCTTACTG	AGGCTAAAGA	GTTGTACAAT	GAGATGATCG	TAAGAGGCTT	AGATCCTAAT	
	810	820	830	840	850	860	870	880	890	900	
BnaA09g45590D	ACCATTACAT	ATGGTTCTCT	GATACATGGG	CTGTGCATGG	AGAATCGCTT	AGATGAGGCC	AACCAGATGT	TGGATCTGAT	GGTTAGCAAG	GGATGCGATC	
	910	920	930	940	950	960	970	980	990	1000	
BnaA09g45590D	CTGATATTTG	GACGTATAAT	ATCCTTATAA	ACGGGTATTG	TAAGGCTAAA	CTGGTTGATG	AAGGTATGAG	ACTTTTCCGC	AAAATGTCTC	TGAGAGGAGT	

	1010	1020	1030	1040	1050	1060	1070	1080	1090	1100
BnaA09g45590D
	GGTTGCAGAT	ACAGTCACTT	ATAGCAGTCT	CATTCAAGGG	TTTGTCAAT	CAGGAAACT	TAAAGTCGCC	AAAGAACTCT	TCCAAGAGAT	GGTCTCTCAA
	1110	1120	1130	1140	1150	1160	1170	1180	1190	1200
BnaA09g45590D
	GGTGTTCATC	CTAATATTGT	AACCTACAAA	ATTTTGATGG	ATGGAGTCGA	TGAAGCTTGG	GATCTATTTT	GTAGCCTCCC	TCTCAAAGAA	GTGAAGCCTG
	1210	1220	1230	1240	1250	1260	1270	1280	1290	1300
BnaA09g45590D
	ATGTCAAAAC	ATACACCATA	ATGATTGGAG	GACTATGTAA	GAAAGGCTCG	ATGCTCTAAAG	CGGGCCTGTT	ATTTAAGAAA	ATGGGAGAGG	ATGGGATTGC
	1310	1320	1330	1340	1350	1360	1370	1380	1390	1400
BnaA09g45590D
	ACCAGATGAT	TGTACATGGA	ACACACTAAT	CCGAGCTCAT	CTCCGAGGTG	GTGACTTAGC	AACATCAGCT	AACTTATAG	AAGAAATGAA	GAGTTGTGGG
	1410	1420	1430	1440	1450	1460	1470	1480	1490	
BnaA09g45590D	
	TTCTCTGCAG	ATGCTTCCAC	GATAAAGATT	GTTATGGATA	TGTTATCGGA	TGGTAGAATG	AAGAAAAGCT	TTCTGGATTT	GCTTCTTAG	

Figure S3. Sequence alignment of single functional *RFL11* gene copy in Westar. The target sequences are shown in underline with the PAM highlighted in red.

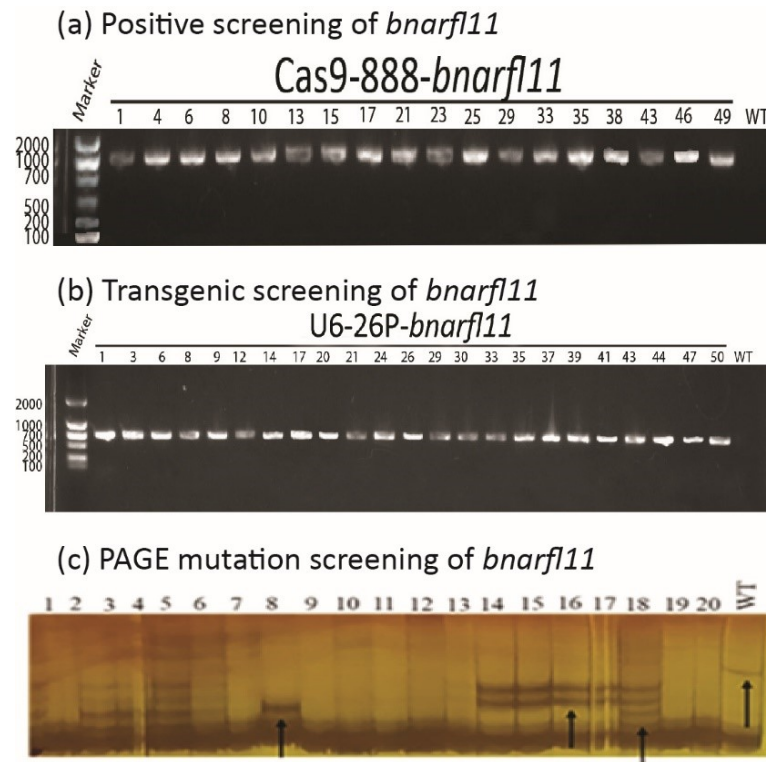


Figure. S4 Positive, Transgenic and mutation screening in *bnarf11*. (a) Cas9 specific primers used for positive screening of *bnarf11*. (b) Universal primers used for transgenic screening of *bnarf11*. (c) Polyacrylamide gel electrophoresis for mutation detection, numerous plants showed various bands that differ from the band on non-denaturing PAGE gels in wild-type plants (WT), displaying target sites with some mutation.

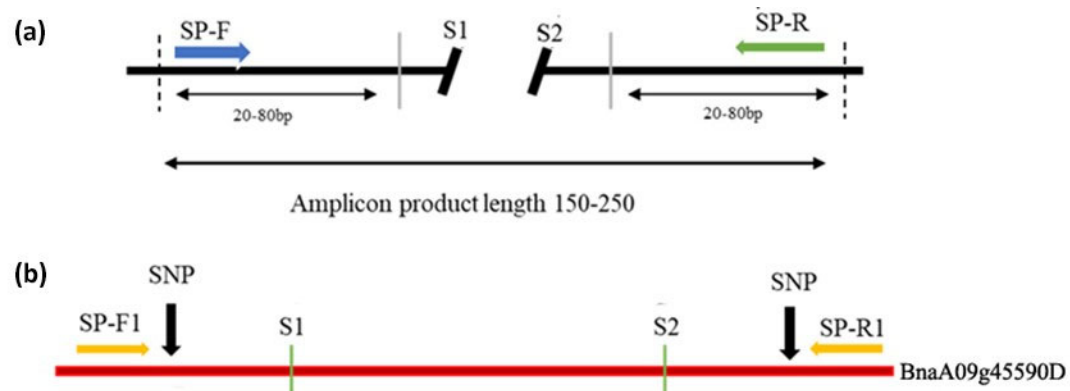


Figure S5 The *BnaRFL11* gene contains a single copy with two target sites S1 and S2. (a) in a single copy, forward and reverse primers covered single target sites, and both primers were designed within 20-80 nucleotides from the target sites. The length of the specific primer pair (SP F/R) was 150-250 bp. **(b)** Site-specific primers were designed for each target based on SNP differences.

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      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL11  TGATAAAATTCGTGGTGTGGATCTAGTTGTGGGTGAAGGGGAAGAAATCATCAAGTCATGAAGATGGGAGTGGAGTTGTTCTTCTTCTGGTTTATTTA 100
RFL6   TGATAAAATTCGTGGTGTGGATGTTTTGTGG-TGAAGGGGAAGAAATCA--AAGTCATGAAGACGGGAGTGGAGTTGTTCTTCTTCTTCTGGTTTATTTA 97
ClustalConsensus ***** * ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** 93

      110      120      130      140      150      160      170      180      190      200
RFL11  GATTTGAAAAAAACAGACAATGTTGTGTCGGAGATTGGTACGTGAGCCTCGTATCCCTCTGAGTCTTGGAGGTACTCTTACAGTTACATGCTTTGT--TA 198
RFL6   GATTTGAAAAAAACAAACAATGTTGTGTCGGAGATTGGTGTCTGCTCTATCCCCCTGAGTCCGTGTTGGTACTCTTGCAACTCCATCTCTCATCTTA 197
ClustalConsensus ***** ***** ***** * * * ***** ***** * * ***** * * * * * * * * * * * * * * * * * * * * * * 175

      210      220      230      240      250      260      270      280      290      300
RFL11  CAAAGAGAGATTGCGAAGTGGGATCGTCGATATTAAAGAAAGATGAAGCTGTGCTCTGTTTCAAACATGATTGTCTCGTCCTCTTCTTACG--CAA 295
RFL6   CAAAGAGAGACTGAGAAGTGGTCTGGTCGATATCAAGAAGAAGGATGCTGTAGCTCTGTTTCAGTCCATGATTAGGTCTCGTCCTCTTCTTACGAAGCAA 297
ClustalConsensus ***** * * ***** * ***** * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * 256

      310      320      330      340      350      360      370      380      390      400
RFL11  ATGGAAATGAATGGGATTGCGCATGACCTCTACACGCTGAGTATTGTATCAACTGCTTCTGCCGTCCGCCGAAACTCGTCACATTCTCAACTTTGATTA 395
RFL6   ATGGAACTGCAAGGGATTGCACATAGCATTTACACGCTGAGTATTATGATCAATTGCTTCTGCCGCTCCAGAACTCGTCACATTCTCAACTTTGATCA 397
ClustalConsensus ***** * * * ***** * * * ***** ***** * * * ***** ***** * * * ***** ***** * * * ***** ***** 340

      410      420      430      440      450      460      470      480      490      500
RFL11  ACGGTTTATGTCTGGGTCGAGTCTCTGAAGCTGTGGAGTTAGTTGATCGTATGGTGGAAATGAAGGTTATTCCAAATCTCATCATCTTAACTCTTGT 495
RFL6   ACGGTTTATGTCTGGGTAGATTTCCAAAGCTGTGGAGTTAGTTGATCATATGGTAGATATGAAGGTTATTCCAAATCTCATCATCTTAACTCTTGT 497
ClustalConsensus ***** ***** * * ***** ***** ***** * * * ***** ***** ***** ***** ***** ***** ***** ***** 432

      510      520      530      540      550      560      570      580      590      600
RFL11  CAATGGACTTTGTCTCCAAGATAGAGTGTCCGAAGCAGTGGCTTTGATAGATCGAATGGTGGCTAATGGATGTCAACCCGATTACCTATGGTCCGATTTT 595
RFL6   CAATGGGCTTTGTCTCCAAGATAGACTCTCTGAAGCAATGTCTTTGATACATCGAATGTTGGCTAATGGGTGCCAACCCGATTACATATGGTCCGGTTTT 597
ClustalConsensus ***** ***** * * ***** * * ***** ***** ***** * * ***** ***** ***** ***** ***** ***** 520

      610      620      630      640      650      660      670      680      690      700
RFL11  GAACAGAATGTGCAATGGAAGATCGAAAGGTCAAGCCTCAAGTATGCGTTGATTGATAGTTTTGTGAAAGAAGGAAAGCTTACTGAGGCTAAAGAGTTG 695
RFL6   GAACAGAATGTGTAATGGAACCTTAGAAAGATCAAGCCTCAAGTATGCTTTGATTGATAGTTTTGTGAAAGAAGGACAGCTTACCGAGGCTAAAAAATTG 697
ClustalConsensus ***** ***** * ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** 610

      710      720      730      740      750      760      770      780      790      800
RFL11  TACAATGAGATGATCGTAAGAGGCTTAGATCCTAATACCATACATATGGTCTCTGATACATGGGCTGTGCATGGAGAATCGCTTAGATGAGGCCAACCC 795
RFL6   TACAATGAGATGATCACAAGAGGCACAGATCCAAATATCATTACATATAACTCTTTGATATATGGGATGTGCATGGACAACGCTAGATGAGGCCAACCC 797
ClustalConsensus ***** ***** ***** * * * ***** ***** ***** ***** ***** ***** ***** ***** ***** ***** 694

      810      820      830      840      850      860      870      880      890      900
RFL11  AGATGTTGGATCTGATGGTTAGCAAGGGATCGCATCCTGATATTTGGACGTATAATATCCTTATAAACGGGTATTGTAAGGCTAAACTGGTTGATGAAGG 895
RFL6   AGATGCTGGATATGATGGTTAGCAAGGGATTCTATCCTGATATTGTGACGTTTAATATCCTTATCAACGGGTACTGTAAGGCTAAACAGGTCGATGAAGG 897

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ClustalConsensus ***** 782
          910      920      930      940      950      960      970      980      990      1000
RFL11      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL6      TATGAGACTTTTCCGCAAAATGCTCTGAGAGGAGTGGTTGCAGATACAGTCACCTATAGCAGTCTCATTCAAGGGTTTTGTCAATCAGGAAAACCTAAA 995
ClustalConsensus ** ***** 871
          1010     1020     1030     1040     1050     1060     1070     1080     1090     1100
RFL11      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL6      GTCGCCAAAGAACTCTCCAAGAGATGGTCTCTCAAGGTGTTTCATCCTAATATTGTAACCTACAAAATTTTGATGGATGGTCGATGAAGCTTGGGATCTA 1095
ClustalConsensus ** ***** 952
          1110     1120     1130     1140     1150     1160     1170     1180     1190     1200
RFL11      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL6      TTTTGTAGCCTCCCTCTCAAAGAAGTGAAGCCTGATGTCAAACATACACCATAATGATTGGAGGACTATGTAAGAAAGGCTCGATGCTTAAAGCGGGCC 1195
ClustalConsensus ** ***** 1039
          1210     1220     1230     1240     1250     1260     1270     1280     1290     1300
RFL11      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL6      TGTATTATAAGAAAATGGGAGAGGATGGGATTGCACCGAGTGATTGTACATGGAACACACTAATCCGAGCTCATCTCCGAGGTGGTGACTTAGCAACATC 1295
ClustalConsensus ***** 1120
          1310     1320     1330     1340     1350     1360     1370     1380     1390     1400
RFL11      ....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|....|
RFL6      AGCTAAACTTATAGAAGAAATGAAGAGTTGTGGGTTCTCTGCAGATGCTTCACATAAAGATTGTTATGGATATGTTATGGATGGTAGAATGAAGAAAAGC 1395
ClustalConsensus ***** 1217
          1410     1420
RFL11      ....|....|....|....|
RFL6      TTTCTGGATTGCTTTCTTAG 1415
ClustalConsensus ***** 1237
          1417
RFL6      TTTCTGGATTGCTTTCTTAG 1417

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Figure S6. Sequence alignment of *BnaRFL11* with *BnaRFL6*.