Supplementary Material



Genetic Transformation of Tobacco Serine Acetyltransferase 4 (NtSAT4) gene in Brassica napus L.

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Received | August 27, 2019; Accepted | October 18, 2019; Published | November 25, 2019 *Correspondence | Muhammad Sayyar Khan, The University of Agriculture Peshawar; Email: sayyar@aup.edu.pk Citation | Rajab, H., M.S. Khan, S.H. Shah and S.M.A. Shah. 2019. Genetic transformation of tobacco serine acetyltransferase 4 (NtSAT4) gene in *Brassica napus* L. Sarhad Journal of Agriculture, 35(4): 1224-1233. DOI | http://dx.doi.org/10.17582/journal.sja/2019/35.4.1224.1233 Keywords | Brassica napus, Transformation, Serine acetyltransferase, Cysteine, Agrobacterium

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		883	890		900		910		920		930		940		950		960	1	970		980		990		1000		1015
» pBinAR_NtSAT4	(883)	ACTACO	AAATTE	TTCTG	TCGA	CCCAGI	TTTCCC	TGAGC	TIGTIT	CTTGC	GCACO	CATTO	CTGA	AAAG	AACTC	CAAAA	TCGG	CGTAA	CGAAG	AGGAA	GACGA:	TTGTO	GCTA	AAAATC	AAAGA	TGAGGC	TAGATCA
« Seg. NtSAT4 Rev																											TAGATCA
																											TAGATCA
																											TAGATCA
Conscribus	(000)	ACIACO	111AAAA	+ +	ICGA	CUCAG.	111000	TGHOC	119111	+	GUALL	CALIC	CIGA	INANO.	AACIC	CAAAA	1000.	+ COTHA	COARO	AGGAA	JACOA.	11010	JUCIAL	IMARA I G	MAAGA	TGAGGC	INGALCE
		1016		10	30	10	040	10	50	1060		107	2	10	080	10	n90	1	100	11	10	11	20	11	30		1148
> pBinAR_NtSAT4																		· · · · · · · · · · · · · · · · · · ·		A							
« Seq. NtSAT4 Rev	(124)	AGACAI	IGAIC	AAGAA	CLLA.	11116	TOTACI	TACIA	CATAAL	TTCAA	TOTTO	GUILA	TGAL	TOTA	IGGAA.	AGGGC	TTTA	SUIAAI	CAILI	TICAA	GAAA.	TCAGI	AAIIC	AAGIC	TTOOL	AGLAGE	ACTINGI
Seq. NtSAT4 355																											
Consensus	(1016)	AGACAI	TGATC	AAGAA	CCCA.	TITIG	ICTACT	TACTA	CATAAC	TTCAA	TCTTS	GCTCA	TGAI	TCTA	IGGAA	AGGGC	TTTA	SCTAAT	CATCT	TTCAA	IGAAA	TGAGI	AATTC	CAAGIC	TICCI	AGCAGC	ACTITGI
		1149		1160		1170		1190	1	190		200		1210		1220		1920		1240		1250		1260		1270	1281
				1100	-			1100		1.50		2.00		1210		1220		12.50		,12-10		16.00		1200		1210	
> pBinAR_NtSAT4	(1149)	TATGAT	CTTTT	CCTAG	GGGT	GCTCA	CAGAGG	ATTGC	TCACAA	GATAT	AATTA	LAAGCI	GTTA	TAGC	TGATT	TAAGG	GCAG:	TTAAAG.	AAAGG	GACCC.	AGCTT	STATT?	GTTAT	IGTACA	CTGTI	TCTTGA	ATTTTAA
« Seq. NtSAT4 Rev																					AGCIT	TATT	GITAI	GTACA	CTGTT	TCTTGA	ATTITAA
» Seq. NtSAT4 35S																					AGCIT	TATTA	GTIAI	IGIACA	CIGII	ICTIGA	ATITIAA
Consensus	(1149)	TATGAI	CITIT	CCTAG	GGGT	GCTCA	CAGAGG	ATTGC	TCACAA	GATAT	AATTA	AAGCI	GITA	TAGC	TGATT	TAAGG	GCAG.	TTAAAG	AAAGG	GACCC	AGCTT	STATIA	GTTAI	GTACA	CIGII	TCTTGA	ATTITAA
		1000	100		100		1.7.1		1 3 9 9		1000				1.00			100			1.70		170	~	140		1414
		1282	1290		130		1310		1320		1330		1340		135			60	137		138		1390		1400		
* pBinAR_NtSAT4																											
« Seq. NtSAT4 Rev																											
» Seq. NtSAT4 35S																											
Consensus	(1282)	AAGGGI	TTTTTA	GCATG	TCAA	GCTCA	TAGGAT	TGCAC	ATAAAT	TATGG	TCAAA	TGGTA	GGCA	AATT	ITGGC.	ACTIT	TGAT	ACAAAA	CAGGG	TATCT	GAAGT	TTTTGG	TGTCG	JACATA	CATCO	TGGTGC	TAAAATI
		1415 1	1420		430	1.1	1440	1	450	14	en.	14	70		1490		1490		1500		1510		520	1	530		1547
						4		1	1.50						1-100				1.500								
» pBinAR_NtSAT4																											
« Seq. NtSAT4 Rev																											
➤ Seq. NtSAT4 35S																											
Consensus	(1415)	TGGTAA	AGGGA	TTTTA	CITG	ATCATO	BCTACT	GGAGT	TGTCGT	TGGTG	AAACI	GCTGI	GATT	GGAA	ATAAT	GIGIC	AATTI	ITGCAT.	AACGT	GACAT	TGGGT	JGAACI	GGCAA	ARATAT	CIGGO	GATAGA	CATCCTA
		1549							2					1.000										1660			1681
				1560		,1570		1580	-	590	1	600		1610		1620		10.30		1040		1600				1670	
» pBinAR_NtSAT4																											
« Seq. NtSAT4 Rev					TTTA	ATTGG	IGCIGG	AACTT	GTGTTC	TTGGA	AATG	TATA	TTGA	AGAT	GGAGC	TAAAA	TTGG	GCAGG	GTCCG	TGGTG	CTGAA	JAAAGI	TCCGG	SCGAGG	ACTAC	CGCCGT	TGGGAAI
➤ Seq. NtSAT4 355																											
Consensus	(1549)	AAATTG	GTGAT	GGGGT	TTTA	ATTGG	IGCIGG	AACTT	GIGITC	TTGGA	AATGI	TATAP	TTGA	AGAT	GGAGC	TAAAA	TTGG	GCAGG	GTCCG	TGGTG	CTGAA	BAAAGI	TCCGG	SCGAGO	ACTAC	CGCCGT	TGGGAAI
					+				+	+	4	+				+			+			+	+		+		+
		1683	1690		1700		1710		1720		1730		1740		1750		176		1770		1780		1790				
» pBinAR_NtSAT4															GGACC	ATACA	TATG	AGTGGT	CTGAT	TATGT	AATTTA	AGCTCO	FACCT				
« Seq. NtSAT4 Rev	(801)	CCGGCG	AGGTT	GCTCG	GAGG	GAAGG	AAAATC	CAAAG	AAACTT	GATAA	GAATO	CTAGI	TIGA	CC													
➤ Seq. NtSAT4 355																											
Consensus	(1683)	CCGGCG	AGGTT	GCTCG	GAGG	GAAGG	AAAATC	CAAAG	AAACTT	GATAA	GATTO	CTAGI	TTGA	CCAT	GGACC.	ATACA	TATG	AGTGGT	CTGAT	TATGT	AATTTA	GCTCO	JACCT				
											+																

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	359	370	380	390	400	410	420	430	440	450	460	470	480	491
	(44) CCATGTO (359) C-ATGTO	CTATAACTAC CTATAACTAC	GTTTAAATTCT GTT-AAATTCT	GTCGACCCAAC GTCGACCCA-C	TTTCCCTGA	SCTIGTITCITG SCTIGTITCITG SCTIGTITCITG SCTIGTITCITG	CGCACCCAT	TCCTGAAAAG	AACTCCAAA AACTCCAAA	TTCGGTCGTA ATCGGTCGTA	ACGAAGAGG	AAGACGATTT	GTGGCTAAAAA GTGGCTAAAAA	ATGAAA
	492	500	510	520	530	540 5	50	560 5	570	580	590	600	610	624
 » pBIN_AR_TKTP « Seq. TKTPNtSA » Seq. TKTPNtSA Conscisus 	(177) GATGAGO (492) GATGAGO	GCTAGATCAG GCTAGATCAG	ACATTGATCAA ACATTGATCAA	AGAACCCATTI -GAACCCATTI	TGTCTACTT	ACTACATAACTT	CAATCTTG -CAATCTTG	GCTCATGATTC GCTCATGATTC	TATGGAAAG	GGCTTTAGCT	AATCATCTT	TCAATGAAAT TCAATGAAAT	IGAGTAATTCZ IGAGTAATTCZ	AAGTCT AAGTCT
	625 63	30 6	40 650	660	670	680	690	700	710	720	730	740		757
 > pBIN_AR_TKTP « Seq. TKTPNtSA > Seq. TKTPNtSA Consensus 	(310) TCCTAGE (625) TCCTAGE	CAGCACTTIG CAGCACTTIG	TATGATCTTTT TATGATCTTTT	CCTAGGGGTGG	TCACAGAGG	ATTGCTCACAAG	ATATAATTA ATATAATTA	AAGCTGTTATA AAGCTGTTATA	GCTGATTTA GCTGATTTA	AGGGCAGTTA	AAGAAAGGG	ACCCAGCTTG ACCCAGCTTG	FATT <mark>-</mark> AGTTAT FATT <mark>T</mark> AGTTAT	TGTACA TGTACA
	758	770	780	790	800	810	820	830	840	850	860	870	880	890
 > pBIN_AR_TKTP « Seq. TKTPNtSA > Seq. TKTPNtSA Consensus 	(443) CTGTTT (758) CTGTTT	CTTGAATTT	TAAAGGGTTTT TAAAGGGTTTT	TAGCATGTCAJ TAGCATGTCAJ	GCTCATAGG	ATTGCACATAAA	ITATGGTCA	AATGGTAGGCJ AATGGTAGGCJ	AATTTT <mark>-</mark> GG AAATTTT <mark>T</mark> GG	CACTTTTGAT	ACAAAACAG	G <mark>G</mark> TATCTGAA G-TATCTGAA	GTTTTTGCT0	GT <mark>-</mark> CGA GT <mark>I</mark> CGA
	891	900	910	920	930	940	950	960	970	980	990	1000	1010	1023
 » pBIN_AR_TKTP « Seq. TKTPNISA » Seq. TKTPNISA Consensus 	(576) CATA-CI (891) CATAACI	ATCCTGG <mark>-</mark> TG ATCCTGG <mark>C</mark> TG	CT <mark>A</mark> AAATT <mark>G</mark> GT CT <mark>TAAATTT</mark> GT	AAA <mark>-</mark> GGGATTI AAA <mark>A</mark> GGGATTI	T <mark>-</mark> ACTIGA <mark>-</mark> T <mark>T</mark> ACTIGA <mark>A</mark>	ICATGCTACIG <mark>G</mark>	AGTIGICGI AGTIGICGI	TGGTGAAACTG TGGTGAAACTG	CTGTGATTG	GAAATAATGI	GTCAATTTT	GCATAACGTG.	ACATTGGGTGG	GAACTG
	1024	1030	1040 1	050 10	60 10	70 1080	10	90 110	0 11	10 11	20 1	130 11	40	1156
★ pBIN_AR_TKTP « Seq. TKTPNtSA > Seq. TKTPNtSA Consensus ((666) GCAAAA (709) GCAAAA (-975)	TATCTGGGGA TATCTGGGGA	TAGACATCCTA TAGACATCCTA	AAATTGGTGAT AAATTGG <mark>T</mark> GAT	GGGGGTTTTA	ATTGGTGCTGGA	ACTTGTGII ACTTGTGTT	CTTGGAAATGI CTTGGAAATGI	TATAATTGA TATAATTGA	AGATGGAGCI AGATGGAGCI	AAAATTGGG	GCAGGGTCCG GCAGGGTCCG	IGGIGCIGAAG	GAAAGT
 > pBIN_AR_TKTP < Seq. TKTPNtSA > Seq. TKTPNtSA (Consensus ((841) TTCCGG((-975)	CGAGGACTAC	CGCCGTTGGGA CGCCGTTGGGA	ATCCGG <mark>C</mark> GAGO ATCCGG <mark>G</mark> GAGO	TTGCTCGGA	1210 SGGAAGGAAAAT SGGAAGGAAAAT SGGAAGGAAAAT	CCAAAGAAA	CITGATAAGAI	TCCTAGTTI	GA		GAGTGGTCTG.	ATTATGTAATT	
				+										

310 CCCTGAGCTTOTTT<mark>C</mark>TTGCGCACCCATTCCTGAAA<mark>A</mark>GAACTCCAAAATCGGTCGTAACGAAGAGGAAGACGATTTGTGGCTAAAAATGAAAGATGAGGCTAGATCAGACATTGATCAAGAACCCATT CCCTGAGCTTGTTT<mark>T</mark>TTGCGCACCCATTCCTGAAA<mark>A</mark>GAACTCCAAAATCGGTCGTAACGAAGAGGAGAGAGAGACGATTGTGGCTAAAAATGAAAGATGAGGCTAGATCAGACATTGATCAAGAACCCATT CCCTGAGCTTGTTTCTTGCGCACCCATTCCTGAAA<mark>A</mark>GAACTCCAAAATCGGTCGTAACGAAGAGGAAGACGATTGTGGCTAAAAATGAAAGATGAGGCTAGATCAGACATTGATCAAGAACCCATT CCCTGAGCTTGTTTCTTGCGCACCCATTCCTGAAAAGAACTCCAAAATCGGTCGTAACGAAGAGGAAGACGATTGTGGCTAAAAATGAAAGATGAGGCTAGATCAGACATTGATCAAGAACCCATT 410 CATCTI AATGAAA AGTAAT TATGAT 513 SHMT N IMTNESA... AAAT ATTATGGTCAAAT TGAT 22220200 0220 MTNESA... (646) AGGATIGCACATAAATTATGGTCAAAT (646) AGGATIGCACATAAATTATGGTCAAAT GTAGGCAAATTTTGGCACTTTTGATAC GGTAGGCAAATTTTGGCACTTTTGATAC CARAACAGGGTATCTGAAGTTTTTGCTGTCGACATACATCC CARAACAGGGTATCTGAAGTTTTTGCTGTCGACATACATCC (533) SHMT N (533) SHMTNESA.... (779) CTACTGGAGITGTCGTTGGTGAAACTGCTGTGATTGGAAATAATGTGTCAATT (779) CTACTGGAGTTGTCGTTGGTGAAACTGCTGTGATTGGAAATAATGTGTCAATT GCATAACGTGACATTGGGTGGAACI GCAAAATAT GGATAGACAT TAAAAT GTGAT GAAATAATGTGTCAATTTTGCATAACGTGACATTGGGT AAAATATCT TAAAATTGGTGATO 912 1000 1010 1020 1030 1044 HMTNESA. (912) 1070 1080 1090 1100 1110 1120 ATTCCTAGTTTGACCATGGACCATACATATGAGTGGTCTGATTATGTAATTTAG (799) GAAAATCCAAAGAAACTTG (495) (4959) (1045) GAAAATCCAAAGAAACTTGATAAGATTCCTAGTTTGACCAIGGACCATACATATGAGIGGICTGATTATGTAATTTAG IMINISA.

Supplementary Figure 1: Cloning and sequencing analysis of NtSAT4 gene overexpression constructs; Sequencing result using 35S forward and NtSAT4 reverse primers for the (A) Cytosolic targeted NtSAT4 (B) Plastid targeted NtSAT4 (C) Mitochondrial targeted NtSAT4 sequences.