

Supplementary Materials:

Genome-Wide Identification, Evolutionary and Expression Analyses of the *GALACTINOL SYNTHASE* Gene Family in Rapeseed and Tobacco

Yonghai Fan ^{1,2,†}, Mengna Yu ^{1,2,†}, Miao Liu ^{1,2}, Rui Zhang ^{1,2}, Wei Sun ^{1,2}, Mingchao Qian ^{1,2}, Huichun Duan ^{1,2}, Wei Chang ^{1,2}, Jinqi Ma ^{1,2}, Cunmin Qu ^{1,2}, Kai Zhang ^{1,*}, Bo Lei ^{3,4,*} and Kun Lu ^{1,2,*}

¹ College of Agronomy and Biotechnology, Southwest University, Chongqing 400715, China; fyh1212@email.swu.edu.cn (Y.F.); yumengna1024@163.com (M.Y.); monky1117@email.swu.edu.cn (M.L.); z2247319@email.swu.edu.cn (R.Z.); reginasw@163.com (W.S.); absorbessence@163.com (M.Q.); dhcspring@163.com (H.D.); cw12345678@email.swu.edu.cn (W.C.); mj2014@email.swu.edu.cn (J.M.); lion4302@163.com (C.Q.)

² Academy of Agricultural Sciences, Southwest University, Chongqing 400715, China

³ Key Laboratory of Molecular Genetics, China National Tobacco Corporation, Guizhou Academy of Tobacco Science, Guiyang 550081, China

⁴ Upland Flue-Cured Tobacco Quality and Ecology Key Laboratory of China Tobacco, Guizhou Academy of Tobacco Science, Guiyang 550081, China

* Correspondence: zhangkai2010s@163.com (K.Z.); leibo_1981@163.com (B.L.); drlukun@swu.edu.cn (K.L.); Tel./Fax: +86-23-6825-1264 (K.L.)

† These authors contributed equally to this work.

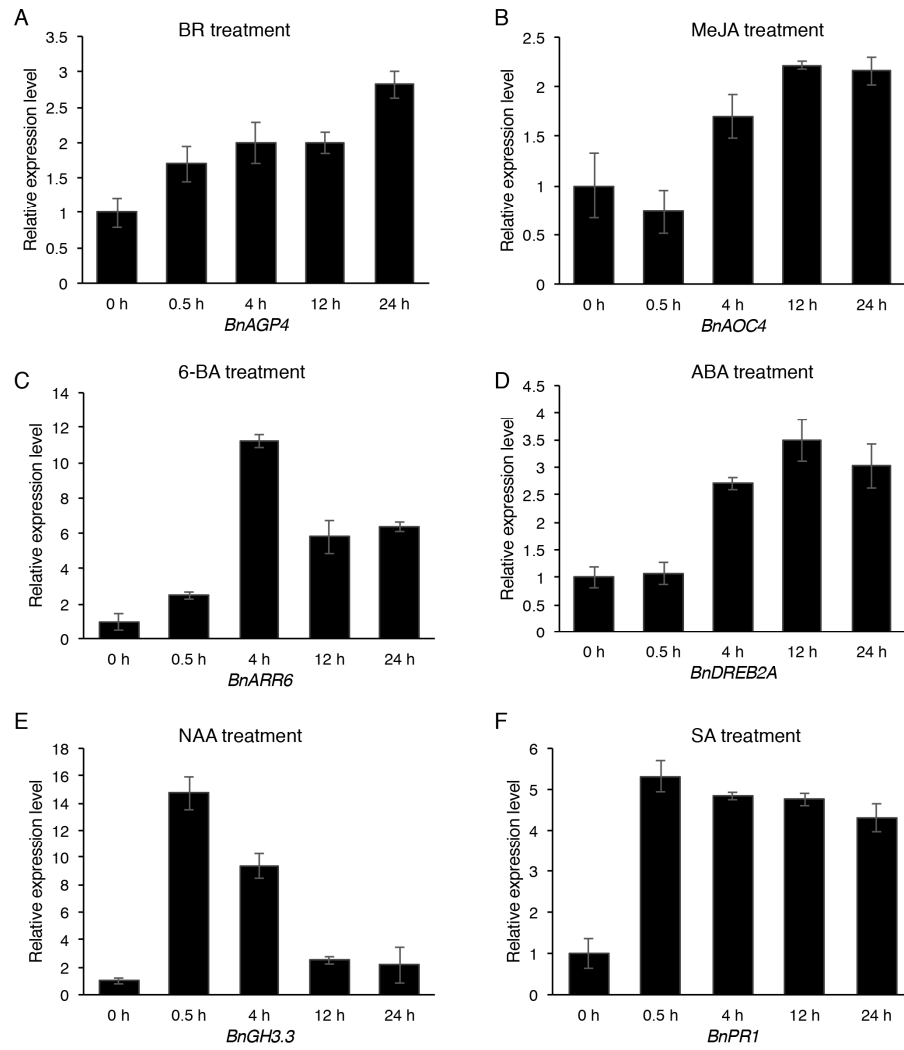


Figure S1. Expression patterns of six hormone inducible marker genes exposed to corresponding hormone treatments.

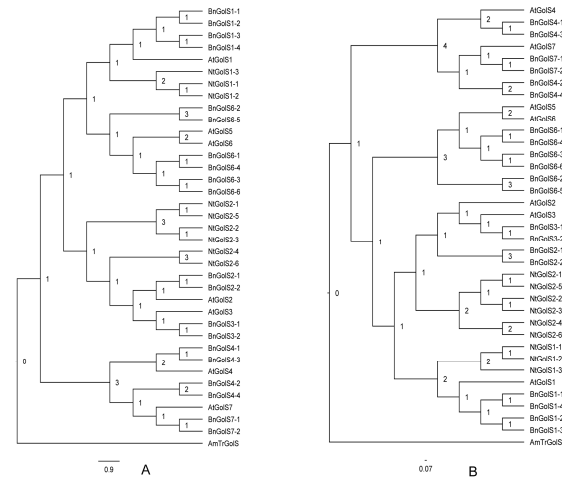


Figure S2. Phylogenetic trees of *GolS* genes in *A. thaliana*, *B. napus* and *N. tabacum*. Proteins sequences of AtGolS, BnGolS and NtGolS were aligned by using the MUSCLE program. The NJ (A) and BI (B) trees were constructed by MGEA7 with bootstrap analysis (1000 replicates) and MrBayes3.2.6. All the trees were displayed using FigTree v1.4.0.

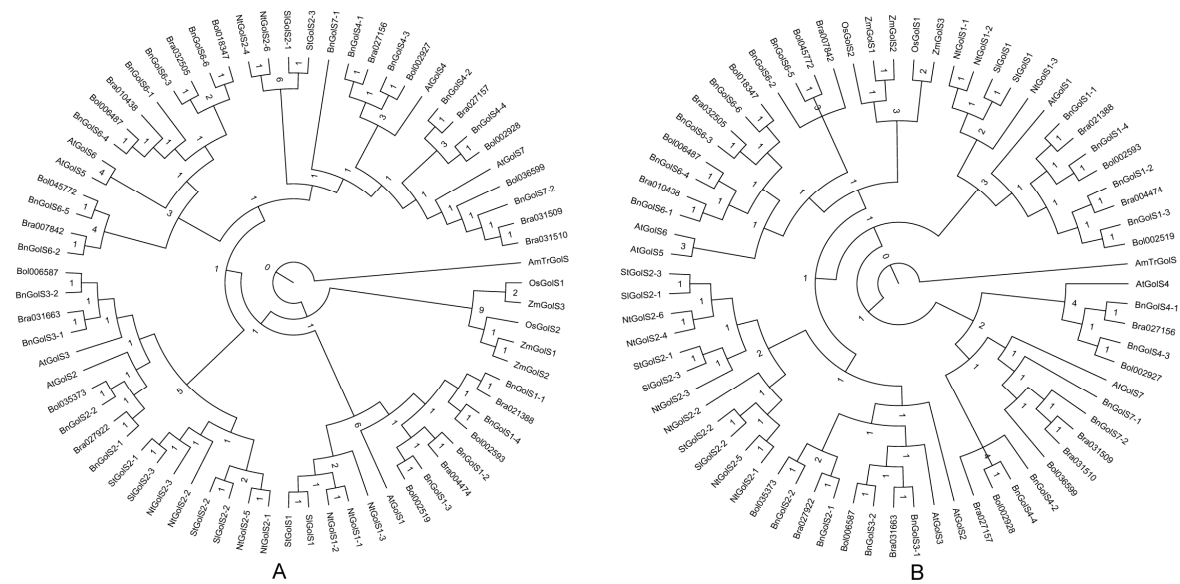


Figure S3. Phylogenetic trees of GolS proteins in plants. The NJ (A) and BI (B) trees were constructed by MGEA7 with bootstrap analysis (1000 replicates) and MrBayes3.2.6, using the GolS proteins in *A. thaliana*, *B. napus*, *B. rapa*, *B. oleracea*, *N. tabacum*, *S. lycopersicum*, *S. tuberosum*, *O. sativa*, *Z. mays* (GolS in *A. trichopoda* as an outgroup). The trees were displayed with FigTree v1.4.0. At, *A. thaliana*; Bn, *B. napus*; Bra, *B. rapa*; Bol, *B. oleracea*; Nt, *N. tabacum*; Sl, *S. lycopersicum*; St, *S. tuberosum*; Os, *O. sativa*; Zm, *Z. mays*; AmTr, *A. trichopoda*.

Table S1. Model-fit Ranking according to all measures

	AIC1	Model	Ln	AIC2	Model	Ln	BIC	Model	Ln
1	19004.56	JTT+G	-9362.28	19424.56	JTT+G	-9362.28	19536.85	JTT+G	-9362.28
2	19019.01	JTT+G+F	-9350.50	19446.56	JTT+I+G	-9370.78	19559.65	JTT+I+G	-9370.78
3	19036.22	JTT+I+G+F	-9358.11	19503.90	WAG+G	-9401.95	19623.54	JTT+G+F	-9350.50
4	19083.90	WAG+G	-9401.95	19516.22	JTT+I+G+F	-9358.11	19637.73	WAG+I+G	-9409.81
5	19101.63	WAG+I+G	-9409.81	19524.63	WAG+I+G	-9409.81	19644.56	JTT+I+G+F	-9358.11
6	19142.89	WAG+G+F	-9412.44	19611.58	Dayhoff+G	-9455.79	19723.87	Dayhoff+G	-9455.79
7	19159.58	WAG+I+G+F	-9419.79	19616.67	LG+G	-9458.33	19728.96	LG+G	-9458.33
8	19191.58	Dayhoff+G	-9455.79	19619.89	WAG+G+F	-9412.44	19736.13	DCMut+G	-9461.91
9	19196.67	LG+G	-9458.33	19623.83	DCMut+G	-9461.91	19744.02	Dayhoff+I+G	-9462.96
10	19203.83	DCMut+G	-9461.91	19630.92	Dayhoff+I+G	-9462.96	19747.42	WAG+G+F	-9412.44
11	19207.92	Dayhoff+I+G	-9462.96	19637.32	LG+I+G	-9466.16	19750.42	LG+I+G	-9466.16
12	19207.93	LG+G+F	-9444.96	19639.58	WAG+I+G+F	-9419.79	19755.05	DCMut+I+G	-9468.48
13	19209.92	Dayhoff+G+F	-9445.96	19641.96	DCMut+I+G	-9468.48	19767.92	WAG+I+G+F	-9419.79
14	19214.32	LG+I+G	-9466.16	19684.93	LG+G+F	-9444.96	19812.47	LG+G+F	-9444.96
15	19218.96	DCMut+I+G	-9468.48	19686.92	Dayhoff+G+F	-9445.96	19814.45	Dayhoff+G+F	-9445.96
16	19221.44	DCMut+G+F	-9451.72	19698.44	DCMut+G+F	-9451.72	19820.16	CPREV+G	-9503.93
17	19222.62	LG+I+G+F	-9451.31	19702.62	LG+I+G+F	-9451.31	19825.97	DCMut+G+F	-9451.72
18	19231.77	Dayhoff+I+G+F	-9455.88	19707.86	CPREV+G	-9503.93	19830.96	LG+I+G+F	-9451.31
19	19238.07	CPREV+G+F	-9460.03	19711.77	Dayhoff+I+G+F	-9455.88	19840.11	Dayhoff+I+G+F	-9455.88
20	19242.41	DCMut+I+G+F	-9461.20	19715.07	CPREV+G+F	-9460.03	19842.60	CPREV+G+F	-9460.03
21	19259.67	CPREV+I+G+F	-9469.83	19722.41	DCMut+I+G+F	-9461.20	19846.22	CPREV+I+G	-9514.06
22	19287.86	CPREV+G	-9503.93	19733.12	CPREV+I+G	-9514.06	19850.75	DCMut+I+G+F	-9461.20
23	19310.12	CPREV+I+G	-9514.06	19739.67	CPREV+I+G+F	-9469.83	19868.01	CPREV+I+G+F	-9469.83
24	19332.66	RtREV+G+F	-9507.33	19784.53	RtREV+G	-9542.26	19896.82	RtREV+G	-9542.26
25	19346.51	RtREV+I+G+F	-9513.25	19799.59	RtREV+I+G	-9547.29	19912.69	RtREV+I+G	-9547.29
26	19356.22	VT+G+F	-9519.11	19809.66	RtREV+G+F	-9507.33	19937.20	RtREV+G+F	-9507.33
27	19363.22	VT+I+G+F	-9521.61	19825.42	VT+G	-9562.71	19937.71	VT+G	-9562.71
28	19364.53	RtREV+G	-9542.26	19826.51	RtREV+I+G+F	-9513.25	19949.54	VT+I+G	-9565.72
29	19376.59	RtREV+I+G	-9547.29	19833.22	VT+G+F	-9519.11	19954.60	BLOSUM62+G	-9571.15
30	19405.42	VT+G	-9562.71	19836.44	VT+I+G	-9565.72	19954.85	RtREV+I+G+F	-9513.25
31	19413.44	VT+I+G	-9565.72	19842.30	BLOSUM62+G	-9571.15	19960.76	VT+G+F	-9519.11

32	19422.30	BLOSUM62+G	-9571.15	19843.22	VT+I+G+F	-9521.61	19968.49	BLOSUM62+I+G	-9575.19
33	19432.39	BLOSUM62+I+G	-9575.19	19855.39	BLOSUM62+I+G	-9575.19	19971.56	VT+I+G+F	-9521.61
34	19450.88	MTREV24+G+F	-9566.44	19927.88	MTREV24+G+F	-9566.44	20055.42	MTREV24+G+F	-9566.44
35	19456.05	BLOSUM62+G+F	-9569.02	19933.05	BLOSUM62+G+F	-9569.02	20060.59	BLOSUM62+G+F	-9569.02
36	19461.82	MTREV24+I+G+F	-9570.91	19941.82	MTREV24+I+G+F	-9570.91	20070.16	MTREV24+I+G+F	-9570.91
37	19466.09	BLOSUM62+I+G+F	-9573.04	19946.09	BLOSUM62+I+G+F	-9573.04	20074.42	BLOSUM62+I+G+F	-9573.04
38	19831.16	JTT+I	-9775.58	20251.16	JTT+I	-9775.58	20363.46	JTT+I	-9775.58
39	19847.61	WAG+I	-9783.80	20267.61	WAG+I	-9783.80	20379.91	WAG+I	-9783.80
40	19849.83	JTT+I+F	-9765.91	20326.83	JTT+I+F	-9765.91	20454.37	JTT+I+F	-9765.91
41	19923.41	WAG+I+F	-9802.70	20400.41	WAG+I+F	-9802.70	20527.95	WAG+I+F	-9802.70
42	19932.91	MtMam+G+F	-9807.45	20409.91	MtMam+G+F	-9807.45	20537.44	MtMam+G+F	-9807.45
43	19982.74	MtMam+I+G+F	-9831.37	20427.42	Dayhoff+I	-9863.71	20539.72	Dayhoff+I	-9863.71
44	20007.21	MtArt+G+F	-9844.60	20440.96	DCMut+I	-9870.48	20553.25	DCMut+I	-9870.48
45	20007.42	Dayhoff+I	-9863.71	20462.74	MtMam+I+G+F	-9831.37	20591.08	MtMam+I+G+F	-9831.37
46	20014.05	CPREV+I+F	-9848.02	20484.21	MtArt+G+F	-9844.60	20605.44	CPREV+I	-9896.57
47	20020.96	DCMut+I	-9870.48	20491.05	CPREV+I+F	-9848.02	20610.67	LG+I	-9899.18
48	20030.05	MtArt+I+G+F	-9855.02	20493.14	CPREV+I	-9896.57	20611.75	MtArt+G+F	-9844.60
49	20044.86	Dayhoff+I+F	-9863.43	20498.37	LG+I	-9899.18	20618.58	CPREV+I+F	-9848.02
50	20058.29	DCMut+I+F	-9870.14	20510.05	MtArt+I+G+F	-9855.02	20638.39	MtArt+I+G+F	-9855.02
51	20073.14	CPREV+I	-9896.57	20521.86	Dayhoff+I+F	-9863.43	20649.40	Dayhoff+I+F	-9863.43
52	20078.37	LG+I	-9899.18	20535.29	DCMut+I+F	-9870.14	20662.83	DCMut+I+F	-9870.14
53	20096.28	LG+I+F	-9889.14	20552.55	MTREV24+G	-9926.27	20664.84	MTREV24+G	-9926.27
54	20132.55	MTREV24+G	-9926.27	20573.28	LG+I+F	-9889.14	20700.82	LG+I+F	-9889.14
55	20173.37	VT+I+F	-9927.68	20598.97	MTREV24+I+G	-9946.98	20712.07	MTREV24+I+G	-9946.98
56	20175.97	MTREV24+I+G	-9946.98	20620.22	BLOSUM62+I	-9960.11	20732.52	BLOSUM62+I	-9960.11
57	20200.2	BLOSUM62+I	-9960.11	20649.61	RtREV+I	-9974.80	20761.90	RtREV+I	-9960.11
58	20200.22	BLOSUM62+I	-9960.11	20649.61	RtREV+I	-9974.80	20761.90	RtREV+I	-9974.80
59	20229.61	RtREV+I	-9974.80	20650.37	VT+I+F	-9927.68	20765.43	VT+I	-9976.57
60	20232.07	RtREV+I+F	-9957.03	20653.14	VT+I	-9976.57	20769.29	JTT	-9981.39
61	20233.14	VT+I	-9976.57	20657.79	JTT	-9981.39	20777.91	VT+I+F	-9927.68
62	20233.45	BLOSUM62+I+F	-9957.72	20674.64	WAG	-9989.82	20786.13	WAG	-9989.82
63	20240.79	JTT	-9981.39	20709.07	RtREV+I+F	-9957.03	20836.61	RtREV+I+F	-9957.03
64	20257.64	WAG	-9989.82	20710.45	BLOSUM62+I+F	-9957.72	20837.99	BLOSUM62+I+F	-9957.72
65	20271.21	JTT+F	-9977.60	20745.21	JTT+F	-9977.60	20871.94	JTT+F	-9977.60
66	20349.25	WAG+F	-10016.62	20823.25	WAG+F	-10016.6	20943.43	Dayhoff	-10068.4
67	20394.38	MTREV24+I+F	-10038.19	20831.94	Dayhoff	-10068.4	20949.98	WAG+F	-10016.6

68	20414.94	Dayhoff	-10068.47	20846.37	DCMut	-10075.6	20957.87	DCMut	-10075.6
69	20418.71	CPREV+F	-10051.35	20871.38	MTREV24+I+F	-10038.1	20998.82	CPREV	-10096.1
70	20429.37	DCMut	-10075.68	20887.33	CPREV	-10096.1	20998.92	MTREV24+I+F	-10038.1
71	20464.80	Dayhoff+F	-10074.40	20892.71	CPREV+F	-10051.3	21019.45	CPREV+F	-10051.1
72	20470.33	CPREV	-10096.16	20917.69	LG	-10111.3	21029.19	LG	-10111.3
73	20479.42	DCMut+F	-10081.71	20938.80	Dayhoff+F	-10074.4	21065.53	Dayhoff+F	-10074.4
74	20500.69	LG	-10111.34	20953.42	DCMut+F	-10081.7	21080.16	DCMut+F	-10081.7
75	20542.69	LG+F	-10113.34	21016.69	LG+F	-10113.3	-10113.34	VT	-10163.8
76	20560.52	VT+F	-10122.26	21022.70	VT	-10163.8	21142.28	BLOSUM62	-10167.8
77	20605.70	VT	-10163.85	21030.78	BLOSUM62	-10167.8	21142.28	BLOSUM62	-10167.8
78	20613.78	BLOSUM62	-10167.89	21030.78	BLOSUM62	-10167.8	21143.43	LG+F	-10113.3
79	20613.78	BLOSUM62	-10167.89	21034.52	VT+F	-10122.2	21152.76	MtMam+G	-10170.2
80	20620.47	MtMam+G	-10170.23	21040.47	MtMam+G	-10170.2	21161.26	VT+F	-10122.2
81	20653.37	RtREV	-10187.68	21070.37	RtREV	-10187.6	21181.86	RtREV	-10187.6
82	20658.90	BLOSUM62+F	-10171.45	-10171.4	MtArt+G	-10203.4	21219.15	MtArt+G	-10203.4
83	20685.89	RtREV+F	-10184.94	21132.90	BLOSUM62+F	-10171.4	21259.63	BLOSUM62+F	-10171.4
84	20686.86	MtArt+G	-10203.43	21159.89	RtREV+F	-10184.9	21275.82	MtMam+I+G	-10228.8
85	20739.72	MtMam+I+G	-10228.86	21162.72	MtMam+I+G	-10228.8	21286.63	RtREV+F	-10184.9
86	20788.85	MtArt+I+G	-10253.42	21211.85	MtArt+I+G	-10253.4	21324.95	MtArt+I+G	-10253.4
87	20797.62	MTREV24+F	10240.81	21271.62	MTREV24+F	-10240.8	21398.35	MTREV24+F	-10240.8
88	21246.87	MtArt+I+F	-10464.43	21709.76	MTREV24+I	-10504.8	21822.06	MTREV24+I	-10504.8
89	21289.76	MTREV24+I	-10504.88	21723.87	MtArt+I+F	-10464.4	21851.41	MtArt+I+F	-10464.4
90	21307.81	MtMam+I+F	-10494.90	21784.81	MtMam+I+F	-10494.9	21912.34	MtMam+I+F	-10494.9
91	21672.25	MTREV24	-10697.12	22089.25	MTREV24	-10697.1	22200.75	MTREV24	-10697.1
92	21685.63	MtArt+F	-10684.81	22159.63	MtArt+F	-10684.8	22286.3 6	MtArt+F	-10684.8
93	21745.35	MtMam+F	-10714.67	22219.35	MtMam+F	-10714.6	22346.09	MtMam+F	-10714.6
94	22302.66	MtArt+I	-11011.33	22722.66	MtArt+I	-11011.3	22834.95	MtArt+I	-11011.3
95	22344.05	MtMam+I	-11032.02	22764.05	MtMam+I	-11032.0	22876.35	MtMam+I	-11032.0
96	22754.74	MtMam	-11238.37	23171.74	MtMam	-11238.3	23283.23	MtMam	-11238.3

The optimal model selection was based on the lowest values of Akaike Information Criterion (AIC) and Bayesian Information Criterion (BIC).

Table S2. Comparison of the topologies of NJ, ML and BI trees

	item	obs	au	np	Bp	pp	kh	sh	wkh	wsh
large tree	ML	-8.7	0.741	0.725	0.725	1.000	0.848	0.917	0.848	0.946
	BI	8.7	0.261	0.275	0.275	0.152	0.152	0.344	0.152	0.271
	NJ	94.9	7e-005	4e-005	0	3e-050	5e-005	5e-005	5e-005	5e-005
small tree	ML	-0.0	0.522	0.502	0.509	0.500	0.848	0.847	0.848	0.946
	BI	0.0	0.513	0.495	0.447	0.500	0.152	0.344	0.152	0.268
	NJ	44.8	0.002	0.002	0.002	2e-020	5e-005	5e-005	5e-005	5e-005

Table S3. The primer list of 16 *BnGolS*, 9 *NtGolS* and 6 external control genes

Gene	Gene ID	Forward Primer (5'-3')	Reverse Primer (5'-3')
<i>BnGolS1-1</i>	BnaA04g26930D	CTTCGAGAAAATCTACAAGCCG	GATAAACGGCTTCAGATTACCC
<i>BnGolS1-2</i>	BnaA05g00720D	CACAAACCACCACAGCTAAATC	CTAAACCAACGACTCCTTTAC
<i>BnGolS1-3</i>	BnaC04g00330D	TCCTCTAAAATCACGTGCTTCT	CTAAACCAACGACTCCTTTAC
<i>BnGolS1-4</i>	BnaC04g51460D	GCATACGTGACATTTCTAGCTG	GGATAAGCGGATTTGACTTTCC
<i>BnGolS2-1</i>	BnaA09g15290D	CGACCATAAGAAGCAGTTTGTC	TCGCAAACCAGTAAAACGAATC
<i>BnGolS2-2</i>	BnaCnng63310D	CGACCATAAGAAGCAGTTTGTC	AATCTCAAGCCGTATCGTACAT
<i>BnGolS3-1</i>	BnaA09g48480D	AGGACTAAGGAAAGCTAAGAGC	CAAACTGAGTTTGTTATCCGG
<i>BnGolS3-2</i>	BnaC08g50010D	CGCCTCAGTATAAGATTGGCTA	CGTTTAGAGTCTCCAAGAGGT
<i>BnGolS4-1</i>	BnaA09g14180D	CGCAGTATTCCATTGGGTATTG	GGCTAGGCTCAAACAAAAACAT
<i>BnGolS4-3</i>	BnaC09g14710D	CGCAGTATTCCATTGGGTATTG	TCTTGTTCTGCAAAAGGTGAAG
<i>BnGolS6-1</i>	BnaA08g14430D	CTTCGTATTTGGGAGTTTGTTG	CGTACAAGTAACCAGAAGGAGT
<i>BnGolS6-3</i>	BnaAnng12190D	CGTGAAGACATAAGAATGCTGG	AGCTAAGACTGCAGTAGTGATC
<i>BnGolS6-4</i>	BnaC08g12130D	ACTTGTACGCGGTTAAAGATTG	AAATACGTTGGAGTGGTGATCT
<i>BnGolS6-5</i>	BnaC08g33920D	CCTACAACCTCGTAATGGCTAT	CCTTTGGCACAGTAATGAACAA
<i>BnGolS6-6</i>	BnaC08g50120D	CTTGACGCGGTTAAAGATTGT	GACAAGATTTGGCTCAAACAGT
<i>BnGolS7-2</i>	BnaC01g28520D	ATGTTTGTGTTTGAGCCTAACC	GTGAACAACCTTGACTGTCTCC
<i>NtGolS1-1</i>	Nitab4.5_0000222g0170	CTTGATCTTGCATGGCTCCGG	CTTAGCCCCTTAGCTAACCCAAC
<i>NtGolS1-2</i>	Nitab4.5_0001013g0090	CATTTTCTTCAGATCACGCAACTGA	CACAATCTCTGGAGCCATGCAGGA
<i>NtGolS1-3</i>	Nitab4.5_0003324g0150	CATCAAACAGGGCGTATGTTACGT	CACGAGCGGATACGCAGTATGA
<i>NtGolS2-1</i>	Nitab4.5_0000136g0290	CCTGTTTATCCTCC-TCAGAATCAGA	TGGGCCTAACTCTTCTTCAGGC
<i>NtGolS2-2</i>	Nitab4.5_0000178g0340	CTTGTTGGTCTTGCAACCAAGGCTAGT	ACCCACTTGTAGTGGCTATTTTGCT
<i>NtGolS2-3</i>	Nitab4.5_0001617g0060	GTTGTTGGTCTAGCAACCAAGGCT	ACCACGCCTTTACGTAGTCACCGT
<i>NtGolS2-4</i>	Nitab4.5_0003044g0080	TGGCTCCTGAAGTACTTGTGAGT	ACCAACGGATACAAAGACT
<i>NtGolS2-5</i>	Nitab4.5_0008397g0020	CCGGTTTATCCTCCTGAGAACC	CTTTGGGCCTAACTCTTCAGCC
<i>NtGolS2-6</i>	Nitab4.5_0011298g0020	GGCATTGCAGTCTGAGACGT	ACCAACGGATACATAGATT
<i>BnAGP4</i>	BnaA07g21900D	GGTGTGCTTGTTCATTCATGA	CAAGCCGCTAATGAATGTAACA

<i>BnGH3.3</i>	BnaA09g42140D	AAGTCTCCGATGATCCATACAC	GAGAAGACAAAATCATGGACCG
<i>BnPR1</i>	BnaC03g45470D	TTAATTTTGGCAGCCCTTGTAG	GGTTGTGAGCGTTTACATAGTC
<i>BnAOC4</i>	BnaA08g24350D	CCCCCTTTTTGGGTTACTCTAG	CACATGGTTCTGAAGAAACCTG
<i>BnDREB2A</i>	BnaC09g49920D	ACTCTTCGGAGATGTTTGATGT	GAAGTCACTACCATCTTTTGCC
<i>BnARR6</i>	BnaC03g51340D	AAGATCTGAGGTTTACCAAGCA	ATAAGCCCTGTTTCTCTGTCAA
