

Impact of mid-season sulphur deficiency on wheat nitrogen metabolism and biosynthesis of grain protein

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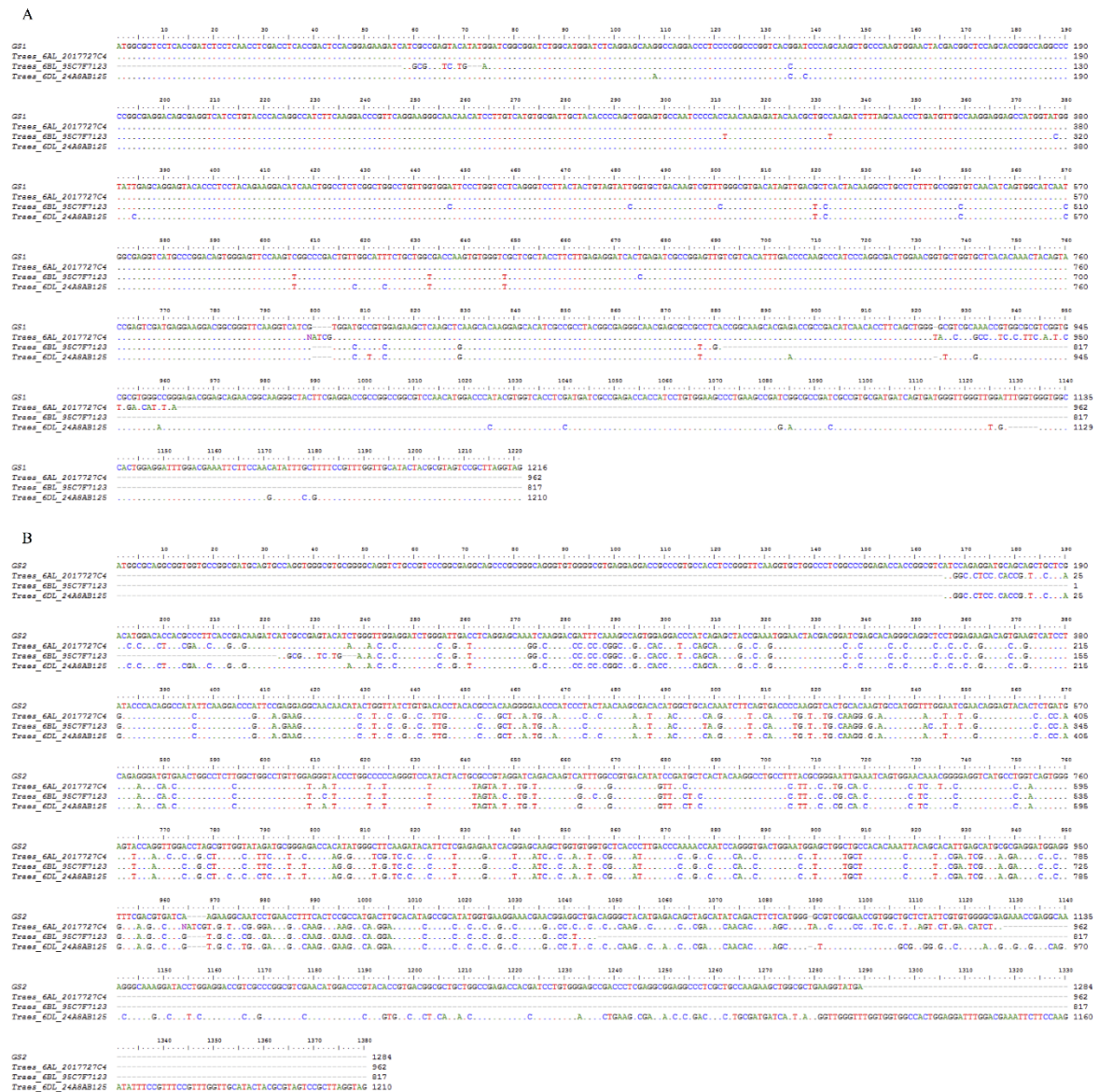
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Supplementary Figure S1

Sequence alignment of *GS* and nitrogen metabolism highly enriched DEGs. (a) *GS1*. (b) *GS2*.



Supplementary Table S1

Table S1. The identification of 63 DEGs from the comparison between S30 and S0 at 7 days post-anthesis

HS vs LS	Gene_ID	Swiss-Prot	Gene_ID	Swiss-Prot
Down-regulation	Traes_1AS_6E5 DECDA1	na	Traes_4AL_205E 88570	na
	Traes_1AS_E832 92840	na	Traes_4AL_A10 FB6994	Bisdemethoxycurcumin synthase, OS= <i>O. sativa</i>
	Traes_1BS_68F7 7FAE7	patatin	Traes_4BS_64FB 912EE	xyloglucan endotransglucosylase/hydrolase
	Traes_1BL_D6E 1CD173	non-specific lipid-transfer protein	Traes_4BL_34FE 2C7F2	Protein DROOPING LEAF, OS= <i>O. sativa</i>
	Traes_1DL_B466 DF47A	protein ECERIFERUM 1, OS= <i>A. thaliana</i>	Traes_4DL_5612 CF456	Glyceraldehyde-3-phosphate dehydrogenase B, chloroplastic, OS= <i>P. sativum</i>
	Traes_1DS_812A FD33F	patatin-like protein 1, OS= <i>O. sativa</i>	Traes_4DL_5C0 CDD31F	apyrase 3, OS= <i>O. sativa</i>
	Traes_1DS_8D70 617FC	patatin	Traes_4DL_B1B 379CB5	Aspartic proteinase Asp1, OS= <i>O. sativa</i>
	Traes_1DS_FA49 5B931	na	Traes_5AL_B446 A07423	non-specific lipid-transfer protein 3
	Traes_2AL_6DE EE3C3E	beta-fructofuranosidase, insoluble isoenzyme 2, OS= <i>O. sativa</i>	Traes_5BS_BA0 EAE97A1	na
	Traes_2AL_CB3 FF487A	ribosomal protein S17, chloroplastic, OS= <i>Z. mays</i>	Traes_5BL_304F AFA26	Lipoxygenase
	Traes_2BS_6762 A1AAE	taxadien-5-alpha-ol O-acetyltransferase, OS= <i>T. cuspidata</i>	Traes_5BL_ED5 21656E1	3-ketoacyl-CoA synthase
	Traes_2BS_B8E7 189CF	basic endochitinase A, OS= <i>S. cereale</i>	Traes_5DS_94D2 1BF47	na
	Traes_2BS_B8E7 189CF1	basic endochitinase A,	Traes_5DL_488F BAEEC	non-specific lipid-transfer

Traes_2BL_032A 12DB3	OS= <i>S. cereale</i> ultraviolet-B receptor UVR8, OS= <i>A. thaliana</i>	Traes_5DL_7575 B5B36	protein 3 Dirigent protein 1, OS= <i>A. thaliana</i>
Traes_2BL_4AA B8719F	ultraviolet-B receptor UVR8, OS= <i>A. thaliana</i>	Traes_5DL_8F68 FBC81	non-specific lipid-transfer protein Glucomannan 4- beta- mannosyltransfer ase 1, OS= <i>O. sativa</i>
Traes_2BL_7911 433A3	alpha- glucosidase, OS= <i>O. sativa</i>	Traes_6AS_3369 0B236	
Traes_2BL_9280 00A2B	ultraviolet-B receptor UVR8, OS= <i>A. thaliana</i>	Traes_6AL_2017 727C4	Glutamine synthetase
Traes_2DL_0489 2661A	Aldehyde dehydrogenase family 3 member F1, OS= <i>A. thaliana</i>	Traes_6AL_38E8 DCF06	Peroxidase
Traes_2DL_1B3 AD9F4E	Beta- fructofuranosidas e, insoluble isoenzyme 2, OS= <i>O. sativa</i>	Traes_6BL_34AF 8C436	Peroxidase
Traes_2DL_3C22 9EB92	protochlorophylli de reductase A, chloroplastic glyceraldehyde-3- phosphate	Traes_6BL_95C7 F7123	Glutamine synthetase
Traes_2DL_80D D45E69	dehydrogenase A, chloroplastic, OS= <i>Z. mays</i>	Traes_6DS_3522 B8EF6	Catalase
Traes_2DL_85A FEAF70	non-specific lipid-transfer protein	Traes_6DS_C035 8414B	ABC transporter G family member 11, OS= <i>A. thaliana</i>
TRAES3BF0044 00040CFD	non-specific lipid-transfer protein	Traes_6DL_0A0 ABD677	Protein TSS, OS= <i>A. thaliana</i>
TRAES3BF0052 00020CFD	domain- containing protein	Traes_6DL_24A8 AB125	Glutamine synthetase, GS1
TRAES3BF0126 00130CFD	esterase/lipase, OS= <i>A. thaliana</i>	Traes_6DL_3EF3 193D0	Peroxidase
TRAES3BF0277 00190CFD	sigma factor binding protein 1, chloroplastic, OS= <i>A. thaliana</i>	Traes_7AL_2914 71D52	Tryptophan synthase beta chain 2, OS= <i>A. aeolicus</i>
TRAES3BF0678	2-alkenal	Traes_7AL_9AE	UPF0481 protein

	00180CFD	reductase (NADP(+)- dependent), OS=N. tabacum	C84938	At3g47200, OS= <i>A. thaliana</i>
	TRAES3BF0763 00030CFD	na	Traes_7BS_5FE2 61AAA	UDP- glucosyltransferas e BX8, OS= <i>Z. mays</i>
	TRAES3BF0883 00010CFD	non-specific lipid-transfer protein	Traes_7DS_71A DD7C7C	na
	TRAES3BF0883 00030CFD	non-specific lipid-transfer protein	Traes_7DS_A934 D9C24	Bisdemethoxycur cumin synthase, OS= <i>O. sativa</i>
	TRAES3BF0884 00020CFD	non-specific lipid-transfer protein		
	Traes_3DS_E756 029E7	glutamate decarboxylase		
Up- regulation	Traes_5BS_B432 6E4BD	Pyrophosphate- fructose 6- phosphate 1- phosphotransferas e subunit alpha, OS= <i>R. communis</i>		

Supplementary Table S4

Table S4. Identification of *cis*-acting element conserved regions in promoter sequence of three DEGs annotated as glutamine synthetase

	Gene ID	Name	Region	Match	Accuracy (%)
AP2/ER EBP	Traes_6AL_2017727C4	AP2/ERE BP	741..744	ACGT	100
	Traes_6AL_2017727C4	AP2/ERE BP	804..807	ACGT	100
	Traes_6AL_2017727C4	AP2/ERE BP	901..906	GCCGCC	100
	Traes_6AL_2017727C4	AP2/ERE BP	complement(741..744)	ACGT	100
	Traes_6AL_2017727C4	AP2/ERE BP	complement(804..807)	ACGT	100
	Traes_6BL_95C7F7123	AP2/ERE BP	744..747	ACGT	100
	Traes_6BL_95C7F7123	AP2/ERE BP	812..815	ACGT	100
	Traes_6BL_95C7F7123	AP2/ERE BP	905..910	GCCGCC	100
	Traes_6BL_95C7F7123	AP2/ERE BP	complement(744..747)	ACGT	100
	Traes_6BL_95C7F7123	AP2/ERE BP	complement(812..815)	ACGT	100
	Traes_6DL_24A8AB125	AP2/ERE BP	726..729	ACGT	100
	Traes_6DL_24A8AB125	AP2/ERE BP	794..797	ACGT	100
	Traes_6DL_24A8AB125	AP2/ERE BP	894..899	GCCGCC	100
	Traes_6DL_24A8AB125	AP2/ERE BP	complement(726..729)	ACGT	100
	Traes_6DL_24A8AB125	AP2/ERE BP	complement(794..797)	ACGT	100
ARF	Traes_6AL_2017727C4	ARF	738..742	GAGAC	100
	Traes_6BL_95C7F7123	ARF	741..745	GAGAC	100
	Traes_6DL_24A8AB125	ARF	723..727	GAGAC	100
bZIP	Traes_6AL_2017727C4	bZIP	740..745	GACGTC	100
	Traes_6AL_2017727C4	bZIP	741..744	ACGT	100
	Traes_6AL_2017727C4	bZIP	804..807	ACGT	100
	Traes_6AL_2017727C4	bZIP	complement(740..745)	GACGTC	100

	Traes_6AL_2017727C4	bZIP	complement(741..744)	ACGT	100
	Traes_6AL_2017727C4	bZIP	complement(804..807)	ACGT	100
	Traes_6BL_95C7F7123	bZIP	743..748	GACGTC	100
	Traes_6BL_95C7F7123	bZIP	744..747	ACGT	100
	Traes_6BL_95C7F7123	bZIP	812..815	ACGT	100
	Traes_6BL_95C7F7123	bZIP	complement(743..748)	GACGTC	100
	Traes_6BL_95C7F7123	bZIP	complement(744..747)	ACGT	100
	Traes_6BL_95C7F7123	bZIP	complement(812..815)	ACGT	100
	Traes_6DL_24A8AB125	bZIP	725..730	GACGTC	100
	Traes_6DL_24A8AB125	bZIP	726..729	ACGT	100
	Traes_6DL_24A8AB125	bZIP	794..797	ACGT	100
	Traes_6DL_24A8AB125	bZIP	complement(725..730)	GACGTC	100
	Traes_6DL_24A8AB125	bZIP	complement(726..729)	ACGT	100
	Traes_6DL_24A8AB125	bZIP	complement(794..797)	ACGT	100
	Traes_6AL_2017727C4	SURE	738..742	GAGAC	100
SURE	Traes_6BL_95C7F7123	SURE	741..745	GAGAC	100
	Traes_6DL_24A8AB125	SURE	723..727	GAGAC	100

Supplementary Table S5Table S5. Real time PCR primer for *GS1* and *GS2*

Genes	Orientation	5'-3' sequence
<i>GS1</i>	Forward	GTGGATGCCGTGGAGAAG
	Reverse	GCTGAAGGTGTTGATGTCG
<i>GS2</i>	Forward	CTCGTCCGCGTCCTTGTCCG
	Reverse	GCCGACCTGCCCCGCACG