

Additional File 3:

Primers for expression analysis by qRT-PCR

DWF4 (At3g50660), *CPD* (At5g05690), *TIP41* (At4g34270), *SAND* (At2g28390), and expressed protein (At4g33380) were selected for expression analysis by qRT-PCR. *DWF4* and *CPD* are involved in brassinosteroid biosynthesis, and their expression is subject to feedback inhibition, i.e. their expression is down-regulated by elevated brassinolide [1, 2, 3, 4]. *TIP41*, *SAND*, and At4g33380 were selected as reference genes based upon their documented stability of expression and successful use in qRT-PCR studies of roots [5, 6, 7, 8, 9]. Primers used, along with qRT-PCR efficiencies and R^2 values determined from serial dilution calibration curves are as follows:

Gene	Primer	Amplicon Size (bases)	bp from 3' end of cDNA	Efficiency	R^2
CPD NM_001036761	CAGGCCCTTCTAATGTGTTCA CCCAACTG^AAGCCTGTCACTA	120	314	96.6%	0.995
DWF NM_114926	CATCGCAAAGCACTCAAAGAT TTATCCAAATGTACGGCTGAGA	95	514	97.8%	0.975
TIP41 NM_119592	GC^GAAAGGGTATCCAGTTGA GGAAGCCTCTGACTGATGGA	65	343	97.3%	0.995
SAND NM_128399	TGATCCACTTGCAGACA^AGG TACCCTTTGGCACACCTGAT	61	310	99.0%	0.990
Expressed Protein NM_119492	TGTTGGCTTAAATAC^GCAGAGA TGCTCAAGCCGTTACAACAC	129	265	96.4%	0.976
^ signifies location of intron in genomic DNA that is spanned by this primer for cDNA					

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