

Title: A cytosolic invertase is required for normal growth and cell development in the model legume, *Lotus japonicus*.

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Supplementary Files

Figures

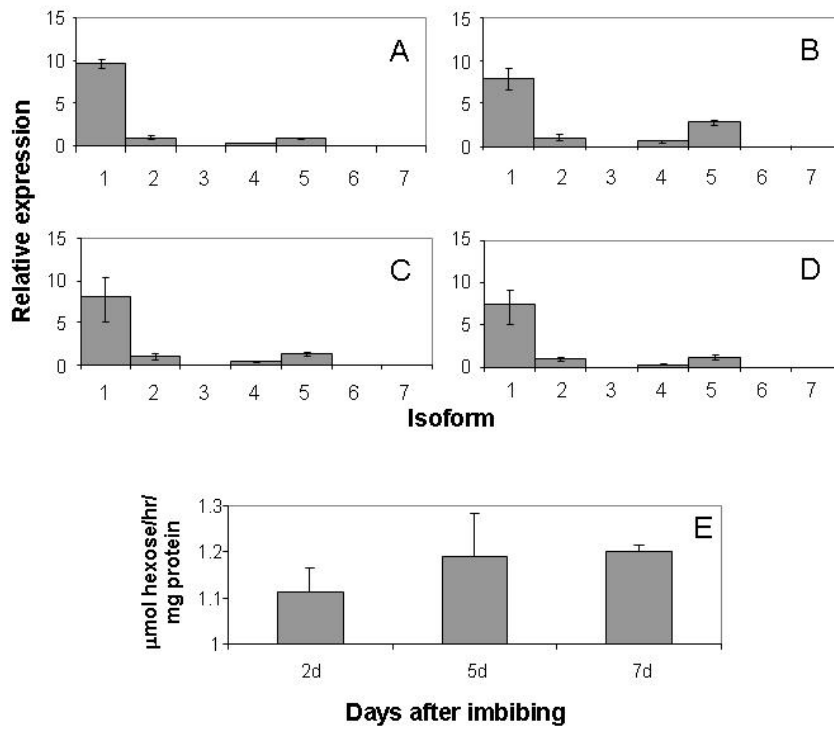


Figure S1. Expression of *LjINV* genes and enzyme activities during seedling growth. Relative abundance of different N/A invertase transcripts. The relative amounts of *LjINV1* to *LjINV7* were analysed by qRT-PCR in whole seedlings (3 replicates) aged: A, 2 days; B, 5 days; C, 7 days; D, 9 days. E, N/A enzyme activity in whole seedlings (3 replicates) at different ages. Transcript levels were expressed relative to the level in *LjINV2* after normalization to the levels of *LjEF-1α*. Note the asymmetrical distribution of the SD caused by the conversion of the exponential process into a linear comparison (Livak and Schmittgen, 2001).

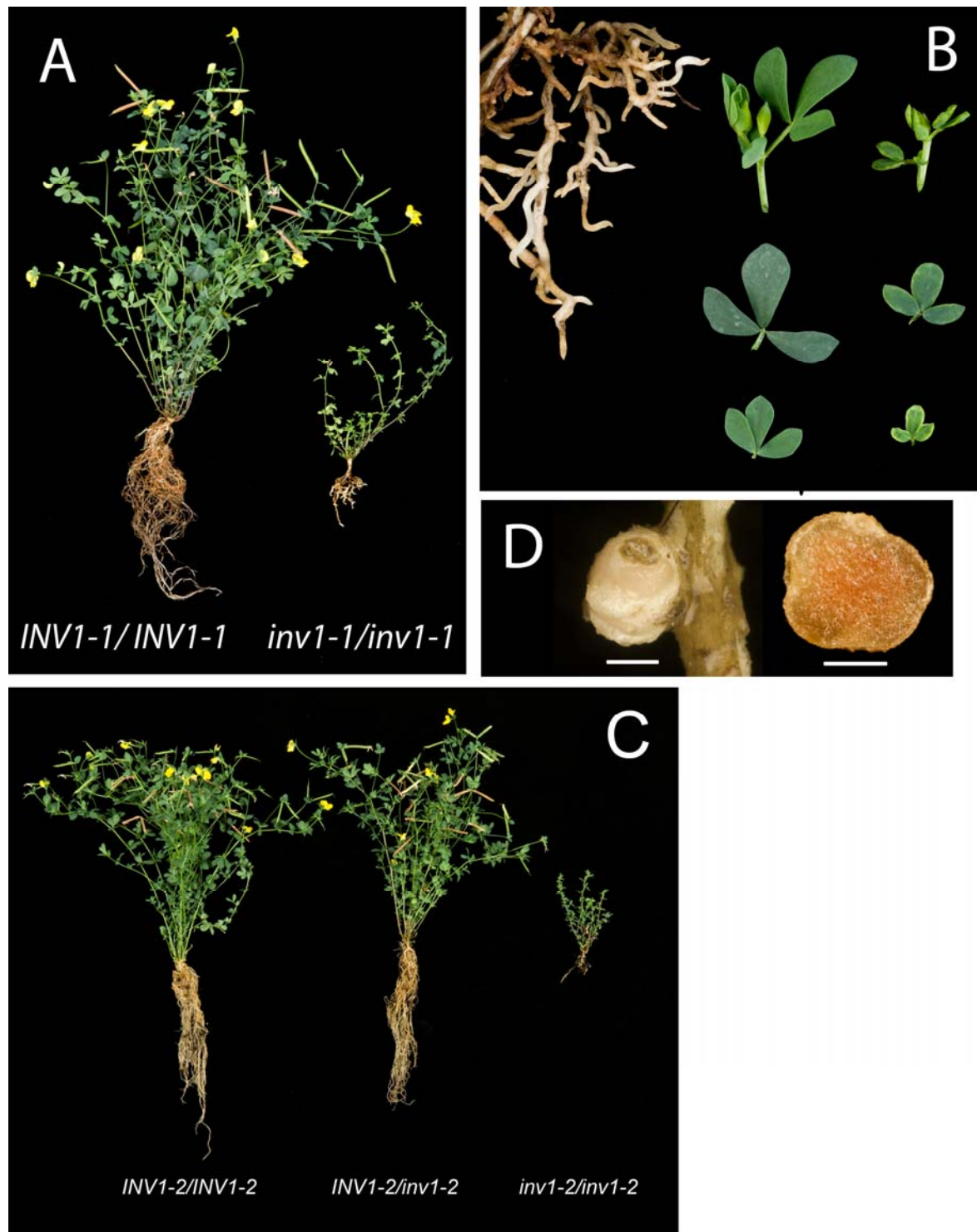


Figure S2. Colour plate of Figure 5.

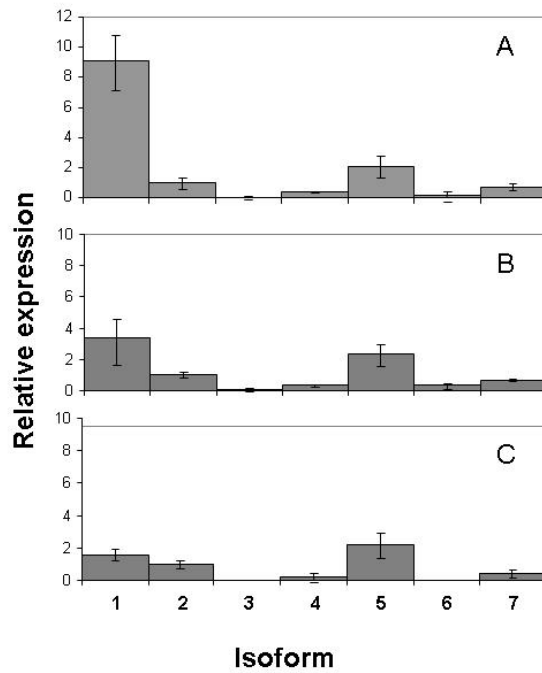


Figure S3. Expression of *LjINV* genes in the *inv1-1* mutant. Relative abundance of different N/A invertase transcripts in the lines segregating for the *inv1-1* mutant allele. A, wild type; B, heterozygote; C, mutant. The relative amounts of *LjINV1* to *LjINV7* were analysed by qRT-PCR in shoot tips (3 replicates). Transcript levels were expressed relative to the level in *LjINV2* after normalization to the levels of EF-1 α . Note the asymmetrical distribution of the SD caused by the conversion of the exponential process into a linear comparison (Livak and Schmittgen, 2001).

Supplementary tables

Table S1. Subcellular location data for *L. japonicus* invertase isoforms. M= mitochondion; P= plastid

Gene	Length (aa)		Prediction Tool					Localization
			Predotar	PSORT	Mitprot II	TargetP	ChloroP	
<i>LjINV2</i>	621	Mitochon.	0.27	0.81	0.99	0.54		M/P
		Plastid	0.02	0.49		0.54	0.56	
<i>LjINV3</i>	668	Mitochon.	0.42		1.00	0.01		M/P
		Plastid	0.46		.	0.95	0.57	
<i>LjINV4</i>	838	Mitochon.	0.03	0.73	1.00	0.07		M/P
		Plastid	0.35	0.52		0.75	0.52	
<i>LjINV5</i>	640	Mitochon.	0.05		0.57	0.03		
		Plastid	0.08			0.24	0.51	
<i>LjINV1</i>	556	Mitochon.	0.01	0.10	0.01	0.07		
		Plastid	0.00	0.10		0.18	0.44	
<i>LjINV7</i>	572	Mitochon.	0.01	0.10	0.047	0.03		
		Plastid	0.01			0.57	0.46	

Table S2. *L. japonicus* *INV1* alleles, their sequence differences to the wild type and Position Specific Scoring Matrix (PSSM) scores

Plant number	Allele	Nucleotide change	Amino acid change	PSSM Difference
SL5864-1	<i>inv1-1</i>	C2047T	Q 329 *	
SL5331-1	<i>inv1-2</i>	G2426A	G 455 E	15.4
SL3337-1	<i>inv1-3</i>	G2168A	G 369 D	13.2
SL3485-1	<i>inv1-4</i>	C2158T	P 366 S	13.7
SL1461-1	<i>inv1-5</i>	G2206A	D 382 N	10.2
SL761-1	<i>inv1-6</i>	C2263T	P 401 S	8.1
SL769-1	<i>inv1-7</i>	C1961T	A 300 V	
SL1178-1	<i>inv1-8</i>	C1604T	intron	
SL4132-1	<i>inv1-9</i>	G2149A	D 363 N	9.9
SL5951-1	<i>inv1-10</i>	C1576T	intron	

Table S3. *L. japonicus* *INV2* alleles, their sequence differences to the wild type and Position Specific Scoring Matrix (PSSM) scores

Plant number	Allele	Nucleotide change	Amino acid change	PSSM Difference
SL174-1	<i>inv2-1</i>	G2533A	W464*	
SL379-1	<i>inv2-2</i>	G2240A	R370K	8.3
SL379-2	<i>inv2-2</i>	G2240A	R370K	8.3
SL1897-1	<i>inv2-3</i>	C2328T	F399=	
SL3388-1	<i>inv2-4</i>	C2598T	T489=	
SL1870-1	<i>inv2-5</i>	G2623A	intron	
SL1959-1	<i>inv2-6</i>	C2626T	intron	
SL1585-1	<i>inv2-7</i>	C1610T	I266=	
SL500-1	<i>inv2-8</i>	G1776A	G322S	12.1
SL717-1	<i>inv2-8</i>	G1776A	G322S	12.1
SL719-1	<i>inv2-8</i>	G1776A	G322S	12.1
SL253-1	<i>inv2-9</i>	1887T	intron	