

Supplementary Material

The role of jasmonate signalling in quinolizidine alkaloid biosynthesis, wounding and aphid predation response in narrow-leafed lupin

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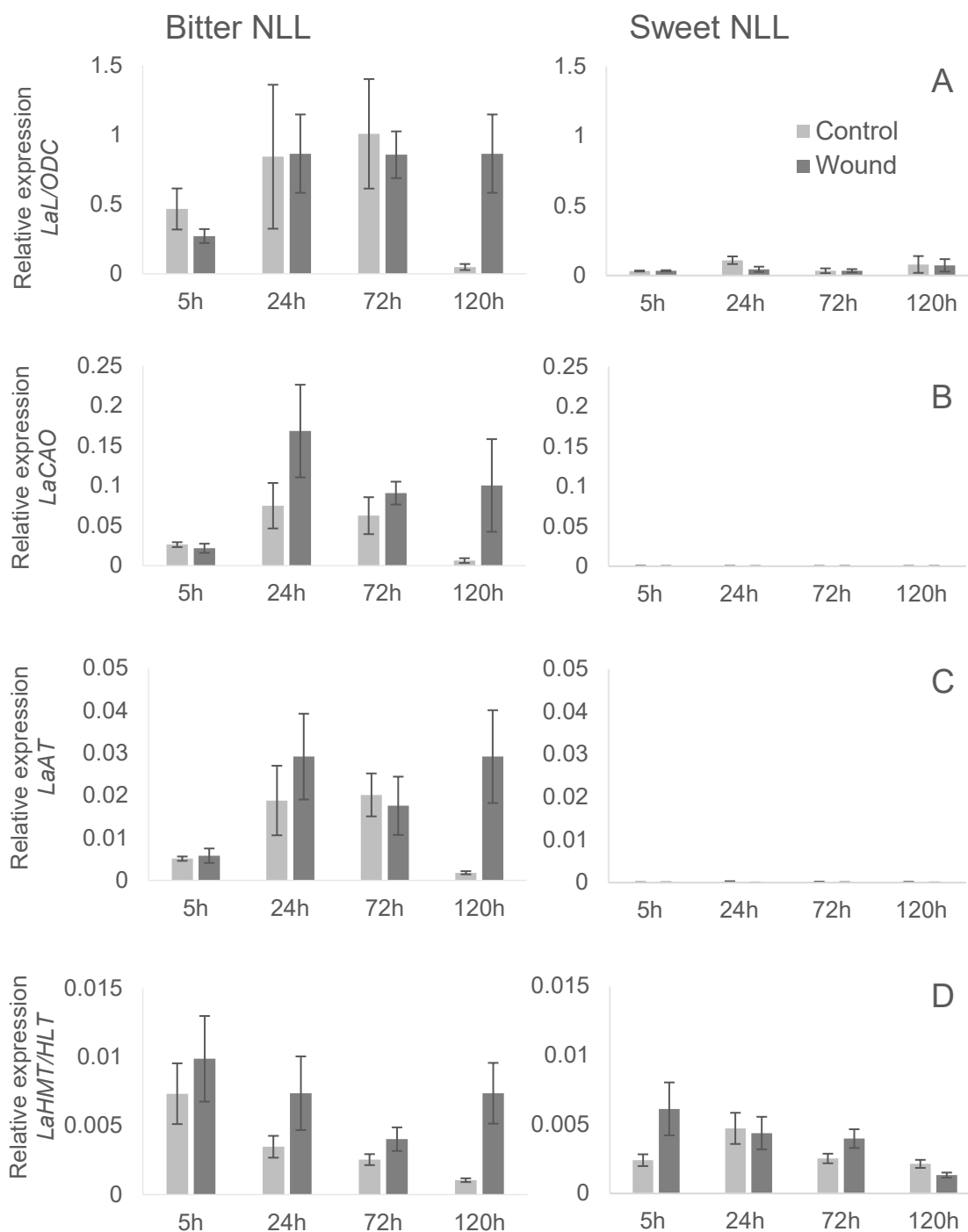


Fig. S1. Relative expression of QA biosynthetic genes (A) *LaL/ODC* (*Lup009726.1*), (B) *LaCAO* (*Lup000530.1*), (C) *LaAT* (*Lup021586.1*) and (D) *LaHMT/HLT* (*Lup022251.1*) in wounded leaf tissue of bitter (P27255; left) and sweet (Tanjil; right) NLL varieties. Three-week-old plants were wounded by rolling a fabric pattern wheel over leaves. The means and standard errors of three biological replicates are presented. One-sided two-sample *t*-test ($P \leq 0.05$) was used to assess whether gene expression increased in wounded leaves compared to controls at a certain time point. No significant differences were found.

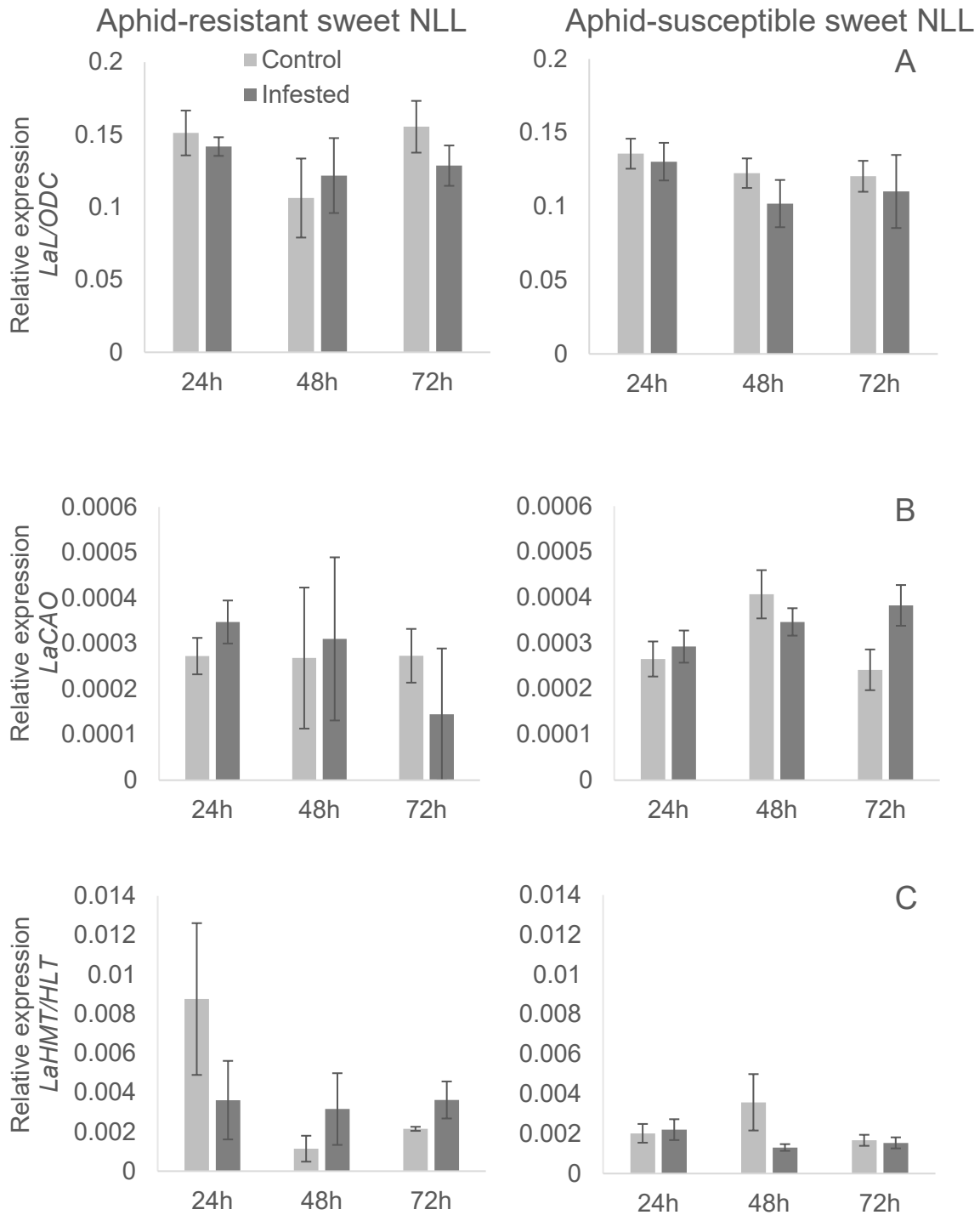


Fig. S2. Relative gene expression of QA biosynthetic genes (A) *LaL/ODC* (*Lup009726.1*), (B) *LaCAO* (*Lup000530.1*) and (C) *LaHMT/HLT* (*Lup022251.1*) in leaf tissue of resistant (Kalya; left) and susceptible (Tallerack; right) NLL cultivars after aphid infestation. Means and standard errors of three biological replicates are presented. Three-way ANOVA was used to assess the effect of aphid treatment, cultivar, time, and their interactions on gene expression. No significant differences in expression between control and infested samples were found.

Table S1. Primers used in this study for qRT-PCR

Gene	Orientation	Sequence	Annealing temp.
<i>LaUBC9-like</i>	Forward	5'-CAAGCAACAATTATGGGTCCTC-3'	57
<i>Lup018255.1</i>	Reverse	5'-AACCTTGGGTGGCTTAAAGG-3'	
<i>LaL/ODC</i>	Forward	5'-TATTGGGAAGCGCGTGAG-3'	57
<i>Lup009726.1</i>	Reverse	5'-TGGGTCCAAACACAGTTGAA-3'	
<i>LaCAO</i>	Forward	5'-CTTGTCCTCTSCCTCCTGT-3'	57
<i>Lup000530.1</i>	Reverse	5'-TCCACTTTTGCCATTCAACA-3'	
<i>LaHMT/HLT</i>	Forward	5'-GATGTTCTGGTTCAGATGGW-3'	57
<i>Lup022251.1</i>	Reverse	5'-AGCAAAGATRAATCCACCACA-3'	
<i>LaAT</i>	Forward	5'-GCACCAGGACCTTCTTCAAC-3'	57
<i>Lup021586.1</i>	Reverse	5'-CAAGTTGTRCTAACCATGAAGG-3'	
<i>LaLOX4-like</i>	Forward	5'-TAGCCCAATCCCTTTGTTCA-3'	57
<i>Lup026769.1</i>	Reverse	5'-ATTCACTCCAGCAATCATCTCTC-3'	
<i>LaPI-like</i>	Forward	5'-TGTTCCAATCCCTTCTTCCA-3'	57
<i>Lup009615.1</i>	Reverse	5'-ACTACTGGTTCAGGTTTCAGATGC-3'	
<i>LaLOX5-like</i>	Forward	5'-ACCAGTGTTTGGCTTTGGAG-3'	57
<i>Lup003340.1</i>	Reverse	5'-GCCTGTTTGTTGCGATGATA-3'	

Table S2. Raw values for levels of QAs in leaf material of sweet NLL cultivar Tanjil treated with methyl jasmonate (MeJA) for 36 h

Limit of quantification is 0.001%

Leaf sample	Angustifoline	α -isolupanine	Lupanine	13-hydroxylupanine	Total
Tanjil 36h control	0.000000	0.000000	0.000222	0.000391	0.000613
Tanjil 36h control	0.000000	0.000000	0.000223	0.000547	0.000771
Tanjil 36h control	0.000000	0.000000	0.000175	0.000329	0.000504
Tanjil 36h control	0.000000	0.000151	0.000178	0.000199	0.000527
Tanjil 36h control	0.000000	0.000000	0.000000	0.000199	0.000199
Tanjil 36h control	0.000000	0.000000	0.000000	0.000351	0.000351
Tanjil 36h MeJA	0.000000	0.000000	0.000000	0.000354	0.000354
Tanjil 36h MeJA	0.000000	0.000000	0.000000	0.001100	0.001100
Tanjil 36h MeJA	0.000000	0.000190	0.000000	0.000465	0.000655
Tanjil 36h MeJA	0.000000	0.000167	0.000000	0.000427	0.000593
Tanjil 36h MeJA	0.000000	0.000000	0.000000	0.000298	0.000298
Tanjil 36h MeJA	0.000000	0.000000	0.000000	0.001148	0.001148

Table S3. Raw values for levels of QAs in leaf material of susceptible (Tallerack) and resistant (Kalya) NLL cultivars after 72 h infestation with aphids

Limit of quantification is 0.001%

Leaf sample	Angustifoline	α - isolupanine	Lupanine	13-hydroxylupanine	Total
Tallerack 72h ctl	0.000000	0.000000	0.000000	0.000185	0.000185
Tallerack 72h ctl	0.000000	0.000621	0.000000	0.000000	0.000621
Tallerack 72h ctl	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h ctl	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h ctl	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h ctl	0.000000	0.000000	0.000000	0.000236	0.000236
Tallerack 72h infested	0.000000	0.000196	0.000000	0.000000	0.000196
Tallerack 72h infested	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h infested	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h infested	0.000000	0.000000	0.000000	0.000000	0.000000
Tallerack 72h infested	0.000307	0.000000	0.000000	0.000000	0.000307
Tallerack 72h infested	0.000000	0.000000	0.000000	0.000000	0.000000
Kalya 72h ctl	0.000000	0.000000	0.000000	0.000448	0.000448
Kalya 72h ctl	0.000000	0.000135	0.000000	0.000685	0.000820
Kalya 72h ctl	0.000000	0.000000	0.000000	0.003191	0.003191
Kalya 72h ctl	0.000000	0.000000	0.000000	0.001909	0.001909
Kalya 72h ctl	0.000000	0.000000	0.000000	0.001379	0.001379
Kalya 72h ctl	0.000000	0.000000	0.000000	0.001877	0.001877
Kalya 72h infested	0.000000	0.000000	0.000000	0.000276	0.000276
Kalya 72h infested	0.000816	0.000324	0.000361	0.004153	0.005654
Kalya 72h infested	0.000000	0.000000	0.000000	0.000490	0.000490
Kalya 72h infested	0.000000	0.000000	0.000000	0.001662	0.001662
Kalya 72h infested	0.000000	0.000000	0.000189	0.001151	0.001340
Kalya 72h infested	0.000000	0.000000	0.000000	0.000706	0.000706