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**Genetic basis of allochronic differentiation in the fall armyworm**

Hänniger, S.; Dumas, P.; Schöfl, G.; Gebauer-Jung, S.; Vogel, H.; Unbehend, M.; Heckel, D.G.; Groot, A.T.

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## Additional file 8.

PCR conditions used in the mentioned experiments.

Experiment	Candidate Gene	PCR components <sup>1</sup>		PCR program		
QTL 1	Vrille	1 $\mu$ l	DNA	2 min	94°C	
		11.92 $\mu$ l	dH <sub>2</sub> O	35x {	94°C	
		2 $\mu$ l	10x Taq buffer		45 s	T <sub>a</sub> <sup>2</sup>
		2 $\mu$ l	2 mM dNTPs		45 s	72°C
		3 $\mu$ l	10 mM primer mix <sup>2</sup>		60 s	72°C
		0.08 $\mu$ l	Taq polymerase		10 min	72°C
Structure analysis of <i>Vrille</i>	Vrille	1 $\mu$ l	DNA	2 min	94°C	
		11.92 $\mu$ l	dH <sub>2</sub> O	45 s	94°C	
		2 $\mu$ l	10x Taq buffer	35x {	T <sub>a</sub> <sup>2</sup>	
		2 $\mu$ l	2 mM dNTPs		45 s	72°C
		3 $\mu$ l	10 mM primer mix <sup>2</sup>		90 s	72°C
		0.08 $\mu$ l	Taq polymerase		10 min	72°C
	Vrille (touchdown PCR for degenerate primers)	1 $\mu$ l	DNA	3 min	94°C	
		11.92 $\mu$ l	dH <sub>2</sub> O	28x {	94°C	
		2 $\mu$ l	10x Taq buffer		30 s	T <sub>a</sub> <sup>2*</sup>
		2 $\mu$ l	2 mM dNTPs	60 s	(decrease by 0,7)	
		3 $\mu$ l	10 mM primer mix <sup>2</sup>		30 s	72°C
		0.08 $\mu$ l	Taq polymerase	23x {	94°C	
					30 s	Lowest T <sub>a</sub> <sup>2</sup>
				60 s	72°C	
Expression analysis	Vrille eIF1 $\alpha$	1 $\mu$ l	cDNA	10 min	90°C	
		10 $\mu$ l	dH <sub>2</sub> O	40x {	95°C	
		1 $\mu$ l	10 mM primer fw		30 s	58°C
		1 $\mu$ l	10 mM primer rv		60 s	72°C
		12 $\mu$ l	SYBR Mix <sup>3</sup>		60 s	95°C
					30s	58°C
					30s	95°C

<sup>1</sup>Taq polymerase, dNTPs, buffer and primers were purchased from Metabion, Martinsried, Germany

<sup>2</sup>Primers and corresponding annealing temperatures (T<sub>a</sub>) can be found in Table S6

<sup>3</sup> ABSolute Blue QPCR SYBR Green Mix from Thermo Fisher Scientific, Schwerte, Germany