

## Additional Tables

**Additional Table 1. Ranking of candidate reference gene tested by BestKeeper, geNorm and NormFinder.** The best gene for housekeeping during *Rhodnius* embryonic stages was chosen by ranking analysis using the BestKeeper, geNorm and NormFinder programs. For the analysis of best candidate genes with BestKeeper, we ranked the genes according to the coefficient of correlation (r) and the p-value between each candidate gene and the BestKeeper index (Pfaffl et al., 2004). The geNorm program estimates gene expression stability (M value). The candidate gene with the lowest M value is considered to have the most stable expression (Vandesompele et al., 2002). In addition, according to NormFinder, the candidate genes that are more stably expressed are indicated by lower expression stability values (Andersen et al., 2004). Considering the whole dataset, *Rp-18S* and *Rp-Efl* were the most stable genes.

Gene ranking	BestKeeper		geNorm	NormFinder
	coef. corr. (r)	p-value	stability value (M)	stability value
<i>Rp-18S</i>	0.965	0.001	0.779	0.136
<i>Rp-Efl</i>	0.930	0.001	0.725	0.172
<i>Rp-gadph</i>	0.773	0.024	0.856	0.495
<i>Rp-mip</i>	0.247	0.558	1.224	0.749

**Additional Table 2. qPCR primer sequences used in this study.**

<i>Rp</i> -Gene	Primers (5'-3')		Amplicon size (bp)
	Forward	Reverse	
<i>dl</i>	TGTCGGTTTAGCAGCTGTTGGT	TCCAGATGGCTTTGAAAAATCCCTG	95
<i>Toll</i>	TCTTGACCCGGAGCTGAAAGC	CCGTAGGATGTGGCAAGGCA	104
<i>cact</i>	GTGCTGGTGCTTGTACGAAA	GGAGTCGGACGATACCTCAA	154
<i>dpp</i>	ACAGCGCAAAAAGGATGGACG	TCGGCCAATGGAATGGGCA	147
<i>sog</i>	TGTTTGCCACAGCCAACCGA	GTCAGCCAAGGAACGCCACT	141
<i>18S</i>	TCGGCCAACAAAAGTACACA	TGTCGGTGTAAGTGGCATGT	104
<i>Efl</i>	GATTCCACTGAACCGCCTTA	GCCGGGTTATATCCGATTTT	92
<i>gadph</i>	TTTTGATGCAAAAAGCTGGAA	AACGACACGATTTGAATAGCC	94
<i>mip</i>	CCTTCTTGGTGGACCCTACA	TCCAACCCAGTACACCCAAT	106
<i>sna</i>	AAACAACATCCTCGTCCGCT	TGTTGATGTTTTGACAGTCCGG	100
<i>twi</i>	GGCTAATGTAAGAGAGCGCC	GATGCCAACCTCAGTGTGTTGT	123

*Rp-gadph* (glyceraldehyde-3-phosphate dehydrogenase) and *Rp-mip* (major intrinsic protein) primers were a gift from Dr. D. Majerowicz. All other genes are referred to in the main text.

**Additional Table 3. pRNAi primer sequences used in this study. Nucleotides in small caps correspond to the T7 sequences.**

<i>Rp</i> -Gene	Primers (5'-3')		Amplicon size (bp)
	Forward	Reverse	
<i>dl</i>	ggcccgggAAGAGGACATATCAGTGCGATTC	cccggggcTGTTGAAAACGTACGGTAGTCG	303
<i>Toll</i>	ggcccgggTCTTGACCCGGAGCTGAAAGC	cccggggcCCGTAGGATGTGGCAAGGCA	718
<i>cact</i>	ggcccgggTGCTTACTCTTTAAGGCAGTCC	cccggggcGGAGTCGGACGATACCTCAA	755

**References:**

Andersen CL, Jensen JL, Orntoft TF: **Normalization of real-time quantitative reverse transcription-PCR data: a model-based variance estimation approach to identify genes suited for normalization, applied to bladder and colon cancer data sets.** *Cancer Res* 2004, 64:5245-5250.

Pfaffl MW, Tichopad A, Prgomet C, Neuvians TP: **Determination of stable housekeeping genes, differentially regulated target genes and sample integrity: BestKeeper-Excel-based tool using pair-wise correlations.** *Biotechnol Lett* 2004, 26:509-515.

Vandesompele J, De Preter K, Pattyn F, Poppe B, Van Roy N, De Paepe A, Speleman F: **Accurate normalization of real-time quantitative RT-PCR data by geometric averaging of multiple internal control genes.** *Genome Biol* 2002, 3:1-11.