

Supplementary Materials

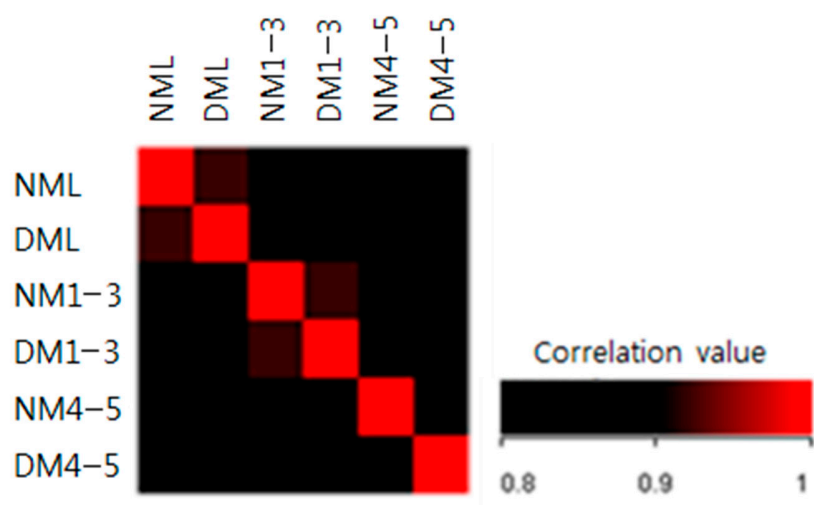


Figure S1. Pairwise correlation of the six libraries; NML (normal morph of late embryo), DML (defensive morph of late embryo), NM1-3 (normal morph of 1-3 instar), DM1-3 (defensive morph of 1-3 instar), NM4-5 (normal morph of 4-5 instar), and DM4-5 (defensive morph of 4-5 instar). Color changes from black to red mean correlation values from low to high.



Figure S2. Hierarchical cluster of the expressed genes in the six libraries; NML (normal morph of late embryo), DML (defensive morph of late embryo), NM1-3 (normal morph of 1–3 instar), DM1-3 (defensive morph of 1–3 instar), NM4-5 (normal morph of 4–5 instar), and DM4-5 (defensive morph of 4–5 instar). Color changes from blue to red represent normalized RPKM values from low to high.

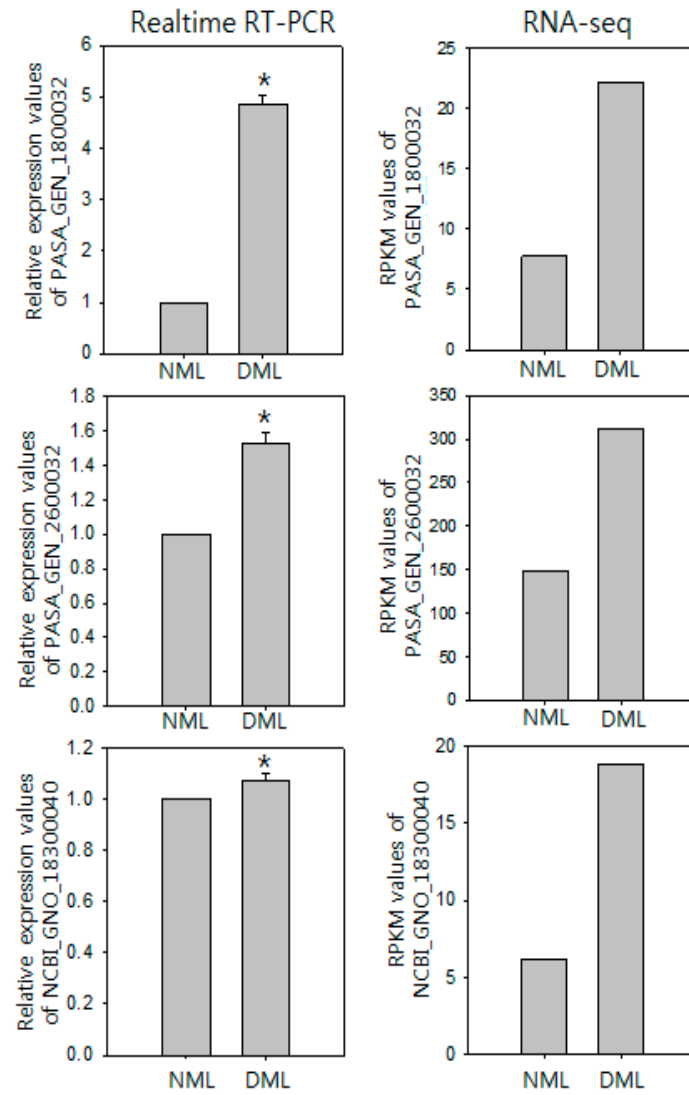


Figure S3. Real-time RT-PCR validation. * represents differentially-expressed genes between normal morph (NM) and defensive morph (DM) with significance ($p < 0.05$). NML and DML represent the normal morph of the late embryo and the defensive morph of the late embryo, respectively.

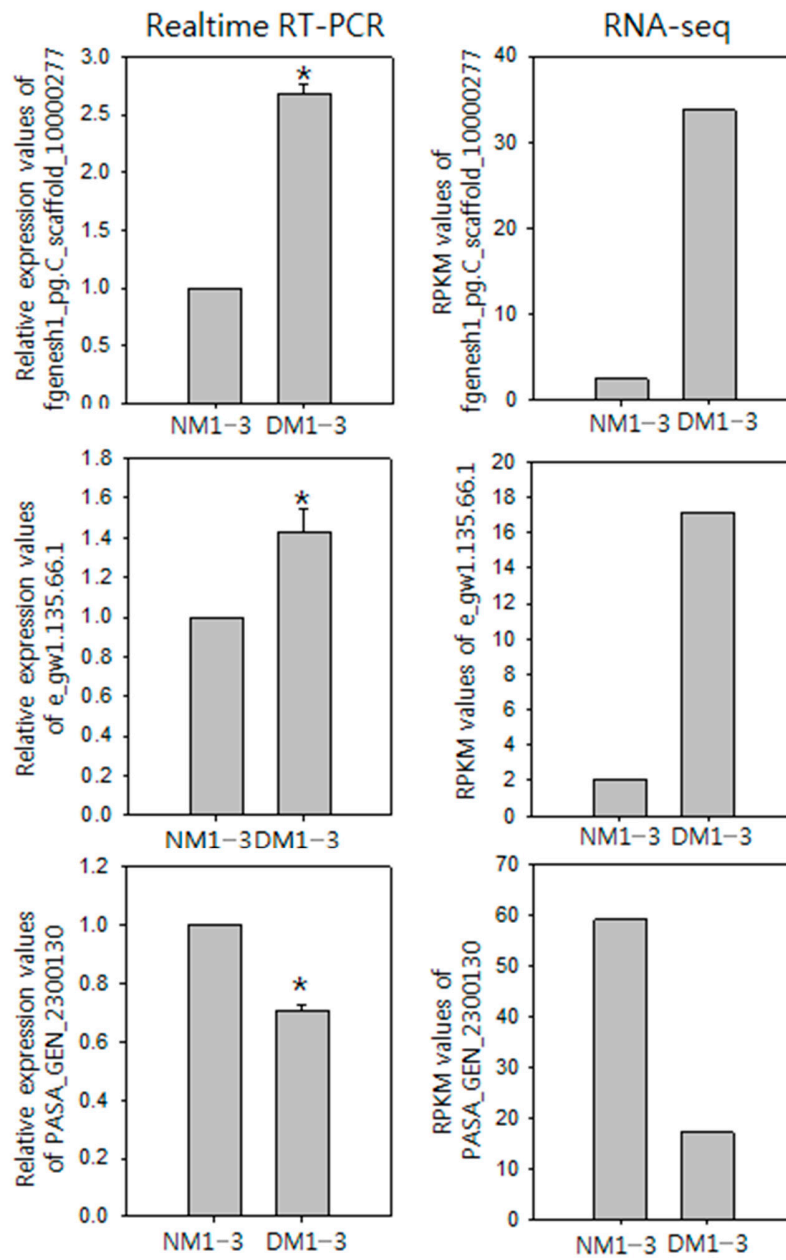


Figure S3. Real-time PCR validation. * represents differentially-expressed genes between normal morph (NM) and defensive morph (DM) with significance ($p < 0.05$). NM1-3 and DM1-3 represent the normal morph of 1-3 instar and the defensive morph of 1-3 instar, respectively.

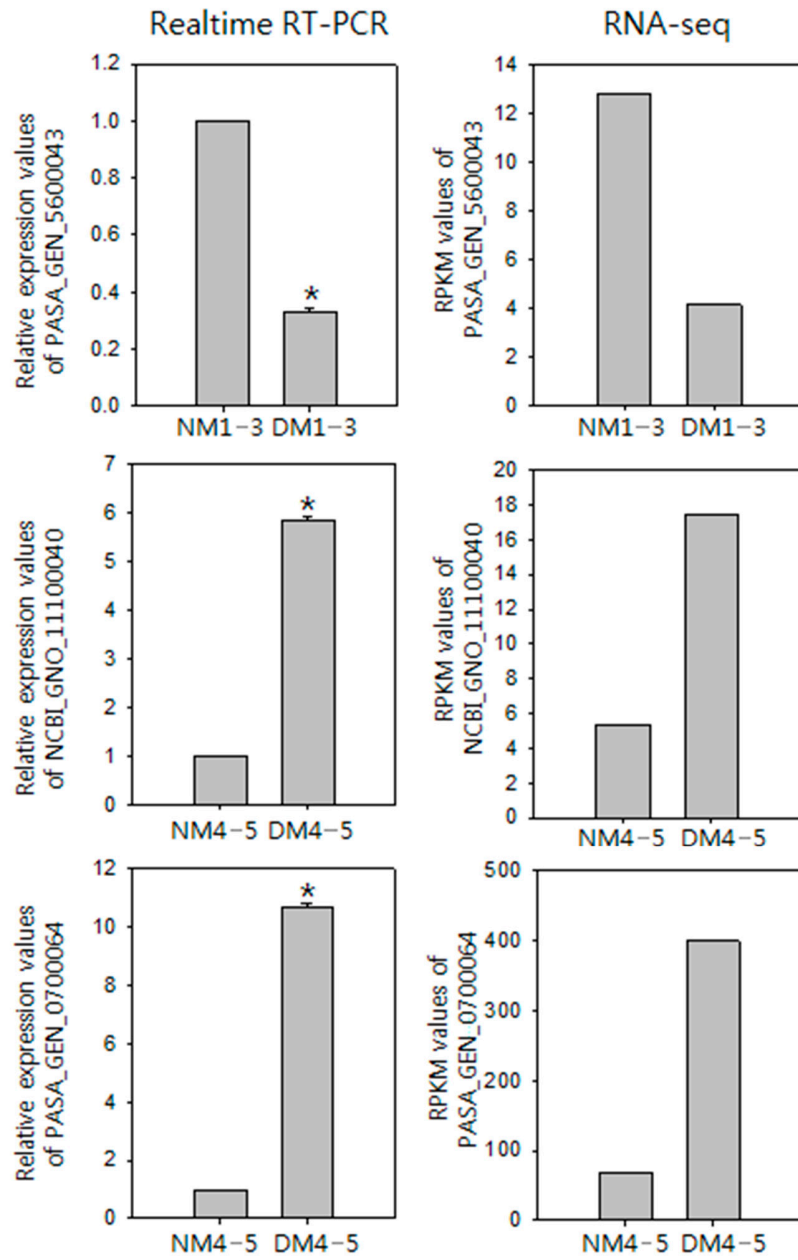


Figure S3. Real-time PCR validation. * represents differentially-expressed genes between the normal morph (NM) and the defensive morph (DM) with significance ($p < 0.05$). NM1-3, DM1-3, NM4-5, and DM4-5 represent the normal morph of 1-3 instar, defensive morph of 1-3 instar, normal morph of 4-5 instar, and defensive morph of 4-5 instar, respectively.

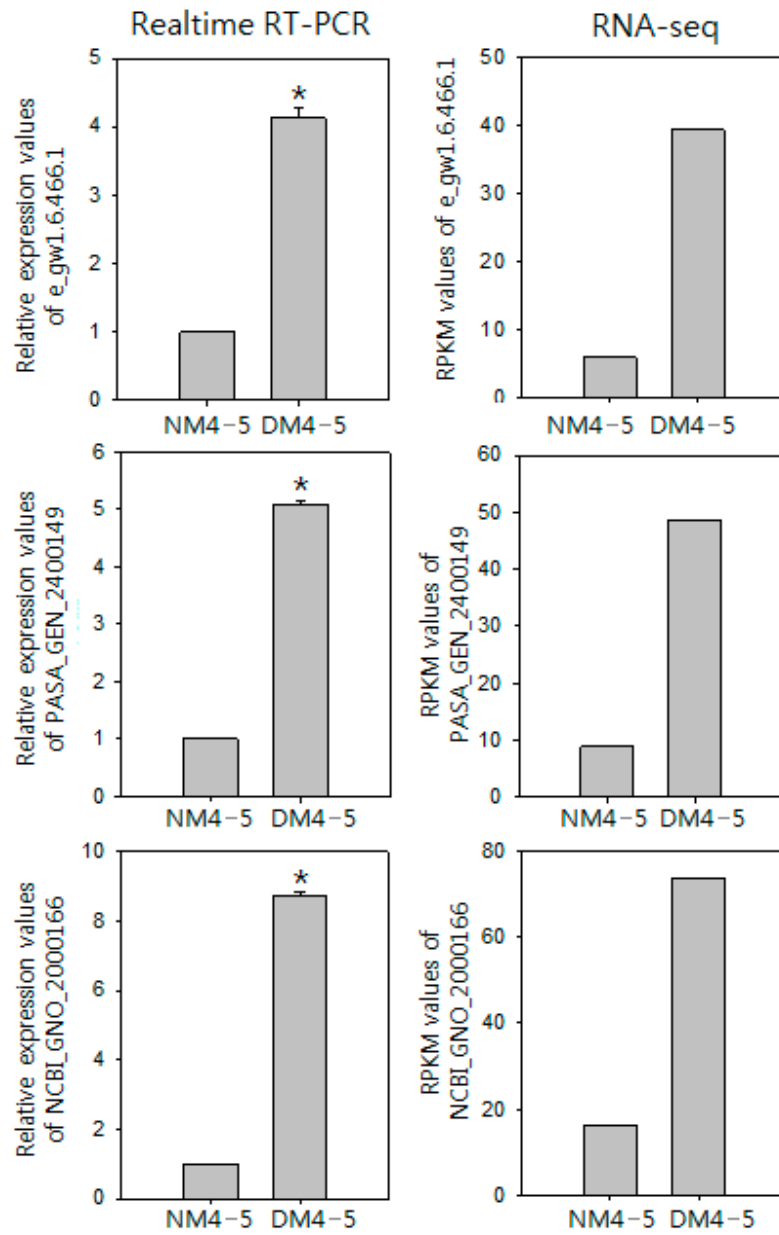


Figure S3. Real-time PCR validation. * represents differentially-expressed genes between normal morph (NM) and defensive morph (DM) with significance ($p < 0.05$). NM4-5 and DM4-5 represent the normal morph of 4–5 instar, and defensive morph of 4–5 instar, respectively.

Table S1. The List of the differently-expressed genes.

See Table S1.

Table S2. Differential expression of neurotransmitter receptors between normal and defensive morphs among developmental stages.

| | Gene ID | Late embryo | 1-3 instar | 4-5 instar | Descriptions |
|-------------------------|----------------------------------|-------------|------------|------------|---|
| Peptide receptor | e_gw1.91.62.1 | Down | - | - | Allatostatin receptor – <i>Periplaneta americana</i> ** |
| | fgenes1_pg.C_scaffold_1416000002 | - | - | Up | Allatostatin receptor – <i>Periplaneta americana</i> ** |
| | gw1.142.6.1 | - | Down | - | Ecdysis triggering hormone receptor subtype B - <i>Aedes aegypti</i> ** |
| | gw1.8.84.1 | - | - | Up | AGAP005229-PA (Myosuppressin receptor) - <i>Anopheles gambiae</i> ** |
| Amine receptor | e_gw1.93.6.1 | - | Down | - | Histamine-gated chloride channel subunit - <i>Culex quinquefasciatus</i> ** |
| | e_gw1.4.443.1 | Up | - | - | Octopamine receptor - <i>Apis mellifera</i> ** |
| Small molecule receptor | fgenes1_pg.C_scaffold_62000026 | - | Up | - | Nicotinic acetylcholine receptor subunit alpha8 precursor - <i>Gallus gallus</i> ** |
| | e_gw1.76.84.1 | - | - | Down | Nicotinic acetylcholine receptor subunit alpha3 - <i>Bombyx mori</i> ** |
| | NCBI_GNO_1500242 | - | - | Down | Kainate receptor subunit GluR5 and related subunits* |
| | fgenes1_pg.C_scaffold_183000025 | - | Down | - | Kainate receptor subunit GluR5 and related subunits* |
| | NCBI_GNO_8300009 | - | - | Up | Kainate receptor subunit GluR5 and related subunits* |
| | NCBI_GNO_1700146 | - | - | Up | Kainate receptor subunit GluR6 and related subunits* |

| | | | | |
|---------------------------------|------|------|----|---|
| NCBI_GNO_2300006 | - | Down | - | Kainate receptor subunit GluR7 and related subunits* |
| NCBI_GNO_8900043 | Down | - | - | Kainate receptor subunit GluR8 and related subunits* |
| NCBI_GNO_0900239 | - | - | Up | Kainate receptor subunit GluR9 and related subunits* |
| NCBI_GNO_18700035 | - | Up | - | Kainate receptor subunit GluR10 and related subunits* |
| NCBI_GNO_8300040 | - | - | Up | NMDA receptor subunit GluN1 and related subunits * |
| e_gw1.374.3.1 | Up | - | - | NMDA receptor subunit GluN2 and related subunits * |
| estExt_fgenes1_pg.C_1 130025 | - | - | Up | NMDA receptor subunit GluN2 and related subunits * |
| NCBI_GNO_3900088 | - | Up | - | AMPA receptor subunit GluR2 and related subunits * |
| PASA_GEN_1700130 | - | Down | - | Metabotropic glutamate receptor subunit GRM2 and related subunits* |

Descriptions of DEGs are referred from wFleaBase (*), uniprot (**).

Table S3. List of primers used for real-time PCR.

| Primers | Sequence (5'-3') | Descriptions |
|--------------------|-----------------------|--|
| GAPDH_F | GGTGCTGCCAGAACATTAT | Glyceraldehyde-3-phosphate dehydrogenase |
| GAPDH_R | GACAGCCTTGATTTTCGTCGT | |
| PASA_GEN_1800032_F | GAGCCATAGACGAGGTCTGC | Insulin-like*** |
| PASA_GEN_1800032_R | CAAGGACGACGACTGATTGA | |
| PASA_GEN_2600032_F | GTGGTGAAGACATGGACGTG | Molecular chaperone (DnaJ superfamily)* |
| PASA_GEN_2600032_R | CAATGGGCGGATCTTGACT | |

| | | |
|----------------------------------|----------------------|--|
| NCBI_GNO_18300040_F | GGTCTCCTCCTCATGTCCAA | Predicted lipoprotein* |
| NCBI_GNO_18300040_R | GACTGGATCAGCTTGTGGGT | |
| fgenes1_pg.C_scaffold_10000277_F | CAGCTCAGTTCAGACAAGCG | Calcium-responsive transcription coactivator* |
| fgenes1_pg.C_scaffold_10000277_R | GGTCCTGCCATCTGATTCAT | |
| e_gw1.135.66.1_F | CTCCGTTTGCCGTTATGTTT | Na+/dicarboxylate. Na+/tricarboxylate and phosphate transporters* |
| e_gw1.135.66.1_R | GCTGCGTTTGAGGCTATTTC | |
| PASA_GEN_2300130_F | AATGCTCGCGTTAATTGGAC | Permease of the major facilitator superfamily* |
| PASA_GEN_2300130_R | CAGCGGTCATTGCAGTTAGA | |
| PASA_GEN_5600043_F | CTGGCATTTCCTTTGGATGT | Transcription factor BLIMP-1/PRDI-BF1. contains C2H2- type Zn-finger and SET domains* |
| PASA_GEN_5600043_R | ACCAGAATGGTGAGAAACGG | |
| NCBI_GNO_11100040_F | GCTCAAGGCTGTGGAAGAAG | Monocarboxylate transporter* |
| NCBI_GNO_11100040_R | AACCAAATGGACGTATGGGA | |
| sPASA_GEN_0700064_F | AGAATCCCACGTCCAGTGTC | Peroxidase* |
| PASA_GEN_0700064_R | CGTTGGTGAGTAGCACGAGA | |
| e_gw1.6.466.1_F | CGGCTGTGAAATCAGTCTCA | Ankyrin repeat* |
| e_gw1.6.466.1_R | TGAGGATTTCTGCAGGCTTT | |
| PASA_GEN_2400149_F | ATTGTGATGAGCACAGCAG | Low-density lipoprotein receptors containing Ca ²⁺ -binding EGF-like domains* |
| PASA_GEN_2400149_R | CATCAACTGAACGCTGAGGA | |
| NCBI_GNO_2000166_F | ATGCTGTCACAGGAGAGGCT | ATP-dependent RNA helicase* |
| NCBI_GNO_2000166_R | TTCAGATGCGATAAGCGATG | |

Descriptions of DEGs are referred from wFleaBase (*), uniprot (**), interpro (***)).