

Supporting Information

TNF signaling mediates cellular immune function and promotes malaria parasite killing in the mosquito *Anopheles gambiae*

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Included supporting information

Supplemental Figures

Figure S1. Silencing efficiency of *Eiger*, *Grnd*, and *Wgn* genes in *An. gambiae*.

Figure S2. Expression of *Eiger*, *Wgn*, and *Grnd* in mosquito hemocyte populations.

Supplemental Tables

Table S1. List of primers used for gene expression and RNAi.

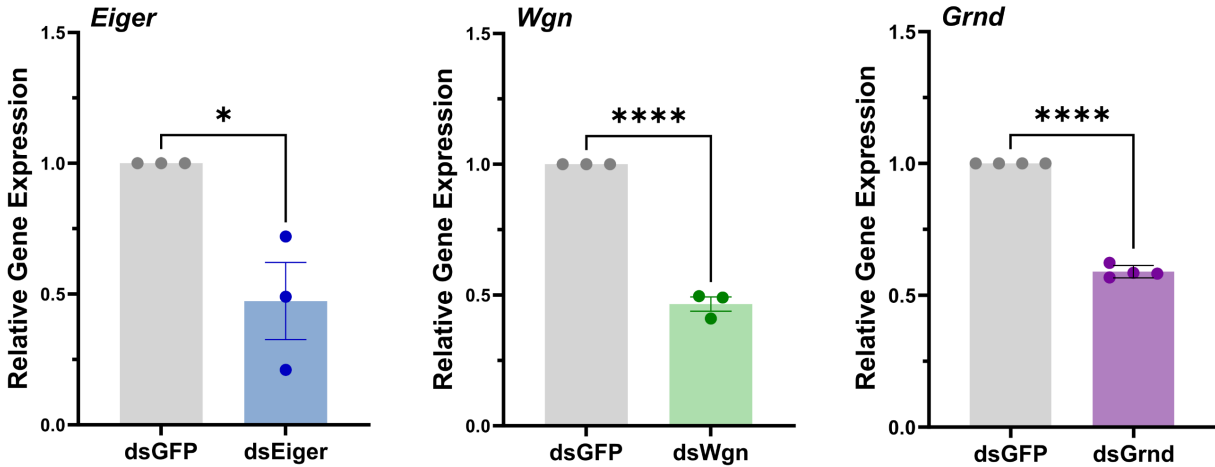


Figure S1. Silencing efficiency of *Eiger*, *Grnd*, and *Wgn* genes in *An. gambiae*. Naïve adult female mosquitoes were injected with dsRNA targeting *GFP* (control), *Eiger*, *Wgn*, or *Grnd*. Two days post-injection, whole-body mosquitoes (10-15 total) were collected for RNA extraction, followed by gene expression analysis using qPCR. Data from three or more independent experiment were examined for statistical significance using an unpaired student's t-test. Asterisks indicate significance (* $P < 0.05$, **** $P < 0.0001$).

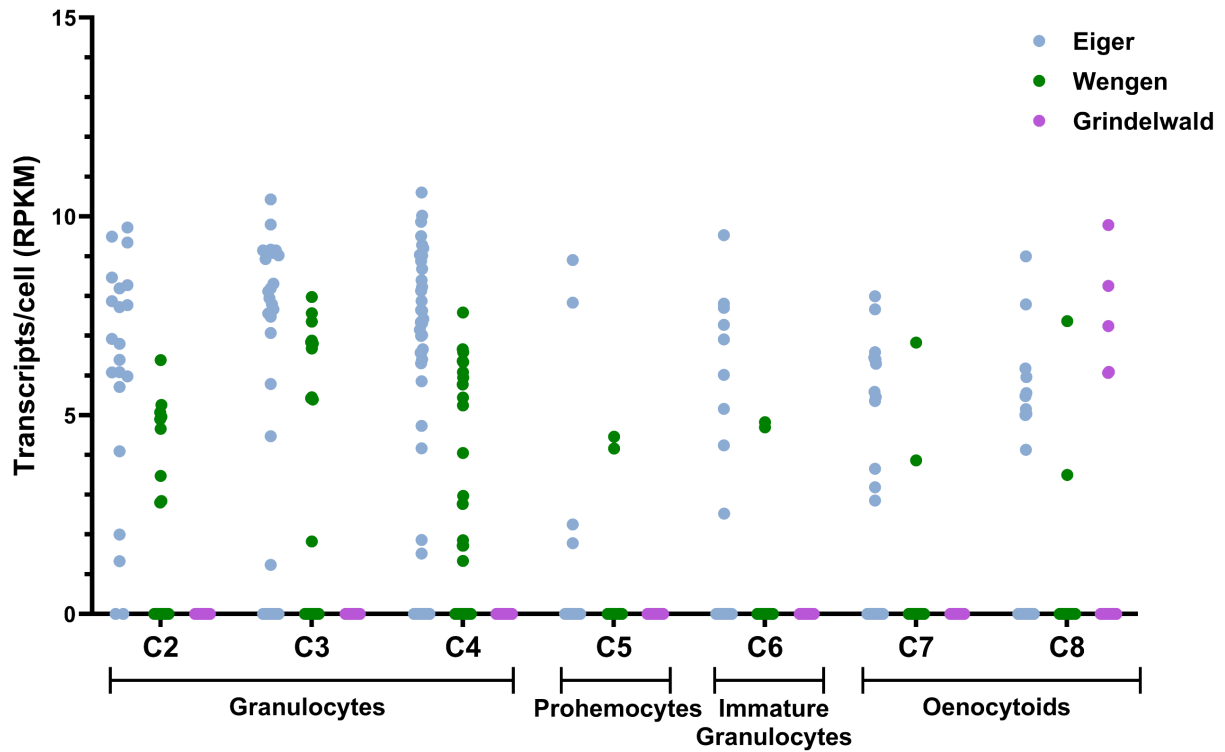


Figure S2. Expression of *Eiger*, *Wgn*, and *Grnd* in mosquito hemocyte populations. Using previously published single-cell RNA-seq data of *An. gambiae* hemocytes [22], the expression of *Eiger*, *Wgn*, and *Grnd* was evaluated for each mosquito immune cell subtype. Each dot represents a cell-specific gene expression value. RPKM, Reads Per Kilobase Per Million.

Table S1. List of primers used for gene expression and RNAi. Small letters indicate the T7 promoter sequence.

| <u>Gene</u> | <u>Forward (5'-3')</u> | <u>Reverse (5'-3')</u> |
|--------------------|-------------------------------------------|---------------------------------------------|
| AgGrnd-qPCR | CAAGGCGGTGCCGAAGAATG | GCTTTCCGTCGAATCTTCCG |
| AgGrnd-T7 | taatacgactcactatagggGTGCGTGTGTGTGTGTTTCAG | taatacgactcactatagggCGGACCCTGTTTCCTTCTTGT |
| AgEiger-qPCR | TCCGCTGGGATGTAGAAAATCG | GGCGTGGTGGTGCTGTGATG |
| AgEiger-T7 | taatacgactcactatagggCAATGAGCTGAACGCTGGAA | taatacgactcactatagggGGCTCGTTGATGGTAAGCTG |
| AgWgn-qPCR | GAGGAGATCCTGTGGGACTG | GCGTCAAAGTGCTTCTCGAT |
| AgWgn-T7 | taatacgactcactatagggAAGCATTATCGGCCAGCTC | taatacgactcactatagggGGGAAGCCAGATTTTGGATCTAG |