Supplementary Material

The in vivo dsRNA cleavage has sequence preference in insects

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Supplementary Figure 1. The REase gene expression level in dsEGFP and dsREase

treatments.



Supplementary Figure 2. Distribution of 19-25 nt and 18-30 nt small RNAs under

different treatment conditions.



Supplementary Figure 3. Statistics regarding small RNA type and amount at different sequence positions.

(A) Small RNA type in three different sequence position of ds*REase-*A, ds*REase-*B, and ds*REase-*C showed in Figure 1B. (B) Small RNA amount in three different sequence position of ds*REase-*A, ds*REase-*B, and ds*REase-*C showed in Figure 1B.



Supplementary Figure 4. Nucleic acid compositions of 5'- and 3'-ends cleavage sites in the top 1.0% of small RNAs after ds*REase* treatment.



Supplementary Figure 5. Relationship between GGU sites in the *REase* sequence and the *in vivo* processing model of ds*REase*. The graph of upper panel: x-axis represents the *REase* sequence, and the y-axis represents the depth of sequencing (amount of mapped small RNA). The sense chain is marked in blue, and the antisense chain is marked in red. The graph of lower panel: red arrow head represents GGU sites on the sense chain, red arrow head represents the GGU sites on the antisense chain.



Supplementary Figure 6. Numbers of different nucleotide compositions of 5'- and 3'-ends cleavage sites in the top 1.0% of small RNAs of *Ostrinia furnacalis* ds*EGFP*-720 and *Helicoverpa armigera* ds*EGFP*-720.