Supplementary Material

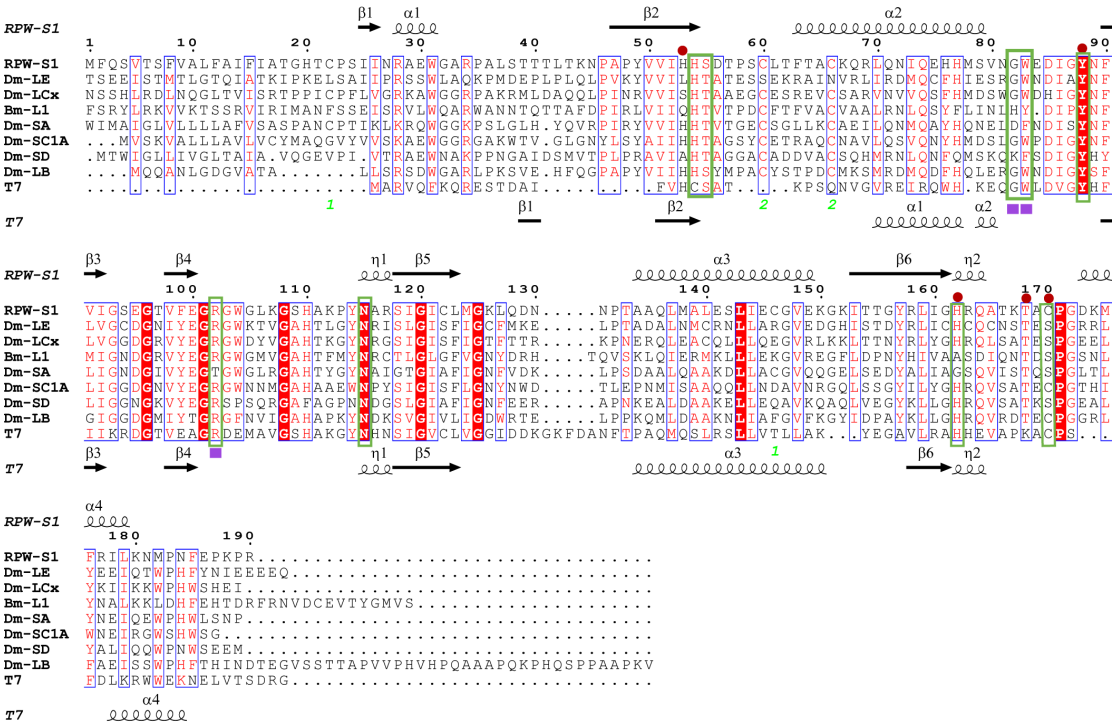

Fig S1: Multiple alignments on the amino acid sequences of RfPGRP-S1 with other insect PGRPs. Insect PGRPs, including Dm-LE (*Drosophila melanogaster*-LE, NP_573078.1), Dm-LCx (*Drosophila melanogaster*-LCx, AAM18530.1), Bm-L1 (*Bombyx mori*, AMR36560.1), Dm-SA (*Drosophila melanogaster*-SA, CAD89125.1), Dm-SC1A (*Drosophila melanogaster*-SC1A, CAD89162.1), Dm-SD (*Drosophila melanogaster*-SD, CAD89197.1), Dm-LB (*Drosophila melanogaster*-LB, AAG23731.1) and *Escherichia* phage T7 lysozyme (AAB32819.1) were retrieved from the Genebank: and are aligned. The five red dots indicate that the amino acids in the green boxes are required for Zn^{2+} binding and amidase activity of T7 lysozyme. Three pivotal amino acid residues for the discrimination of DAP-PGN from Lys-PGN are highlighted with the purple squares.

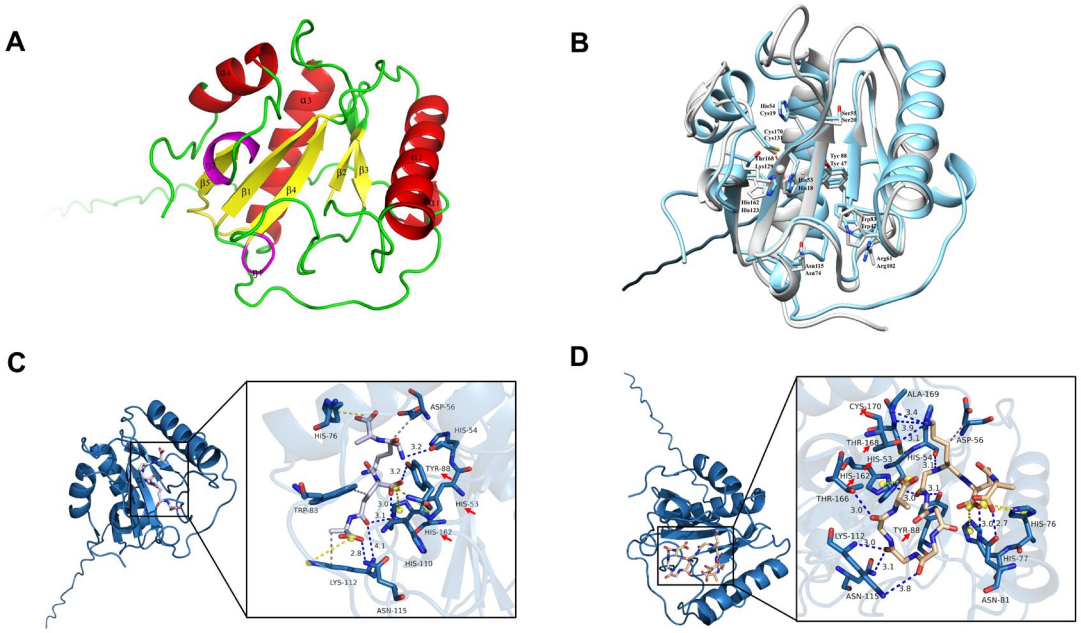

Fig. S2: Prediction of the 3D structure of RfPGRP-S1 and its binding with peptidoglycan. The computational procedures for protein structure prediction, molecular docking, and visualization are summarized as follows: Protein Structure Prediction: The three-dimensional structure of the target protein was generated using AlphaFold3 (A). The predicted model with the highest confidence score (pLDDT) was selected and used as the receptor for all subsequent docking simulations. Molecular docking analyses were performed with AutoDock Vina 1.2.3. The docking grid was defined to encompass the known active site, and the exhaustiveness parameter was set to 32. The resulting docking poses were analyzed based on their binding affinity (kcal/mol). All molecular graphics, including the visualization of docking complexes and analysis of non-covalent interactions (e.g., hydrogen bonds, hydrophobic contacts), were visualized by running UCSF Chimer 1.19. (B) Superposition of RfPGRP-S1(blue) and T7 lysozyme (gray, its accession number in the PDB database is 1LBA). Amino acid residues in the PGN-binding pocket of RfPGRP-S1 (blue) and T7 lysozyme (light gray) are represented as stick models. The amino acid residues of RfPGRP-S1 and T7 lysozyme that are involved in binding with PGN are located at the bottom of the L-shaped groove. The figures (C) and (D) show the different amino acid residues in the L-shaped binding domain of RfPGRP-S1 that interact with DAP-PGN and Lys-PGN, respectively. The red arrows in figure (D) indicate that all five conserved amino acid residues are involved in binding Lys-PGN.