

Molecular relatedness and diversity of insect antimicrobial defensin genesY I N Silva Gunawardene¹ and R S Dassanayake^{2*}¹*Molecular Medicine Unit, Faculty of Medicine, University of Kelaniya, Thalagolla Road, Ragama*²*Department of Biochemistry & Molecular Biology, Faculty of Medicine, University of Colombo, Colombo*

Insect defensins are cationic, inducible antibacterial peptides involved in humoral defence against pathogens. To study the origin, relatedness and diversity of these antimicrobial peptides, similar sequences from various genomic and protein databases were retrieved using well characterized *An. gambiae* defensin. These were found to be derived from the orders Diptera, Lepidoptera, Coleoptera, Hemiptera to Hymenoptera and also from diverse molecular contexts, such as fungi. Analysis of the retrieved sequences alignments revealed three well-defined regions, signal peptide, prodefensin and the mature defensin. Of these, former two deemed to be diverse. Six cystein residues of the defensin, which were found to be well conserved amongst diverse molecular taxa, diverged only in the lepidopteran at 2nd cystein residue. Further, the only examined lepidopteran and fungi defensins were seen to cluster together with the dipteran defensins. Furthermore, the hypothetical protein derived from fungi, *A. nidulans* formed an orthologous pair with the dipteran *Phlebotomus*. Similarly, the dipteran *Glossina* shared another defensin orthologous pair with the Hemiptera cluster of defensins. These imply that fungi and hemipteran defensin orthologous share well conserved immune functions with dipterans *Phlebotomus* and *Glossina* respectively. Cladogram topology revealed dipteran defensin to be diverse suggesting diverse immune mechanism amongst the dipteran insects. There were individual dipteran defensins which did not shuffle with any of the other clusters in phylogram and rather remained as genetically distinct entities and most of the latter were derived from the same species (e.g. *An. gambiae*) indicating their independent evolutionary origin. The defensin peptides of the order Coleoptera exhibited two distinct clades with mixture of different species. The two clusters of Coleopteran defensins showed ancestry to orders Diptera and Hymenoptera. An orthologous group was evident between the *Drosophila* and one of the Coleopteran clusters, representing species-specific expansions of the defensin in these two groups. The only Hemipteran cluster observed in the dendrogram implied closer phylogenetic origins to the other Coleopteran cluster. In conclusion, phylogenetic analysis indicated marked diversification of mosquito vector defensins relative to non-mosquito defensins, possibly implying the presence of finely tuned immune responses to multiple pathogenic environments distinct in these two groups of organisms. Investigations into molecular diversity of mosquito defensins in particular and insects in general will greatly facilitate insights into vector insect immunity and its relationship to insect-parasite interactions. This knowledge is essential for designing future strategies for controlling mosquito-borne diseases through genetic manipulations of vectors.

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