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[P54] X-ray crystal structure and binding studies of the Odorant Binding Protein 5 from the malaria vector *Anopheles gambiae*

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ABSTRACT

Anopheles gambiae is the primary mosquito vector responsible for the transmission of malaria, causing more than 1 million deaths each year. Like other insects, mosquitoes rely on olfaction to find mates, food and sources of blood meals. Odorant Binding Proteins (OBPs) that mediate the initial step in the transduction cascade of olfactory signals in insects have been suggested to play an essential role in the detection and transportation of semiochemicals to Odorant Receptors (ORs), and thus, they constitute promising targets for the design of new repellent/attractant molecules [1-4,7]. Therefore, a detailed knowledge of the 3D structures and functionality of OBPs may provide a valuable tool for the structure-based discovery of novel olfactory disruptors of insect host seeking behavior to be used in more effective mosquito control strategies.

Among a subset of 10 *Anopheles gambiae* OBPs that found to exhibit strong female-specific expression and so are likely to be involved in host-seeking bahavior [5], the Odorant Binding Protein 5 (AgamOBP5) displays the highest expression levels in the female antenna. Its expression levels are also appear to be affected by the circadian cycle as they dramatically reduced in dark (DD) compared to light dark (LD) circles [6].

Herein, we present the novel 3D crystal structure of AgamOBP5 at 1.43Å resolution (***Figure 1***). The AgamOBP5 structure comprising of six α-helices connected by 3 disulfide bridges, closely resembles the known structure of AgamOBP4 (73 % homology, 62.4 % identity). Although their structural similarity, Differential Scanning Calorimetry (DSC) studies at different pH values showed that these proteins exhibit different thermodynamic profiles. We found that AgamOBP5 is stable at 2 different pH values (pH 5 & pH 8) while AgamOBP4 demonstrates stability only at pH 8. It has been previously proposed that OBPs undergo pH-induced structural changes in the area of ORs (low pH) causing odor release [8]. Our findings suggest that the two proteins utilize different release mechanisms of odors.

Furthermore, fluorescence displacement experiments in the presence of various volatile compounds of natural and synthetic origin [7] indicate that AgamOBP5 and AgamOBP4 exhibit different binding affinities for certain molecules suggesting that they may recognize and bind different classes of odors and in all likelihood are involved in different pathways of host recognition. The binding specificity of AgamOBP5 in comparison to AgamOBP4 is underway.

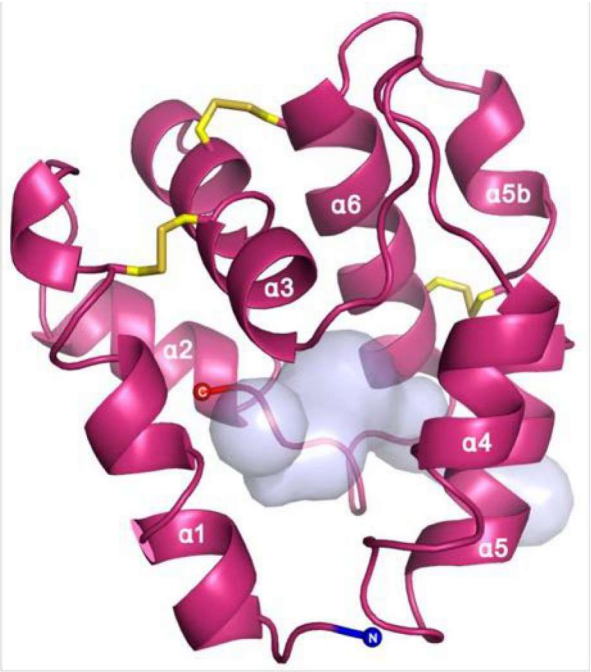

Figure 1. *Cartoon representation of AgamOBP5. The binding site is located at the center of a long hydrophobic tunnel represented as a surface. The site is accessible to the solvent via two protein mouths.*

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