MOLECULAR SEQUENCE AND CELLULAR LOCALIZATION OF AN ANTENNAL-SPECIFIC PROTEIN IN ADULT *LYGUS LINEOLARIS* (TARNISHED PLANT BUG)

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Abstract

The tarnished plant bug, *Lygus lineolaris* (Hemiptera, Heteroptera: Miridae) is a serious pest of cotton. The pests' ability to survive on a wide variety of host plant species, numbering at least 400, and its complex biology, involving transition through five different immature stages, are practical obstacles to integrated approaches for control of the pest. Trapping or monitoring for proper timing of insecticidal control is not simple because the behavioral cues, such as sex pheromones and food/oviposition-site odors, have not been identified for the plant bug. In order to develop timely and effective, yet environmentally sound, control measures for this pest, we have undertaken research to understand the physiological and molecular mechanisms of chemical reception in the plant bug.

The behavior of insects, including the plant bug, is largely a response to volatile chemical signals. Such odors are detected by the insect's antenna, specifically via the hairs or sensilla covering the antenna. Carrier proteins within the sensilla are thought to bind and transport the chemical signals to specific receptor cells. Our previous electrophysiological and morphological studies showed that sensilla with receptor cells for plant or insect odors appear during the final molt from fifth instar nymph to adult stage in the plant bug. We have utilized fifth instar nymphs versus adults as a developmental system to identify antennal gene products correlated with this transition to adult olfactory function.

Here we summarize results from an interdisciplinary approach, which characterizes a *Lygus* antennal-specific protein (LAP) of 15 to 16 kDa most likely involved in chemical reception in the plant bug. An antibody, which we previously developed for specific tagging of LAP, was employed to show that LAP was found in antennae of *Lygus* species but was not detectable in distant relatives such as *Podisus* or *Nezara* species. Immunocytochemical analyses localized LAP to the sensilla of the antenna. Immunogold labeling of thin sections revealed that LAP was associated with the extracellular, sensillar lymph. LAP was associated with specific subtypes of sensilla, specifically the multiporous olfactory sensilla. Using primers designed from LAP N-terminal amino acid sequence, we isolated and cloned the cDNA encoding LAP via reverse transcription and polymerase chain reaction. The full-length cDNA sequence indicated that LAP is related to Odorant Binding Proteins previously found in the fruit fly, *Drosophila melanogaster*. *In situ* hybridization of the LAP cDNA localized the mRNA for LAP to specific cells beneath the antennal cuticle near the sensilla. Thus, LAP is most likely synthesized in these cells and secreted into the sensillar lymph. As a whole, these physiological, structural, and anatomical studies indicate that LAP is an Odorant Binding Protein of the tarnished plant bug; as such, LAP is the first biochemical step in the pathway that recognizes behaviorally important olfactory signals, including mating pheromones and feeding attractants.

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