

# HISTOCHEMICAL STUDY OF LECTIN BINDING SITES IN FOURTH AND FIFTH INSTAR GYPSY MOTH LARVAL MIDGUT EPITHELIUM

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## ABSTRACT

There is evidence that the gypsy moth, *Lymantria dispar*, midgut epithelial brush border membrane has membrane-bound glycoconjugates, such as BTR-270 and aminopeptidase N (APN), which function as high affinity binding sites (receptors) for the insecticidal proteins produced by *Bacillus thuringiensis* (*Bt*). As gypsy moth larvae become older, they become resistant to the entomocidal activity of *Bt*, suggesting there may be a change in the expression of these toxin-binding glycoconjugates in older larvae.

To examine the possibility that the expression of cell surface glycoconjugates may be altered in older larvae, paraffin-embedded midgut tissues of fourth and fifth instar gypsy moth larvae were probed with a panel of lectins. Lectin binding to the tissue sections was assessed using fluorescence microscopy. Lectins are sugar-binding proteins that bind to specific carbohydrate structures in tissues. Specific and very small changes in the carbohydrate moiety of glycoconjugates attached to the midgut epithelial cells can be identified using lectins.

In this study, striking differences were found between fourth and fifth instar gypsy moth larvae with several lectins, including peanut, *Arachis hypogaea* (PNA), ricin, *Ricinus communis* (RCA<sub>120</sub>), and wheat germ, *Triticum vulgare* (WGA). In contrast to fifth instar larvae, cell surface molecules on the microvilli of fourth instar gypsy moth larvae did not interact with peanut (PNA) and ricin (RCA<sub>120</sub>) lectins, which have high affinity and specificity for terminal  $\beta$ -galactose residues on glycoproteins and glycolipids. In addition, the fourth instar microvilli and the basal lamina were labeled with WGA, but WGA lectin binding is restricted to the basal lamina in the fifth instar larval gut tissue. These results provide evidence that there are ontogenetic changes in the expression of specific carbohydrates on the surface of brush border microvilli during larval development that may have functional significance to the decrease in susceptibility to *Bt* insecticidal proteins in older insect larvae.