

DEVELOPMENT OF AN ARTIFICIAL DIET FOR THE GLASSY-WINGED SHARPSHOOTER

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ABSTRACT

The intent of this project is to develop an artificial rearing system for the glassy-winged sharpshooter (*Homalodisca coagulata*) (GWSS), the primary vector of Pierce's Disease (*Xylella fastidiosa*) (PD). In order to accomplish this, a diet delivery system will first be developed and then used to test artificial diets. Diet formulations will be based, in part, on previous studies performed by Cohen (2002) using GWSS, as well as on artificial diets developed for other Hemiptera (Mitsuhashi, 1979; Coudron *et al.*, 2002) and on the xylem chemistry of GWSS host plants (Andersen, *et al.*, 1992). Diets will be evaluated based on their effects on life history analyses, reproductive rate and intrinsic rate of increase of GWSS. Another aspect of our project involves investigating nitrogen source(s) for GWSS, as that may represent a nutrient limitation for xylem feeders. Two potential sources for nitrogen, i.e. proteins or peptides, will be studied by determining the fate of dietary proteins/peptides (Brandt, *et al.*, 2004) and the ability of salivary and midgut proteolytic enzymes to digest proteins/peptides (Wright, *et al.*, 2004). In this way, we will identify the role(s) proteins and peptides play in GWSS nutrition and their potential uses in artificial diet formulations.

INTRODUCTION

The formulation of an artificial diet for GWSS will greatly enhance the ability of researchers to rear this insect. Presently, the rearing of GWSS is labor-intensive and costly because of its dependence on the propagation of appropriate host plants, with researchers often needing to propagate several species of plants to enable them to rear GWSS under optimal conditions. The development of an artificial diet would likely be more cost effective and portable, increasing the availability of high quality insects for Pierce's disease researchers and decreasing the costs and time-constraints associated with maintaining the insect in culture. The increased accessibility of GWSS to researchers can lead to more rapid developments in novel control measures for this major vector of PD, with these new measures being directly applied by growers. Furthermore, the coupling of an artificial diet with a suitable delivery system can lead to an improved understanding of the relationship between GWSS nutrition and other PD-related issues (including GWSS' varying abilities to acquire/maintain/transmit infectious *Xf* under different circumstances, e.g., via artificial membranes vs. plants, Redak *et al.*, 2004). In addition, the diet delivery system alone would have other potential uses such as in studying the interactions between GWSS, *Xf*, and the host plant, as well as in testing potential anti-GWSS and anti-*Xf* control agents. This could be accomplished by incorporating into the feeding system: 1) selected host plant-associated compounds; 2) media containing the causative agent of PD (*Xylella fastidiosa*, *Xf*) (although some studies have suggested that *Xf* acquired via an artificial membrane by GWSS may not be infectious, Redak *et al.*, 2004); 3) control agents including anti-GWSS or -*Xf* compounds (such as proteins to be engineered into host plants to control either GWSS or *Xf*; Dandekar *et al.*, 2003; Lin, 2003; Meredith and Dandekar, 2003; Reisch *et al.*, 2003) or anti-GWSS microbials (Kaya, 2003; Mizell & Boucias, 2003). In summary, the development of an artificial diet and a corresponding delivery system for GWSS could lead to insights that can be used to generate improved methods for controlling GWSS and, therefore, Pierce's disease.

An important part of our project also involves gaining a better understanding of the digestive physiology of GWSS. This will be investigated by focusing on the role proteins and peptides play in GWSS nutrition, as these or similar compounds have been isolated from some xylem fluids (Cohen, 2002; Jain and Basha, 2003; Rep *et al.*, 2003). We will accomplish this by determining the extent to which GWSS can digest proteins and peptides, as well as elucidating the fate of specific ingested proteins in GWSS. This information will be directly used in the generation of an optimal artificial diet for GWSS. Furthermore, GWSS' ability to degrade proteins/peptides will also shed light on the degree to which GWSS can disable defensive proteins/peptides in plants, which is important when dealing with salivary enzymes that are secreted into plant tissues and could alter anti-*Xf* defense components (e.g., either naturally occurring or genetically engineered proteins/peptides; Lin, 2003; Meredith and Dandekar, 2003; Reisch *et al.*, 2003). This knowledge could be used when modifying target plants such as grapevines to improve their resistance against Pierce's disease (PD). Therefore, our investigation into nutritional requirements will not only aid us in the development of a suitable artificial diet for GWSS, but

will also provide insights into the potential efficacies of anti-PD plant modifications.

OBJECTIVES

1. Develop an artificial diet delivery system for rearing the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata*.
2. Formulate and evaluate an artificial diet for the development and reproduction of GWSS.
3. Investigate the utilization of proteinaceous components in the food stream of GWSS in order to refine and improve the artificial diet using physiological and proteomic/genomic approaches.

RESULTS AND CONCLUSIONS

This project has just been funded. Preparation of quarantine facilities is complete and the identification of insect cultures to be used in our studies is underway. The process to hire an additional researcher has been initiated. Preliminary experiments, in collaboration with Jones and Setamou at ARS in Weslaco, have demonstrated continuous feeding by adult GWSS for over 30 days on artificial diets presented through a specialized feeding tube. Additionally, differences in survival have been noted as a result of changes in amino acid concentration and composition within the diet.

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