2006 Beltwide Cotton Conferences, San Antonio, Texas - January 3 - 6, 2006

INNOVATIVE STRATEGY FOR DETERMINING STINK BUGS AND STINK BUG DAMAGE

Ahmad Khalilian
Clemson University
Blackville, SC
Sam Turnipseed
Clemson University
Blackville, SC
Young Han
Clemson University
Clemson, SC

Abstract

Stink bugs have become a major problem in current B.t. cotton varieties, because of reductions in insecticides that also provided coincidental control of these bugs. Current scouting and monitoring techniques for bug pests are time-consuming and unreliable. In addition, growers and consultants lack ready access to effective, efficient and affordable decision-making and management tools, making them particularly susceptible to unneeded insecticides use. The main focus of this study was to develop more accurate and effective diagnostic and management strategies for detecting stink bugs and their damage. An electronic nose (Cyranose 320) was used for this purpose and its performance was evaluated in laboratory and field conditions. The E-nose accurately predicted damaged bolls, interior walls of bolls and locks with lint and seed approximately 95 percent of the time under laboratory conditions. In a field setting the E-nose was able to identify stink bug damaged bolls 67% of the time. Under laboratory conditions, the E-nose identified presence of stink bugs 100 percent of the time. There was a strong correlation (R^2 = 0.9486) between the number of sting bugs in a sample and the Cyranose sensor response.

Introduction

Cotton (Gossypium hirsutum L.) is a major crop in the United States with an estimated production value of $6 billion. Pest and insect damage cost farmers millions of dollars each year. Recently, sucking bugs have become a major problem in current B.t. cotton varieties (e.g. Boll-Guard) because of reductions in insecticides that also provided coincidental control of these bugs. From 1995 to 2001, insecticide application for stink bug control increased from 0 to 4 million applications at a cost of $27 million (Williams, 2002b). In 2000, crop losses from these pests exceeded $50 million (Williams, 2002a).

The two types of sucking bugs that are detrimental to cotton are lygus and stink bugs. In South Carolina yield losses due to stink bugs are significantly higher than those caused by lygus (five times more in 2003). According to a 2002 survey by the South Carolina Cotton Board in which growers were asked to rank 60 production problems in order of importance, stink bug problem ranked number one. In addition, the southeastern region of USA experienced more than 65% of the stink bug related losses in 2003 (Williams, 2004). Therefore, the main focus of this study is on stink bugs and stink bug damage.

Current intervention thresholds for sucking bugs in cotton are based on field-sampling with a beat cloth (Greene, 2001a) to determine population levels and/or hand-picking of ½ grown bolls to assess internal damage in the form of punctures, warts and seed/lint staining. Since growers and consultants are reluctant to routinely use these cumbersome and time consuming techniques, applications of insecticides are often made before bugs are present or after economic damage has occurred. An effective sensing system which could access stink bug levels to improve management strategies is needed to reduce pesticide use and economic losses and potentially increase production is needed.

Recent advances in electronic nose technology has developed commercially available detectors that employ several sensing elements and pattern recognition software to detect volatile compounds at the parts-per-million (ppm) to parts-per-billion (ppb) threshold levels. Stink bugs release volatile compounds that can be sensed by biological organisms and sophisticated technology such as an electronic nose. Similarly cotton plants and bolls release chemicals as part of a defensive mechanism against pests. Plants, such as cotton, also produce chemicals that are used as a defense against damage from plant feeders and pathogens, and have evolved over millions of years of natural selection. These volatiles can be released at the site of damage as well as systemically throughout the plant.

These sensors could be used to detect the enzymes and/or the resultant gas evolution from the damaged seeds and boll tissue. The aggregation of sucking bugs within a field could be documented using advanced sensing techniques as well.

The objectives of this study were: A) to determine stink bug presence/damage using an electronic nose; and B) to determine stink bug population densities with E-nose.

Materials and Methods

Chemical Sensor (E-nose)

A commercially available portable electronic nose (Cyranose 320) made of an array of carbon polymers was used in this study. With this system, an air stream is drawn across the array of 32 sensors and the change in resistance to the sensors is measured. The resistance change creates a "smellprint" for the compounds. When trained, the Cyranose 320 can accurately predict the presence of stink bugs or plant compounds that correspond to those given off by the plant when damaged. Before the Cyranose 320 can be used to identify the presence of stink bugs, stink bug damage, or plant damage it has to be trained.

Figure 1- Cyranose 320

Assessment of Boll Damage

Forty cotton bolls were caged in the field using 470-mL Styrofoam cups with nylon stockings stretched over the outside of the cup. Rubber bands were used to hold the cups to the plant as well as securing the nylon stockings over the top of the cup. Twenty were marked as undamaged (red) and 20 were marked as damaged (black). The bolls were approximately the size of a quarter which is the same size that a cotton scout will look at when determining damage. In the cups marked 'damaged' one stink bug was added to each cup. The stink bugs were allowed to feed for four to five days to ensure that the volatile chemicals were released by the plant. The bolls were removed from the plant and 10 samples from each group of 20 were used for training of the C-320. The remaining bolls were used to see if the C-320 could correctly identify the difference between damaged and undamaged bolls. Also, the C-320 was used to identify damaged locks with lint and seed and interior walls of bolls.

Verification of Stink Bug Compounds Using Cyranose 320

Known compounds from stink bugs (trans-2-decenal and trans-2-octenal) were purchased and were compared to the volatiles released by stink bugs. Each compound was diluted in water at concentrations of 0.67, 1, 1.33, 2, and 4 mg/L. A 0.5-mL sample from each concentration was put into a 250-mL Erlenmeyer flask for testing with the C-320. These corresponded to 1.3, 2, 2.65, 4, and 8 µg/L. The similar techniques were used to identify smellprints of the stink bugs.

Stink Bug Population Densities

The C-320 was used to determine the stink bug population densities. Different numbers of stink bugs (1 to 10) were placed into the 250-mL flask and the smellprints for each group were identified using the E-nose. Three samples were taken with the C-320 for each number of insects. The C-320 was also put through a series of caged field tests to see if it could determine the number of stink bugs present in a contained outdoor environment. Sixteen field cages (1.2 m wide X 1.2 m deep X 1.5 m high) covered with an insect mesh were placed in a cotton field.

Results and Discussion

Table 1 shows the prediction percentages of the C-320 compared to the actual boll condition looking at the whole boll, boll wall, and lint. The E-nose accurately predicted damaged bolls, interior walls of bolls and locks with lint and seed approximately 95 percent of the time.

Table 1. Prediction of damaged and healthy boll materials using C-320

Boll Material	Actual Condition	E-Nose Prediction		
		Right	Wrong	Unknown

Whole Boll	Good	80%	15%	5%
	Damaged	90%	5%	5%
Boll Wall	Good	95%	0%	5%
	Damaged	95%	0%	5%
Lint	Good	80%	5%	15%
	Damaged	100%	0%	0%

The C-320 was able to verify the results of Gilby and Waterhouse (1964) that trans-2-decenal and trans-2-octenal are present in the volatile chemicals associated with stink bugs. The best correlation to stink bug response and tran-2-decenal was at a concentration of 1.3µg/L (Figure 2). Similar responses were obtained with trans-2-octenal was at a concentration of 2µg/L. The sensor response of the C-320 showed similar magnitude and identity to trans-2-decenal and trans-2-octenal at these concentrations. Only five sensors (#18, 20, 26, 28, and 29) responded to both chemicals and stink bugs with peak magnitude of 5000 ΔR/R. However, there were numerous cases that the response from sensor number 18 was not significant.

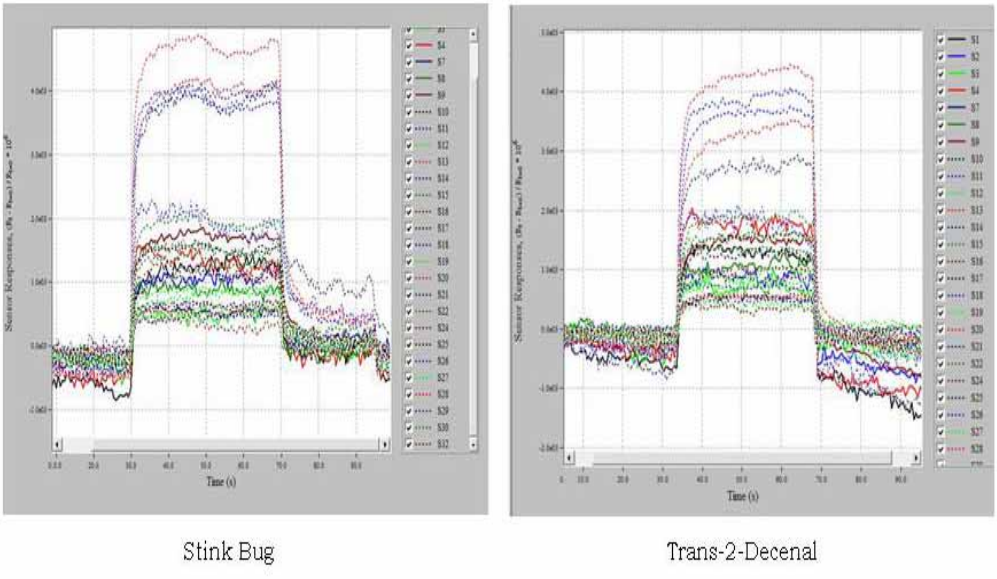

Figure 2. C-320 Response of Stink Bug Compared to Trans-2-Decenal

Under laboratory conditions, the E-nose identified presence of stink bugs 100 percent of the time. The magnitude of the C-320 sensors response increased with the number of stink bugs. Another important discovery was the development of a curve that correlates the sensor response of the C-320 to the number of stink bugs present. Only the responses from the four major sensors (sensor numbers 20, 26, 28, and 29) were used in the development of the curve. The maximum response for each of the four sensors was recorded. The three samples were average to give the average sensor response for the specified number of insects. This was plotted to give the stink bug curve. There was a high correlation (R^2>0.94) between the sensor response and the number of stink bugs.

After averaging the four sensors output for each specified number of insects the Average Stink Bug Curve was developed and fit with a 95% confidence interval (Figure 3) There was also a high correlation

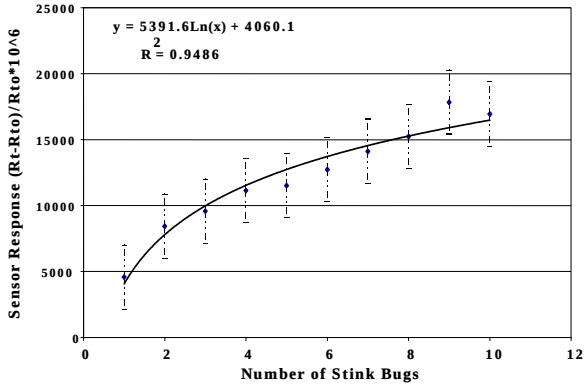

Figure 3. Correlation of stink bug population densities with C-320 sensor responses.

(R^2=0.9486) between sensor response and the stink bug population densities. The process of determining the number of stink bugs present under field conditions did not show promise under field conditions.

Conclusions

- The Cyranose 320 showed extreme promise as an instrument in determining stink bug damage by external properties.
- The E-nose accurately predicted damaged bolls, interior walls of bolls and locks with lint and seed approximately 95 percent of the time.
- Under laboratory conditions, the E-nose identified presence of stink bugs 100 percent of the time. There was a strong correlation (R2 = 0.9486) between the number of sting bugs in a sample and the Cyranose sensors response. However, utilizing insect cages under field conditions, the E-nose failed to determine stink bug population densities accurately.
- Using C-320 the volatile compounds given off by stink bugs were verified to be trans-2-decenal and trans-2-octenal. Only four sensors, out of 32 available, responded to volatile chemicals produced by bugs. The same sensors showed identical responses (smell prints) to trans-2-decenal and trans-2-octenal as compared to those obtained from sting bugs.

Acknowledgements

The authors acknowledge the support of the South Carolina Cotton Board, EPA, and Southern IPM.

Disclaimer

Mention of a trade name does not imply endorsement of the product by Clemson University to the exclusion of others that might be available.

References

Gilby, A. R., D. F. Waterhouse. 1964. The composition of the scent of the green vegetable bug, Nezara viridula. Division of Entomology, C.S.I.R.O., Canberra, Austrilia. June 16, 1964.

Greene, J. K., G. A. Herzog, and P. M. Roberts. 2001a. Management decisions for stink bugs, pp. 913-917. *In* C. P. Dugger and D. A. Richter [eds.], Proceedings, Beltwide Cotton Production Research Conferences, January 2001, Anaheim, CA, National Cotton Council of America, Memphis, TN.

Williams, M. R. 2004. Cotton insect losses – 2003. Proceedings, Beltwide Cotton Production Research Conferences, January 2004, San Antonio, TX.

Williams, M. R. 2002a. Cotton insect losses - 2000, [web page]. *In* J. McRae and D. A. Richter [eds.], Proceedings, Beltwide Cotton Production Research Conferences, January 2002, Atlanta, GA, National Cotton Council of America, Memphis, TN.

Williams, M. R. 2002b. Cotton insect losses - 2001, [web page]. *In* J. McRae and D. A. Richter[eds.], Proceedings, Beltwide Cotton Production Research Conferences, January 2002, Atlanta, GA, National Cotton Council of America, Memphis, TN.