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Journal of Bionic Engineering Suppl. (2008) 172-180

## **Identification of Stink Bugs Using an Electronic Nose**

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#### **Abstract**

Stink bugs are recognized as pests of several economically important crops, including cotton, soybean and a variety of tree fruits. The Cyranose 320 was used for the classified investigation of stink bug. Stink bugs including males and females of the southern green stink bugs, *Nezara viridula*, were collected from crop fields around College Station, TX. Results show that the released chemicals and chemical intensity are both critical factors, which determine the rate that the Cyranose 320 correctly identified the stink bugs. The Cyranose 320 shows significant potential in identifying stink bugs, and can classify stink bug samples by species and gender.

Keywords: electronic nose, detection, stink bugs, gender, species

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#### 1 Introduction

"Electronic noses" are instruments that mimic the sense of smell. Consisting of olfactory sensors and a suitable signal processing unit, they are able to detect and distinguish odors precisely and at low cost. This makes them useful for a variety of applications in the food and pharmaceutical industry, in environmental control, and clinical diagnostics and more. Many insects show a high sensitivity and selectivity for specific plant odors, and are able to perceive volatiles released by damaged plants in order to find food sources or mating partners<sup>[1]</sup>. In order to use the highly developed olfactory sense of insects for analytical purposes, the "biological nose" of insects has to be combined with some electronic instrument via a bio-electronic interface to yield an "electronic nose". The development of electronic nose technology in recent years has stimulated interest in the use of characteristic volatiles and odors as a rapid, early indication of deterioration in cereal grain quality<sup>[2]</sup>. The potential for rapid and remote grain classification and future prospects for the use of such technology as a major descriptor of quality are discussed. Börjesson et

al. [3] used an electronic nose to classify grain samples based on neural network analysis. The samples were divided into one of four odor classes and used to predict the degree of moldy/musty odor. The samples were also assigned score by two grain inspectors. Headspace samples from heated grain were used to describe their intensity of moldy/musty odor. These values exceeded the infrared detector monitoring CO<sub>2</sub>. The sensor signals were evaluated for corresponding percentages of agreement between two grain inspectors with a pattern-recognition software program based on artificial neural classification of the grain. Wheat at five storage ages and at 15 degrees of insect damage were evaluated and classified by the static-headspace sampling method using an electronic nose (E-nose). The results obtained indicated that the E-nose could discriminate successfully among wheat of different age and with different degrees of insect damage<sup>[4]</sup>.

In addition to grain, detection of insect infestations in crops in field is an application for E-nose. Rains *et al.*<sup>[5]</sup> reported the insects release chemical containing rich bio-information which can be used to develop a device for detection of insect odors. The chemical

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released is specific to the type of insect attacking the plant. Henderson et al. [6] used a commercially available electronic nose (Cyranose 320) to develop effective and affordable tools for detecting stink bugs and stink bug induced damage in cotton production under laboratory and field conditions. The volatile compounds produced by stink bugs were identified as trans-2-decenal and trans-2-octenal. The E-nose was trained to identify stink bugs' (presence) smell prints. Under laboratory conditions, the E-nose identified presence of stink bugs 100 percent of the time. There was a strong correlation ( $R^2$  = 0.95) between the number of sting bugs in a sample and the response of Cyranose sensors. Under field conditions, the E-nose was able to identify stink bug damaged bolls 67% of the time. Recent studies have indicated that many insect species have the ability to recognize volatile chemical compounds associated with food or other resources. These insects present a novel approach to volatile chemical detection that could provide a highly sensitive, inexpensive, flexible, and portable sensor. One characteristic of insects that makes them desirable as a potential chemical detector is their ability to detect extremely low levels of chemical compounds. A parasitoid wasp, Microplitis croceipes, was used as the model insect for determining the threshold of response to four compounds: 3-octanone, a compound found in many fungal pathogens; myrcene, a volatile constituent released by cotton plants fed on by cotton bollworms; and putriscene and adaverine, two products of the breakdown of dead animal protein by microorganisms<sup>[7]</sup>. Compounds causing off-odors in foods can be measured using gas chromatography and mass spectrometry<sup>[8–10]</sup>.

Henderson *et al.*<sup>[5]</sup> and Rains *et al.*<sup>[7]</sup> reported the feasibility of E-nose to detect stink bugs. However, there was no information available using E-nose to discriminate the gender and species, which is critical for the effective application of E-nose to pest detection and crop protection in the field. Therefore, the overall objective of this study was to determine the feasibility of detecting gender and species of stink bugs utilizing an E-nose.

#### 2 Material and method

#### 2.1 Cyranose 320-E-nose

The Cyranose 320 (Model 40101, Cyrano Sciences

Inc., Pasadena, California) is a commercially available, portable electronic nose made of an array of carbon polymers and is capable of simulating the human olfactory sense. 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. The Cyranose 320 also proved to be very easy to operate and train and provided a substantial software package of analytical tools for developing methods for detecting volatile chemicals. The Cyranose is reliable up to that limit of detection and could be used for accurately detecting odors when properly trained.

Before the Cyranose 320 can be used to identify the presence of insects, it has to be trained. Additionally, it is important to develop a method specific to the application for which the C-320 will be used. Seven factors were determined in order to create an accurate method. The details are given as follows:

- (1) The system set up was accomplished by connecting the C-320 to a PC using a serial (RS-232) cable.
- (2) The substrate temperature was typically set as low as possible, but at least 7 °C higher than the highest expected ambient temperature during normal operation.
- (3) The pump speeds for baseline and sample draw(s) were set to medium; however, a high setting was used for all purges.
- (4) The charcoal filter was used for all tests in the lab and in the field to eliminate the variability from organic contaminants.
- (5) Baseline purge, sample draw(s), and purge(s) times were setup as shown in Fig. 1.
- (6) Digital filtering smoothes the resistance reading from the sensors. Digital filtering was set to "ON" to improve the signal to noise ratio.
  - (7) Collecting data and reviewing the C-320 output.

#### 2.2 Stink bugs

Male and female stink bugs of the southern green stink bug, *Nezara viridula*, were collected around College Station, TX as shown in Figs. 2 and 3. At least two of the adults were very soft, likely to be very young, and not willing to release the alarm pheromone. Older stink bugs released the alarm pheromone (i.e., defensive secretion) when handled.

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