

# Electroantennogram Obtained from Honeybee Antennae for Odor Detection

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**Abstract.** Electroantennogram and spiking trains were obtained from honeybees' antennae for detection using an apparatus described in this paper. Different odor stimuli were applied on the antennae and got different EAG responses. The spikes of neurons were observed with this system too.

**Keywords:** Electroantennogram; EGA; Insect antenna; Honeybee; Stimulus.

## 1 Introduction

Gas detecting technology has a variety of application, the researches main focus on the gas sensors. But most of them are not sensitive enough, and it may take a long time to be as sensitive as a dog or an insect. Insects' antennae are very sensitive, can diagnose the tiny odors within a complex chemical background. For example, Honeybees had already been used in the airport as detectors to search the explosive. So the scientists begin to extract the signals from the antennae of the honeybees with the electrodes [1][2].

The sum of the responses of thousands of olfactory receptor neurons on insect antennae comprise the whole antennal response to odor is named as the electroantennogram (EAG). Ever since it was invented by Schneider in 1957, EAG recording has become one important techniques in insect olfaction and pheromone bioassay research [3]. After olfactory training, the electrophysiological data reveal that training to different odors induced different effects on the antenna sensitivity. These effects were found only in the learners' group [4].

The goal in this study was to build up an EAG detecting system to detect the different response of the honeybees to different stimulus for further odor discrimination or recognition.

## 2 Experiment

### 2.1 Experimental Animals

A Chinese honeybees (*Apis cerana*) hive is kept in the institute, and feed with honey in the winter. The adult drones were collected from the top of the hive or outside of the hive, and kept in Pasteur pipettes.

## 2.2 Set-Ups and Operation

The set-ups consists of 4 parts as shown in Fig.1: micro-manipulators, a differential pre-amplifier (ADA400A, Tektronix Inc., USA), a stimulus application system and an oscilloscope (TDS5054B, Tektronix Inc., USA). The diagram of the differential pre-amplifier is shown in Fig.2. Artifacts caused by background signals in the immobilized bees were greatly reduced by thoroughly designed faraday cage. The antennae were cut off from drone together with its head using micro-scissors, to keep the antennae to be alive for longer time. Clip off a few segment from the tip of the antennas. On the micro-manipulators, antennae were connected to one Ag/AgCl recording electrode by the electrically conductive gel and the head was connected to another Ag/AgCl electrode as the reference. If the resistance between the two electrodes was below 2 MOhm, the contact of the electrodes was sufficient. The surface of the manipulator is covered by the gold film to increase the connection. We used multiple antennae in parallel to improve signal-to-noise ratio in EAG responses [5]. There were two tubes in the stimulus set-up; each was connected with one conical flask, and two tubes join together to be a mix-odor tube. The mix-odor tube is directly face to the antenna, the distance between them is about 1cm. A flow meter was adapted to measure and control the flow. The flow was usually adjusted about 0.6L/min. Use a switch pedal to apply test puff. The sample rate of the oscillator was set as 5K sample/s to continue recording 40s by one channel, while the other channel was used to mark the starting and the end time of the stimulus synchronously. Up to now, two kinds of the gas were used to stimulate the antenna in the experiments. One is saturated honey vapor in the top of the conical flask, and the other is water vapor. The gas kind can be easily extended by adding the conical flasks.

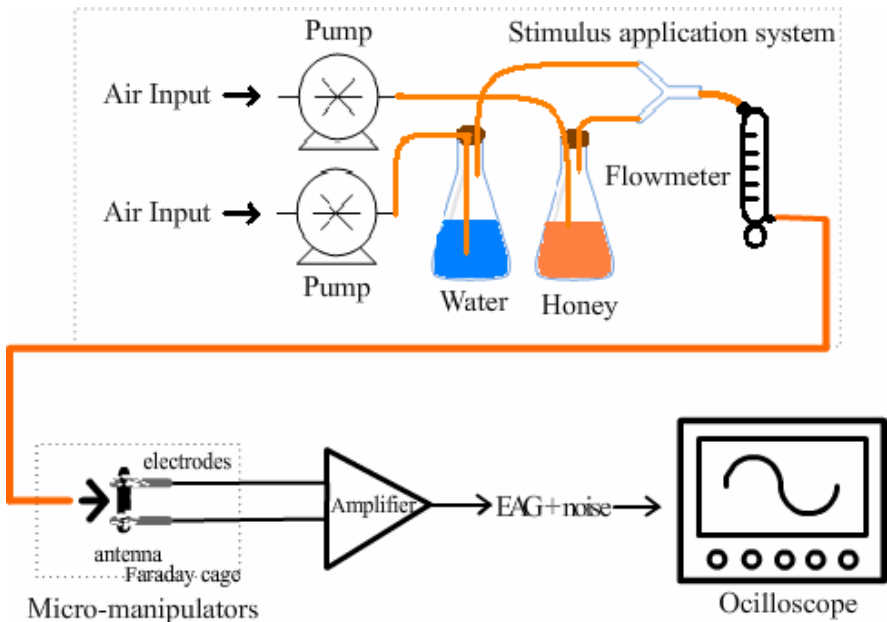


Fig. 1. Experimental apparatus

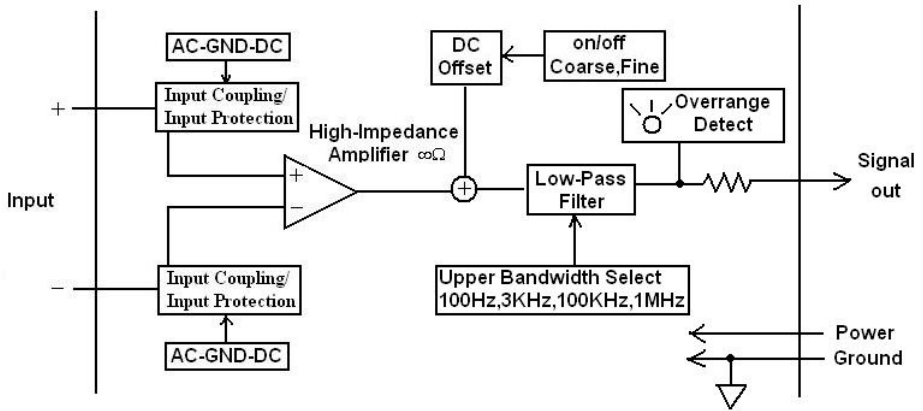
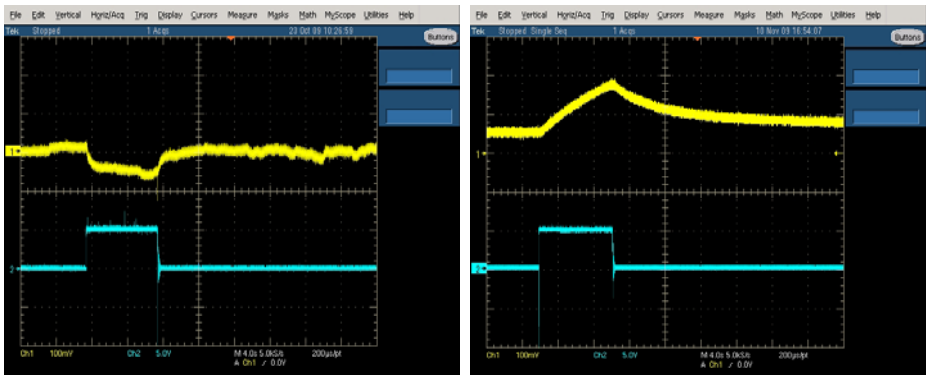


Fig. 2. Diagram of the Differential Preamplifier

### 3 Results and Discussion

Typical EAGs corresponding to water and honey vapors are shown in Fig.3. The blue line marks the duration of the stimulus, while the yellow line (above) is the response of the antenna to the different stimuli. Without any training on the honeybee, the EAG responses were not guaranteed when a stimulus applied. The rate of obtaining EAG is 49.5% for honey vapor, and 94.1% for the water vapor.

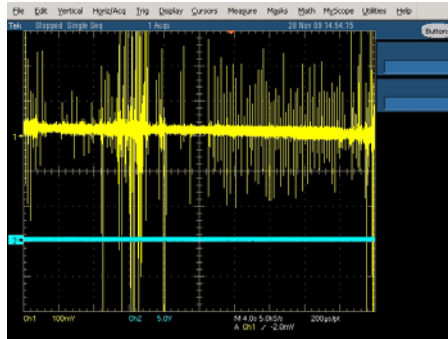
Besides the EAG waveforms, the spiking trains of the antenna as shown in Fig.4. They may provide more information for odor discrimination for further study.



(a)

(b)

Fig. 3. (a) The EAG of the honey vapor (b) The EAG of the water vapor. The blue lines (below) mark the start and the end of the stimulus, the yellow lines (above) are the response of the antennae to different stimuli.



**Fig. 4.** The spiking train of an antenna

## 4 Summary and Discussions

In order to utilize insect antenna as an ultra-sensitive sensor for odor discrimination, an EAG measurement system was built up. Based on the set-up, the EAG signals of the honeybees' antennae corresponding to different odor stimuli were recorded. Based on the EAG datasets collected, the experiments for odor discrimination will be carried out.

The spiking trains of the honeybee antenna were obtained by carefully adjusting appropriate parameters of the system. There should be more detailed odor information hidden within them in comparison with EAG. Hopefully, the spiking trains corresponding to different odor stimuli will provide efficient information for odor recognition.

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## References

- [1] Barsan, N., Koziej, D., Weimar, U.: Metal oxide-based gas sensor research: How to? *Sensors and Actuators B: Chemical* 121, 18–35 (2007)
- [2] Harris, S.: The honey trap, *Engineering & Technology*, pp. 24–26 (December 2007)
- [3] Schneider, D.: Elektrophysiologische Untersuchungen von Chemound Mechanoreceptoren de Antenne des Seidenspinners *Bombyx mori* I. *Zeitschrift für vergleichende Physiologie* 40, S8–S41 (1957)
- [4] Jong, R.D., Pham-Delègue, M.-H.: Electroantennogram responses related to olfactory conditioning in the honey bee. *Journal of Insect Physiology* 37, 319–324 (1991)
- [5] Park, K.C., Baker, T.C.: Improvement of signal-to-noise ratio in electroantennogram responses using multiple insect antennae. *Journal of Insect Physiology* 48, 1139–1145 (2002)