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Bioelectronic Artificial Nose Using Four-Channel Moth Antenna Biopotential Recordings

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Abstract— **The use of insect antennae as an odor sensor array was evaluated as a means to advance the current capabilities of "artificial nose" technology. A given species is highly sensitive to odors of survival interest (e.g. species-specific pheromones), but also to a broad range of other natural and anthropogenic compounds. The sensitivity of the antennae to some odors extends to the parts per billion range [1]. In contrast, the best current artificial nose technology is able to detect compounds in the parts per million range. Here, a system designed to utilize four antenna biopotential signals suitable for field use and a computational analysis strategy which allows discrimination between specific odors, and between odor and background or unknown compounds, with high fidelity and in real time, is described. The automated analysis measures three parameters per odor response. Following a training period, a K nearest-neighbor (KNN) approach is used to classify an unknown odor, assuming equal prior probabilities. The algorithm can also declare an odor as "unknown". System responses to single filaments in an odor plume can be analyzed and classified in less than one second.**

I. Introduction

Detection and identification of chemical compounds have many potential applications in military, industrial, clinical and research areas. A device used to detect and discriminate among airborne volatile compounds (i.e. odors) is generally referred to as an artificial nose. As the name implies, artificial noses assume a bio-mimetic theme, generally consisting of an array of differentially selective sensors whose output is interpreted by a computer. A variety of computing schemes have been applied to the classification problem that arises, a common method being the K-nearest neighbor technique (KNN) [2]. Fig. 1 illustrates the concept of distinguishing odors from the unique response pattern available from an array of different sensors. Such arrays have been constructed using several sensor types. [3] Sensors that change in resistivity in response to odorants include conducting polymer, metal oxides, carbon black/polymer, intrinsically conducting polymers. Devices that detect minute changes in mass due to adsorption of various compounds include surface acoustic wave and quartz crystal devices.

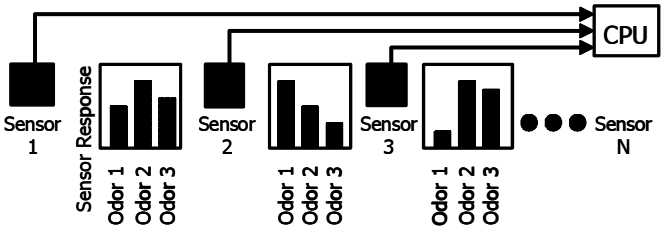

Fig. 1

Other sensors include coated optical fiber sensor arrays, and porous gate MOSFETs.

The electroantennogram (EAG) is the biopotential developed between two points on an insect antenna, due to the massed response of the olfactory receptor neurons (ORNs) in response to an odor stimulus; the response to a single filament of odor is a depolarization approximately 200 ms in duration. Although not entirely dedicated to olfaction, the antennae are the appendages containing the ORNs in insects [4]. Volatiles enter the sensillar lymph through pores in the cuticle, which are approximately 0.1 microns in diameter. The presence of a particular compound is detected by the sensory neurons that project dendrites into the sensilla. In the moth *manduca sexta*, each antenna has approximately 10^5 sensilla associated with about 3×10^5 olfactory neurons.

Chemisensory information is transmitted by various classes of olfactory nerves, which are defined by characteristic responses to various volatile compounds. The response of the nerve to a particular compound is dependent on the olfactory receptors, specialized odor sensing proteins, expressed on its cell membrane. In moths, sensillar lymph has been found to contain high concentrations of odorant binding proteins and pheromone binding proteins. [5] The exact role of these unbound proteins in communicating information to the receptors is unknown.

The morphology and mechanisms of olfaction are similar across many insect species [6]. Information obtained by the olfactory nerves is organized and transmitted to distinct glomeruli within the insect's brain. The number of glomeruli in *Drosophila* is approximately the same as the number of receptor types. The male moth *Agrotis ipsilon* contains 66 glomeruli, which gives an idea of the number of olfactory receptor types and thus sensor types available to this insect. Indeed, access to the different individual olfactory neuron types or glomeruli would be desirable. In fact, recordings from 21 individual glomeruli of honeybees has recently been used to classify several odors using principal component analysis (PCA) [7]. The problem with this arrangement is obviously the difficulty in the recording setup.

The antennae of different species of insects exhibit differential sensitivity to a given compound. Therefore, an array of antennae from various species can be used as a multi-channel detector.

For the past several years, a hybrid system for odor detection based on the olfactory organs of insects has been under development [8, 9]. Biological noses are capable of

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detecting many odors of no possible evolutionary usefulness, including explosives and drugs. Under controlled circumstances, some compounds can be detected down to 1 part per billion using EAG recordings [1]. Properties of the EAG sensor include short response time, low detection threshold, and sensitivity to a broad number of distinguishable compounds.

II. System Design

derivative at the maximum.

The primary feature, trough to peak magnitude, is then compared to a threshold. Events with amplitudes larger than a user adjustable threshold are kept, while those smaller are thrown away. The purpose of this is to remove data for marginal signal to noise ratios.

Finally, events are constructed by linking peak-trough events across channels that are within 160 ms. Each event contains 12 features (4 channels x 3 features).

In order to associate the responses of the antennae with a particular odor, the system must be trained first. Training is accomplished by collecting data while exposing the antennae to various known odor sources. After training has been completed, each feature in the training set is scaled so that its standard deviation is equal to 1 across all odors. Classification is accomplished by implementing (3). for each odor and selecting the maximum probability estimate.

The classifier has the additional capability to classify odors that don't look like any of the training odors as "unknown". The user may enter a "confidence" parameter, which sets a threshold for how close to an odor an unclassified event must be in order to be declared that odor. This is accomplished by calculating the probability estimates (3) of membership of each training point to its own class. These are then sorted. The confidence parameter then selects a value within the sorted list as the threshold for class membership. A high value of the confidence parameter will minimize false negatives (i.e. an odor classified as unknown), but maximize false positives.

III. Methods

The electroantennogram (EAG) was simultaneously recorded from excised antenna obtained from four insects of different species and sex, yielding a four-channel response to selected volatile compounds. Two species configurations were employed: Experiment 1 were used antennae from male insects of the following species: 1. *Platynota ideusalis* (tufted apple budmoth) 2. *Heliocoverpa zea* (corn earworm) 3. *Ostrinia nubilalis* (European cornborer) and 4. *Cydia pomonella* (codling moth). Experiment 2 (repeated twice) utilized antennae from the following species: 1. Male *Trichoplusia ni* (cabbage looper) 2. Male *Helicoverpa zea* 3. Female *Helicoverpa zea 4.* Female *Trichoplusia ni*. Each antenna was fixed to the preamplifier; electrical contact was made with a conductive gel. The antenna array was positioned 3 m downwind from the odor source in a wind tunnel; flow rate ~1.5 m/sec. The odor source consisted of 100 µg of a chosen compound in solvent (hexane) applied to a piece of filter paper and placed in the tunnel after the solvent had evaporated. The compounds used in experiment 1 [(Z)-11-hexadecenal (Z11-16:Ald), (Z)-11-tetradecenyl acetate (Z11-14:Ac), (E,E)-8,10-dodecadien-1-ol (E8, E10-12:OH), (E)-11-tetradecen-1-ol (E11-14:OH)] are major components in the pheromones of the insects used in this study. Compounds used in experiment 2 consisted of some pheremonal components [(Z)-8-Dodecenyl acetate (Z8-12:Ac), Z11-16:Ald, Z11-14:Ac] and some components present in manure [dimethyl disulfide, butyric acid, putrescine and trimethyl-amine].

Raw data consisted of voltage vs. time records from the 4 channel-probe under each experimental condition. EAG odor filament response events are classified using the modified K-nearest neighbor (KNN) technique. Each classification result is then weighed by the KNN estimate of the probability that the odor filament is correctly classified. This gives the user a measure of how certain the algorithm is of its classification. During the testing phase, the probability estimates for each class are accumulated over time and presented as a histogram. Each testing phase contains on the order of 100 events per odor. The accuracy of the classification is the sum of the correct classification probability estimates over all the classification estimates during the presentation of a particular odor.

IV. Results

Raw EAG data from experiment 2 are shown in Fig. 3, where each trace plots the biopotential activity recorded on one antenna. Panel A is typical of pheromonal responses. It can be seen that Male *H. Zea* (Red) responds strongly to Z11-16:Ald, which is present in Female *H. Zea* pheromone. Male *T. Ni* responds as well, but less strongly. The female antennae respond slightly if at all. The EAG response to a non-pheromone stimulus is shown in panel B. Responses are much lower in magnitude and include a greater fraction of the response from female antennae (note the scale). After the collection of training data, the user may view 2D projections of a feature type in the feature space. Training data for the experiment 1 trough to peak size feature is shown in Fig. 4. In these data, each odorant is clearly distinguishable from the 2D projections alone.

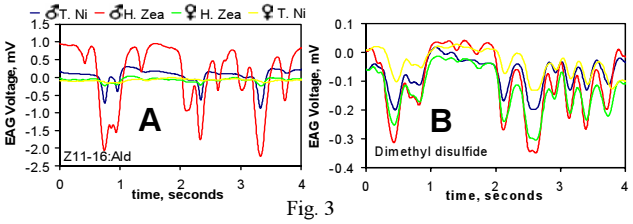

Fig. 3

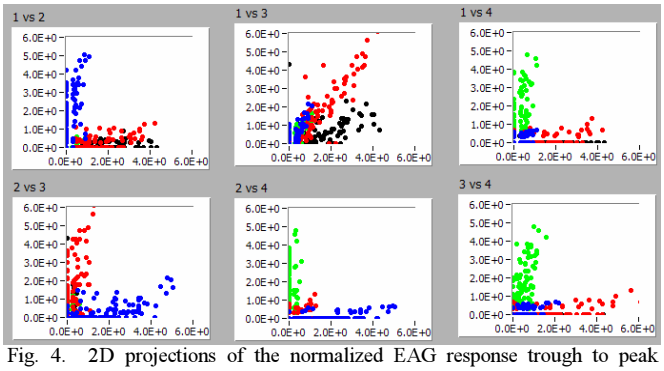

Fig. 4. 2D projections of the normalized EAG response trough to peak amplitudes for experiment 1 training data. The amplitude of this feature on one channel is plotted vs. the amplitude on every other channel; each channel (1-4) represents the response of a different antenna. Odor present during each event is coded by color: Blue = Z11-16:Ald; Red = Z11-14:Ac; Green = E8, E10-12:OH; Black=E11-14:OH.

In an effort to demonstrate the robustness of the approach, and to optimize the classifier, the overall accuracy of the classifier was tested as a function of several parameters including peak/event threshold (Fig. 5A), number of training points (Fig. 5B), number of nearest neighbors per odor (Fig. 5C), and the confidence parameter (Fig. 5D). When testing one parameter, the others remained at the default values, which were 100 μV for peak/event threshold, 100 for number of training points, 10 for number of nearest neighbors per odor, and 0.8 for the confidence parameter. Each parameter was evaluated in three experiments, represented as the three symbols in each panel of Fig. 5.

Increasing the peak threshold had the general effect of ignoring weak EAG responses. In evaluating this parameter, it was set to the same value for both the training and testing phases. Fig. 5A shows that in both experiments of type 2, where small events are frequent, the optimal threshold appears to be around 100 μV. As the threshold is increased, non-pheromonal odors, to which the system is generally less sensitive to, will tend to be classified as pheromonal odors, which dominate the feature space for larger amplitudes. Experiment 1 involves larger depolarization events; thus, elimination of low-amplitude events near the noise level continues to improve the accuracy over the investigated range.

The speed of training the system is determined in part by the number of training points required. The overall accuracy of the classifier as a function of the number of training points is shown in Fig. 5B. For all experiments, the accuracy continues to grow with increasing training set size, but in Experiment 1 the trend begins to asymptote near 70.

The accuracy as a function of nearest neighbors per odor is shown in Fig. 5C. For our data, it appears that smaller numbers of neighbors are optimal.

The accuracy as a function of the confidence parameter is shown in Fig. 5D. As the confidence parameter is decreased, the allowable distance between the unknown point and the centroid of the training data for a given odor is minimized, and so the classification accuracy increases, as expected.

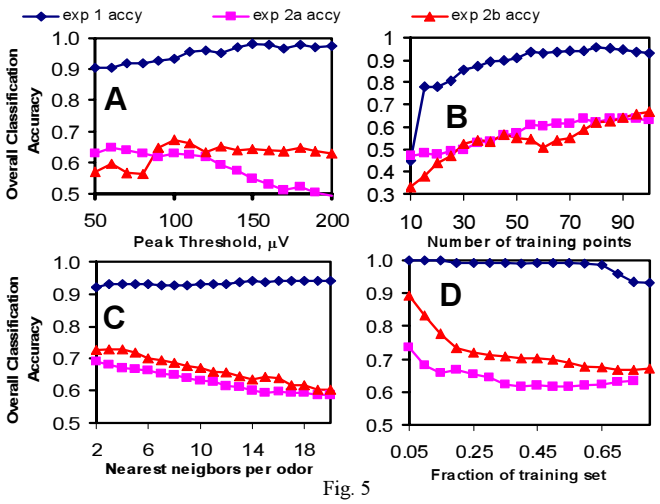

Fig. 5

V. Discussion

A real-time bioelectronic artificial nose system utilizing insect antenna as the odor sensor has been implemented using the KNN technique for classification. The technique has been modified to declare odorants as unknown using a "confidence" parameter entered by the user, selected based on the relative cost of false positives or false negatives in a given application. Further, the effects of several parameters on the accuracy of the system have been evaluated, including peak threshold, number of training points per odor, nearest neighbors per odor and a confidence parameter. The overall accuracy of the system ranges from 50% - 100% in the experiments and conditions evaluated here; 50% accuracy is significantly better than chance with five possible classes (four odors or unknown). Optimal classifier parameter values vary for different experimental conditions (species, odors), but general trends were consistent across experiments.

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