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Towards a biosynthetic infochemical communication system

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Abstract

By exploiting recent advances in the understanding of both pheromone biosynthesis pathways and the detection of molecules in the insect nervous system, we are developing a new class of technology for biosynthetic infochemical communication. Here we present a prototype instrument that comprises an integrated chemotransmitter, an odour pathway and a chemoreceiver controlled by a laptop computer. Our initial studies show that the system can transmit, receive and decode a set of volatiles commonly found in fruits. The system is capable of ratiometrically encoding information by releasing complex blends of compounds in specific ratios, detecting the transmitted blends with a sensor array and decoding the infochemical information using neuromorphic algorithms representing ratio processing in the macroglomerular complex (MGC) of the antennal lobe of the moth (e.g. *Manduca sexta*).

Keywords: ratiometric encoding, neuromorphic, biosynthetic, infochemical

1. Introduction

The complex form of information exchange in invertebrates mediated by chemicals represents an un-explored form of communication and labeling technology which is yet to be harnessed. Eusocial insects in particular rely heavily on chemical communication, for example to organise the colony, find food, conspecific mate location, and divide labour.

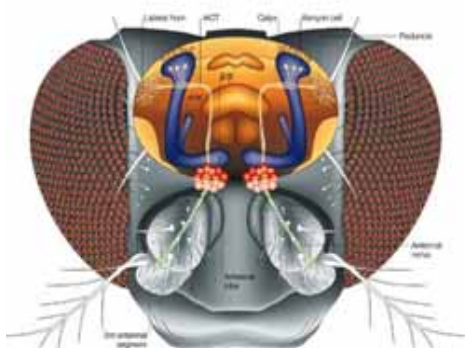


Figure 1. Schematic representation of the olfactory pathways of an insect in terms of the antennal lobe and mushroom body³

In this paper a novel approach to information and communication technology is introduced through the design of a first prototype instrument inspired by the biosynthesis pathways of infochemical production in insects. This innovative system capable of generating and detecting a precise mix of predefined synthesised compounds in programmable ratios of concentration will be capable of information transmission using chemicals alone and it will form the basis of a new technological field for labelling, information transmission and biochemical interfacing. The initial work concentrates in particular on the implementation of a so-called chemoreceiver, detector of volatile chemical mixtures, based upon principles of molecular detection in the receptor and antennal lobe neurons of an insect (shown in Figure 1), which is able to recover the ratiometric information deployed in the world. A member of the Lepidoptera, the sphinx or hawk moth (*Manduca sexta*) was chosen to model blend processing in the insect's antennal lobe. A considerable advances in the understanding of insect olfaction in terms of functioning of both the antennal lobe and the mushroom body; projection from receptor neurons to the mushroom body¹, in particular work on the glomerular representation of plant volatiles and sex pheromones in *Spodoptera littoralis*, as well as behavioural studies of moths in wind tunnels, have been exploited in this work.

2. Methodology

The prototype infochemical system, shown in Figure 2, comprises of three main units: the chemoemitter module, odour chamber and chemoreceiver module. The chemoemitter module is used to deliver the pre-synthesized analyte to the odour chamber in precise concentration and ratio's. The odour chamber provides a well-defined and controlled environment where the volatile molecules mix and travel to the chemoreceiver. The chemoreceiver consists of an array of gas sensors used to detect and differentiate the analytes introduced by the chemoemitter.

2.1. The Chemoemitter Module

The chemoemitter unit comprises of two high precision microlitre glass syringes mounted and controlled by a fully automated NeMESYS syringe pump system (Cetoni GmbH, Korbussen, Germany). Through this setup nanolitre accuracy in delivering analyte in precise concentrations and ratios with pulsating flow was achieved.

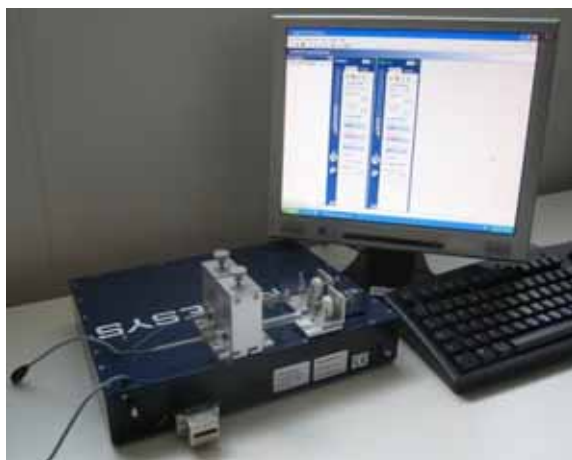


Fig. 2: NeMESYS syringe pump system with two independent, computer controlled dosing units.

2.2. The Chemoreceiver Module

The chemoreceiver module, shown in Figure 3, comprises an array of four 30 MHz polished Quartz Crystal Microbalance (QCM) sensors interfaced to a laptop via a USB port. The frequency shift generated by a change in mass on the surface of the QCMs is ca. 7.2 Hz/ng according to Sauerbray's equation³. Thus the practical detection limit of chemoreceiver unit was ca. 100 pg. Temperature and humidity were monitored using sensirion SHT-75 sensor. A gas sensitive polymer coating was created on both surfaces of the sensor by dip coating the crystals in 0.3 %wt polymer solution followed by an overnight bake at 60°C. The sensor was coated with four different polymer solutions namely poly(methyl methacrylate) (PMMA), poly(styrene-co-butadiene) (PSB), poly(ethylene glycol) (PEG), Polysulfone(PSF). The thickness of the gas sensitive coating was found to be approximately 100nm⁶. The sensors response to the analyte injection was recorded using commercial Multisens software (JLM Innovation, Tübingen, Germany).

2.3. The Odour Chamber

The odour chamber, see Fig 3, provides an environment for the volatile molecules to mix and travel to the chemoreceiver module attached to its far side. A circular design was adopted for the odour chamber. Due to the chemically resistive nature, glass was used for the main body. Due to the symmetric nature of the design we were able to minimize the dead space and record dynamic measurements using air flow. The chamber also houses the QCM sensors which are arranged in a circular manner with temperature and humidity sensor occupying the central position, see Fig 3.

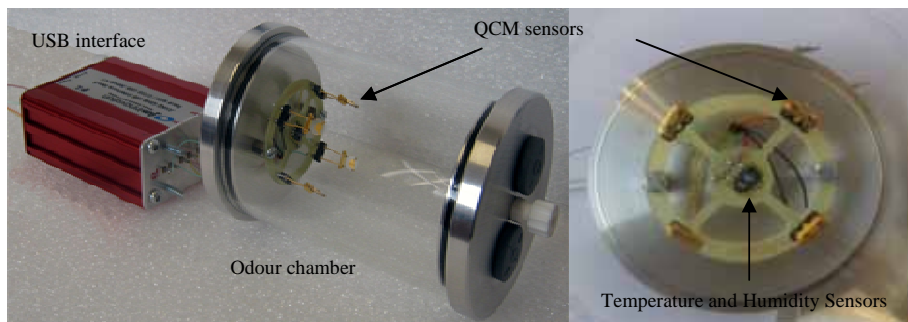


Fig 3 Cylindrical prototype odour chamber with four QCM micro-chemoreceivers (and temperature and humidity sensors) in a circular arrangement

2.4. Experimental Procedure

In order to assess performance of the system, experiments were performed using set of four fruit volatiles (butyl hexanoate (BH), hexyl butanoate (HB), 3-methylbutan-1-ol (3M) and ethyl acetate (EA)) commonly found in fruit blends such as apple, hawthorn and dogwood.⁴ These common plant compounds are behaviorally relevant and are detected by many species of insects (e.g. *Rhagoletis pomonella*). These four volatiles were injected into the chamber in three randomly ordered sequences. The aim of this experiment was to classify the four volatiles blends. In order to test chemo transmission, we have attempted to encode ratiometric information with the chemoemitter, and then decode this information with the chemoreceiver. This was realized by creating 9 mixtures of 3-methylbutan-1-ol and ethyl acetate of varying concentrations (1, 2 and 3 units) and ratios (3:1, 1:1 and 1:3). Each injection was repeated four times in a random sequence to eliminate the memory effects of the polymer coatings. For the experiments three different polymers i.e. PEG, PSB and PCL were chosen as coatings of three QCMs while the fourth QCM was left uncoated and used as a reference point. Fig 5(a) shows the target results of the measurements. The signals are analysed using both linear classifiers and a neuronal model of MGC processing with forward projection and lateral inhibition. This biomimetic models shown in Fig 5 was used to discriminate between different ratios.

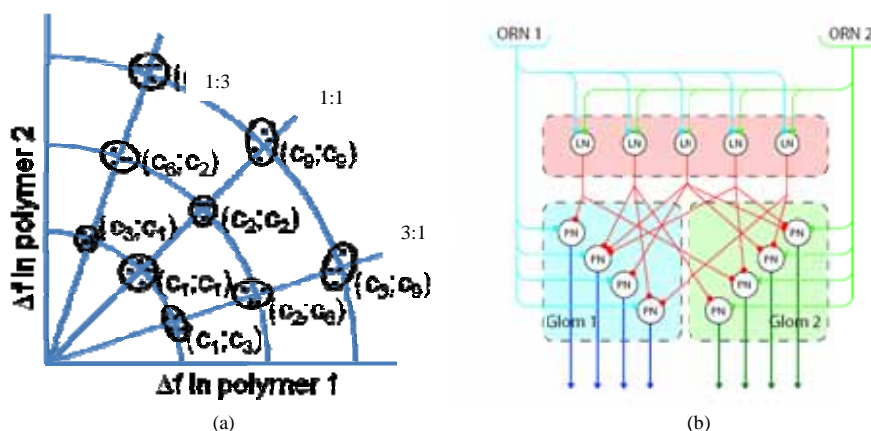


Fig. 4: (a) Target result of the ratiometric experiments showing three points tested for each ratio and each test point consisting of a cluster of four random experiments; (b) Biological model of ratio processing in the macroglomerular complex of the insect antennal lobe.

3. Results

3.1. Volatile Detection

For the classification of the four fruit blends Linear Discriminant Analysis (LDA) was performed on the signals extracted from the four polymer coated QCM's. LDA technique employed is a statistical multivariate data analysis technique that is used to detect linear combination of features which best separate two or more classes of objects or events. It assumes that the data are multi-normally distributed and separable in linear feature space. From the LDA analysis shown in Figure 5 it can be seen that 3-methylbutan-1-ol and ethyl acetate can be discriminated, however, the clusters for hexyl butyrate and butyl hexanoate overlap.

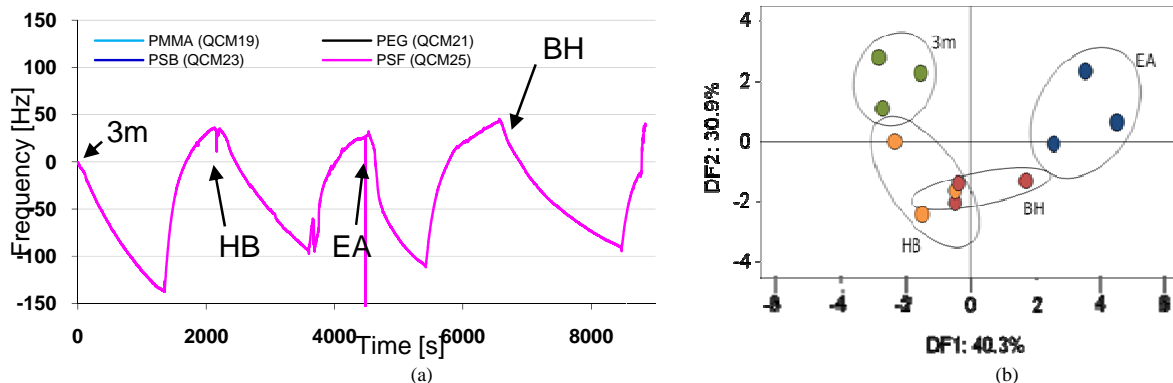


Fig. 5: (a) Frequency response of four quartz crystal microbalances coated with PMMA, PEG, PSB or PSF (b) LDA analysis of the four fruit volatiles forming four distinct clusters.

4. Conclusion

A new computer-controlled prototype of an infochemical communication system consisting of an odour flow chamber, a syringe pump as the chemoemitter at one end, and polymer coated quartz crystal microbalances as the chemoreceiver at the other has been constructed in order to prove the concept of infochemical based communication. The method by which polymer coated QCM sensors can be used to detect low volumes (<100 nl) of analytes in vapour phase has been successfully demonstrated. More experiments are underway and more polymer coatings are being sought based on non-polar solvents that should be more superior for the detection of non-polar compounds.

Once the concept of chemical infocommunication is demonstrated by static measurements, the feasibility of using the prototype in dynamic mode will be investigated with a new odour chamber designed to facilitate such measurements.

Acknowledgements

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