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Detection of VX Simulants Using Piezoresistive Microcantilever Sensors

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Abstract: Piezoresistive microcantilever sensors may be used in a variety of sensing applications, including chemical analytes and some types of biological species. These sensors employ a tiny piezoresistive microcantilever functionalized with a “sensing material” that acts as a probe for the desired analyte. In this study, the microcantilever was partially embedded into the sensing material, producing a sensor element that is highly rigid and resistant to shock, making it suitable for portable or handheld operation. The sensing material matrix used was Hypol, a hydrogel capable of preserving the bio-functionality of molecules embedded into it. This matrix was combined with acetylcholinesterase to form the composite sensing material. Results of exposing these sensors to a VX simulant, Malathion, are presented for both vapor and liquid environments. *Copyright © 2011 IFSA.*

Keywords: Piezoresistive microcantilever sensor, VX.

1. Introduction

Piezoresistive microcantilever sensors in which the microcantilever is partially embedded into the sensing material have been referred to as embedded piezoresistive microcantilever (EPM) sensors [1]. These EPM sensors operate by embedding or partially embedding a small piezoresistive microcantilever into a “sensing material”. The sensing material is synthesized in such a way as to respond volumetrically to the presence of a particular analyte species. In many cases, chemical, physical or other reactions with the sensor material may produce the desired volumetric shift. Sensing materials used in EPM applications may include common organic polymers, composite

polymer/biomolecule materials, or polymers functionalized with other active particles or chemicals. When they are exposed to the analyte species, the sensing material volume change causes a bending or strain in the embedded cantilever, which is subsequently measured as a resistance change by the sensor electronics (Fig. 1). Volume changes in the sensing material may be due to diffusion of the analyte molecules into the sensing material, probe-target binding on the material surface or bulk, or surface or bulk chemical reactions between the analyte and sensing material. In many cases, only tiny volumetric changes are needed to successfully detect the target species, as microcantilever strains of only a few angstroms are measurable.

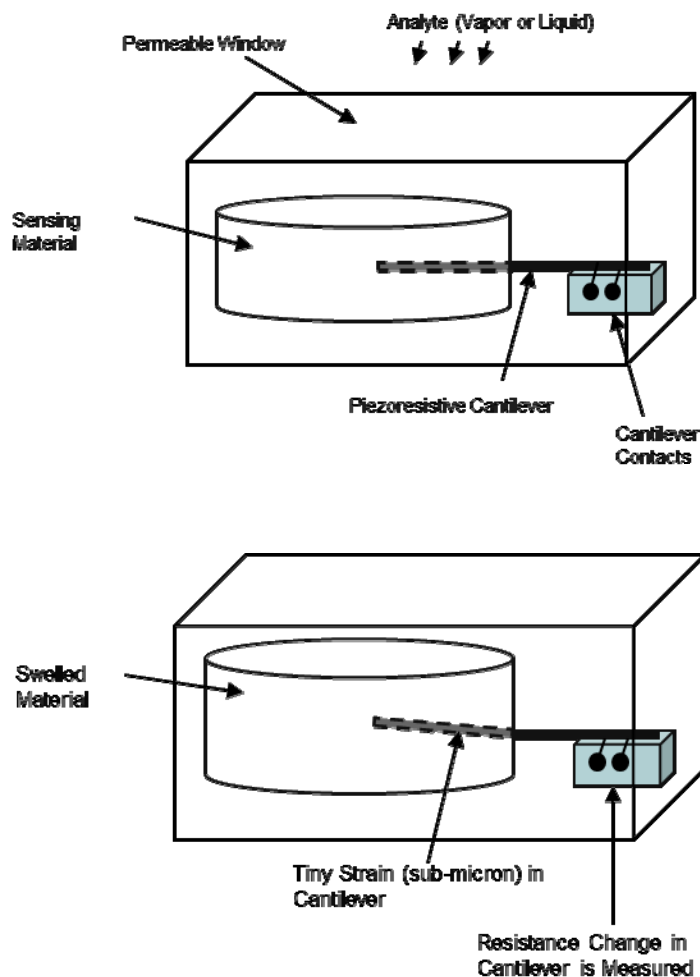


Fig. 1. Basic operation of EPMA sensor. Analyte molecules react with the sensing material causing the material to change volumetrically. This volume change in the sensing material is measured as a strain in the embedded microcantilever.

Electronics packages for these sensors are generally simple and inexpensive, as only the resistance of the microcantilever is measured. Fully assembled sensors are tiny in size, portable, and highly resistant to shock or vibration as the entire assembly is contained in a single, rigid unit. Previously, these types of sensors have been used to detect volatile organic compounds [1], bacteria in solution [2, 3], viruses [4], carbon monoxide [5], cyanide [6], hydrogen fluoride, and other analytes.

V-series nerve agents are part of the larger group of organophosphate nerve agents, first studied as pesticides in the 1920's. The V series itself (organophosphate esters of substituted aminoethanethiols) were studied intensively beginning in the 1950's by British scientists. Several V-series compounds were eventually produced, including VG, VE, VM, VR, VS, and the well-known VX. Many of these deadly compounds function by binding to and thus inhibiting the enzyme acetylcholinesterase (AChE),

which is present at the synapse between nerve and muscle cells. V-series and other organophosphate nerve agents disrupt the nervous system by inhibiting the AChE by forming a bond with the site of the enzyme where acetylcholine normally undergoes hydrolysis. Exposure to V-series nerve agents leads to rapid contraction of pupils, excess salivation, convulsions, and eventual death by asphyxiation as control is lost over respiratory muscles. As little as ten milligrams of VX on the skin will kill a person. Many of these nerve agents are easily deployable in aerosol form, with subsequent human exposure through the respiratory system. They may also be absorbed through the skin if exposure occurs in liquid form.

2. Experimental

VX simulant sensors were fabricated by immobilizing the enzyme acetylcholinesterase (AChE) in a hydrogel matrix. The gel host used was Hypol, a biologically compatible material from Dow Chemical. Hypol has been shown to immobilize many different types of biological molecules while preserving the biological activity of the immobilized species. The enzyme was loaded into the liquid Hypol host at approximately 10% ratio by mass. The material was then deposited onto a Si substrate, and the microcantilevers partially inserted into the compound with the use of a micromanipulator. The material with cantilevers embedded in it was allowed to cure in air at room temperature for 72 hours prior to use.

The piezoresistive microcantilevers (PMC) used for these VX experiments were produced by Cantimer, Inc., Menlo Park, CA (Fig. 2). The microcantilevers are contained in a die, and are approximately 200 microns in length, and 40 microns wide. The nominal resistance of an unstrained cantilever is approximately 2.2 k Ω . Each cantilever extends into a small circular area on the die in order to contain the sensing material and also to partially protect the cantilever. In addition, each cantilever die also contains an integrated thermistor if temperature correction is needed. The EPROM on each circuit board may be used to store unique temperature or humidity correction coefficients if very precise corrections are needed in a particular application.

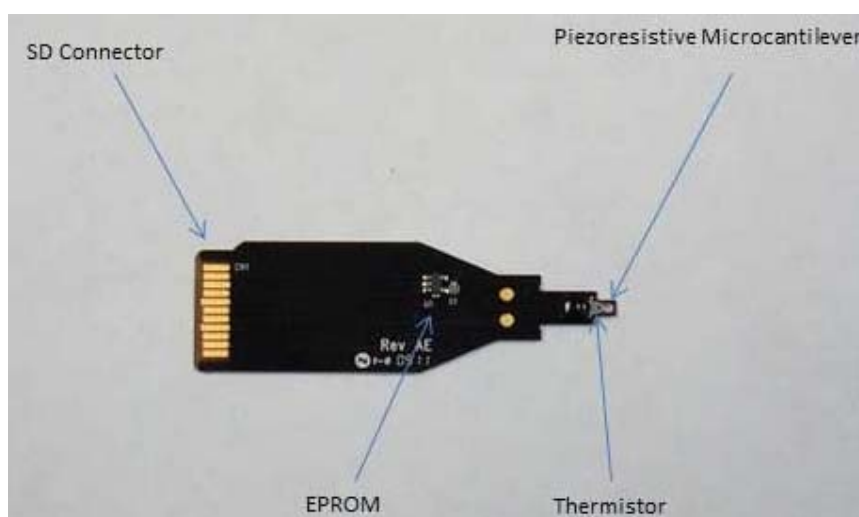


Fig. 2. Piezoresistive microcantilever die (far right) attached to a plastic circuit board for plug-in to a standard mini-SD connector.

For the VX sensing experiments, a single-chip AD7793 24-bit A-D converter which functions as a 6½ digit multimeter was used to directly measure the cantilever resistance. The AD7793 contains two

independent 24-bit AD conversion channels, and two independent precision current sources. For these experiments, the current sources were set to 200 μ A current through the microcantilevers. The AD7793 chip was interfaced to a laptop computer through the USB interface provided by an USBmicro 421 chip. The USB 421 chip is used to translate the SPI commands to and from the AD7793 to standard USB signals for a personal computer. The user interface and data collection was performed on the laptop computer using LabView. Experiments were performed in a chemical hood using a plexiglass test chamber with flow monitoring capability (Fig. 3).



Fig. 3. Test chamber with LabView interface to laptop computer.

3. Results and Discussion

Fig. 4 shows the sensor response for a large exposure to the VX simulant, Malathion. Malathion is commonly used as a simulant for VX. Here, the test chamber was set up with a dry nitrogen flow at a rate of approximately 1 l/min. 0.5 ml of Malathion in liquid form was then injected into the nitrogen flow line. The time of the Malathion injection is indicated in Fig. 4 by the arrow. The Malathion exposure within the test chamber was simply the vapor from the evaporating Malathion as it flowed through the chamber. Based on the slow rate of evaporation of the Malathion liquid entering the test chamber, we estimate that the maximum sensor exposure experienced by the sensor was approximately 100 ppm.

This 100 ppm exposure level was essentially continuous, and throughout the time period indicated in Fig. 4. Evaporation then continued for many more minutes after data recording was stopped. The sensor response to the simulant occurred within a few seconds of initial exposure. Also, the sensor continued to react to the simulant for over 300 seconds after the first exposure. After approximately 350 seconds of exposure, the sensor began to saturate, indicating that no additional response to the

Malathion was occurring. This saturation effect was most likely due to all of the probe molecules in the sensing material, acetylcholinesterase, that were accessible to the analyte had fully reacted. In cases such as this, the sensor is no longer capable of detecting the analyte, and must be replaced. This general type of sensor operation is sometimes referred to as a chemical fuse.

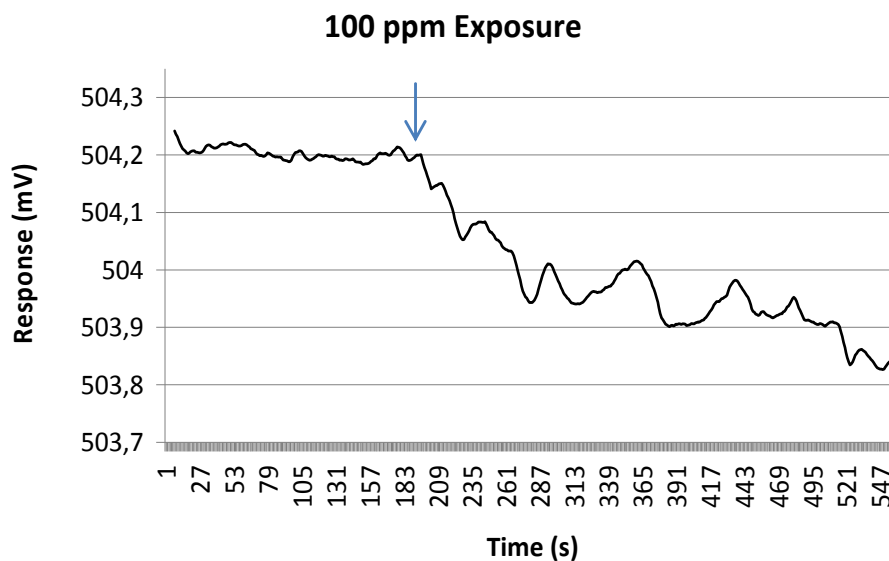


Fig. 4. Sensor response to 100 ppm Malathion exposure. The initial sensor response to the simulant occurred within a few seconds of initial exposure. The sensor continued to react to the simulant for over 300 seconds after the first exposure. After approximately 350 seconds of exposure, the sensor began to saturate, and was rendered unusable. At such high doses, or doses even higher, this sensor functions essentially as a chemical fuse.

For the next exposures, the sensor was replaced with a new unit, and the dose of Malathion was reduced by a factor of 10. The surface area of the injected Malathion was reduced by this same factor. The corresponding vapor concentration was approximately 10 ppm throughout the course of the evaporation. The nitrogen flow conditions of 1 l/min were kept the same as the previous exposure levels. The time of Malathion injection is indicated by the arrow in Fig. 5. At this low exposure level, the sensor response is seen to be near the background noise level of the sensor itself, although some sensor response is indicated. The average sensor voltage prior to exposure is seen from Fig. 5 to be approximately 504.07 mV. After exposure, the average voltage level is approximately 504.03 mV. This low sensor response is partially obscured by the background noise levels in the sensor package. We conclude that exposures at this very low level are generally difficult to detect with the current sensor configuration, although some response is certainly indicated here.

It is possible that these sensors would be reasonable devices for the sensing of this analyte at minimum exposures of around 20 ppm with no additional changes to the sensor or the electronics. In this case, the sensors would also most likely be reusable for two or more individual sensing operations. We note, however, that the data shown in Fig. 5 is raw data, and some additional sensor sensitivity gains would also be possible using various types of electronic noise reduction. One possibility would be to use a simple smoothing algorithm in the LabView data acquisition program used to display the data. Here, data points were taken once every second. These electronics we are using are capable of taking measurements at a much higher rate. At a data collection rate of 10 per second, we could then take an average of every 10 measurements, and update the display once every second. Many other types of electronic filtering are also possible, and we will report on these improvements in a later publication.

In addition to improvements to the current sensor configuration possible through lowered electronics noise levels, we could also envision changes to the sensor itself including different matrix/enzyme ratios. These changes and their results will be reported in a future publication for very low exposure levels.

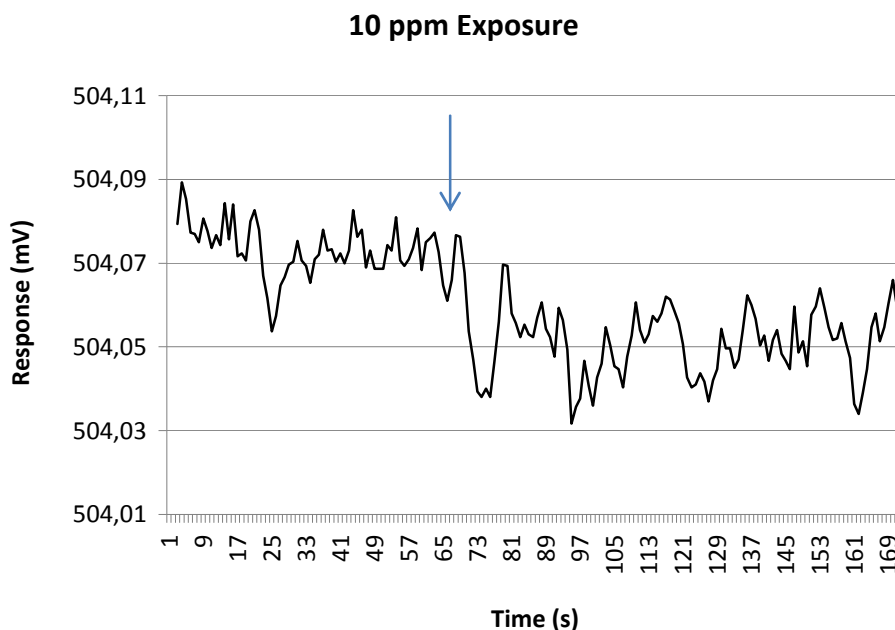


Fig. 5. Sensor response to 10 ppm exposure to Malathion. Here, the sensor response is seen to be at or near the minimum level of detection for this analyte. Improvements in the sensor sensitivity at these low levels of exposure may be possible by employing electronic noise reduction schemes in the data collection.

In addition to improvements to the current sensor configuration possible through lowered electronics noise levels, we could also envision changes to the sensor itself including different matrix/enzyme ratios. While the enzyme loading into the hydrogel host might be increased somewhat, it is relatively easy to significantly degrade the structural integrity of the composite sensing material by over-loading with the probe material. These hydrogel/enzyme changes and their results will also be reported in a future publication for very low exposure levels.

These EPM sensors are also capable of being submersed in liquids for the detection of various analytes in water or other non-corrosive solvents. The sensor components themselves do not react to liquid environments, and while exposed electrical connections may be subject to corrosion over time, they may simply be coated with a waterproof epoxy. We have previously used EPM sensors in solution for the detection of single-strand DNA, bacteria, viruses, and chlorinated hydrocarbons in water. Sensors with protected electrical connections have been operated by us in aqueous environments for several weeks with no degradation in functionality.

For this test in solution, we immersed a new sensor in a beaker containing 250 ml of nanopure water. The solution was held at room temperature, and was magnetically stirred. The sensor was allowed to come to equilibrium prior to addition of the VX simulant. This equilibrium step is required as most polymer materials, including Hypol, will absorb water to some extent, and subsequently swell. For this experiment, the sensors were seen to come to equilibrium with the solution in 15 minutes or less. After reaching equilibrium, Malathion was added to the water. The concentration of the Malathion in the water was adjusted to provide a sensor exposure of 100 ppm. Here, the sensor response was rapid and large (Fig. 6). The total sensor response is approximately 0.35 mV (voltage drop across the cantilever),

indicating a large direct exposure when in aqueous environment. Also, in this liquid environment, the sensor is seen to reach saturation very quickly when compared to the vapor-phase trials. Here, the aqueous delivery of the analyte to the active binding sites in the sensing material is greatly enhanced by the solution, resulting in the observed rapid sensor saturation.

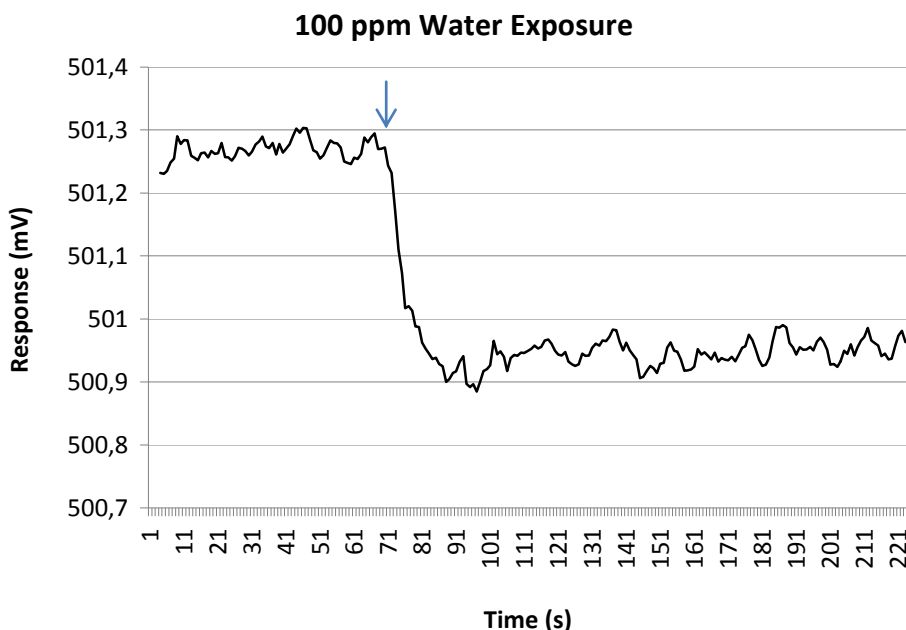


Fig. 6. Sensor response to aqueous exposure to Malathion at 100 ppm. Here, the sensor response was both rapid and large compared to the vapor-phase exposures.

4. Conclusions

The sensor responses in vapor-like or aqueous environments indicate that this sensor material may be suitable for detection gases such as VX as a chemical fuse, essentially for one-time use is the exposure level is on the order of 100 ppm. At these levels, the sensor essentially integrates the total exposure, eventually reaching saturation. At very low exposure levels, such as 10 ppm, the sensor response, in its current configuration, is near its detection limit. For future experiments, we will be studying improvements to the sensor electronics package, as well as different formulations to the sensing material itself.

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