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**SELECTED VAPOR PHASE SENSING OF SMALL MOLECULES
USING BIOFUNCTIONALIZED FIELD EFFECT TRANSISTORS**

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**Non Metallic Materials
Nanostructured and Biological Materials**

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Selective vapor phase sensing of small molecules using biofunctionalized field effect transistors

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ABSTRACT

This work details a proof of concept study for vapor phase selective sensing using a strategy of biorecognition elements (BRE) integrated into a zinc oxide field effect transistor (ZnO FET). ZnO FETs are highly sensitive to changes to the environment with little to no selectivity. Addition of a biorecognition element retains the sensitivity of the device while adding selectivity. The DNA aptamer designed to bind the small molecule riboflavin was covalently integrated into the ZnO FET and detects the presence of 116 ppb of riboflavin in a nitrogen atmosphere by a change in current. The unfunctionalized ZnO FET shows no response to this same concentrations of riboflavin, showing that the aptamer-binding strategy may be a promising strategy for vapor phase sensing.

Keywords: DNA aptamer, biological recognition element, field effect transistor, sensor, zinc oxide, riboflavin

1. INTRODUCTION

Development of sensor elements and devices requires the ability to determine the presence and concentration of target molecules of interest. A number of strategies are used to achieve this, such as mass spectrometry^{1,2}, biomolecular interaction analysis (BIA)^{3,4,5}, and mass based resonance sensing^{6,7}. Recent advances in sensor development highlight a selective sensing strategy using biorecognition elements (BRE)⁸, such as DNA/RNA aptamers^{9,10} or peptides^{11,12}. Instead of using complex separations or algorithms for data interpretation, the BRE offers a point based detection where the specific molecule of interest is bound directly by the BRE. The binding event can then be transduced into a reportable signal based on the sensor platform in which it is integrated. A number of different sensing platforms have been developed with BRE integration such as electronic¹³, optical^{14,15}, and mechanical devices¹⁶. One of the more promising platforms is the biofunctionalized field effect transistor (BioFET). The semiconductor in the FET is particularly sensitive to surface effects or changes, with little to no selectivity. These surface changes can be due to environmental effects such as the presence of liquids or vapors. Attaching a BRE to the semiconductor adds selectivity while maintaining the inherent sensitivity of the surface and has been successfully shown on various semiconductors such as carbon nanotubes^{12,17}, silicon nanowires¹⁸, and zinc oxide¹⁹. However, a majority of the work with BRE-FETs is in the liquid sensing environment.

Previous work in this group involved using a zinc oxide (ZnO) FET with an integrated DNA aptamer that was selected for the small molecule riboflavin¹⁹. Selective detection was achieved below nano-molar levels in the liquid phase. The work presented here describes the initial studies

on the feasibility to detect riboflavin in the vapor state using the same ZnO aptamer-FET (AptaFET) strategy.

2. EXPERIMENTAL

2.1 Device geometry and experimental setup

The ZnO FETs were designed for ease of handling and testing while maintaining the active area presented in previous work. The interdigitated electrode distance is 10 μm and has extended contact pads (400 μm^2). Three sets of three FETs were fabricated on a diced chip which is 20 mm x 7.5 mm as shown in Figure 1 below. The devices were fabricated in a 3 mask design on a 3" silicon wafer. The first mask was used for patterning thick SiO_2 insulator paths along the extended contact lines, the second for patterning the ZnO only in the active area (green region in Figure 1a right), and third for the top electrode patterning. The ZnO was deposited using pulsed laser deposition described previously²⁰. The dry state device performance shows an excellent on/off ratio of 10^6 (A) shown in Figure 1b.

The next step is attaching the DNA aptamer, which is functionalized with a thiol group as specified and purchased from IDT Technologies. The sequence and structure of the riboflavin binding aptamer is well known²¹, and is used in these experiments as: 5'-Thiol-AGAGAGGAACGACGGTGGTGGAGGAGATCGTTCC-3'. The thiol group on the aptamer provides the linkage to the ZnO via a silane linker as described previously¹⁹. The surface morphology of a covalently attached aptamer on ZnO is shown in the micrograph in Figure 1c.

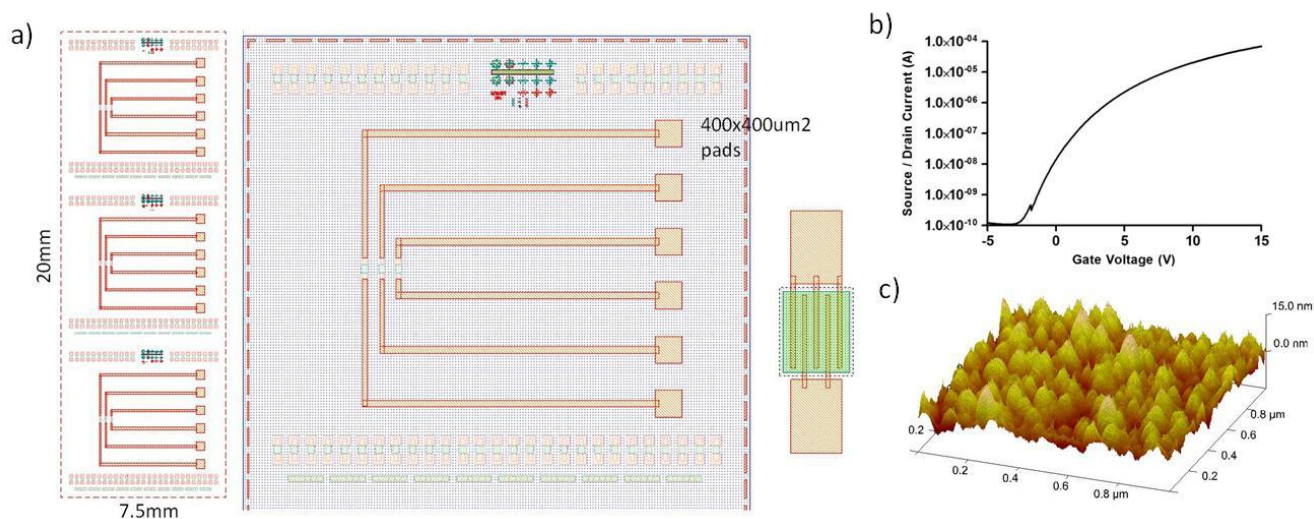


Figure 1. ZnO AptaFET

a) Schematic of device geometry with full chip image on the left, zoom in of single set of FETs in center, and individual FET right, b) Dry state device performance, c) Atomic force micrograph of aptamer functionalized ZnO semiconductor

2.2 Device testing – Bias stressing

Initial testing of the devices in a nitrogen atmosphere showed a significant downward drift of current when a constant voltage ($V_G = 5V$, $V_{SD} = 0.2V$) was applied. A bias stressing study was done based on work by Marks et al.²² to reduce this drift and have a steady starting current. Bias stressing is done by applying a large voltage (5V) to the source/drain electrodes to help clear any charges that are trapped between the insulator and semiconductor layers. This can be done in cycles, and is complete once a steady charge state is achieved. A bias stressing study is shown in Figure 2 below, where 5V was applied for 500 seconds in 5 cycles. An additional 6th cycle was tested after the device sat in the nitrogen environment for 2 hours. Even after the 2 hour delay, the bias stressing remained effective with a largely decreased drift.

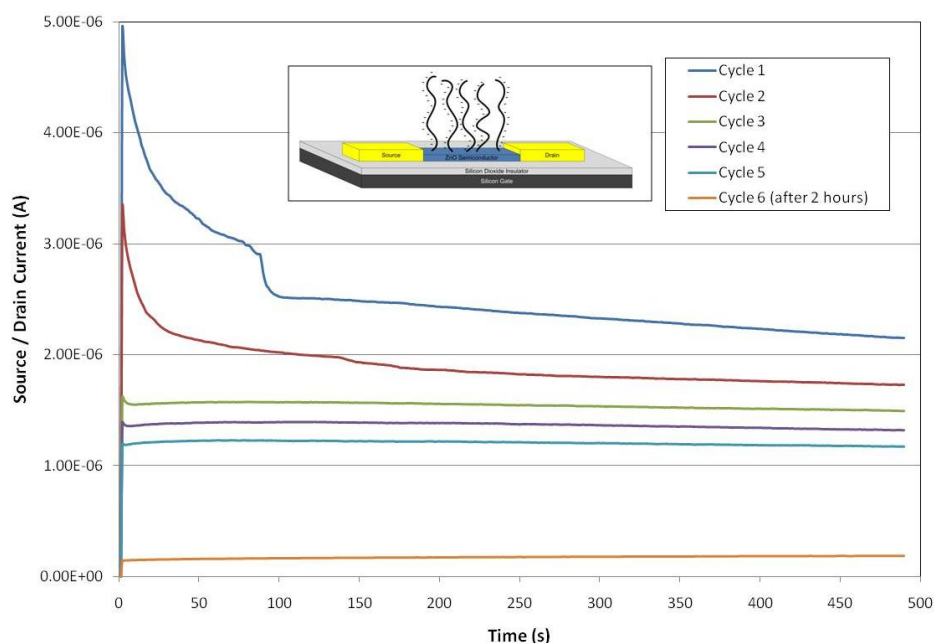


Figure 2. Source/Drain Current (A) versus Time: Bias stressing study of ZnO AptafET with $V_{SD} = 5V$

2.3 Non-selective vapor testing

For baseline performance testing, the unfunctionalized ZnO FET was evaluated in a nitrogen atmosphere as well as adding additional vapors to the nitrogen stream. The first evaluation was the effect of nitrogen flowrate on the device. Figure 3 below shows that varying the nitrogen flowrate from 50 to 100 mL/min had no effect on the current flowing through the FET.

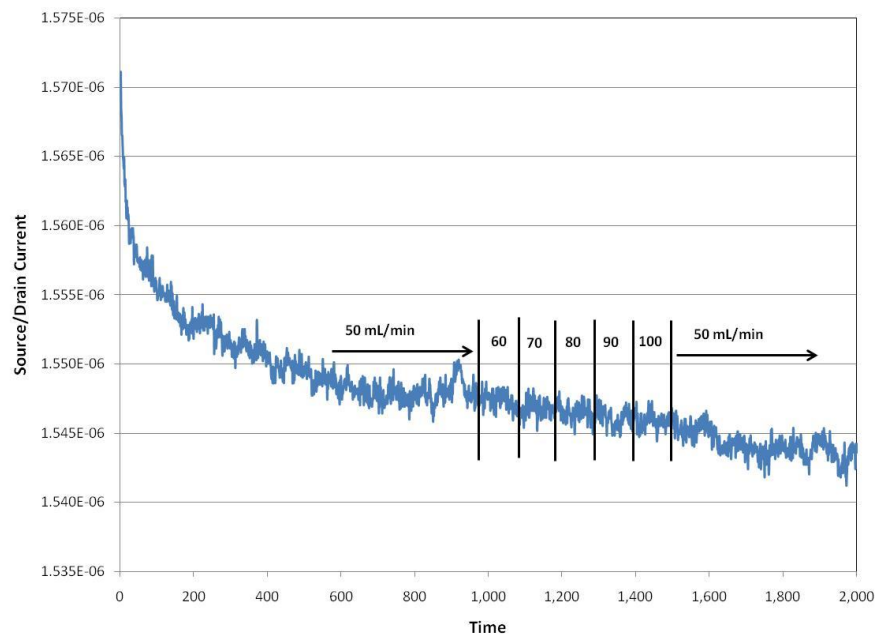


Figure 3. Source/Drain current (A) versus Time with varying nitrogen flowrates

As mentioned previously, unfunctionalized FETs commonly respond to a wide variety of vapors. Many vapor molecules non-selectively interact with the surface causing the current to modulate. The fact that the devices can respond to a variety of vapors is promising, but again there is no selectivity. These interactions are shown in Figure 4 below, where high concentrations of ethanol and acetone were exposed to the unfunctionalized ZnO FET. The vapors were generated by bubbling nitrogen through the liquids. Figure 4a shows high concentration exposure of ethanol, and 4b acetone. In both cases, the current is significantly increased upon exposure. In the case of ethanol, the vapor seems to desorb as the current decreases and approaches the initial level. Acetone does not appear to desorb where a constant elevated current is maintained.

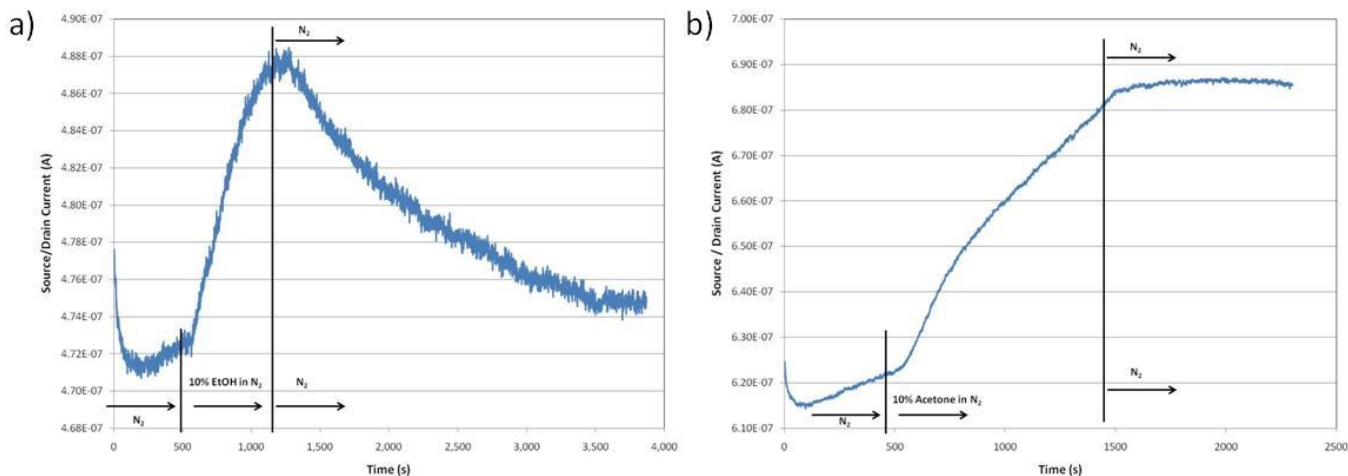


Figure 4. Source/Drain Current (A) versus Time (s) for exposure of the ZnO FET to a) ethanol and b) acetone

2.4 Selective vapor testing – exposure to riboflavin

The negative aspect of using surface sensitive FETs in vapor sensing applications is the lack of selectivity upon exposure to various vapors. This can be partially overcome by incorporating selective recognition elements, such as the riboflavin DNA aptamer, which was evaluated upon exposure to riboflavin vapor. Since riboflavin is a solid with very low vapor pressure, an Owlstone vapor generator was used to create the vapor stream. Riboflavin was placed in a permeable tube and inserted into the oven of the vapor generator. A constant nitrogen flowrate (50 mL/min) was flowed through the oven, and a calibration curve was established to determine the amount of riboflavin that was vaporized. This corresponded to a riboflavin vapor flowrate of 116 ppb (309 nM) in 50 mL/min of nitrogen. A negative control experiment was run which included exposure of a non functionalized ZnO FET exposed to 116 ppb riboflavin, and is shown in Figure 5. A constant nitrogen flow with and without riboflavin vapor was cycled back and forth to establish whether any recognition was evident.

Upon functionalization of the ZnO FET with the riboflavin aptamer, detection of riboflavin in the vapor phase is possible, shown in Figure 6. When riboflavin is introduced into the nitrogen stream, the current quickly begins to decrease, which corresponds to the mechanism described previously¹⁹. After the riboflavin flow is stopped (while maintaining nitrogen flow), the current continues to decrease, likely due to the slow absorption transport to the surface and residual riboflavin in the chamber which has not been cleared. After purging with nitrogen, the riboflavin appears to desorb from the surface shown by the increase in current at ~1200 s. Additional cycles between pure nitrogen and riboflavin vapor shows continued detection of riboflavin that is not seen in the unfunctionalized device.

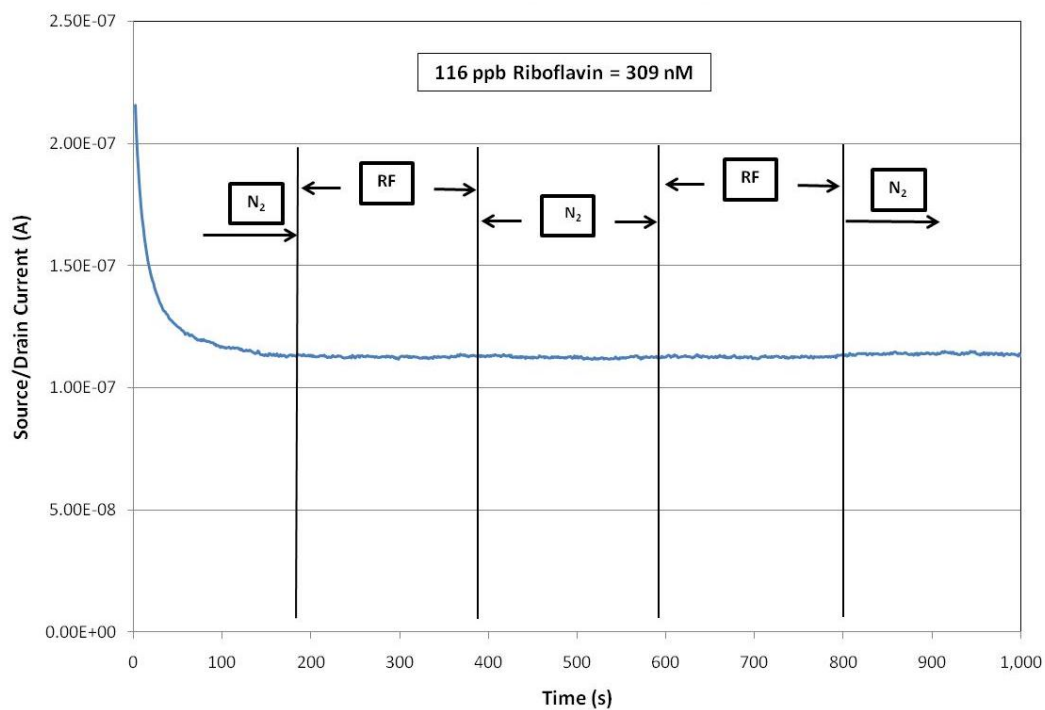


Figure 5. Source/Drain Current (A) versus Time (s) for an unfunctionalized ZnO FET and exposed to 116 ppb riboflavin vapor in nitrogen

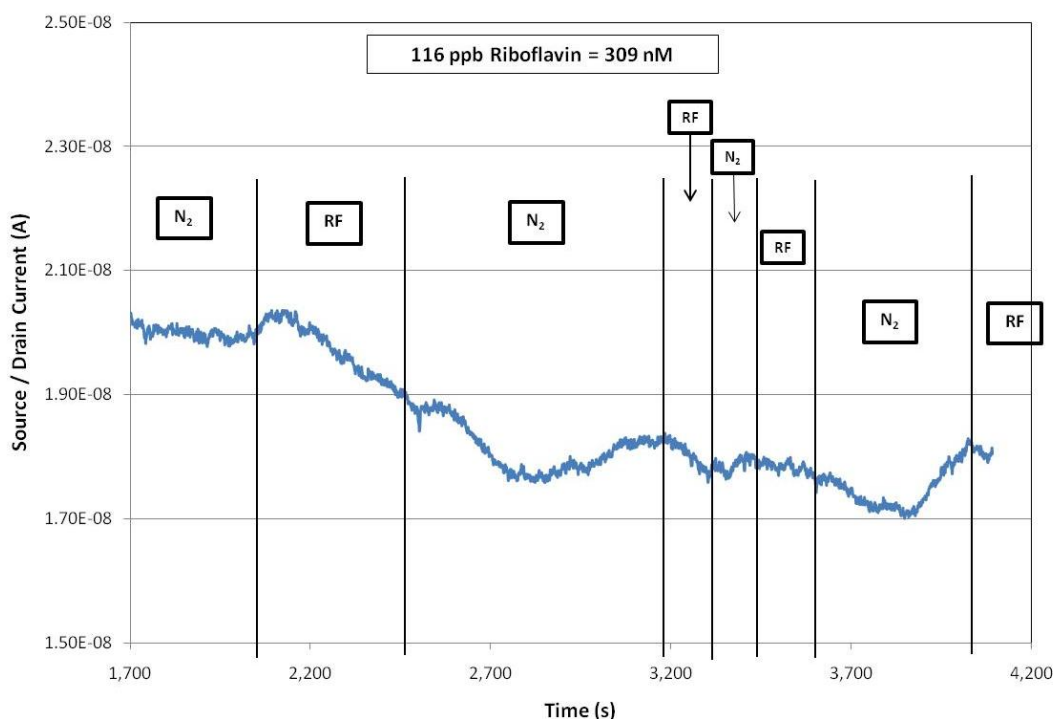


Figure 6. Source/Drain Current (A) versus Time (s) for a riboflavin-Aptamer functionalized ZnO FET and exposed to 116 ppb riboflavin vapor in nitrogen

3. CONCLUSIONS

Field effect transistor sensors can be incorporated into standard integrated electronic systems which can rapidly transmit sensing data to a computer, handheld sensor, or even wirelessly. A great deal of research has been, and is currently being pursued for liquid phase FET sensor systems with added selectivity provided by biorecognition elements. The preliminary data established here shows a promising route for selective sensing in the vapor phase. By incorporating the riboflavin binding aptamer to a ZnO FET, we show that riboflavin can be detected at ppb levels in the vapor phase. No detection occurs with an unfunctionalized ZnO FET.

The next steps in this research include testing of additional targets using both aptamers and peptides, testing these in more complex vapor environments, and coupling with microchannel technologies for increased surface exposure to the vapors.

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