A Cost-efficient, Camera-based Electronic Nose

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***Abstract* - Human's olfactory system has a large number of receptor types that respond only to a limited number of molecules. Since human noses are subject to fatigue and inconsistency and lack the sensitivity to low-concentration molecules, scientists have developed the Electronic noses. E-nose is a device that identifies the specific components of volatiles and analyzes the chemical makeup to identify it. Current applications include detection of odors specific to diseases for medical diagnosis, and detection of gas leaks for environmental protection. An E-nose consists of both, a mechanism for chemical detection through an array of electronic sensors and mechanism for pattern recognition similar to that of the neural network. Relating to the previous work of our fellow colleagues and major development of the E-nose by scientists at Tufts University, our E-nose developed here consists of a Biosensor, which is a protein extracted from animals and a labeling component that is a chemical dye that is suitable to the protein used. As for the electronic part of the prototype that is responsible for the detection of color change of the dye during the interaction with the odors, it is made up of an application programmed for this specific purpose that ensures proper detection of RGB (red, green and blue) value color change.**

Keywords - E-nose; Volatiles; Biosensors; Chemical dye; RGB values

I. INTRODUCTION

The olfactory system is one of the most intelligent sensory systems to be developed in the mother's womb. The primary pathway consists of two components, the olfactory epithelium and the olfactory bulb. Olfaction means the sense of smell. The main organ involved in olfaction is the nose, which has millions of receptors for smelling that are present within olfactory epithelium. This epithelium has three kinds of cells: basal, supporting, and olfactory receptor cells [1]. Olfactory receptors are bipolar neurons that are made up of dendrites and an axon that ends at the olfactory bulb. The most important part of this receptor is the olfactory hairs that respond to chemicals that are breathed in. When odorants enter the nose, the olfactory hairs that are present inside the nose become activated, generating the potential that then initiates the response [2]. Signals move from the olfactory cells to the olfactory bulb and move on to different parts of the brain, depending on what kind of signals. Figure 1 illustrates the process by which olfactory information is transmitted to the brain.

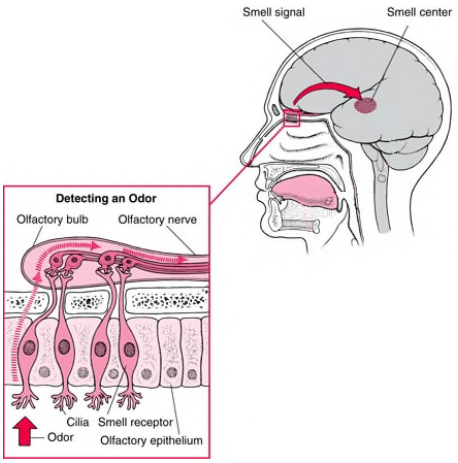

Figure 1. Olfaction procedure where signals move from the olfactory cells to the olfactory bulb and move on to different parts of the brain [3]

Human sniffers are costly when compared to electronic noses. Electronic noses are quick and use reliable new technology of gas sensors. One major point is detection of hazardous or poisonous gas that is not possible with human sniffers that could be overcome by electronic devices.

Scientists thought of coming up with a device to overcome these complications. The solution to these difficulties would also advance the relationship between biology and technology. An electronic nose (e-nose) is a device that identifies the specific components of an odor and analyzes its chemical makeup to identify it. Electronic noses have been around for several years, but have typically been large and expensive. Current research is focused on making the devices smaller, less expensive, and more sensitive. [4]

A next-generation artificial nose developed by Tufts neuroscientists [4] uses DNA (Deoxyribonucleic Acid) to detect odors, and possible applications range from medical to commercial to defense. Researchers at Tufts have pioneered the use of DNA molecules to detect millions of odors. In early versions of the electronic nose, airborne odors passed over a square of silk screen treated with a mixture of a reactive polymer (a large molecule comprising

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a chain of smaller ones) and a fluorescent dye. If some property of the odor, its molecular shape, polarity or charge—interacted with the polymer, the fluorescent dye would glow in response. The trouble was, for each odor they wanted to detect, the researchers had to find, mainly through trial and error, the specific polymer that could serve as a sensor. Over 15 years, the Tufts team and researchers elsewhere discovered 20 to 30 polymers capable of detecting a handful of odors. Figure 2 shows Tuft's University device and it could be tailored to be used in the food and beverage industry, ensuring high quality products and detecting possible contaminants [5].

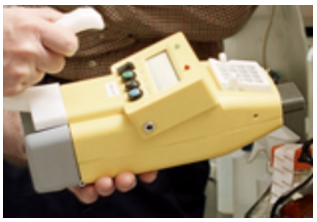

Figure 2. Tufts University Electronic Nose [6]

As for the medical field, other researchers have come up with a device later called Nano Artificial Nose as shown in figure 3 [6]. In the absence of clear surrogate clinical markers that could discriminate between various sources of respiratory infections, over-treatments with antibiotic prescriptions are evidenced in a large portion of the treated cases. There has been an increasing interest in recent years in improved methods for diagnosis of many metabolic and infectious diseases. These new methods are expected to be non-invasive and inexpensive, while allowing: (1) screening of high-risk populations for emerging diseases; (2) early detection and prediction of diseases; and (3) evaluation and monitoring of therapy efficacy.

Figure 3. Nano Artificial Nose

A prototype of cross-sensitive nanowire-based sensors to be integrated in the 'Nano Artificial Nose' were trained to detect target disease related mixtures of biomarkers. Advanced development of the Nano Artificial Nose disease detection capabilities are present for the detection of the following indications from exhaled breath: Streptococcus; Methicillin resistant (MRSA); Staphylococcus; etc. Nano Artificial Nose technology detects specific disease biomarkers based on a change in the blood chemistry or metabolic activity (which is reflected in the chemical composition of the exhaled breath and cell/tissue headspace) rather than by other forms of imaging or invasive blood analysis [7].

Relating to the previous work of our fellow colleagues and major development of the E-nose by scientists at Tuft's University, our E-nose consists of a Biosensor, which is a protein extracted from animals and a labeling component that is a chemical dye that is suitable to the protein used. To detect color change, an application is programmed and used to validate the interaction occurring between odors and proteins.

The rest of the paper is structured as follows. In section 2, represent the materials used, the requirements and device design, the project working process and the methods used. Afterwards, in section 3, experiments, results and data analysis are described. Then, section 4, results are discussed and explained. Finally, in section 5, a conclusion summarizes the requirements of this project and enhancements that meet the market requirements.

II. MATERIALS AND METHODS

E-Nose technology joins several analysis techniques that make way to understanding the structures and composition of odors. Figure 4 illustrates the diagram of the device that consists of a chamber containing the fan and battery, biopolymer sensor, and odor receiving duct that is analyzed using a programmed application that recognizes the color change.

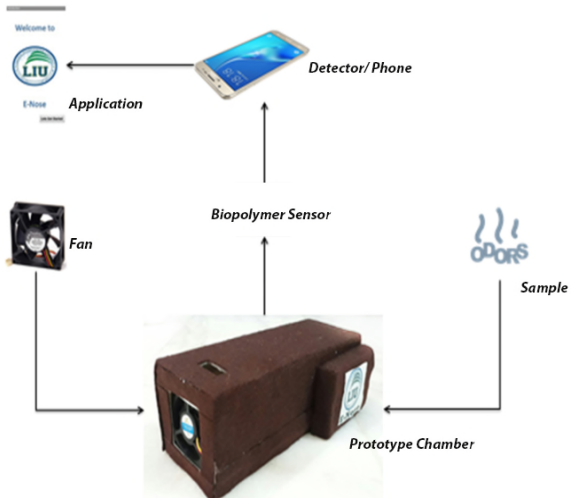

Figure 4. LIU (Lebanese International University) E-Nose Block Diagram

Materials and Preparation Steps:

1) Use an Analytical Balance to weigh 3.5mg of ALP (alkaline phosphatase) protein
2) Dilute with 0.67ml distilled water at 33ºC to obtain 5.2g/l concentration
3) Take 10ul prepared ALP and add to it 5ul coomassie dye and 5ul tea tree oil and pour in 1.5ml eppendorf tubes
4) Prepare reference tubes containing:
   a) 10ul ALP protein with 5ul coomassie dye (control tube)
   b) 5ul coomassie dye with 5ul tea tree oil
   c) 5ul coomassie dye
5) Mix the combinations for 2 min using a vortex mixer

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6) Place the sampled tubes in a water bath at 33ºC for 30 min
7) Record the change in color at the end of the experiment, each tube with respect to the reference tube containing the protein-dye mixture, using the phone camera
8) Analyze and compare the results using the LIU E-Nose application

III. RESULTS

LIU E-nose application serves as the image processing software programmed using MIT (Massachusetts Institute of Technology) App inventor [8]. The software makes use of a typical phone with 13-megapixel rear autofocus, a wide aperture for extra light reaching image sensor, and an LED (light emitting diodes) flash. Software attains RGB values at fixed distance from the samples as well as exact focused point in all the tubes to minimize light differences and have more precise results.

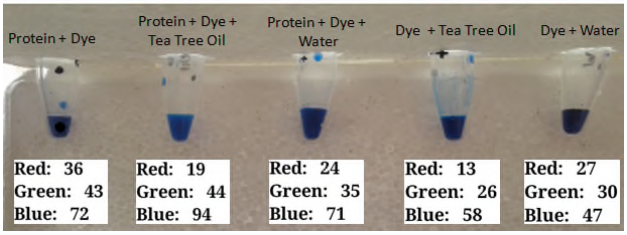

Figure 5. RGB Values of the Prepared Samples in Liquid Phase

Figure 5 shows the different results obtained from the experiment at the same time. The figure shows the prepared samples with a label with its content above each tube, while the RGB values are listed below the tubes. Detailed explanation of the results is further mentioned in the discussion.

We keep the following conditions of the samples constant:

- Temperature using water bath
- pH levels using ALP; stable pH
- Volume and concentrations using pipettes, balance, and dilution factor

We used alkaline phosphatase as the protein due to its key features:

- 4 hydrophobic pockets are necessary to interact with the acquired odors
- Stable pH levels reduce odors' pH influence
- Temperature stability (up to 80 degrees) [9].

We choose tube 1 to the left as the reference and control to compare the color change of the rest of the tubes to it. Following is the protein and dye interacting with tea tree oil, transparent and low viscosity organic oil with a specific chemical structure.

To eliminate the effect of dilution that might be the reason of the lighter blue color of the sample, we add in the 3rd tube with the same volume of the tea tree oil than that of tube 2 and notice that there is difference in color as compared to the mixture with the organic oil. Thus, another factor is studied, and this showed positive results.

In the following tubes, 4 and 5, we tested for the interaction between the organic oils and dye. It is required to validate that the interaction is with the protein and not the dye. Comparing the results of the tube 4 to tube 2, and tube 5 to tube 3, the dye did not quite change its color, which made us conclude that there is minimal interaction between the oils and dye.

IV. DISCUSSION

Electronic noses were originally used for quality control applications in the food, beverage and cosmetic industries. Current applications include detection of odors specific to diseases for medical diagnosis, and detection of gas leaks for environmental protection [10].

The advancement of e-noses may be coupled with different sensor technologies, such as optical sensors, conductive polymers, and in our case biopolymers.

Initiating our analysis, a color sensor is used, combined with an arduino that controls and acquires data received by the sensor. First experiments showed detection of color change of the RGB values while stabilizing surrounding conditions to prevent artifacts from influencing our results. Yet, after maintaining constant light, temperature, and sample settings, inadequate results were obtained. Any little light disruptions, as well as inappropriate distance of sensor from samples caused false results. In addition, reflections off of objects placed in the surroundings, either sample or sensor, did have a wrong impact on the results. Thus, another detection method is used after several trials performed earlier. CCD (charge-coupled device) camera is considered more convenient than the color sensor due to the high efficiency of this camera, as well as the broad availability of this sensor. Combined with a microcontroller or analyzing unit, this arrangement makes way to detecting the RGB value changes more accurately than that of the color sensor, as well as proper analysis of the results.

It is validated that DNA interacts with odors as mentioned in Tuft's University experiments. The experiments conducted here are through using proteins as biopolymers instead of DNA. It is known that protein normally binds lipids. Most odors are hydrophobic, which makes them candidates to bind to the hydrophobic pockets available in the protein structures used here [11].

The specific influences of conditions that might affect the interaction of the protein with the samples are avoided. Temperature is maintained constant at 37 degrees, using a water bath. The first experiments included the use of LDH (lactate dehydrogenase) and BSA (bovine serum albumin) proteins. Several trials were conducted on these proteins. In addition, to prevent any pH fluctuations, ALP is used

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instead of the previous proteins for its high affinity to bind lipids without changing pH levels. Setting pH at a specific value is required, for we are not probing the change in pH, but we shall see odor sensitivity.

In order to visualize color change, a labeling substance is used. The marker used here is the coomassie blue dye. To detect the color change after an interaction between the biopolymer and certain odors, the dye fluoresces and changes its degree that facilitates the visualization of the interaction occurring between the protein and volatiles. The coomassie blue dye associates with basic and aromatic amino acids, thereby causing shift in absorbance during protein determination [12].

The volatiles used in this experiment are coconut oil, rose water oil, cloves oil and tea tree oil. Coconut oil has high viscosity that prevented proper mixing with proteins. Rose water and cloves oils already have a non-transparent color, thus they will affect the RGB values analysis. We used the tea tree oil, a transparent, low viscosity, and essential organic oil with a specific chemical structure is then used.

Binding of hydrophobic entities like odors may induce structural changes to protein due to the presence of hydrophobic pockets that may change their optical characteristics. Such interaction with lipids will induce structural changes down to the helical structure of proteins (Angstroms measurement unit) that affect their color nature, e.g.: globin proteins that change color from dark red to light violet based on structural change; cytochromes, hemoglobin, etc.

Optical activity of substance changes based on its modification in structure impacts transparency vs. opacity as well as color vs. color change.

The detected difference of the RGB values can be visualized both using the naked eye, as well as by using the application. The application uses the camera that is set to measure RGB values so there is no need for any pre-processing.

V. CONCLUSION

As a conclusion, the focus of this project is to mimic the functionality of the olfactory system using materials available in every lab. This makes way to producing a cost-effective and easy to use device to perform the necessary function. The experiments were performed on similar materials as that of Tuft's University then deviated the attention onto using proteins instead of DNA as the sensors for odor identification. It was validated that each gaseous and liquid phased molecules and odors interact differently when in contact with different proteins. This variance is used to check for better repeatability, sensitivity, and stability. This is analyzed by using a developed LIU E-nose application that records and displays the values of change in color post protein-odor interaction. Further practice shall lead to a better understanding of the specific interaction with given odors that can lead to notable discoveries in the field of diseases' testing and identification as well as in other daily-life fields.

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