An Electronic Nose using a Protein-Biosensor and a Camera-Based Application

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***Abstract* – Human's olfactory system has a large number of receptors that respond only to a limited number of molecules. Since human noses are subject to fatigue, inconsistency and lack the sensitivity to low-concentration molecules, scientists have developed the Electronic nose (E-nose). E-nose is a device that identifies the specific components of volatiles and analyzes the chemical makeup to identify them. Regarding the applications of this device, they include, but not limited to, detection of odors specific to diseases for medical diagnosis and are used for the 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 like that of the neural network. As for chemical detection and 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 compatible 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. Several trials were, and will be, further performed to validate the interaction between Biosensors and odor samples, in both liquid and solid phases.**

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][2]. 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 [3]. 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.

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 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, and 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 a chain of smaller ones) and a fluorescent dye. If any 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. Tuft's University device where it could be tailored to be used in the food and beverage industry, ensuring high quality products and detecting possible contaminants [5].

As for the medical field, other researchers have come up with a device, later called Nano Artificial Nose (Na-Nose) [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.

A prototype of cross-sensitive nanowire-based sensors to be integrated in the 'Nano Artificial Nose' was trained to detect target disease related mixtures of biomarkers. Advanced development of the Nano Artificial Nose disease detection capabilities is present for the detection of the following indications (bacteria and other pathogens) from exhaled breath: Streptococcus; Methicillin resistant (MRSA); Staphylococcus; etc. Na-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 [6].

Relating to Tuft's University scientists' work, our prototype was initially intended to mimic the structure and functionality of the earlier projects. During experimentations, some modifications were performed that made way into what advancements in materials used in this project and composition of testing procedures along with the results.

The rest of the paper is structured as follows. Section II represents the materials used, the requirements and device design, the project working process and the methods used. Afterwards, in Section III, experiments, results and data analysis are described. Then, in Section IV, results are discussed and explained. Finally, in Section V, 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 1 below shows the simplicity of the system; it is composed of the color sensor (middle), Arduino circuit (bottom) that acts as the brain of the system, and the required sample (upper images) to be tested. Following, Figure 2 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 3 describes the LIU (Lebanese International University) E-nose Application screens.

The following diagram describes the initial method of detecting change in color of the Biopolymer. It consists of the Arduino Uno microcontroller, which acts as the brain of the system and communicates between the color sensor and computer.

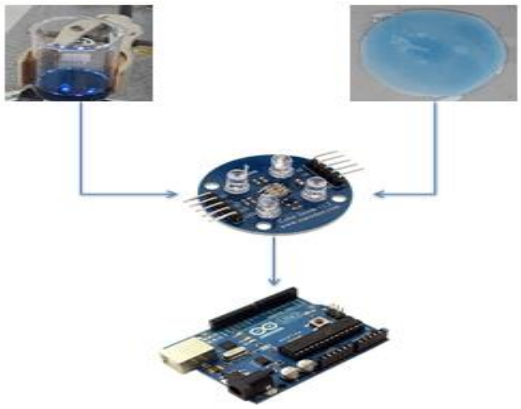

Figure 1. Color Sensor and Arduino Block Diagram.

The color sensor identifies the RGB spectrum of colors of the biopolymer, and then the results are visualized on the computer after being converted and processed by the Arduino. The experiment conducted is performed in both, the liquid and solid phase of the biopolymer.

This experiment was a proof of concept of the experiment done by Tufts University and the initial step of the upcoming experiments.

The diagram illustrated in Figure 2 describes the second and main method for detecting color change. The fan acts in drawing the odors from one part of the chamber with the least amount of turbulence towards the biopolymer sensor. After the biosensor and odor interact, change in color of the sensor composition is recorded by the phone camera. The phone flash light acts in transmitting visible light towards the sensor and the camera in receiving the light reflected. The data is analyzed and processed by an application programmed for this specific project. The application identifies the RGB spectrum, records the received data through an image taken, and saves the results in the phone library.

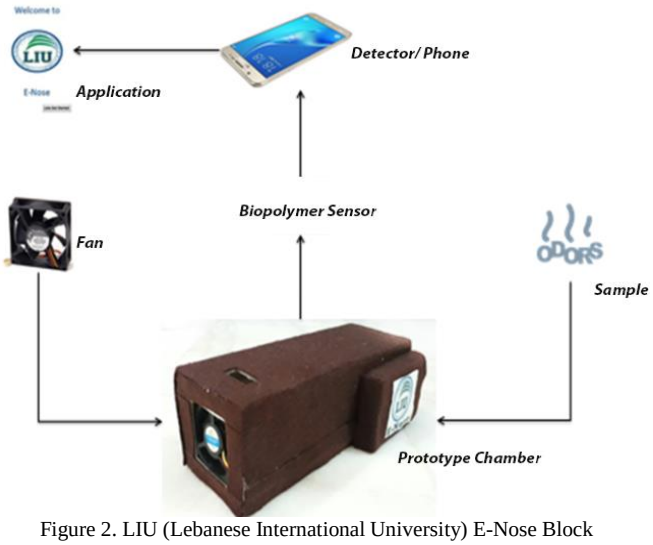

Figure 2. LIU (Lebanese International University) E-Nose Block Diagram.

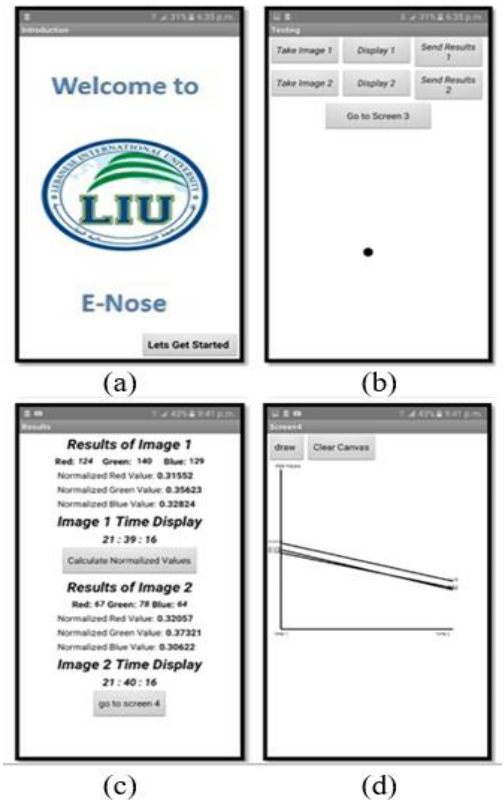

Figure 3. LIU E-Nose Application Stages: (a) Screen 1, (b) Screen 2, (c) Screen 3, (d) Screen 4.

Throughout the experiments performed on both methods, it was found more specific, accurate, and easier to choose the phone camera to identify and detect the color change.

The application is divided into four main screens. Screen 1 is the cover page of the application, which we can get started from. Screen 2 is the testing phase of the application. The user inputs the two images; the first one is that of the biosensor prior to interaction with the odor, then image two is taken after the fan draws the odor towards the biosensor and interacts with it. We display the images, one and two, and the ball is dragged to a pre-specified point that relates to the location of the biosensor in the chamber. The specified point relevant to the ball location is where the RGB values are extracted and are then saved. Then the results are sent to screen 3; the results screen. Each image taken on screen two has its RGB values saved here and displayed. In addition to the RGB values and normalized values, the time at which each image was taken is saved and displayed on this screen. Finally, in screen 4, a graphical diagram displays the change in RGB values before and after the interaction took place.

LIU E-nose application serves as the image processing software programmed using MIT (Massachusetts Institute of Technology) App inventor [7]. 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.

A. Proteins

One major factor that describes these chosen proteins is their amphipathicity. Amphipathicity describes the surfaces on a protein, particularly the alpha helix, which mean that one surface of the alpha helix has hydrophilic amino acids while the opposite face has hydrophobic (or lipophilic) amino acids.

Bovine Serum albumin (BSA) is the most abundant plasma protein in mammals. SA is a multifunctional protein with extraordinary ligand binding capacity. Albumins exhibit a high degree of cross-reactivity due to significant sequence and structure similarity of SAs from different organisms [8].

LDH (Lactate Dehydrogenase) is an enzyme found in nearly all living cells and is composed of four subunits (tetramer). LDH in humans uses His(193) as the proton acceptor, and works in unison with a coenzyme, and substrate binding residues [11]. The His(193) active site, is not only found in the human form of LDH, but is found in many different animals, showing the convergent evolution of LDH. A dehydrogenase is an enzyme that transfers a hydride from one molecule to another [9].

ALP (Alkaline phosphatase), from bovine intestinal mucosa, is most stable in the pH range 7.5-9.5. The enzyme has a broad specificity for phosphate esters of alcohols, amines, pyrophosphate, and phenols and it requires zinc, and magnesium or calcium divalent ions for activity. These characteristics give rise for the stability and structure suitable for the project's main application [10].

B. Dye

In this experiment, it is required to monitor color change. This factor is not present for the protein alone but requires an additional color label that facilitates monitoring this change. The label used here is the coomassie blue dye that is used to glows after an interaction between the biopolymer and certain odors takes place. The coomassie is the most suitable and commercially available colorant for this experiment.

C. Organic Oils:

To limit the specificity of the odors used in this experiment, we used three types of odors that are: a) widely abundant in all homes, b) cheap and available, and c) are considered essential oils that minimize the quantity of these oils used. In the beginning of the experiments, coconut and rose water oils are used, but since they have certain limitations, (high viscosity of coconut oil, rose water being non-colorless) those factors influenced false results. To avoid these aspects, we used tea tree oil that is colorless, has low viscosity like that of water's, and has concentrated scents.

III. Preparation Steps

A. *Experiment 1: LDH – Coomassie using LIU E-Nose Application:*

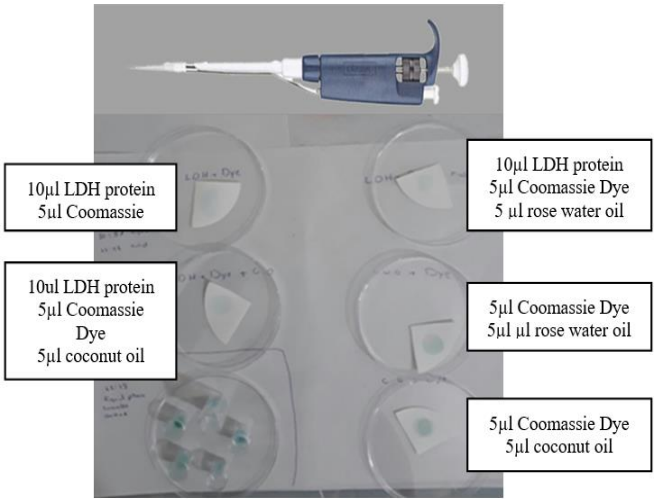

Figure 4. LDH - Coomassie - Oils (liquid and solid) Preparation.

B. *Experiment 2: ALP – Coomassie Solid State against Exhaled Volatiles*

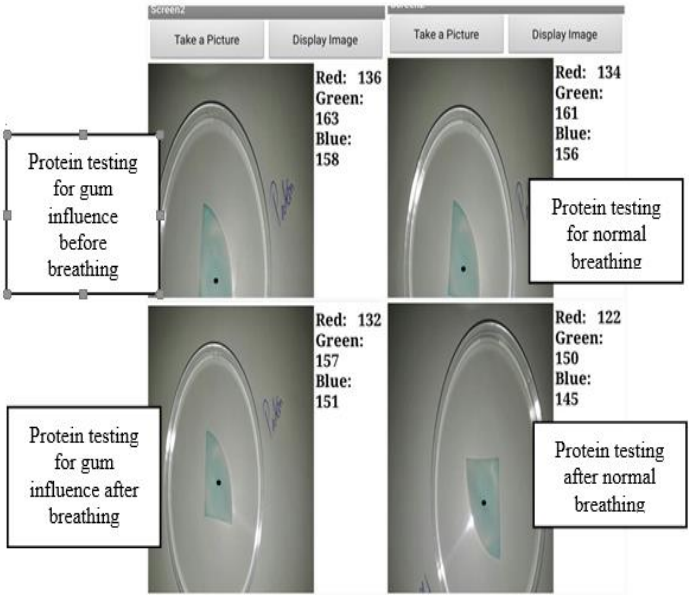

Figure 5. ALP vs Exhaled Volatiles in Solid Phase.

In this experiment, a different approach was followed. Using the same procedure as the ones studied earlier, but without adding the organic oils to the samples, instead the Whatman papers only contained 5g/l concentration of ALP protein and 5 ul dye volume.

The odors now used are human breaths. The approach here was to monitor the change in color of the protein-dye mixture due to their interaction with chemical gases exhaled from a certain individual. Prior to any exhale, the color of the paper was recorded and noted down. Then, at one hand, the individual was asked to exhale with ordinary breath and noted down the difference in the color as shown in Figure 5, right column. On the other hand, the same patient was asked to chew a gum for 3 min then exhale with the same pressure and volume as that of the first trial and recorded the results afterwards.

The two trials showed positive results to the interaction between the protein and the exhaled gases. Additional experiments were held using the prototype and the LIU E-Nose application to validate the previous results.

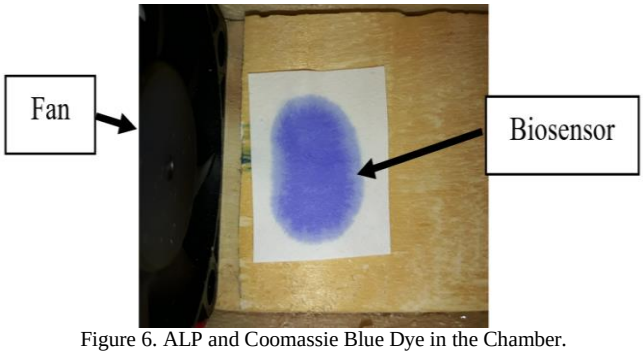

Figure 6. ALP and Coomassie Blue Dye in the Chamber.

The biosensor was situated in the specified location on the biosensor-odor slider inside the chamber, as shown in Figure 6. Using the application, the first image was taken before any interaction with the biosensor and then the second image was taken after an individual exhaled on the biosensor; knowing that the same person was chosen to perform both previous experiments; data was received and saved by the application.

C. *Experiment 3: ALP – Coomassie Interaction with Tea Tree Oil in Liquid Phase*

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
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

IV. RESULTS

Combining the previous installments and procedures, several experiments were performed to identify and allocate the appropriate combination of ingredients. And in order to assess the most suitable experiment, comparison was performed between the different proteins, odors, and even phases of mixtures. Different proteins give rise to which is most suitable to use. From different odors, we can clear out some artifacts and specify which has the least effect on other components. Whereas from different phases, liquid or solid, we can somehow reach a state of testing very similar to how the natural nose works.

A. Experiment 1 results:

1) Results in liquid phase:

Using a 5-50 µl pipette, prepare the previous mixtures with the values of each paper listed beside it; noting that the liquid prepared solutions are the same as those of the solid phase ones.

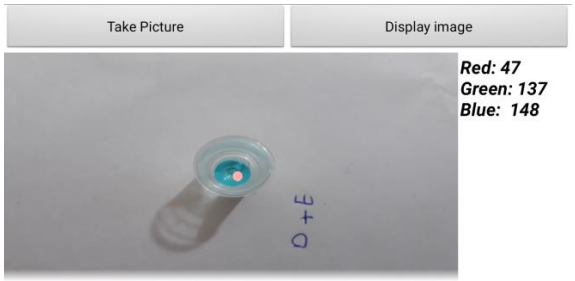

Figure 8. Testing in liquid phase (LDH protein + Coomassie Blue Dye).

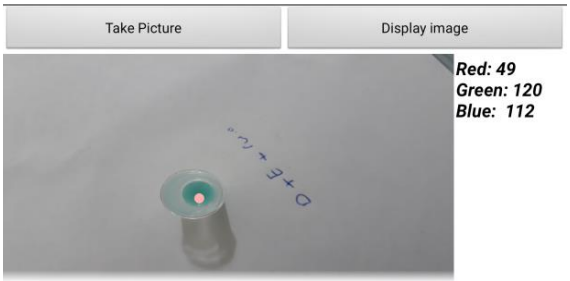

Figure 7. Testing in liquid phase (LDH + Dye+ Rose Water).

As for the LDH protein and dye, it is the main biosensor through which results of the other added odors are compared. The lighter mixture, rose water, that is added to the previous biopolymer mixture, shows different RGB values, both visualized by the application as well as the naked eye. These results give rise to verifying the different interactions between the protein-dye mixture and different odors. But what if the change in color was due to the oil interacting with the dye? The previous question is answered through taking other prepared dye samples and adding the oils to them. The results of these two different samples are visualized while comparing Figures 7 and 8.

Figure 9 shows that the interactions between the dye and organic samples are different than those with the protein-dye mixture. This verifies that the original color of oils factor is well studied, and error possibility is significantly minimized that allows studying the interaction between the protein-dye mixtures with added organic oils.

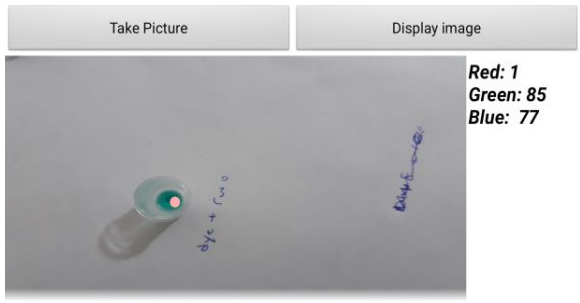

Figure 9. Rose Water Oil + Dye in Liquid Phase.

2) Results in solid phase:

Another experiment was conducted in the dried-up phase of the protein and dye on Whatman paper. The procedure is described as adding the given quantity of protein onto the paper, then dropping the quantity of dye onto the protein. Following, the organic oils are finally added to the previous mixture.

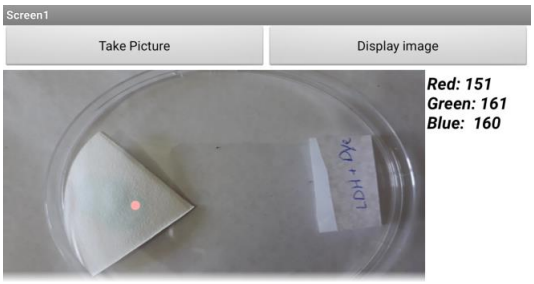

Figure 10. LDH + Dye in Solid Phase.

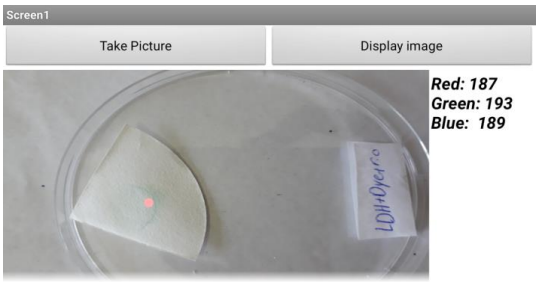

Figure 11. LDH + Dye + Rose Water.

Consider the previous preparations in Figures 10 and 11, having the same volume and concentration of the protein and dye, oil added after a period of 5 min shows different recorded RGB values between the two. Whereas, when adding different organic oils, each composition reacts in a specific manner with respect to the oil used.

B. Experiment 2 results:

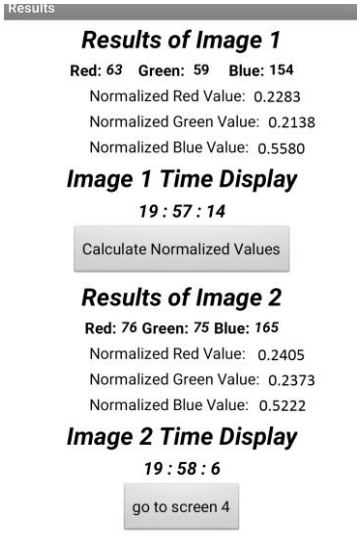

Figure 12. RGB Values before and after the Exhaled Gases.

As shown in Figure 12, the results screen from the application, the trials showed positive outcome to the interaction between the biosensor and the exhaled gases. In the following graphical diagram of Figure 13, we can visualize the change in color as enlisted in the results screen previously.

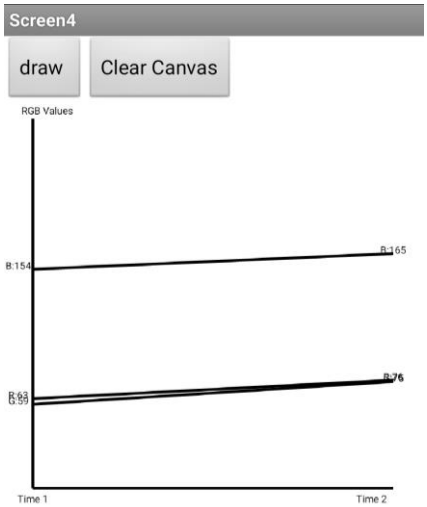

Figure 13. Graphical Diagram Displays the Change in RGB Values before and after the Interaction.

In this experiment, the same preparation steps, conditions and materials of that of the previous experiment were used. The approach was to perform the test 30 times on the same biosensor and by the same given individual. After every trial, the setup was flushed by a reference gas in order to return the RGB values to the baseline. Following that, a period of 2 minutes was taken (recovery period) to make sure the biosensor returns to the initial state (baseline) before the interaction with the odors.

TABLE I. STATISTICS OF EXPERIMENT PERFORMED 30 TIMES.

	before exhale			after exhale		
	Red	Green	Blue	Red	Green	Blue
Trial 1	63	59	154	76	75	165
Trial 2	62	58	154	73	72	164
Trial 3	62	58	156	72	71	165
Trial 4	63	57	153	75	69	162
Trial 5	64	58	153	77	70	163
Trial 6	64	59	152	78	73	160
⋮	⋮	⋮	⋮	⋮	⋮	⋮
Trial 25	62	60	152	71	76	166
Trial 26	63	57	154	75	74	164
Trial 27	65	58	153	74	73	162
Trial 28	63	58	154	76	75	167
Trial 29	64	59	155	70	76	170
Trial 30	63	59	154	71	72	168
Average	63.16667	58.33333	153.6667	74	73	164.6667
Mean Average	63	58	154	74	73	165
% error	4.761905	5.172414	2.597403	10.81081	8.219178	6.060606
STD	0.897527	0.849837	1.105542	2.483277	2.198484	2.687419

Pre-exhalation:

Repeatability of RGB values before exhalation was consistent with an error of less than or equal to 5% → this validates that:

- the baseline of RGB values was maintained after each exhalation
- the odors absorbed by the proteins are rid due to flushing reference gas for a recovery time of 2 minutes

Post-exhalation:

Error percentage was too high since breathing wasn't consistent (pressure, volume, temperature, and pH of exhaled breath) were not the same in all exhalations.

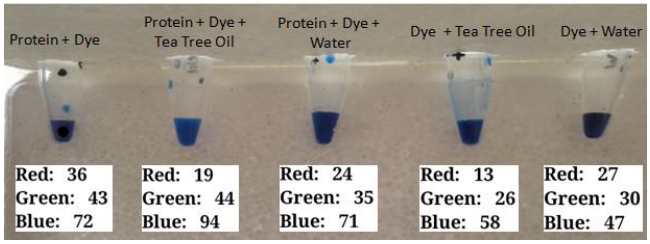

Figure 14. RGB Values of the Prepared Samples in Liquid Phase.

C. Experiment 3 results:

Figure 14 shows the different results obtained from the experiment at the same time. It 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) [11].

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 the same volume of the tea tree oil same as 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.

V. Related Work

Recent advances demonstrate that proteins, ORs (olfactory receptors) or OBPs (odorant-binding proteins), can be employed as molecular recognition units in gas-phase biosensors. The interactions between odorant molecules and these proteins are a source of inspiration for designing peptides with tunable odorant selectivity [12].

Peptides represent a simple and low-cost option for biosensors that can be biologically or synthetically produced. Initially, OBP biosensors were tested against volatiles and odor solutions, with measurements of analytes in gas phase only reported by the end of the 2000s. These peptides are considered easier to place and stabilize on sensor surfaces than ORs [12].

Some methods for testing of VOC (Volatile Organic Compounds) were used. Each had a certain significance in respect to the peptides used as biosensors. The following table, Table II, summarizes the strategy used, from relevant biosensor to the equivalent VOC and the type of transducer specific for this method. Each testing group had specific support; each had a specific-to-biosensor material to be mounted on.

The different methodologies mentioned in the following table are relevant to the different techniques of testing VOCs throughout the years of advancements of volatiles' identification.

Table II: Biosensors and Transducers for a Specific VOC

Protein	Protein Origin	Transducer	VOC	Support	Reference
pOBP	Pig OBP	Si-substrate with interdigitated electrodes (EIS)	Ethanol; methanol	Silicon	[13]
AaegOBP22	Mosquito Aedes aegypti	ZnO film bulk acoustic resonators (FBAR)	N,N-diethyl-meta-toluamide (DEET)	Gold	[14]
mOR174-9	Mouse OR	CNT (carbon nanotubes) transistors, Current-gate Voltage	Acetophenone, Others	Carbon Nano-Tubes	[15]
NQLSNLSF SDLC (dORp61)	Dogs OR	Piezoelectric multiarray analyzer	Trimethylamine (TMA); ammonia; acetic acid; ethyl acetate; methanol	Gold Surface	[16]

Following are some explanations to the different transducers used above:

- Dielectric spectroscopy measures the dielectric properties of a medium as a function of frequency. It is based on the interaction of an external field with the electric permittivity of the sample [17].
- A thin-film bulk acoustic resonator (FBAR) is a device consisting of a piezoelectric material sandwiched between two electrodes and acoustically isolated from the surrounding medium [18].
- A carbon nanotube field-effect transistor (CNT) refers to a field-effect transistor that utilizes a single or array of carbon nanotubes as the channel material instead of bulk silicon structure [19].
- A piezoelectric sensor is a device that uses the piezoelectric effect to measure changes in pressure, acceleration, temperature, strain, or force. Then these factors are converted to electrical signals for identification [20].

Using biological recognition units such as insect/ animal-based proteins in sensing devices improves their selectivity and sensitivity towards defined targets, with proven pronounced expression in samples analyzed in liquid states. The behavior of these biosensors in testing analytes in the gas phase is also recommended to attempt and compare. Means of mimicking the actual olfactory system functionality gives rise to complementing real-life scenarios.

Based on the techniques discussed in similar projects, there are several challenges present that inspire future developments. These challenges may include drawbacks of low stability and high promiscuity towards VOC molecules, resulting in low selectivity. The difficult, expensive, and time-consuming handling of membrane proteins may have led to the pursuit of simpler and more robust biomolecules as recognition agents; therefore, gas-sensing biosensors based on the soluble proteins [21].

Some similarities in application were found in the technology used in the previous experiments while being compared to the project under study.

A) OR response to VOCs may be distorted due to the presence of the lipidic fraction of the membrane such as phospholipids. LIU e-nose proteins used act in binding odors to the phospholipids of proteins used.

B) OBPs can withstand high temperatures before experiencing denaturation, and after unfolding they frequently refold, restoring their initial structure. Stability of pOBP increases at higher temperatures [22]. Researchers used animal proteins to test for the affinity of interaction between biosensors and volatiles and studied their influence while relating to different temperatures. On the other hand, our project used alkaline phosphatase protein that incorporates temperature stability prior to and after interaction with volatiles.

C) A detailed analysis of the VOC-binding pocket from OBPs was tested, even though the target VOC is different. Here, testing included the study of binding pockets from OBPs while ours discusses the pockets of ALP structure and their interaction with and immersing of VOCs at the site of structural pockets.

Over the last 5 years, there has been an increase in OBP- and peptide-based biosensors. Certainly, using membrane protein receptors as ORs is more challenging, expensive, and time consuming, when compared with soluble and robust proteins such as OBPs. An investment in the expansion of tested VOCs for the development of gas biosensors is required, as recent examples report a very limited number and chemical diversity of VOC targets.

VI. 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 [23].

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 objects placed in the surroundings, either sample or sensor, did have a wrong impact on the results. Thus, another detection method is to be used after several trials performed earlier. CCD (charge-coupled device) camera is considered more convenient than the color sensor due to its high efficiency, 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 that make them candidates to bind to the hydrophobic pockets available in the protein structures used here [24].

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 and BSA proteins. Several trials were conducted on these proteins. In addition, to prevent any pH fluctuations, ALP is used 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 [25].

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.

VII. 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.

Current research is focused on making the devices smaller, less expensive, and more sensitive. The previously mentioned are crucial factors to the analysis and odor identification. The smaller the device, the more user friendly it is. Being more affordable increases the number of users, as well as ensures better testing of daily-life products, and more importantly better disease identification. Thus, 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. The possibilities are almost endless.

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