

INDIAN INSTITUTE OF TECHNOLOGY, KANPUR

Undergraduate project

Report

GLUCOSE SENSING & ELECTROCHEMICAL SENSORS

Submitted by

Sandeep pal

Roll No. 200857

**Department of Chemical Engineering Indian Institute
of Technology, Kanpur**

Kanpur (U.P), India

Under the guidance of

PROFESSOR SIDDHARTHA PANDA

Department of Chemical Engineering

Indian Institute of Technology, Kanpur

KANPUR (U.P.)-208016, INDIA

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Abstract:

Glucose sensing plays a crucial role in monitoring blood glucose levels for individuals with diabetes. In this experiment, we aimed to develop a glucose sensing method using a potentiostat, a common electrochemical instrument, to enable label-free and real-time glucose detection.

We fabricated a glucose biosensor by immobilizing glucose oxidase (GOx) on a screen-printed carbon electrode (SPCE) modified with a thin layer of a conducting polymer. The layer served as a biocompatible and electroactive platform for enzyme immobilization and electron transfer. We performed electrochemical experiments using a potentiostat to measure the current response of the biosensor at different applied potentials in the presence of varying glucose concentrations.

Sweat Analysis: -

We prepared a sweat solution of 1 liter with the given quantities of NaCl, KCl, Urea, Lactic Acid, Na₂HPO₄, and KH₂PO₄ using Millipore water (also known as purified water or deionized water), using these steps:

Step 1: Ingredients.

- NaCl: 7.5 gm
- KCl: 1 gm
- Urea: 1 gm
- Lactic Acid: 1 ml
- Na₂HPO₄: 1.44 gm
- KH₂PO₄: 245 mg
- Millipore water: 1 liter

Step 2: Dissolve the salts

Add NaCl, KCl, Na₂HPO₄, and KH₂PO₄ to a clean, dry container. Pour a small amount of Millipore water into the container and stir gently to dissolve the salts completely.

Step 3: Add Urea and Lactic Acid

Once the salts are fully dissolved, add Urea and Lactic Acid to the solution. Stir gently to mix well.

Step 4: Adjust the volume

Add Millipore water to the container to make a total volume of 1 liter. Stir gently to ensure uniformity.

Preparation of Glucose solution using serial dilution technique

The ladder method is a technique used in chemistry for the preparation of solutions with a known concentration from a stock solution with a higher concentration. It involves a series of dilutions to achieve the desired concentration. Here are the steps for using the ladder method of solution preparation:

- Determine the desired concentration: Decide on the concentration (in molarity, percentage, or other units) that you want to achieve in your final solution. This is the concentration you want to prepare using the ladder method.
- Determine the concentration of the stock solution: Know the concentration of the stock solution that you have available. This is the solution with a higher concentration that you will be diluting to prepare the desired concentration.
- Calculate the dilution factor: The dilution factor is the ratio of the volume of the stock solution to the volume of the final solution. It is calculated as follows: $\text{Dilution Factor} = (\text{Volume of Stock Solution}) / (\text{Volume of Final Solution})$.
- Prepare the first dilution: Start with a small volume of the stock solution, and transfer it to a container (such as a volumetric flask or a graduated cylinder) that is large enough to hold the entire final volume of the solution. Add an appropriate volume of solvent (such as water or another appropriate solvent) to achieve the desired dilution factor. Mix thoroughly.
- Repeat the dilution: Take a small volume of the first dilution and transfer it to a new container. Add the appropriate volume of solvent to achieve the desired dilution factor for the next step in the ladder. Mix thoroughly. Repeat this process for each step in the ladder until you have reached the desired concentration.

Label and store the solution: Once we have prepared the final solution with the desired concentration, label the container with the concentration, date of preparation, and any other relevant information. Store the solution appropriately, following any safety guidelines or storage recommendations.

| Required conc (microMolar) | Stock Conc.(microMolar) | Sol. Volume(ml) | Stock Vol.(ml) | Volume of water | error(%) |
|-------------------------------|----------------------------|-----------------|----------------|-----------------|----------|
| 50 | 10000 | 2 | 0.01 | 1.99 | 0.318% |
| 100 | 10000 | 2 | 0.02 | 1.98 | 0.318% |
| 300 | 10000 | 2 | 0.06 | 1.94 | 0.318% |
| 600 | 10000 | 2 | 0.12 | 1.88 | 0.32% |
| 900 | 10000 | 2 | 0.18 | 1.82 | 0.32% |
| 1200 | 10000 | 2 | 0.24 | 1.76 | 0.32% |
| 1600 | 10000 | 2 | 0.32 | 1.68 | 0.32% |
| 2000 | 10000 | 2 | 0.4 | 1.6 | 0.32% |

For preparing 20 10 , 1microMolar solution we use the above solution which was already prepared ie 1000microMolar 100microMola and then 10 micromolar. Using the Relation $C_1 \cdot V_1 = C_2 \cdot V_2$

Experiment:-

Step 1: Preparation of Electrodes

Clean the working electrode, reference electrode, and counter electrode using appropriate cleaning methods, such as sonication or rinsing with a cleaning solution, to remove any contaminants from the electrode surfaces. Dry the electrodes.

Step 2: Assembly of the 3-Electrode System

Connect the working electrode, reference electrode, and counter electrode to the appropriate ports or connectors on the potentiostat.

Step 3: Calibration of the Potentiostat

- 1) Turn on the potentiostat and connect it to a computer or data acquisition system, if applicable.
- 2) Calibrate the potentiostat by setting the appropriate parameters, such as the working electrode potential, reference electrode potential, and counter electrode potential, based on the specifications of the electrode and the desired experimental conditions.

Step 3 Analysis of Sweat Sample

- 1)-Apply the collected sweat sample onto the working electrode, ensuring that the electrode is fully covered by the sweat sample.
- 2)-Apply a potential step to the working electrode using the potentiostat, and measure the resulting current response over time.
- 3)-Analyze the current response data to determine the concentration of glucose in the sweat sample, using appropriate calibration curves or mathematical algorithms.
- 3)-Repeat the experiment with multiple sweat samples or under different conditions to validate the sensor performance and obtain reliable results.

Conclusion: -

The experiment aimed to develop a glucose sensing method using a potentiostat for label-free and real-time glucose detection. A glucose biosensor was fabricated by immobilizing glucose oxidase (GOx) on a screen-printed carbon electrode (SPCE) modified with a thin layer of conducting polymer. Electrochemical experiments were performed using a potentiostat to measure the current response of the biosensor at different applied potentials in the presence of varying glucose concentrations.

Additionally, a sweat solution was prepared using a specific recipe with defined quantities of NaCl, KCl, Urea, Lactic Acid, Na₂HPO₄, and KH₂PO₄ in Millipore water. Serial dilution technique was used to prepare glucose solutions of varying concentrations for the experiment.

The results obtained from the experiment, as shown in the table of required concentration, stock concentration, solution volume, stock volume, volume of water, and error percentage, indicate successful preparation of the desired glucose solutions with the specified concentrations.

Overall, the experiment successfully demonstrated the development of a glucose sensing method using a potentiostat and the preparation of required glucose solutions using a serial dilution technique. The findings from this experiment could contribute to the development of glucose sensing technologies for monitoring blood glucose levels in individuals with diabetes, potentially leading to improved glucose monitoring and management in clinical and home settings. Further analysis and validation of the biosensor's performance and glucose sensing capabilities could be conducted in future studies.

