PORTABLE ELECTROCHEMICAL DEVICE FOR RAPID ON-SITE DETECTION AND QUANTIFICATION OF BACTERIA IN WATER

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Introduction:

Safe drinking water scarcity globally necessitates rapid, cost-effective microbial detection, especially *E.coli*, a key contamination indicator. Traditional methods are slow, expensive, and lab-dependent, hindering field use. Electrochemical biosensors offer a faster, sensitive alternative by monitoring bacteria-electrode interactions via chronoamperometry. Their portability suits remote areas. However, enzyme-based biosensors lack durability, and costly potentiostat / complex electrodes limit adoption, highlighting the need for robust, affordable sensors with simpler materials and instrumentation. Our research tackles this by creating a novel nickel electrode-based electrochemical biosensor for quick, affordable *E.coli* detection in water, aiming for field-based water quality monitoring. To bypass expensive commercial potentiostat, we designed a custom PCB-based potentiostat optimized for our three-electrode system. This integrated solution offers a more accessible, scalable tool for microbial detection, ultimately enhancing water safety and public health.

Objectives:

- Develop a portable electrochemical device for rapid, on-site detection and quantification of *E.coli* in water samples.
- Integrate a miniaturized potentiostat and a three-electrode system into a handheld device to achieve precise electrochemical measurements for accurate bacterial analysis.
- Enhance the accessibility and usability of water testing through a device with simple operation and easy-to-understand results.

Methodology:

For electrochemical detection of *E.coli*, a three-electrode system was employed, featuring a nickel working electrode, platinum counter and pseudo-reference electrodes, connected to a potentiostat for precise electrochemical control and measurement. A specific *E.coli* strain was cultured in lysogeny broth at 37 °C and 200 rpm, concentrated via centrifugation at 5,000 rpm at an optical density of 0.6, and resuspended in sterile water. Standard solutions, including a 0.858 M KOH solution for nickel electrode activation and a 1:99 dilution of KOH in sterile water as a baseline for chronoamperometry, were prepared. Varying bacterial concentrations, created through serial dilutions and verified by standard plate counts, were used for testing. Cyclic voltammetry (CV) was utilized to activate the nickel electrode by scanning the potential between 0.20 and 0.57 V in 0.858 M KOH, forming a catalytic $Ni(OH)_2$ layer through the oxidation of nickel to Ni^{2+} and subsequent conversion in the alkaline solution. This active surface was stabilized by 300 CV cycles. Chronoamperometry was then employed to monitor the current response of the pre-treated nickel electrode in a solution of 99 parts *E.coli* suspension and 1 part 0.858 M KOH, with the KOH concentration optimized for bacterial viability. Before applying a potential, the open-circuit potential was stabilized. A potential of 0.58 V was applied for 5 seconds to oxidize $Ni(OH)_2$ to NiOOH, followed by a 10-second reduction at 0.10 V to regenerate $Ni(OH)_2$, with this redox cycle repeated for each sample to ensure consistent electrochemical measurements for accurate *E.coli* detection.

We have developed a miniaturized potentiostat, where each individual component and operational amplifier was simulated using PSPICE to validate its functionality. The components were then assembled on a breadboard, and the output was verified using an oscilloscope, which matched the expected results. Additionally, we have written and implemented the necessary code to perform both cyclic voltammetry and chronoamperometry.

Result and Conclusion:

The sensor's performance, tested with serially diluted *E.coli* (1:10, 1:100, 1:1000 in sterile water, then 99 parts sample + 1 part 0.858 M KOH, compared to a sterile water/KOH baseline), showed an inverse correlation: higher bacterial concentrations correlated with significantly lower current densities (Fig.1). This suggests that the lowest *E.coli* concentration (1:1000 dilution) produced the highest current, and vice versa. This current decrease with more bacteria is likely due to several factors. Firstly, the physical presence of *E.coli* cells impedes the diffusion of hydroxide ions (OH^-) crucial for forming NiOOH, the active species during oxidation, by blocking electrode sites. Secondly, bacterial metabolic activity near the electrode may further hinder OH^- transport, slowing the conversion of $Ni(OH)_2$ to NiOOH, thus reducing the current. Lastly, the presence of bacteria might indirectly affect water concentration at the electrode, influencing the reduction of NiOOH back to $Ni(OH)_2$, potentially contributing to the lower current observed at higher bacterial loads. These interconnected factors provide a mechanistic explanation for the sensor's response to different *E.coli* concentrations.

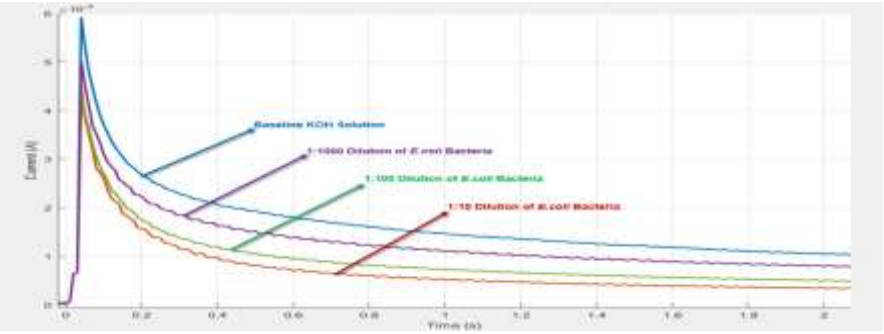

Fig.1. Chronoamperometry plot representing the average of current densities of four samples with known *E.coli* concentrations over three trials of oxidation at a fixed potential of 0.58 V vs. Pt.

Project Outcome & Industry Relevance

This project is a foundation for advanced microbial detection, envisioning a modular platform with interchangeable electrodes for diverse contaminant detection (heavy metals, nitrates, pH). Future work will optimize electrode materials (nanostructures, composites) for enhanced sensitivity and integrate user-friendly smartphone interfaces for data handling. Automated sampling/microfluidic systems for continuous testing and AI/ML for refined data analysis and prediction are planned. Applications could expand to food safety, medicine, agriculture, and environmental monitoring, with potential for large-scale sensor networks and solar power for sustainability. Engaging the public through citizen science is also considered. Crucially, future development will focus on improving durability, reusability, cost-effectiveness, and regulatory compliance for real-world impact in diverse water quality safeguarding.

Working Model vs. Simulation/Study:

A working prototype was developed featuring a three-electrode system mounted on a custom 3D-printed frame designed to securely position the electrodes within bacterial water samples. It uses a nickel working electrode and platinum electrodes as counter and reference, all connected to a custom potentiostat for precise potential control and current measurement, enabling effective *E.coli* detection in water.

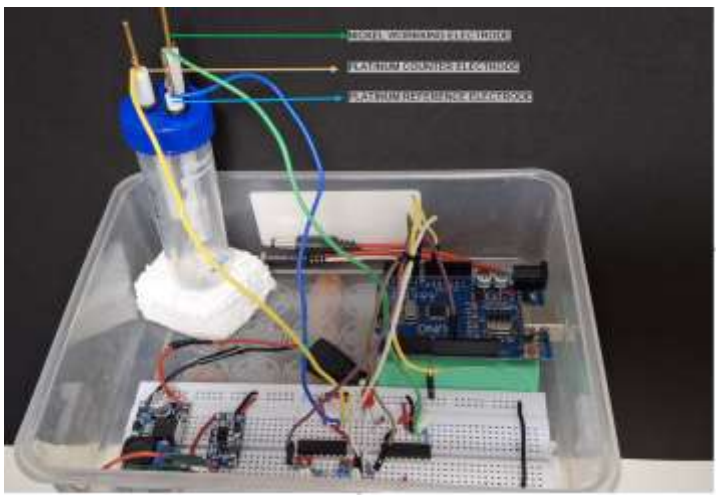

Fig.2. Miniaturized potentiostat with Three- Electrode Setup

Project Outcomes and Learnings:

A sensitive and reliable nickel-based electrochemical sensor was developed for detecting *E.coli* in water. Cyclic voltammetry and chronoamperometry were used to form a stable $Ni(OH)_2$/NiOOH redox layer, enabling effective bacterial interaction and measurable current response. The study highlighted key aspects of electrode surface modification, electrochemical optimization, and bacterial quantification. It also reinforced the value of precision, standardization, and reproducibility in developing practical sensing technologies..

Future Scope:

This project lays the foundation for advanced microbial detection and aims to become a versatile platform for monitoring various water contaminants such as heavy metals, nitrates, and pH levels. Future developments will focus on using improved electrode materials like nanostructures and composites to increase sensitivity. A user-friendly smartphone interface will be integrated for easy data handling. To support continuous monitoring, automated sampling and microfluidic systems will be added. Advanced data analysis and prediction will be achieved using AI and machine learning. The technology could be applied in fields such as food safety, healthcare, agriculture, and environmental monitoring. Importantly, future work will aim to enhance durability, reusability, cost-effectiveness, and compliance with regulations to ensure real-world impact.