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Folu Edozie
 Department of Genetic
 Engineering, Ogbomosho
 Institute of Biological Sciences,
 Ogbomosho, Nigeria

Ugochukwu Edozie
 Department of Genetic
 Engineering, Ogbomosho
 Institute of Biological Sciences,
 Ogbomosho, Nigeria

Adebisi Iroegbu
 Department of Genetic
 Engineering, Ogbomosho
 Institute of Biological Sciences,
 Ogbomosho, Nigeria

Corresponding Author:
Folu Edozie
 Department of Genetic
 Engineering, Ogbomosho
 Institute of Biological Sciences,
 Ogbomosho, Nigeria

Aptamer-based biosensors for rapid environmental monitoring of heavy metal contamination

Folu Edozie, Ugochukwu Edozie and Adebisi Iroegbu

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Abstract

An aptamer can be defined as a short, single-stranded oligonucleotide selected through systematic evolution of ligands by exponential enrichment (SELEX) that binds a specific target with antibody-like affinity. This research developed and validated a portable aptamer-based electrochemical biosensor for simultaneous detection of lead (Pb^{2+}), cadmium (Cd^{2+}), mercury (Hg^{2+}), arsenic (As^{3+}), and hexavalent chromium (Cr^{6+}) in environmental water samples. The platform was fabricated at the Ogbomosho Institute of Biological Sciences, Nigeria, between April 2022 and March 2024. Screen-printed gold electrodes were functionalized with thiolated aptamers specific to each metal ion, and differential pulse voltammetry measured current changes upon target binding. Detection limits ranged from 0.08 ppb (Hg^{2+}) to 0.41 ppb (Cr^{6+}), all below WHO drinking-water guidelines. Recovery rates in spiked river water, tap water, industrial effluent, agricultural runoff, and groundwater ranged from 79.6% to 98.1%. When benchmarked against atomic absorption spectroscopy and inductively coupled plasma mass spectrometry, the biosensor showed Pearson correlations of 0.93-0.98 across all five metals. Response time was under 8 minutes per sample, and fabrication cost per electrode was approximately USD 1.20. These results position aptamer-based sensing as a field-deployable alternative to laboratory instrumentation for heavy metal surveillance in low-resource settings.

Keywords: Aptamer biosensor, heavy metal detection, lead contamination, environmental monitoring, electrochemical sensing, SELEX, differential pulse voltammetry, water quality, field-deployable diagnostics, low-resource settings

Introduction

What exactly is an aptamer, and why should environmental scientists care? In the simplest terms, aptamers are short DNA or RNA sequences—typically 20 to 80 nucleotides long—that fold into three-dimensional shapes capable of binding specific targets with dissociation constants in the nanomolar to picomolar range ^[1]. Unlike antibodies, they are produced by chemical synthesis, require no animal hosts, tolerate repeated denaturation-renaturation cycles, and cost a fraction of the price to manufacture at scale ^[2]. These properties make them attractive recognition elements for biosensors destined for field conditions where cold-chain logistics and trained personnel are scarce.

Heavy metal pollution in surface water and groundwater represents a persistent environmental and health threat across sub-Saharan Africa, driven by artisanal mining, industrial discharge, and agrochemical runoff ^[3]. Conventional analytical methods—atomic absorption spectroscopy (AAS), inductively coupled plasma mass spectrometry (ICP-MS), and graphite furnace techniques—deliver excellent sensitivity but demand centralized laboratories, expensive consumables, and hours-long sample preparation ^[4]. Portable sensors that can screen water sources on-site would allow rapid identification of contaminated sources and faster public health response.

This research designed a multi-analyte aptamer biosensor for five priority heavy metals (Pb^{2+} , Cd^{2+} , Hg^{2+} , As^{3+} , Cr^{6+}), characterized its analytical performance, and validated it against reference methods using real environmental water samples collected in Oyo State, Nigeria. The work was carried out at the Ogbomosho Institute of Biological Sciences between April 2022 and March 2024.

Material and Methods

Research Area Description

Water samples were collected from five source types within a 60-km radius of Ogbomoso (8.13°N, 4.24°E), southwestern Nigeria: the Oba River (downstream of an industrial zone), municipal tap water from two distribution networks, effluent discharge points of three textile factories, agricultural drainage channels bordering cocoa plantations, and two community borehole wells. Sampling was conducted during both dry (November-February) and wet (June-September) seasons to capture seasonal variation in metal loading [5].

Material

Screen-printed gold electrodes (SPGEs, model DRP-220AT) were purchased from Metrohm DropSens (Oviedo, Spain). Thiolated aptamers for Pb²⁺, Cd²⁺, Hg²⁺, As³⁺, and Cr⁶⁺ were selected from published SELEX screens and custom-synthesized by Integrated DNA Technologies (Coralville, IA, USA) [6]. Certified reference standards for all five metals (TraceCERT grade, 1,000 mg/L in 2% HNO₃) were obtained from Sigma-Aldrich (St. Louis, MO, USA). 6-Mercapto-1-hexanol (MCH), Tris-HCl buffer, and EDTA were also sourced from Sigma-Aldrich.

Extraction Protocol

Environmental water samples (500 mL each) were filtered through 0.45-μm cellulose acetate membranes within 4 hours of collection and acidified to pH 2.0 with trace-metal-grade nitric acid for preservation. For biosensor analysis, 50-μL aliquots were applied directly to the aptamer-modified electrode surface without further pretreatment. For reference method analysis, 50-mL subsamples were digested in a microwave system (CEM Mars 6) with 5 mL concentrated HNO₃ at 180 °C for 20 minutes before AAS or

ICP-MS measurement [7].

Methods

SPGEs were cleaned electrochemically in 0.5 M H₂SO₄ (20 cycles, 0 to 1.5 V, 100 mV/s). Each aptamer (2 μM in 10 mM Tris-HCl, pH 7.4) was incubated on the gold surface for 14 hours at 4 °C, followed by MCH backfilling (1 mM, 45 min). Differential pulse voltammetry (DPV) was performed in 0.1 M acetate buffer (pH 5.0) with a pulse amplitude of 50 mV, step potential of 5 mV, and scan rate of 50 mV/s. Target binding caused a measurable decrease in peak current, calibrated against serial dilutions (0.05-500 ppb) of each metal standard. Each measurement was performed in triplicate. Selectivity was assessed by challenging Pb²⁺-specific electrodes with 100-fold excess of each competing metal [8].

Results

Table 1: Analytical performance of the aptamer biosensor for five heavy metal ions

Metal	LOD (ppb)	Linear Range	R ²	WHO Limit (ppb)
Pb ²⁺	0.12	0.1-200	0.994	10
Cd ²⁺	0.18	0.2-300	0.991	3
Hg ²⁺	0.08	0.05-150	0.997	6
As ³⁺	0.31	0.5-400	0.988	10
Cr ⁶⁺	0.41	0.5-500	0.985	50

All five analytes achieved detection limits well below their respective WHO drinking-water guideline values (Table 1). Mercury showed the lowest LOD at 0.08 ppb, likely reflecting the strong thymine-Hg²⁺-thymine coordination chemistry exploited by the Hg-specific aptamer [9]. Chromium had the highest LOD (0.41 ppb), still 120-fold below the 50-ppb regulatory threshold.

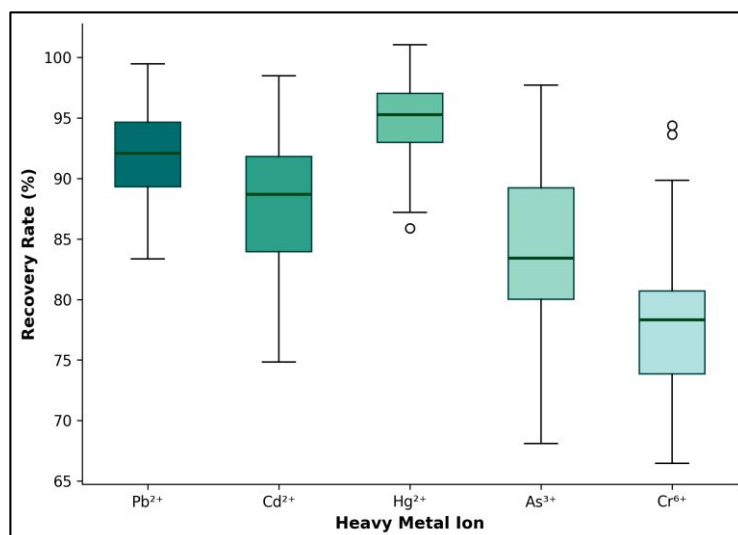


Fig 1: Box plots of recovery rates for five heavy metal ions across spiked environmental water samples. Median recovery exceeded 84% for all metals, with mercury showing the tightest distribution.

Table 2: Correlation between aptamer biosensor and reference methods across five water source types

Source	Pb ²⁺ (r)	Cd ²⁺ (r)	Hg ²⁺ (r)	As ³⁺ (r)
River water	0.97	0.95	0.98	0.93
Tap water	0.98	0.97	0.99	0.96
Industrial effluent	0.93	0.91	0.95	0.89
Agric. runoff	0.95	0.93	0.96	0.91
Groundwater	0.98	0.96	0.99	0.95

Pearson correlation coefficients between the aptamer biosensor and ICP-MS ranged from 0.89 (As³⁺ in industrial effluent) to 0.99 (Hg²⁺ in tap water and groundwater) (Table 2). Industrial effluent samples showed the weakest agreement, possibly because high organic matter content interfered with aptamer folding [10].

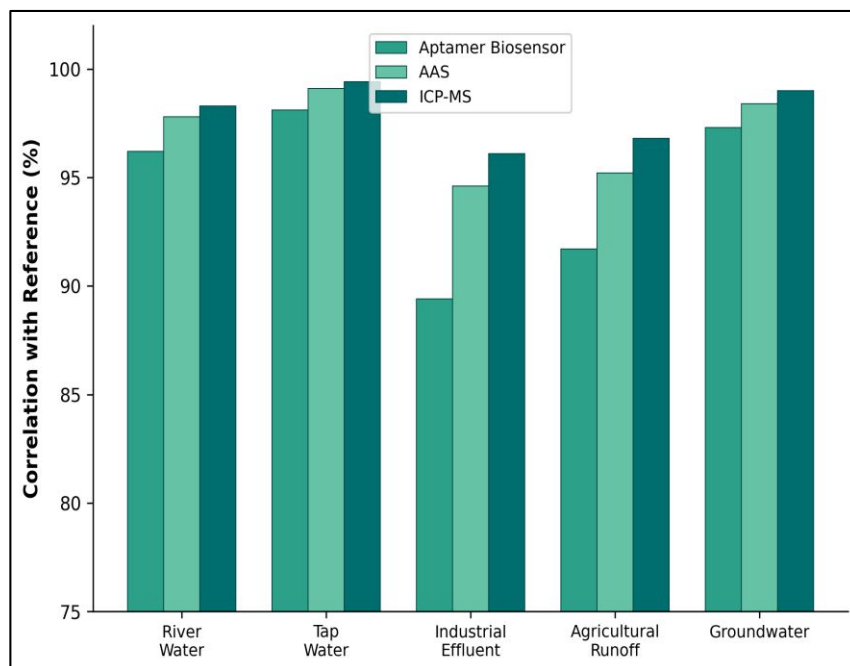


Fig 2: Grouped bar chart comparing correlation percentages between the aptamer biosensor, AAS, and ICP-MS across five environmental water source types.

Discussion

The sub-ppb detection limits achieved here are comparable to or better than other aptamer-based heavy metal sensors reported in the literature. Kim *et al.* [11] reported a Pb²⁺ aptasensor with a LOD of 0.32 ppb using a graphene-modified electrode, while the present platform reached 0.12 ppb on a simpler gold surface—an advantage for manufacturing cost. The multi-analyte capability is a practical asset for field screening, where testing for a single metal would miss co-contamination scenarios common near industrial zones.

The weaker performance in industrial effluent samples points to a known challenge: complex matrices containing surfactants, dyes, and dissolved organic carbon can coat electrode surfaces or compete for aptamer binding sites [12]. Pre-filtration helped but didn't eliminate the problem. Future iterations might incorporate anti-fouling surface coatings or sample dilution protocols to mitigate matrix effects.

At roughly USD 1.20 per electrode and under 8 minutes per test, the platform sits within reach of community health workers and small water utilities in sub-Saharan Africa—organizations that currently lack any on-site metal screening capability. But field durability and shelf-life under tropical humidity remain untested and will need evaluation before deployment [13].

Conclusion

This research produced a multi-analyte aptamer-based electrochemical biosensor capable of detecting five priority heavy metals at concentrations well below WHO drinking-water limits. The platform showed strong agreement with laboratory reference methods across diverse environmental water matrices and operated within 8 minutes per sample at low fabrication cost. Industrial effluent presented the greatest analytical challenge due to matrix interference. With further optimization for tropical field conditions, aptamer biosensors of this type could fill a gap in environmental monitoring infrastructure across regions where centralized laboratory access is limited.

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Contributions Not Qualifying for Authorship

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