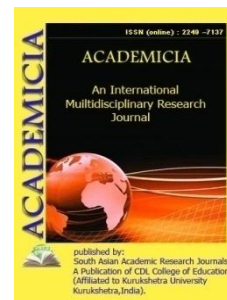


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DETECTION AND SPATIAL MAPPING OF MERCURY CONTAMINATION IN WATER SAMPLES USING A SMART-PHONE

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ABSTRACT

The majority of bulky and expensive analytical equipment are used to detect environmental pollution such as trace-level hazardous heavy metal ions. However, there is a significant worldwide need for portable, quick, specific, robust, and cost-effective detection methods that can be utilized in resource-constrained and field environments. We present a smart-phone-based hand-held platform that enables for the measurement of mercury (II) ions in water samples with a sensitivity of parts per billion (ppb). We developed an integrated upto-mechanical connection to a smart phone's built-in camera module to digitally measure mercury content utilizing a magnetic gold powder (Au NP) and aptameric based colorimetric transmission assay applied in disposable test tubes for this purpose. We quantified mercury(II) cu(ii in water samples using a two-colourradiometric method using light-emitting diodes (LEDs) at 523 and 625 nm and a custom-developed smart application to process each acquired transmission image on the same phone to actually accomplish a limit of detection of 3.5 ppb with this 40-gram smart-phone attachment.

KEYWORDS: *Detection, Light, Measurement, Smartphones, Techniques.*

1. INTRODUCTION

The development of detection techniques for real-time and long-term monitoring of mercury contamination in environmental and biological samples has been a major priority since the discovery of mercury's severe neurotoxic effects in the 1960s¹³. The organic form of mercury, methylmercury (MeHg), which is known to accumulate in the food chain⁴ and pass the blood brain barrier after human consumption, has been primarily blamed for various neurological

consequences of mercury poisoning[1]. While these discoveries have raised awareness of the dangers of organic mercury pollution, inorganic mercury, namely mercury (II) ions (Hg^{2+}), should not be overlooked. In reality, owing to bacteria-assisted biotransformation processes, mercury (II) ions constitute the main mercury pollution in the aquatic system and the “precursor” form of methylmercury. Furthermore, since it accumulates mainly in the kidney's proximal tubule cells, inorganic mercury is known to be more nephrotoxic than its organic counterpart[2].

As a result, detecting and quantifying mercury(II) ion pollution in water systems is critical, and this information may be utilized to help prevent mercury ions from entering the food chain. To meet this requirement, spectroscopic techniques such as atomic absorption spectroscopy (AAS), inductively coupled plasma mass spectrometry (ICP-MS), and atomic fluorescence spectrometry (AFS). These methods, on the other hand, require time-consuming sample preparation, costly and cumbersome equipment, and professionally trained people to conduct the tests. As a result, they are not well adapted for on-site mercury detection, and they may not even be accessible in poor nations. Recent developments in microfabrication and nanoscience, on the other hand, have allowed the creation of portable detection assays that are integrated with lab-on-a-chip systems, showing tremendous promise for application in resource-constrained settings. Figure 1 shows the Design of the ratio metric optical reader on a smart-phone[3].

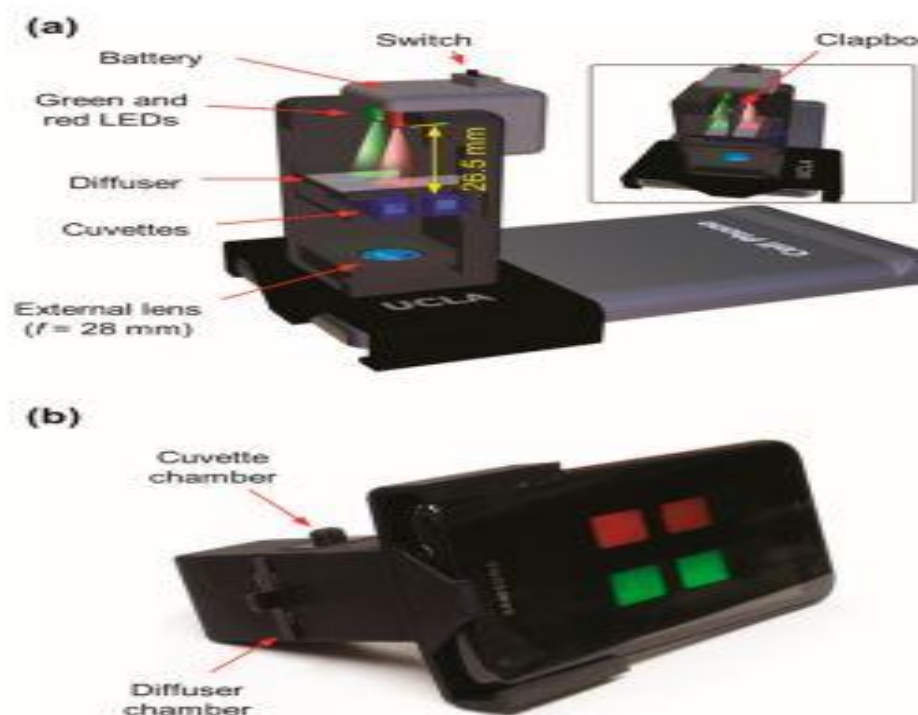


Figure 1: The above figure shows the Design of the ratio metric optical reader on a smart-phone.

However, owing to their relatively large equipment, higher prices, and lack of wireless connection, which is critical for distributed sensing and spatiotemporal mapping of

contamination in distant places and field settings, these current systems that use NPs are still restricted. The detection of subppm quantities of mercury (II) ions has recently been shown utilizing dye-embedded polymer sheets as colorimetric substrates that are digitized using, for example, smart-phone cameras as an alternative to Au NP-based plasmonic methods. We created a mercury contamination map using our smart-phone-based detection technology by assessing water samples from municipal tap water sources, rivers, lakes, and beaches in California (USA). This sensitive and accurate heavy metal detection platform running on cellphones could be quite useful for shared sensing, tracking, and sharing of increased pollution personal data as a function of both space and time, thanks to its cost-effective design, size, and wireless data connectivity[4]–[6].

However, because of inevitable changes in ambient light conditions and human operation and/or alignment during the picture capture process, this current method does not take use of the phone's processing/computational capacity, and thus has limited detection sensitivity and repeatability. To provide a field-portable, cost-effective, and wirelessly connected platform for sensitively quantifying heavy metal ion concentration in water samples, we present a battery-powered mobile sensing device that consists of a lightweight (37 g) upto-mechanical attachment to a smartphone, as well as a custom-developed Android application for quantification, reporting, and sharing detection results. This lab-on-a-phone gadget uses dual-wavelength illumination using light-emitting diodes (LEDs) at 523 and 625 nm to measure mercury-induced modest transmission variations in a colorimetric assay using citrate-stabilized plasmonic Au NPs and aptamers (Apt) combined in disposable test tubes. We demonstrated sensitive detection of mercury contamination in water samples with a limit of detection (LOD) of 3.5 ppb, which is the same order of magnitude as the maximum contaminant level (MCL).

Mercury (II) recommended for drinking water, i.e., 2 and 6 ppb, as established by the shift in the plasmonic resonance wavelength of dispersed and aggregated Au NPs in response to mercury (II) ions. We also showed geographic mapping of mercury(II) pollution in California using our cellphones-based colorimetric detection technology by analyzing water samples taken at more than 50 sites, including tap water sources as well as natural sources such as rivers, lakes, and beaches. This heavy metal detection system for smartphones may be a useful supplement to existing mobile-phone-based imaging, sensing, and diagnostics devices²⁵⁴², and it has a lot of promise for distributed sensing, spatiotemporal mapping, and mercury pollution monitoring across the world. Indeed, by the end of 2013, global cellphones subscribers had surpassed 7 billion, and smartphone penetration is on the rise, with estimates of more than 60 percent, 45 percent, and 25 percent in North America, Europe, and Africa, respectively, by the end of 2015. As a result, using mobile phones for bioanalytical measurement science, as well as reporting and sharing results, provides widely scalable, cost-effective, and yet rather powerful/competitive solutions to implement various tests and measurements even in resource-constrained and field settings, which is a major motivation for this research[7]–[9].

1.1 The Smart-Phone-Based Mercury Reader's Optical Design:

Using a colorimetric nanoparticle and aptamer assay, we developed an optical imaging interface that is mechanically connected to the current camera module of a smartphone to measure

mercury content. This attachment includes two button cells (3 V) that are utilized to power two LEDs.

These LEDs are spaced far enough apart (26.5 mm) from the rectangular test tubes (which hold the sample and control solutions) and are diffused by optical diffusers to guarantee uniform lighting of both tubes (Figure 1a). These LEDs' emission wavelengths were chosen to be 523 and 625 nm, respectively, to match the change in the extinction wavelengths of dispersed and aggregated Au NPs. The green LED lit the bottom half of each cuvette while the red LED illuminated the top half in order to gather multispectral information from a particular water sample (Figure 1a). To prevent crosstalk between the green and red lights, the optical pathways of the two LEDs were separated by an opaque clapboard before reaching the cuvettes (Figure 1a, inset) and went via two rectangular apertures (6.6 5 mm, one for each hue) in front of each cuvette. The transmitted light from the sample and control cuvettes was then gathered via two additional rectangular apertures of the same size and photographed using a plano-convex lens ($f = 28$ mm) on the smart-digital phone's camera. The demagnification factor of this external lens was selected to be 7 so that the two 6.6-mm wide sample cuvettes could be photographed concurrently inside the active region of the smart-phone CMOS imager chip[10].

Plasmonic Colorimetric Assay and Mercury-(II) Ion Concentration Measurement Spherical Au NPs have previously been investigated as new mercury (II) ion sensing probes. 1921 The Au NP-based colorimetric detection test is based on the distinctive color shift of Au NPs from red to purple or blue following aggregation, which is caused by mercury (II) ion binding events. Most Au NP-based probes, on the other hand, require a surface modification step to bind mercury (II)-specific ligands to Au NPs, and the LOD varies depending on the capturing ligand used. We use a different strategy here, relying on the thymine-rich aptamer sequence's strong affinity for mercury (II) ions and citrate-stabilized Au NPs as colorimetric signal transducers to achieve high detection sensitivity. The use of Au NPs in this technique eliminates the requirement for surface functionalization stages, making it much easier to employ in the field. The probe solution is made up of 0.64 nM Au NPs (50 nm diameter) and 3 mM aptamer in 20 mM Tris-HCl buffer (pH 8.0) in a conventional mercury detection experiment. The probe solution is then mixed with 4 liters of water sample solution and incubated for 510 minutes (see Methods section for details). Even in a high-salt environment like 10 mM NaCl, aptamer creates a protective coating on the surface of Au NPs, preventing them from aggregating. The presence of mercury(II) ions, on the other hand, will strip away this aptamer layer, resulting in the creation of more stable T-Hg₂-T complexes. 50,51 As a consequence, in the presence of NaCl, unprotected Au NPs may undergo an unique color change from red to blue (Figure 2a), which can be detected using our dual-wavelength smart-phonebased colorimetric reader to measure mercury concentration.

1.2 A smart tool for mercury testing based on Android.

We developed a bespoke Android app that enables mobile assessment and sharing of mercury quantification findings. The user may hold the smartphone horizontally after connecting the colorimetric mercury measuring equipment to the smart-phone camera unit and then conduct mercury tests using this smart application. The user may start a new test, build a device-specific calibration curve, examine previously performed tests, exchange test results, and study the operating instructions from the application's main menu. By photographing, for example,

chemical control samples at established quantities, the user may calibrate the application for attachment-specific changes. Various devices/attachments may save and reuse these calibration curves. After recording a colorimetric transmission picture of the sample, the user may see it on the screen before digitally analyzing/processing. For processing/testing, the app may alternatively utilize an image file already saved on the phone's memory. Following the image-processing procedures described in the preceding section, the transmission signal ratios between the sample and control areas will be automatically computed on the phone after clicking the "Process" button. The computed signal ratio is converted into the mercury concentration level of the sample (in ppb) using a previously saved calibration curve, and the findings are then shown on the phone's screen. On the Android phone (Samsung Galaxy S II), it took 7 seconds to calculate the mercury concentration. The final test results may be stored on the phone's memory with a time stamp and the test's GPS coordinates, and they can also be shared with a secure server for spatiotemporal mapping using a Google Maps-based interface, for example. The findings may also be seen as a function of time per place using the same Android application's graph-based interface.

Calibration and Specificity Tests are two types of tests. Each normalized G/R ratio calculated from a recorded RGB picture correlates to a particular mercury concentration measurement in our cellphone-based mercury detection technology (ppb). The default calibration curve in the Android app was created by measuring the normalized G/R ratios of a series of known concentration mercury(II) solutions with concentrations ranging from 0 to 5 M. (see Figure 4). As the concentration of mercury(II) ions climbed beyond 10 nM, the values of these normalized G/R ratios increased, reaching saturation at >1000 nM. (Figure 4). The aggregation of Au NPs, which is induced by the mercury(II) ion concentration, is mostly responsible for the signal rise in the 101000 nM range. This Au NP aggregation process increases the extinction at red wavelengths (e.g., 625 nm) while decreasing the extinction at green wavelengths (e.g., 523 nm), as shown by our UV is spectroscopic measurements. This Plasmon-resonance-based phase shift occurred quickly after approximately 5 minutes (Supporting Information, Figure S2), showing that the NP/aptamer-based colorimetric test has a fast reaction time, making it suitable for application in field settings. The transmission signal of the red channel dropped because of these plasmonic changes caused by NP aggregation, while the transmission signal of the green channel rose.

2. DISCUSSION

The author has discussed about the detection and spatial mapping of mercury contamination in water samples using a smart-phone. However, there is a considerable global need for portable, fast, specific, robust, and cost-effective techniques that can be used in resource-limited and field settings. We demonstrate a smart-phone-based hand-held platform for measuring mercury (II) ions in wastewater sample with an accuracy of parts per billion (ppb). Using a magnetic gold powder (Au NP) and aptameric-based colorimetric transmission assay used in disposable test tubes, we created an integrated upto-mechanical link to a smart phone's built-in camera module to digitally detect mercury level. As an alternative to Au NP-based plasmonic techniques, the detection of subppm amounts of mercury (II) ions has recently been shown using dye-embedded polymer sheets as spectrofluorometric boards that are digitized using, for illustration, smart-phone cameras. We assessed water samples from potable water sources, rivers, lakes, and

beaches in California to produce a mercury toxicity map using our smart-phone-based detection technique (USA). Because of its cost-effective design, location, and wireless data connection, this sensitive and precise heavy metal detection platform operating on smartphones may be very helpful for shared sensing, monitoring, and sharing of increasing pollution private information as a function of both place and time.

3. CONCLUSION

In conclusion, the author presented a sensitive and cost-effective smart-phone-based mercury(II) ion sensor platform that employs a battery-powered opto-mechanical reader connected to a smart-existing phone's camera module to digitally quantify mercury concentration using a plasmonic Au NP and aptamer-based colorimetric assay. On the same phone, we used a two-colourratio metric detection technique with LEDs at 523 and 625 nm, as well as a custom-developed Android application for fast digital image processing of the recorded transmission pictures. This mobile device has a mercury(II) ion LOD of 3.5 ppb, which is on the same order of magnitude as the maximum allowed amount of mercury(II) ions in drinking water specified by the US EPA (2 ppb) and WHO (2 ppb) (6 ppb). Furthermore, we created a geospatial mercury(II) pollution map by analyzing over 50 samples obtained in California from a variety of sources, including tap, river, lake, and ocean water samples. The sensitive heavy metal detecting gadget incorporated into cellphones cost-effective design, mobility, and internet connection may be very helpful for widespread sensing, tracking, and sharing of water pollution information as a function of both location and time, according to a new study.

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