

# Development of high sensitivity portable test kit for multiple detection of metal ions with minimal amount analysis towards environmental awareness

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## **Abstract**

Heavy metal ions contamination in the environment is a crucial issue to be concerned about. However, the methods for metal analysis require high-priced instruments and are usually limited to laboratories. Therefore, we aim to develop a paper-based sensor device portable kit which is sensitive and selective to metal ions and can be used on the field for rapid and effective detection of heavy metals in water resources. Herein, the fluorescence properties of cadmium telluride quantum dots (CdTe QDs) in the presence of L-cysteine were employed to study the quenching results of copper and mercury ions. A mixture of CdTe QDs, Britton-Robinson buffer and L-cysteine hydrochloride was coated on the device. A calibration curve of  $\text{Hg}^{2+}$  and  $\text{Cu}^{2+}$  detection was generated. By using sodium diethyldithiocarbamate and ammonium hydroxide,  $\text{Hg}^{2+}$  and  $\text{Cu}^{2+}$  can be exclusively detected in the existence of one another. The device portable kit is designed to be used in the on-field detection. After applying the sample, the result is visible under the UV-blacklight and the color intensity can be interpreted in Adobe Photoshop. Therefore, the results of each ion can be precisely analyzed. Our method also uses low-cost materials and is suitable for heavy metals analysis in the environment.

## **Keywords**

CdTe quantum dots, Paper-based sensor device portable kit, Fluorescence quenching, Color intensity, Copper ions and mercury ions

## **1 Introduction**

In the present, industrial sectors are expanding widely in order to meet the exponentially growing demand of people. These industry sectors release a huge amount of heavy metal-contaminated waste water which could easily pollute the environment, especially water sources. However, heavy metals are not naturally degradable, causing hazards to living things either directly or indirectly.

Copper is one of the most used metals in the world. Research found that up to 24.4 million metric tons of copper was used in 2019 and the usage trend is also

steadily increasing [1]. Because of its properties, copper is being used in many industrial sectors such as electricity, machinery, and vehicles. As a result, copper contamination in the environment, especially water sources, is hard to control. Consuming more than 1 milligram of copper per 1 kilogram of food could lead to stomach pain, diarrhea, headaches, and in severe cases, kidney failure.

Mercury is used in many products. For example, it is used in dental Amalgam, house paint, batteries, scientific materials, and pesticides [2]. Although mercury has a variety of usages, its contamination in water and food could cause severe effects to humans. Mercury may have toxic effects on the nervous, digestive and immune systems, and on the lungs, kidneys, skin and eyes. It is even considered by the World Health Organization as one of the top ten chemicals or groups of chemicals of major public health concern [3].

There are many ways to analyze heavy metals contamination. Examples include Inductively Coupled Plasma Mass Spectrometry (ICP-MS), Inductively Coupled Plasma - Optical Emission Spectrometry (ICP-OES) and Atomic Absorption Spectroscopy (AAS). However, these methods require high-priced equipment and are usually limited to laboratories. Therefore, analyzing heavy metals in water sources is overly complicated.

A possible alternative to these methods is to use quantum dots. These are “man-made nanoscale crystals that can transport electrons. When UV light hits these semiconducting nanoparticles, they can emit light of various colors. These artificial semiconductor nanoparticles have found applications in composites, solar cells and fluorescent biological labels” [4]. Moreover, as quantum dots can be quenched by heavy metals, they can be used as a sensor for analysis of heavy metals [5,6].

Because of their low cost of production, convenient usage and rapid analysis, paper-based device sensors are an alternative way of analyzing chemical contaminations in the environment. They could also be designed to be microfluidic devices that can detect multiple substances at a time [7, 8, 9].

## 2 Content

### 2.1 The purpose of the investigation

In order to rapidly and effectively analyze heavy metals contamination in the environment by using a minimal amount of reagent, we chose to develop quantum dots to paper-based sensor devices with portable kits to examine heavy metals contamination in the field while keeping the costs low. The heavy metals that we have chosen to be multi-detected by cadmium telluride quantum dots are copper and mercury.

### 2.2 Method of the investigation

#### 2.2.1 Paper-based sensor device preparation

The paper-based sensor device was designed using Adobe Photoshop. The device was divided into two areas which are hydrophilic and hydrophobic area.

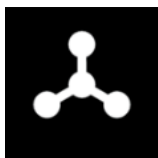


Figure 1: Image showing mimetic design of paper-based sensor devices

Wax-Printing using Fuji Xerox ColorQube 8570 and heat the wax-printed paper at 100 °C for 2 minutes 30 seconds.

#### 2.2.2 Paper-based sensor device portable kit preparation

Measure the size of the paper to be assembled into different sides of the box. Determine the position to place the paper-based sensor device on the box and measure it to be in the center. Drill a hole on the top of the box directly to the position of the paper-based sensor device for the image processing method. Assemble all the parts and put a UV-blacklight inside the box on the top edge.



Figure 2: Images showing the mimetic design of the portable kit (left) and the portable kit

#### 2.2.3 Experimental methods

##### Determining the most suitable ratio of CdTe QDs : B-R buffer : L-cysteine hydrochloride

Mix 0.5 mg/ml CdTe QDs, 0.5 M B-R buffer and 10 mM L-cysteine hydrochloride at 1:3:5 1:2:2 and 1:1:1. Wait 10 minutes, then drop the solution on paper-based sensor device 1  $\mu$ L on each area. After 30 minutes, observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

##### Determining the most suitable concentration of L-cysteine hydrochloride

Mix 0.5 mg/ml CdTe QDs and 0.5 M B-R buffer with 0.5, 1, 2, 2.5, and 3 mM L-cysteine hydrochloride. Wait 10 minutes, then drop the solution on paper-based sensor device 1  $\mu$ L on each area. After 30 minutes, drop 10 ppm  $\text{Cu}^{2+}$  solutions,  $\text{Hg}^{2+}$  solutions and distilled water. Then wait for 15 minutes and observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

##### Determining the most suitable concentration of Sodium diethyldithiocarbamate (DDTC)

Prepare 15, 25 and 35  $\mu$ M DDTC and drop the solution on one of the reagent-coated detection zones. After 30 minutes, drop 10 ppm  $\text{Cu}^{2+}$  solutions and  $\text{Hg}^{2+}$  solutions to determine the selectivity of the solutions for  $\text{Cu}^{2+}$  and  $\text{Hg}^{2+}$ .

##### Determining the most suitable concentration of Ammonium hydroxide ( $\text{NH}_4\text{OH}$ )

Prepare 0.5, 1.0 and 1.5  $\mu$ M  $\text{NH}_4\text{OH}$  and drop the solution on one of the reagent-coated detection zones. After 30 minutes, drop 10 ppm  $\text{Cu}^{2+}$  solutions and  $\text{Hg}^{2+}$  solutions to determine the selectivity of the solutions for  $\text{Cu}^{2+}$  and  $\text{Hg}^{2+}$ .

##### Determining the masking agent ability of DDTC for $\text{Cu}^{2+}$ (tolerance)

Prepare 0, 1, 5, and 10 ppm  $\text{Cu}^{2+}$  solutions to drop on the reagent-coated paper-based device. Then coat 25  $\mu$ M DDTC on the paper. After 15 minutes, observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

##### Determining the masking agent ability of $\text{NH}_4\text{OH}$ for $\text{Hg}^{2+}$ (tolerance)

Prepare 0, 1, 5, and 10 ppm  $\text{Hg}^{2+}$  solutions to drop on the reagent-coated paper-based device. Then coat 0.5  $\mu$ M  $\text{NH}_4\text{OH}$  on the paper. After 15 minutes, observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

##### Determining the selectivity of Cys-CdTe QDs for other heavy metal ions

Prepare 10 ppm  $\text{Fe}^{3+}$ ,  $\text{Cd}^{2+}$ ,  $\text{Pb}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Ni}^{2+}$ ,  $\text{Ca}^{2+}$  and  $\text{Mn}^{2+}$  to drop on the paper-based sensor device which was coated by CdTe QDs, B-R buffer, L-cysteine hydrochloride. After 15 minutes, observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

### Generating the calibration curve of Hg<sup>2+</sup>

The calibration curve was generated under the suitable condition using Hg<sup>2+</sup> solutions at the concentrations of 0, 0.1, 0.5, 1, 2, 4 and 8 ppm on the paper-based sensor device. Observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

### Generating the calibration curve of Cu<sup>2+</sup>

The calibration curve was generated under the suitable condition using Cu<sup>2+</sup> solutions at the concentrations of 0, 0.1, 0.5, 1, 2, 4 and 8 ppm on the paper-based sensor device. Observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6.

### Determining the efficiency of the device

Generate Hg<sup>2+</sup> calibration curve and Cu<sup>2+</sup> calibration curve by applying Cu<sup>2+</sup> solutions and Hg<sup>2+</sup> solutions at the concentrations of 0, 3, 6 and 9 ppm on the paper-based sensor device to obtain the conditions which match the environment. Prepare Cu<sup>2+</sup> solutions and Hg<sup>2+</sup> solutions at the concentrations of 3 and 6 ppm in the synthetic freshwater and test with reagent-coated paper-based sensor devices. After 15 minutes, observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop CS6. Then replace the color intensity in the calibration curve equation and calculate recovery percentages.

### Generating the calibration curve of the portable kit

Generate Hg<sup>2+</sup> calibration curve and Cu<sup>2+</sup> calibration curve by applying Cu<sup>2+</sup> solutions and Hg<sup>2+</sup> solutions at the concentrations of 0, 4, 8 and 10 ppm on the paper-based sensor device in the portable kit to obtain the conditions which match the environment. Then observe the fluorescence under UV-blacklight and analyze the color intensity in Adobe Photoshop.

## 2.3 Results of the experiment

### Determining the most suitable ratio of CdTe QDs : B-R buffer : L-cysteine hydrochloride

The results from color intensity interpretation using Adobe Photoshop are as follows

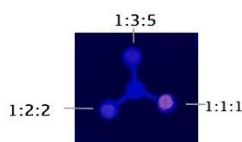


Figure 3: Image showing the results from the process to find the most suitable ratio of CdTe QDs: B-R buffer : L-cysteine hydrochloride

Table 1: Table showing color intensity of used reagent at 1:3:5, 1:2:2 and 1:1:1 ratio

ratio (QDs : L-cys : BR)	color intensity
1:3:5	71.86
1:2:2	77.92
1:1:1	101.69

We can conclude that the ratio of CdTe QDs : B-R buffer : L-cysteine hydrochloride at 1:1:1 results in the most suitable fluorescence that can be seen with bare eyes under UV blacklight and the result after color intensity interpretation using Adobe Photoshop CS6 also shows that the ratio of 1:1:1 provides the most suitable results. Therefore, we decided to use a 1:1:1 ratio in further testing.

### Determining the most suitable concentration of L-cysteine hydrochloride

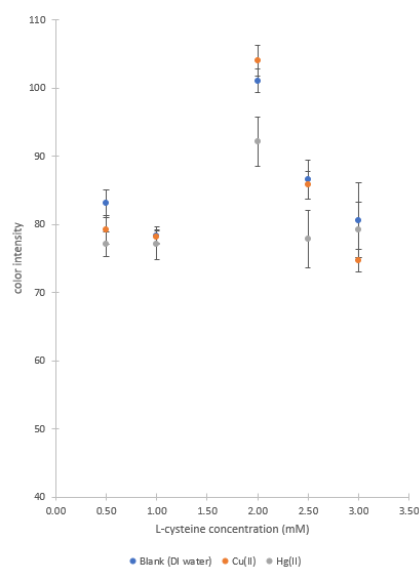


Figure 4: Graph showing the results from the process to find the most suitable concentration of L-cysteine hydrochloride

We can conclude from color intensity interpretation at each concentration that 0.5 mM L-cysteine hydrochloride results in the biggest difference of Cu<sup>2+</sup> and Hg<sup>2+</sup> color intensity compared to controlled group, meaning that CdTe QDs is quenched by Cu<sup>2+</sup> and Hg<sup>2+</sup> ions. Therefore, we decided to use L-cysteine hydrochloride at 0.5 mM concentration.

### Determining the most suitable concentration of Sodium diethyldithiocarbamate (DDTC)

Table 2: Table showing the results of the process to find the most suitable concentration of DDTC

ddtc concentration (μL)	15.00		25.00		35.00	
	no ddtc	ddtc	no ddtc	ddtc	no ddtc	ddtc
blank (DI water)	72.29	74.48	83.15	80.79	83.82	82.05
Cu(II)	70.77	69.38	77.26	77.87	79.02	78.96
Hg(II)	74.02	73.58	80.53	78.48	77.82	77.12
Cu(II)+Hg(II)	70.53	70.07	76.42	78.76	78.64	76.54

We can conclude from color intensity interpretation at each concentration of DDTC that at 25 μM, the color intensity at the DDTC coated area is higher than the color intensity of controlled area when Cu<sup>2+</sup> and Cu<sup>2+</sup> mixed with Hg<sup>2+</sup> solution is applied, meaning that the DDTC at 0.5 μM is the most suitable masking agent for Cu<sup>2+</sup>.

### Determining the most suitable concentration of Ammonium hydroxide (NH<sub>4</sub>OH)

Table 3: Table showing the results from the process to find the most suitable concentration of NH<sub>4</sub>OH

NH <sub>4</sub> OH concentration (μM)	0.50		1.00		1.50	
	no NH <sub>4</sub> OH	NH <sub>4</sub> OH	no NH <sub>4</sub> OH	NH <sub>4</sub> OH	no NH <sub>4</sub> OH	NH <sub>4</sub> OH
blank (DI water)	68.92	68.65	69.15	69.58	74.57	74.65
Cu(II)	73.72	76.15	73.84	75.24	74.80	77.98
Hg(II)	71.66	72.98	72.44	69.69	71.55	72.40
Cu(II)+Hg(II)	70.52	68.56	71.32	71.89	72.44	72.04

We can conclude from color intensity interpretation at each concentration of NH<sub>4</sub>OH that at 0.5 μM, the color intensity at the NH<sub>4</sub>OH coated area is higher than the color intensity in the controlled area when Hg<sup>2+</sup> and Cu<sup>2+</sup> mixed with Hg<sup>2+</sup> solution is applied, meaning that NH<sub>4</sub>OH at 0.5 μM concentration is the most suitable masking agent for Hg<sup>2+</sup>.

### Determining the masking agent ability of DDTC for Cu<sup>2+</sup> (tolerance)

Table 4: Table showing the results from the process of finding DDTC tolerance

Cu(II) concentration (ppm)	no ddtc	with ddtc
0	84.80	83.06
1	84.90	86.68
5	77.02	83.49
10	78.84	77.70

We can conclude from color intensity interpretation at each concentration of Cu<sup>2+</sup> solution that at 0, 1 and 5 ppm, the color intensity at the DDTC coated area is higher than the color intensity of the controlled area and at 10 ppm the color intensity at the controlled area is only slightly higher, meaning that the DDTC at 25 μM is the most suitable masking agent for Cu<sup>2+</sup> at 5 ppm concentration and lower.

### Determining the masking agent ability of NH<sub>4</sub>OH for Hg<sup>2+</sup> (tolerance)

Table 5: Table showing the results from the process to find NH<sub>4</sub>OH tolerance

Hg(II) concentration (ppm)	no NH <sub>4</sub> OH	with NH <sub>4</sub> OH
0	70.83	72.02
1	75.51	76.78
5	76.30	76.61
10	72.39	71.21

We can conclude from color intensity interpretation at each concentration of Hg<sup>2+</sup> solution that at 0, 1 and 5 ppm, the color intensity at the NH<sub>4</sub>OH coated area is higher than the controlled area and at 10 ppm the color intensity at the controlled area is only slightly higher, meaning that the NH<sub>4</sub>OH at 0.5 μM is the most suitable masking agent for Hg<sup>2+</sup> at 5 ppm concentration and lower.

### Determining the selectivity of Cys-CdTe QDs for other heavy metal ions

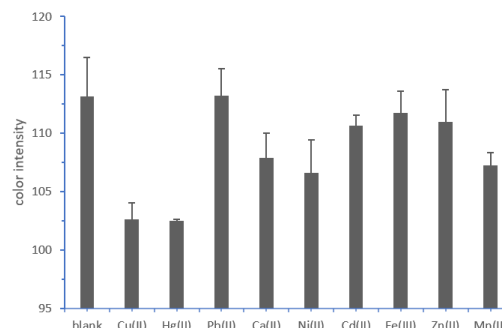


Figure 5: Graph showing the results from the process of finding the selectivity of Cys-CdTe with heavy metals

The results from color intensity interpretation when tested with various metal ions shows that the color intensity sorted in decreasing order are as follows; Pb<sup>2+</sup>, Fe<sup>3+</sup>, Zn<sup>2+</sup>, Cd<sup>2+</sup>, Ca<sup>2+</sup>, Mn<sup>2+</sup>, Ni<sup>2+</sup>, Cu<sup>2+</sup> and Hg<sup>2+</sup>. Therefore, we can conclude that Cys-CdTe is most specific with Hg<sup>2+</sup> and Cu<sup>2+</sup>.

### Generating the calibration curve of Hg<sup>2+</sup>

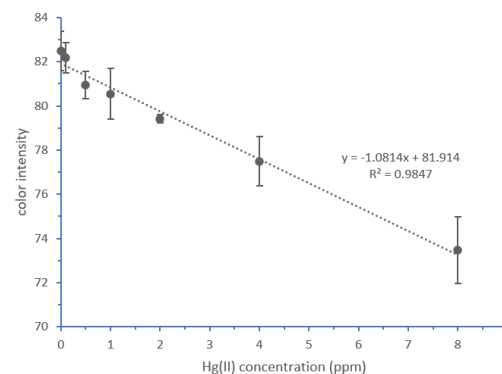


Figure 6: Graph showing the results from Hg<sup>2+</sup> calibration curve generation

We can conclude from color intensity interpretation when tested with  $\text{Hg}^{2+}$  at different concentrations that our reagent can detect the presence of  $\text{Hg}^{2+}$  at 0.1 - 8 ppm concentration.

### Generating the calibration curve of $\text{Cu}^{2+}$

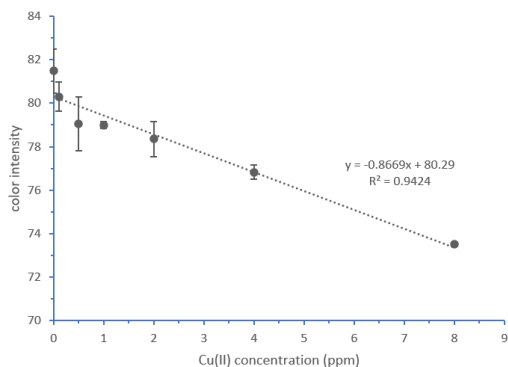


Figure 7: Graph showing the results from  $\text{Cu}^{2+}$  calibration curve generation

We can conclude from color intensity interpretation when tested with  $\text{Cu}^{2+}$  at different concentrations that our reagent can detect the presence of  $\text{Cu}^{2+}$  at 0.1 - 8 ppm concentration.

### Determining the efficiency of the device

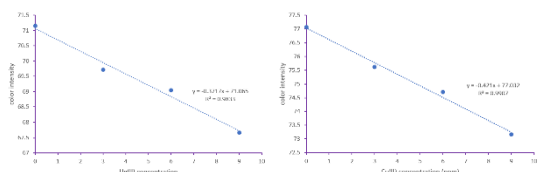


Figure 8: Graph showing the simple calibration curve of  $\text{Hg}^{2+}$  (left) and  $\text{Cu}^{2+}$  (right) for efficiency testing

Table 6: Table showing the results of the efficiency test with  $\text{Hg}^{2+}$

Hg(II) concentration (ppm)	n1	n2	n3	average (y)	stdev	concentration (x)	recovery %
3	69.78	70.09	69.85	69.91	0.13	3.12	103.88
6	68.63	68.28	69.33	68.75	0.44	6.24	103.95

We can conclude that when the device is tested for efficiency with  $\text{Hg}^{2+}$  solution at 3 and 6 ppm concentration and by substituting the color intensity into the linear equation of the calibration curve resulted in 3.12 and 6.24 concentration with the recovery percentage of 103.88 and 103.95. Therefore, we can conclude that the device has high efficiency and can be used to test for  $\text{Hg}^{2+}$  in a real environment.

Table 7: Table showing the results of the efficiency test with  $\text{Cu}^{2+}$

Cu(II) concentration (ppm)	n1	n2	n3	average (y)	stdev	concentration (x)	recovery %
3	77.51	75.36	74.65	75.84	1.22	2.83	94.38
6	74.32	74.89	73.79	74.33	0.45	6.41	106.84

We can conclude that when the device is tested for efficiency with  $\text{Cu}^{2+}$  solution at 3 and 6 ppm and by substitution the color intensity into the linear

equation of the calibration curve resulted in 2.83 and 6.41 concentration with the recovery percentage of 94.38 and 106.84. Therefore, we can conclude that the device has high efficiency and can be used to test for  $\text{Cu}^{2+}$  in a real environment.

### Generating the calibration curve of the portable kit

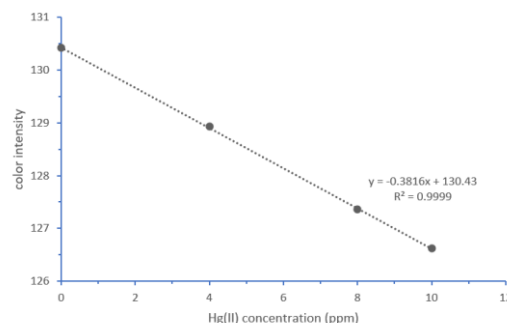


Figure 9: Graph showing the results of  $\text{Hg}^{2+}$  calibration curve generation in a portable kit

We can conclude from color intensity interpretation when tested with  $\text{Hg}^{2+}$  at different concentrations that the portable kit can be used to detect the presence of  $\text{Hg}^{2+}$  with linear tendency.

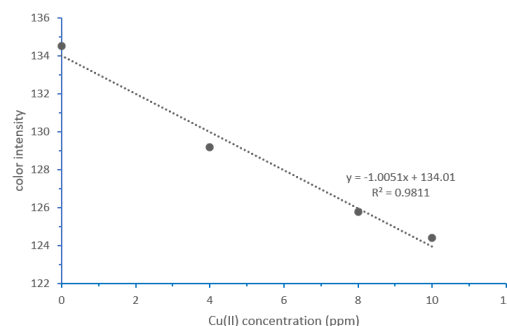


Figure 10: Graph showing the results of  $\text{Cu}^{2+}$  calibration curve generation in a portable kit

We can conclude from color intensity interpretation when tested with  $\text{Cu}^{2+}$  at different concentrations that the portable kit can be used to detect the presence of  $\text{Cu}^{2+}$  with linear tendency.

## 3 Conclusion

Cys-CdTe QDs, which is composed of 0.5 mg/mL CdTe QDs, 0.5 mM L-cysteine hydrochloride, 0.5 M Britton-Robinson Buffer, 25  $\mu\text{M}$  DDTC and 0.5  $\mu\text{M}$   $\text{NH}_4\text{OH}$ , enable us to detect the amount of  $\text{Hg}^{2+}$  and  $\text{Cu}^{2+}$  on a paper-based sensor device by using the quenching property of CdTe Qds and analyzing the color intensity through Adobe Photoshop CS6. Our reagent is suitable for the individual detection of  $\text{Hg}^{2+}$  and  $\text{Cu}^{2+}$ , and for multiple detection when the masking agents are applied.

DDTC and  $\text{NH}_4\text{OH}$  have the ability to be masking agents of  $\text{Cu}^{2+}$  and  $\text{Hg}^{2+}$  respectively. It was found

that 25  $\mu\text{M}$  DDTC can bind with  $\text{Cu}^{2+}$  ions not less than 5 ppm and 0.5  $\mu\text{M}$   $\text{NH}_4\text{OH}$  can bind with  $\text{Hg}^{2+}$  ions not less than 5 ppm.

It can be concluded from the  $\text{Hg}^{2+}$  and  $\text{Cu}^{2+}$  calibration curve that the solution of Cys-CdTe QDs, L-cysteine hydrochloride, Britton Robinson Buffer with DDTC is able to detect 0.1-8 ppm of  $\text{Hg}^{2+}$  and the same solution with  $\text{NH}_4\text{OH}$  is able to detect 0.1-8 ppm of  $\text{Cu}^{2+}$ .

According to the efficiency test, the recovery percentage is relatively high, which means that the device is not interfered with by ions in the water sources and is able to be used in the field. The result is close to the actual concentration. The simple calibration curve must be generated before each on-site use for a precise result.

The paper-based device portable kit can be improved by finding new optimal concentrations for the reagents, which would help widen the detection range especially on the lower concentration side. Moreover, the position and power of UV-blacklight in the portable kit could be modified to enable the fluorescence of CdTe QDs to be seen more clearly, which would result in higher accuracy.

The interpretation process can be improved in order to conveniently analyze metal ion contamination in the environment by developing smartphone software that can interpret color intensity and immediately convert it to concentration.

Lastly, this idea and process of analyzing copper and mercury concentrations could be adapted to detect other metal ions in the environment

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