Detection of Hexavalent Chromium Concentration in Aqueous Solution Using Lab Developed Colorimeter

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Abstract. The detection of hazardous carcinogenic heavy metals in aqueous solutions is important for biological health and environmental protection. There are various techniques to ascertain these oncogenic chemicals in an aqueous solution, like a potentiometric electrochemical sensor, spectrophotometer sensor, colorimeter sensor, and chemical sensors. As compared to other available methods, the colorimeter sensor has advantages such as low-cost instrument, qualitative sample detection, and higher precision. In this paper, the colorimeter is developed in the lab to detect hexavalent chromium [Cr (VI)] in the aqueous solution. Moreover, the absorbance principle is used to calibrate and measure the known sample concentration using amplified optical power light-emitting diodes (OP-LED). The estimation of Cr (VI) concentration in an aqueous solution is evaluated and programmed in a low-cost Arduino UNO development board. The concentration value is displayed on the touch screen. The readings displayed on the lab developed instrument annotate that the concentration percentage of the known sample can be used to determine the unknown concentration. The concentration of the known solution is determined and compared with commercially available colorimeters concerning the lab-developed colorimeter.

Keyword Cr (VI), Colorimeter, Arduino Uno, Thin Film transistor (TFT) touch screen, OP-LED

1. Introduction
Heavy metals (HMs) tend to be serious threats to environmental sustainability, which have increased in the ecological cycle due to industrialization growth. Mercury, cadmium, chromium, arsenic, and palladium tend to be highly poisonous and oncogenic even at micro-levels. These heavy metals are non-biodegradable and get accrued in the food chain [1]. Thus, it has become crucial for monitoring HMs in drinking water sources, the environment, and industrial effluents. Methods for detecting these heavy metals commonly include ultraviolet spectroscopy, atomic absorption spectroscopy inductively coupled spectroscopy (ICP-MS) [2]. These techniques are highly sensitive and accurate but are tedious, expensive,
and require handling expertise making them ineffective to be portable and easy [3]. Moreover, optical /RGB sensors are suitable as on-site models for the detection of heavy metals.

The usage of chemosensors for selective sensing Cr (VI) using Zr4+metal organic framework (MOF) with a very low detection limit was reported [4]. A recent study suggests the gallic acid capped gold nanoparticles were used to detect Cr (VI). These chemosensors have limitations like less selectivity, standard synthetic process, and high detection variation with pH [5]. Detection of Cr (VI) with photochemical sensors has been reported due to its advantages like low cost, easy operability. However, if the analysis concentration exceeds the device's maximum limit, no further detection response can be noticed, which is the greatest limitation of these sensors [6]. The real-time sensing capability can ensure adequate time to take safety measures if maximum limits of contaminant metal ions are exceeded [7]. Graphene-based optical sensors have also been reported and can meet the goal of rapid in situ measurement of metals in water [8]. A multi-cation terpyridine-based design was reported to have colorimetric and fluorometric principles for detecting Fe3+, Co2+, Hg2+, and Sn2+22 [9].

Similarly, a paper-based colorimeter sensor was designed using a color sensor and Arduino microcontroller to determine the known concentration of cyanide. Still, calculating unknown cyanide concentration was not achieved [10]. Another report sheds light on detecting cobalt Co2+ detection in Hela cells, giving visual detection of pollutants using a fluorescent probe [11]. Further advancement of the optical color sensor's reflectivity was demonstrated in one of the findings with a low-cost model with narrow and wideband measurements [12]. Detection of Cr (VI) selectively was also reported in the range of 1.1x10^-5 with an optical membrane sensor [13]. Moreover, metal cladding optical sensor with hollow-core was fabricated on the principle of extinction reaction coefficient for DPC and chromium ion [14]. However, most of these studies only explain the proof of concept for the detection of HMs in aqueous solutions [15].

In this context, the current research focuses on rapid detection of one HM, i.e., Hexavalent chromium, in an aqueous state using a lab-developed colorimeter. The need for sensors that can measure metals in water has triggered concerns about the effect of metal ions on human health and the environment [16]. For example, sensors that can make real-time measurements of metals in water at low concentrations are desirable for maintaining safe water supplies. The lab's fabrication developed a colorimeter sensor consists of the true-green light source (wavelength 530nm), quartz cuvette, and light-to-frequency photodetector [17]. The photodetector sensor senses the absorbed light and converts it into pulsed frequency, and subsequently, it is converted into transmittance (T) and absorbance (ABS), respectively [18]. The absorbed frequency detected from the analysis becomes the absorbance value, and then it can be converted into concentration using Beer-Lambert’s law.

2. Material and Methods

2.1 Glassware's and chemical
Analytical grade reagents and chemicals procured from Sigma Aldrich were used in the experimentation. The chemicals were diluted in double-distilled deionized water. A stock solution of Cr (VI) (100mg/L) was prepared by dissolving appropriate quantities of potassium dichromate (K2Cr2O7). Working standards were freshly prepared as per requirement from the working stock solution. Dye solution 1,5 - diphenyl carbazide (DPC) was prepared freshly by dissolving 0.5gm of DPC powder in ethanol and stored in dark conditions. Hydrochloric acid was used for adjusting the pH. All the glassware was washed using 10% nitric acid followed by water before each set of experiments.

2.2 Hexavalent chromium detection protocol
Working standards of chromium were prepared from stock solution in concentration 0.2 to 1.0mg/L and taken into separate volumetric flasks. 1ml hydrochloric acid (1:1) was added in each of the respective flasks. The 1ml of 0.5% of DPC were further added to each of the flasks and thoroughly mixed and kept for 5 minutes. The bright pink color appears in the mixture representation formation of hexavalent chromium. The absorbance was taken on a standard colorimeter and lab-developed colorimeter at 530nm light exposure.

2.3 Experimental reaction parameter optimization (pH and DPC concentration)
To optimize the pH for the conversion of trivalent chromium to hexavalent chromium, a better absorbance range of pH from 0.5 to 3.5 was adjusted. All the experiments were performed in triplicates for each of the pH, and the mean standard deviation was found to be negligible. All measurements were taken at 530nm light exposure. DPC dye concentration was optimized using a concentration range from 0 % to 1.5%, and the concentration showing the highest absorbance at 530nm was selected as the optimum concentration for all further experiments.

2.4 Lab developed colorimeter
As described in Figure 1, the lab-developed colorimeter consists of the enhanced thin-film optical power true-green LED (λ = 530 nm), lens optics, quartz cuvette, photodetector, Arduino UNO microcontroller module, TFT touch screen, and fiber casing is used to fabricate the prototype of colorimeter sensor. The scattered wave from the true-green light source (530nm) is passed through the lens optics, which converts the scattered light into focused light and lets it fall on the quartz cuvette. The photodetector captures the transmitted true-green light and it converts the true-green light-to-frequency, and subsequently, the frequency is converted into measurable PWM pulses. The 100% total transmittance voltage of the true-green light source is 5V. Hence, the frequency of green light changes under the absorbance, which means that the photodetector senses the total absorbance value of the analytic to be tested from the total transmitted frequency of the true-green light source.

![Figure 1: Schematics of the instrumental setup of lab-developed colorimeter sensor consists of the enhanced thin-film optical power true-green LED (λ = 530 nm), lens optics, quartz cuvette, photodetector, Arduino UNO microcontroller module, TFT touch screen and fiber casing](image)

The filter present in the detector compares the value of the true-green color of wavelength 530 nm with the green 16×2 matrices mesh at the surface of the detector. The instrument is calibrated using distilled water, and after that absorbance of the analyte to be tested was determined. The absorbance measurement is referred to as the change in total absorbed frequency concerning total transmitted
frequency passed through a cuvette containing analyte [Cr (VI)]. The quantity of absorbance is sent to the micro-controller, which saves the data, and the value is displayed on the TFT touch screen.

2.5 Experimental setup

As represented in Figure 2 experimental setup consist of nitrile gloves, micro tips, micropipette, pH strip, volumetric flask containing Cr (VI) solution in the range of 0.2mg/l, 0.4mg/l, 0.6mg/l, 0.8mg/l, and 1.0mg, hydrochloric acid (HCL) 1:1 solution, 1, 5 - diphenyl carbazide, Cr (VI) stock solution (10ppm), quartz cuvette, power supply and lab developed colorimeter.

The volumetric flasks contain Cr (VI) with a red-violet colored compound, which is achieved by a combination of chromium ions, hydrochloric acid, and DPC dye. Once stable color is formed, the solution is transferred to quartz cuvette, and absorbance is taken on a lab-developed colorimeter and standard colorimeter simultaneously. The maximum correlation is achieved after 1 minute of reaction and stability in 5 minutes. All the readings for absorbance of analyte were taken in triplicates and the mean was calculated, the mean, the standard deviation was found to be negligible.

Figure 2: Representation of experimental setup consists of nitrile gloves, micro tip, micropipette, pH strip, a volumetric flask containing Cr (VI) solution in the range of 0.2mg/l, 0.4mg/l, 0.6mg/l, 0.8mg/l, and 1.0mg, hydrochloric acid (HCL) 1:1 solution, 1, 5 - diphenyl carbazide, Cr(VI) stock solution (10 ppm), quartz cuvette, power supply, and lab-developed colorimeter.

3. Results and Discussion

3.1 Optimization of reaction parameters

For optimization of the reaction, we first examined the absorbance for the pH range, i.e., from 0.5 to 3.5. As shown in Figure 3, the absorbance has been increased from pH 0.5 and the maximum response was noted at pH 2.0, after which it has shown a sudden drop in absorbance. Thus, the reaction shows enhanced results at acidic pH. Following findings of chromium removal mechanisms and effect of pH variation, as the pH reaches near acidic, i.e., 2.0, the detection increases as compared to the higher pH. When pH was below 2.0, the shift of color from colorless to red-purple was not visualized, and eventually, the absorbance values were less. As the pH value increases beyond 1.8, there was a sudden shift of color observed, giving maximum absorbance values at 530nm. Moreover, the optimum pH of 2.2 for monitoring Cr (VI) also justifies our findings.
The concentration of detection dyes, i.e., DPC, is the second important parameter influencing the color intensity and absorbance. Different concentrations of DPC (0.5 to 1.5%) were used for detecting the highest absorbance. As per Figure 3b the highest absorbance was achieved at 0.5% concentration. Thus, 0.5% DPC was taken as an optimized concentration for all further experiments. Similarly, DPC concentration stability was reported for 7-14 days at a concentration of 0.5%, and it was found to give greater absorbance compared to other concentrations consistently. Color saturation starts at a concentration higher than 0.5%, ultimately reducing the absorbance of Cr (VI). This annotates that at 0.5% DPC the optimum absorbance value is 0.54 at 530 nm.

![Figure 3: highest absorbance was achieved at 0.5% concentration](image)

**Figure 3** responses represent five samples of the known concentration of the Cr (VI) analyte (a) the optimization of pH in the range of 0.5 - 3.5 at 530 nm. This annotates that at 2.0 pH the optimum. The absorbance value is 0.07234 at 530 nm (b) optimization of DPC concentration in the range of 0 % to 1.5 %.

### 3.2 Estimation of Concentration of analyte

The optical power green LED emits a dispersed wavelength of 530 nm towards lens optics. It targets the beam of light towards the sample compartment and the detector. The transparent quartz cuvette is a small vessel container having a square cross-section of 10 mm length. It is filled with aqueous solution and Cr (VI), which is placed at the equidistant path of the lens optics and the detector. The Arduino UNO controller module is used to program the programming logic of the sensor setup and to estimate the transmittance and absorbance of the light source. The controller converts the frequency into PWM pulses to count the number of pulses at each rising and falling edge of the transmitted wavelength.
When the sample is calibrated with transparent water in a cuvette, the transmittance of the source shows 100 %. Therefore, the intensity of true-green light at 530 nm is \( I \) to the intensity of the observed sample \( I_0 \). Initially, the controller measures the transmittance (T) and absorbance (ABS) in a sequence while calibrating water and stores the data in it to compare the measured data with calibrated data.

The 100 % transmittance and absorbance of the light can be represented as,

\[
T = 100 \times \frac{1}{I_0} \tag{1}
\]

\[
ABS = - \log_{10}(T/100) = \log_{10}(I_0/I) \tag{2}
\]

From eq. (1) and (2), the reference value of transmittance is set to 100 % (where, ABS = 0) initially while calibration is selected. Wherein, according to Beer-Lambert’s Law [1],

\[
ABS = x \times len \times Con \tag{3}
\]

Where \( x \) is the dependent constant on molar coefficient of absorptivity on wavelength of true-green light, absorbing solution of the cuvette and the medium; \( len \) is the total path length of the light from source to detector and \( Con \) is the total concentration of the solution.

From eq. (1), (2) and (3), when a dye of 1, 5 – diphenyl carbazide is added to the aqueous solution to determine the Cr (VI) content, the color of it changes from transparent white to lavender-pink. The absorbance value can be calculated more suitably by using true-green light having wavelength 530 nm.

At 530 nm, the red dye at 0.8 ppm has the known absorbance of 0.0997 in the cell of path length 1 cm. From Figure 4 (a), Absorbance readings for 5 samples of Cr (VI) solution is taken. Using Beer-Lambert’s law, the plot generated with the equation is identical to the basic slope equation, which is, \( Y = m \times X + C \). The Cr (VI) solution of known concentration is found to have an absorbance of 0.0997. It can be calculated by setting \( y = 0.0997 \) and solving for \( X \). Also, by the visual comparison to the plot shown in the Figure 4 (a), the concentration of the known absorbance becomes 1.0280 mol/L.

3.3 Comparison of lab-developed colorimeter response with standard colorimeter

The absorbance of Cr (VI) with optimized reaction parameters obtained from standard colorimeter at 530 nm and the correlation coefficient (R²-value) was reported to be 0.94. The slope response on the graph fits the best line, which passes across the origin. The Y-Intercept represents the absorbance value which is close to zero. Similarly, the value of unknown concentration of Cr (VI) in aqueous solution can be found out, which is as shown in the Figure 4 (b).

Therefore, according to the equation of slope, the absorbance of unknown solution is 0.0992. Hence, the concentration of the unknown sample can be determined. From calculation the solution of known and unknown concentration is found to have an absorbance of 0.0997 and 0.0992, respectively. Thus, the concentration of the known absorbance becomes 1.0280 mol/L and that of unknown concentration is 1.0140 mol/L, which validates the lab developed colorimeter efficacy for the Cr (VI) concentration determination.
4. Conclusion
In this paper, the fabrication of colorimeter sensor was developed for the detection of hexavalent chromium concentration in aqueous solution. The detection parameter like pH and DPC concentration were optimized. By using Beer-Lambert’s law, the developed algorithm has been implemented so as to get the estimation of the unknown concentration of the analyte. The conversion of frequency from source to photodetector and absorbed frequency to PWM pulses is achieved. The optical power green LED emits a dispersed wavelength of 530 nm towards lens optics. It targets the beam of light towards the sample compartment and the detector. The transparent quartz cuvette is a small vessel container having a square cross-section of 10 mm length. Thus, the experimental results show that the colorimeter sensor can detect the concentration of the unknown sample, which makes this sensor an excellent alternative for commercially available expensive colorimeter sensor methods for detection of heavy metal in the aqueous matrices.

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Conflicts of Interest

There are no conflicts of interest.

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