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# Green Silver Nanoparticle for Colorimetric Determination of Cyanide in Water Samples

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#### **Abstract**

Cyanide ion is highly toxic to human. Cyanide ion is mainly used in processes like electroplating and extraction of silver and gold. Therefore, it can enter the environment and pollute soil and water. In the present work, a colorimetric method based on silver nanoparticle (AgNP) was proposed for detection and determination of cyanide ion. Silver nanoparticle was prepared by carbon dots as reducing agent. Volume of AgNP and concentration of sodium hydroxide were optimized for determination of cyanide ion by AgNP. It was observed that greenly synthesized AgNP can serve as reagent in detection and determination of cyanide ion. In the presence of sodium hydroxide, a method was optimized and a robust model with linear range of 4.0-100.0  $\mu$ M, limit of detection of 3.8  $\mu$ M and limit of quantification of 12.7  $\mu$ M was obtained. In the method presented, the color change of AgNP from yellow to colorless in the presence of cyanide ion was observed. Environmental water samples including spring, well and wastewater were successfully analyzed by this method, which is simple and inexpensive.

**Keywords:** Silver Nanoparticle, Cyanide Ion, Colorimetric, Water.

## 1. Introduction

Cyanide is one of the most toxic compounds to human beings (Wishart and Dutton, 1975). It is highly hazardous to humans even at very low amounts of 0.5 mg per kilogram of body weight, which results in asphyxiation of cells and death (Holland and Kozlowski, 1986).

Food sources like potato, cassava, bamboo shoots, apple seed, bitter seed and almond contain trace amounts of cyanide. Even trace amounts of cyanide ion in food

samples can result in serious health effects (Cheng et al., 2016).

Cyanide ion is used in processes like electroplating, gold extraction and processing of polymers (Gong et al., 2013). It is estimated that half of the cyanide produced each year is used in extraction of gold and silver in the mines (Gimeno et al., 2008). Moreover, cyanide as a chemical reagent in large quantities is produced for mining, metal, drug, dye, leather and textile industries (Gimeno et al., 2008).

By forming complexes in the cell mitochondrion, cyanide ion induces the accumulation of chemicals in the bloodstream. Since cyanide ion can bind to the active site of cytochrome oxidase, it inhibits cellular respiration and exhibits extreme toxicity to mammals (Vennesland et al., 1981). Therefore, the maximum contaminant level of cyanide ion in drinking water was regulated to be 2 ppm (76 µM) by the U.S. Environmental Protection Agency<sup>2</sup> (U.S. Environmental Protection Agency, 1999).

Potentiometry, titrimetry and spectroscopy are traditional methods which have been applied to determine cyanide ion (Blaedal et al., 1971; Breuer et al., 2011; Egekeze and Oehme, 1979; Nagashima and Ozawa, 1981). Recently, introducing organic based probes and materials for selective detection of anions has been a topic of many researches (Anzenbacher et al., 2002; Bao and Zhou, 2010; Ding et al., 2014; Gale, 2008; Huang et al., 2012; Jiang et al., 2016; Kumar et al., 2018; Lin et al., 2015; Lou et al., 2009; Lv et al., 2011; Ngulube et al., 2017; Piątek and Jurczak, 2002; Suganya and Velmathi, 2015; Wang et al., 2015; You et al., 2010; Zhang and Jiang, 2012).

In these works, derivatives of organic molecules such as rhodamine, indole, carbazole and coumarin have been used. On the other hand, developing probes for fluorescence and colorimetric detection of cyanide is the main field of these researches because of their prominent properties (Chen et al., 2012; Yang et al., 2013). Naked eye detection is the main advantage of fluorescent and colorimetric probes compared with various traditional approaches (Cheng et al., 2012; Erdemir and Malkondu, 2020; Huang et al., 2012; Li et al., 2019; Princy et al., 2021; Wu et al., 2021; Zheng et al., 2014).

However, preparation and synthesis of sensing molecules is complex and expensive. In the literatures, few reports in utilizing silver nanoparticles<sup>3</sup> for colorimetric detection and determination of cyanide can be found (Dong et al., 2017; Princy et al., 2021). Therefore, in the present work, an AgNP was synthesized by green method and applied in detection and determination of cyanide ion. Procedure for preparation of AgNP was simple and silver ions have been reduced without any chemical reductant, as reducing agent, carbon dot was employed (Shariati-Rad et al., 2018). Clearly, a green reducing agent like carbon dot is superior to chemical ones. For preparation of AgNPs, chemical reducing agents like sodium borohydride (Divsar et al., 2015; Rahim et al., 2019; Yao et al., 2010), sodium citrate (Wang et al., 2017) and other chemicals (Salman et al., 2012) have been used. On the other hand, using surface plasmon property of AgNP, it is possible to detect different analytes (Garg et al., 2016; Kumar et al., 2010; Phoonsawat et al., 2018; Sasikumar and Ilanchelian, 2017; Dong et al., 2017).

# 2. Experimental

# 2.1. Instruments and software

All absorbances were recorded using an Agilent 8453 UV-Vis spectrophotometer with a diode array detector equipped with 1 cm path-length quartz cells.

Transmission electron microscopy<sup>4</sup> images were taken by a Zeiss EM900 TEM.

#### 2.2. Materials and solutions

Silver nitrate (99.0%), sodium hydroxide ( $\geq$ 98%, Merck), ethanol (99.5%), ammonia (25%), potassium cyanide ( $\geq$ 96%) and deionized water were used.

Water samples including well and spring waters were taken in different locations of Kermanshah city, Iran. Random sampling was followed for sampling of well and spring water in different times in the summer. Polyethylene terephthalate<sup>5</sup> bottles were used to place water samples and the samples were stored at room temperature till analysis (USEPA Office of Science and Technology, March 2007). A sample of electroplating was also collected and analyzed by the proposed method. Stock solution of cyanide ion with a concentration of 0.01 M was prepared in 0.001 M sodium hydroxide.

# 2.3. Preparation of AgNP

For synthesis of carbon dot as reducing agent in preparation of AgNP (Jin et al., 2015), firstly, 20 g of dried green walnut bark was ground, 20 mL of deionized water and 20 mL of ethanol were added successively and it was heated at 170 °C for 2 h. After that, it was filtered by addition of 20 mL of ethanol and 20 mL of deionized water. The yellow mixture remained after filtering contains carbon dot. Solid carbon dot was obtained by freeze drying the filtered mixture.

In preparation of AgNP, 100 mL of deionized water was boiled by heater and while boiling, 100.0  $\mu$ L of the prepared carbon dot solution (50.0 mg/mL) was added. After 15 min, 1 mL ammonia solution (10%, w/w), fresh silver nitrate solution (5 mL, 20.0 mM) were added while stirring for 2 min. The reaction was continued for 50 min at 90 °C. Finally, the yellow solution of AgNP was obtained.

# 3. Results and discussion

# 3.1. Characterization of AgNP

TEM image of the synthesized AgNP can be seen in Fig. 1. A uniform size of the spherical AgNP can be observed in Fig. 1. The mean diameter of the prepared AgNP is 10.0±1 nm. Uniformity in the size of the synthesized

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<sup>&</sup>lt;sup>1</sup> Maximum Contaminant Level (MCL)

<sup>&</sup>lt;sup>2</sup> Environmental Protection Agency (EPA)

<sup>&</sup>lt;sup>3</sup> Silver Nanoparticles (AgNPs)

<sup>&</sup>lt;sup>4</sup> Transmission Electron Microscopy (TEM)

<sup>&</sup>lt;sup>5</sup> Polyethylene Terephthalate (PET)

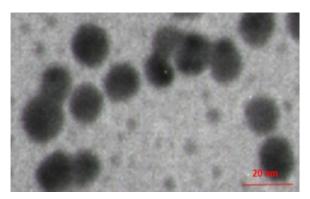


Fig. 1. TEM image of the synthesized AgNP

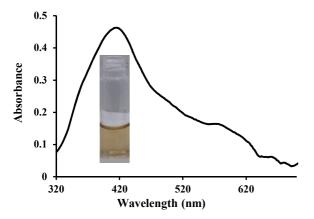


Fig. 2. UV-Vis spectrum of the synthesized AgNP. Inset shows the image of the corresponding solution

AgNP has been reflected in the low value of the standard deviation of the mean of their diameter.

In order to further study about the synthesized AgNP, UV-Vis spectrum of the nanoparticle was recorded (Fig. 2). As can be seen in Fig. 2, the synthesized AgNP has a major peak with maximum located at 415 nm. The AgNP solution appears as yellow.

#### 3.2. Optimization

Color of the synthesized AgNP solution disappears in the presence of cyanide ion (Fig. 3). At the same time, the peak absorption of AgNP located at 415 nm was decreased (see Fig. 3). Therefore, synthesized AgNP can serve as a reagent in determination of cyanide ion.

Because of the high affinity of cyanide ion to silver ion and complex formation between them, AgNP dissolves in the presence of cyanide ion. Therefore, species like AgCN and Ag(CN)<sub>2</sub> can be formed. This results in AgNP dissolving and the solutions yellow color fading.

# 3.3. Volume of AgNP

In this work, AgNP serves as the reagent. Therefore, its amount should be optimized. In this way, different amounts of AgNP as volume were explored.

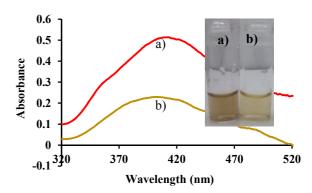


Fig. 3. a) Absorption spectrum of AgNP in the absence and b) in the presence of cyanide ion (20.0 µM). Inset shows the corresponding solutions

Optimization of nanoparticle volume was performed between volumes of 0.0 and 600.0 µL in seven pairs of test tubes. The total volume in the tubes is 2000.0 µL. For observing a clear color change, concentration of cyanide ion was selected to be 40.0 µM.

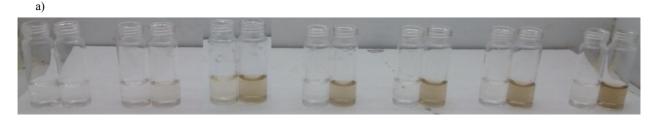
Finally, according to the color and UV-Vis spectrum, the volume of 600.0 µL was selected as optimal one (Fig. 4). It can be seen that by increasing the volume of AgNP, the color change between blank and standard solution is more distinct and absorbance changes are higher (Fig. 4). However, because of the restrictions in the total volume of the solutions and saving the AgNP, 600.0 µL of AgNP was selected as optimal one.

#### 3.3.1. Medium optimization

Since toxic hydrogen cyanide is delivered in acidic medium containing cyanide ion, it was decided to explore the system in alkaline medium. Moreover, it is desired to observe color changes which can be seen by the naked eye. Preliminary tests showed that in alkaline medium and in the presence of cyanide ion, the color of the AgNP solution disappears.

Therefore, different volumes of 0.01 M sodium hydroxide and a fixed 600.0 µL volume of the synthesized AgNP were examined in the presence of a known concentration of cyanide (40.0 μM). The results have been shown in Fig. 5. In all experiments, a clear color change between sample and blank solutions can be observed (Fig. 5a).

Absorbance of these solutions was recorded and the difference between absorbance of each pair was calculated at 415 nm. Variation of the absorbance changes with volume of sodium hydroxide can be seen in Fig. 5b. It can be seen that by increasing the volume of sodium hydroxide up to 100.0 µL, absorbance changes increase and after that a gradual decrease in absorbance changes occurs (Fig. 5b). Based on the plot, volume of sodium hydroxide for analyzes of cyanide was selected to be 100.0 µL.



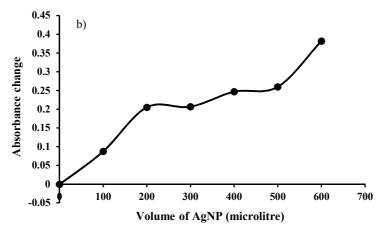


Fig. 4. Optimization of the volume of AgNP. a) Image of the solutions with volumes of 0.0, 100.0, 200.0, 300.0, 400.0, 500.0 and 600.0  $\mu$ L of AgNP and b) absorbance changes at 415 nm between blank and standard solution. In each pair of sample tubes, the left ones contain standard solution (AgNP+Sodium hydroxide+ cyanide ion) and the right ones are blank solutions (AgNP+Sodium hydroxide). Conditions: sodium hydroxide = 0.1 M, cyanide ion = 40.0  $\mu$ M

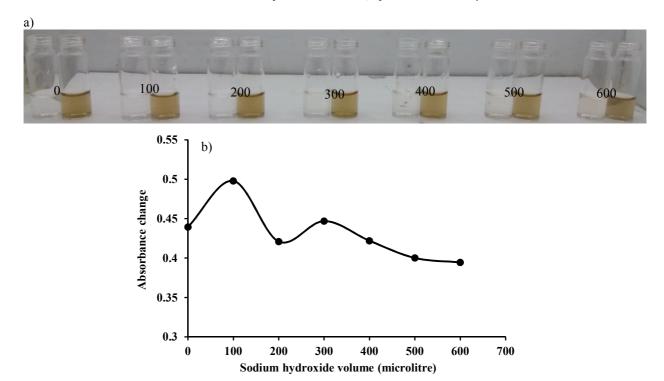


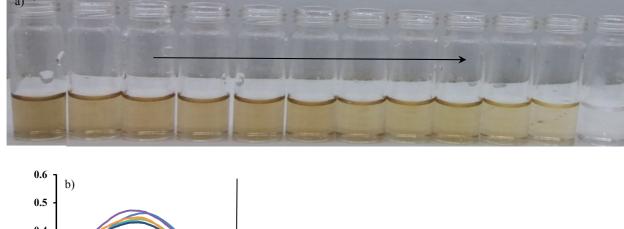
Fig. 5. Optimization of the volume of sodium hydroxide (0.1 M). a) Image of the solutions with volumes of 0.0, 100.0, 200.0, 300.0, 400.0, 500.0 and 600.0  $\mu$ L of sodium hydroxide and b) absorbance changes at 415 nm between blank and sample solutions. In each pair of sample tubes, the left ones contain sample solution (AgNP+Sodium hydroxide+ cyanide ion) and the right ones are blank solutions (AgNP+Sodium hydroxide). Conditions: AgNP = 600.0  $\mu$ L, cyanide ion = 40.0  $\mu$ M

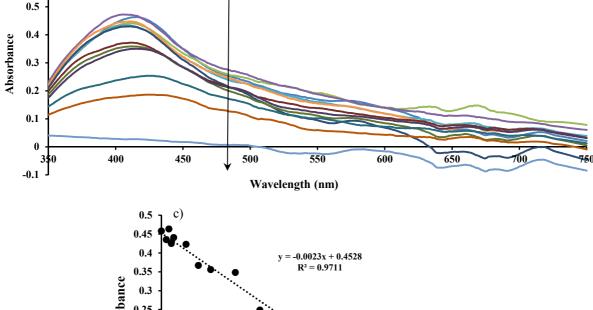
# 3.4. Calibration

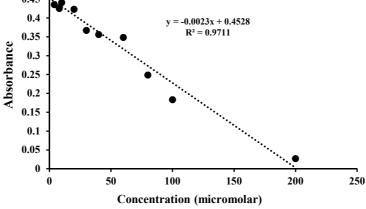
After finding the optimal conditions for detection of cyanide ion by AgNP, a series of solutions containing different concentrations of cyanide ion with 600  $\mu$ L of AgNP and sodium hydroxide were prepared and color and absorbance changes were explored and recorded. The results can be seen in Fig. 6. As can be observed in Fig. 6a, the color of AgNP fades by increasing cyanide ion concentration. Absorbance changes can also be

observed (see Fig. 6b). Therefore, gradual color changes are accompanied by gradual variations in absorbance.

In order to find a relation between absorbance of synthesized AgNP and concentration of cyanide ion, absorbance of the prepared solutions shown in Fig. 6a, at 415 nm were plotted against cyanide ion concentration (Fig. 6c). Results of the statistical analyzes of the calibration curve have been included in Table 1. A nearly linear relation betweeen absorbance of AgNP and







**Fig. 6**. a) Image of the calibration samples containing cyanide with concentration of 4.0, 6.0, 8.0, 10.0, 20.0, 30.0, 40.0, 60.0, 80.0, 100.0 and 200.0 μM, b) corresponding spectra and c) calibration curve. Arrows show the direction of increase in concentration. Conditions: sodium hydroxide =  $600.0 \mu L$  (0.1 M), AgNP =  $600.0 \mu L$ 

Statistical parameter	Value		
R	0.9854		
$R^2_{adj}$	0.9682		
Slope	$-0.0023\pm0.0001$ , $t = -18.3$		
Intercept	$0.4528\pm0.0089, t=50.9$		
LOD	3.8 μΜ		
Linear range	4.0-100.0 μΜ		
F statistics	336.44		

**Table 1.** Statistics of the calibration based on the proposed method

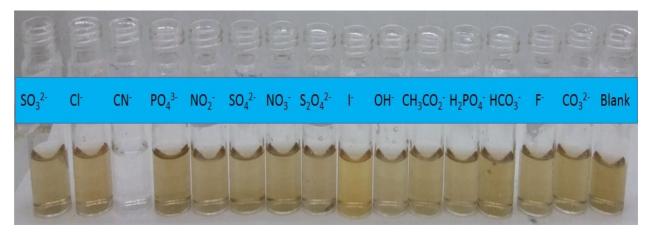


Fig. 7. Effect of different potential interferences in colorimetric detection of cyanide ion by the proposed method in optimal conditions (sodium hydroxide =  $600.0~\mu L$  (0.1 M), AgNP =  $600.0~\mu L$ ). Concentration of cyanide ion and potential interferences are 40.0~Mm

concentration of cyanide ion is confirmed based on the statistics in Table 1. This can be interpreted by inspection of correlation coefficient values (r) which are close to unity. Moreover, a high value of F statistics is obtained for the calibration curve. This illustrates that the calibration model adequately explains the absorbance changes of AgNP in the presence of cyanide ion and the relation between absorbance and concentration of cyanide ion is systematic.

Very low values of standard error calculated for slope and intercept of the calibration model also indicate the reliability of the model. Large values of t statistics for these parameters indicate that the model is robust.

Linear range of the calibration is relatively wide and detection limit is acceptable. Since the MCL of cyanide in drinking water is 0.2 ppm (7.7  $\mu$ M) based on (USEPA, 1999), the proposed method is advised for testing water samples for cyanide ion.

## 3.5. Interference study

In order to investigate selectivity of the proposed

method, a series of solutions containing AgNP and sodium hydroxide in optimal volumes were prepared and different potential interferences were added. The image of these solutions can be seen in Fig. 7. As can be seen from Fig. 7, disappearance of the color of the synthesized AgNP can only be observed in the presence of cyanide ion. However, in the presence of iodide ion, the color changes to a brighter yellow.

# 3.6. Real samples

Applicability of the proposed method was confirmed by analyzes of two water and one wastewater samples. For each water sample, six replicates were measured and the mean and relative standard deviation of the results have been collected in Table 2. Accuracy of the method was explored by injection of a known amount of cyanide ion to the real samples. As can be seen from the results in Table 2, accuracy and precision of the method are satisfactory.

<sup>&</sup>lt;sup>1</sup> Relative Standard Deviation (RSD)

	Added (μM)	Found (µM)	RE(%)	RSD(%)
Spring water				
	0.0	$ND^a$	-	12.7
	40.0	43.0	7.5	10
Well water				
	0.0	$ND^a$	-	1.4
	40.0	40.9	2.2	8.1
Electroplating wastewater				
	0.0	40.7	-	11.3
	40.0	75.4	13.3	3.9

**Table 2.** Results of the analyzes of different real water samples by proposed method. The results are for six replicate determinations of the samples

# 4. Conclusions

Various methods such as titration, voltammetry, electrochemistry and chromatography have been developed to diagnose cyanide, but unfortunately most of these techniques are expensive, time consuming and laborious.

A simple, fast and inexpensive colorimetric method for determination of cyanide in various water samples using a spectrophotometer was proposed. According to the obtained data, the proposed method has satisfactory accuracy and precision. Greenly synthesized AgNP served successfully as the reagent in a colorimetric way.

# 5. Acknowledgment

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