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Determination of Iron Content in Water

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By

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Abstract

An easy, efficient and safe method was developed to determine iron in water samples. The method is an Iron Cell Test kit from Spectroquant in which firstly all iron ions are reduced to iron (II) ions by ascorbic acid. In a thioglycolate buffered medium, iron (II) reacts with a triazine derivative to form a purple complex that is determined photometrically. Calibration curve of iron standards was done with concentrations of 0.50, 1.0, 2.0, 3.0 and 4.0 ppm and it gave a R^2 value of 0.9989 and straight line equation y=0.4749x-0.046. Iron analysis was done on two sets of water samples. Named as set I samples 1, 2, 3, 4, 5, and 6 and set II samples 1, 2, 3, 4 and 5, they were acidified with 0.1% HNO_3 and the absorbance was measured in a UV-Visible Spectrometer at 565 nm. The concentrations were found as 0.45, 0.13, 3.84, 5.64, 6.72, 5.78 ppm for set I samples and 0.11, 0.11, 0.14, 0.12, and 0.11 ppm, for set II samples respectively. The limit of detection (LOD) is 0.10 ppm, and, the limit of quantification (LOQ) is 1.0 ppm.

Note: The water samples were provided by Carolyn Johnson, Environmental Safety Officer at Governors State University.

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1. Introduction

"Iron is the second most abundant metal in the earth's crust. Dissolved iron in water, causes the water to taste metallic".[1] The water may also be discolored due to suspended solids containing minerals of iron that appear brownish in color.[2] Iron will leave red or orange rust stains in the sink, toilet and bathtub. It can build up in your dishwasher and discolor ceramic dishes. It can also enter into the laundry equipment and cause stains on clothing. "Even though the EPA says that the iron in the drinking water is safe to drink, the iron sediments, other trace impurities may support bacteria that are harmful, and these bacteria are mostly found in wells where the water has not been chlorinated".[3]

"Elemental iron is rarely found in nature, as the iron ions Fe^{2+} and Fe^{3+} readily combine with oxygen and sulfur containing compounds to form oxides, hydroxides, carbonates, and sulfides, so, dissolved iron more commonly exists in the form of its oxides".[4] To provide safe drinking water to the public, both government and private organizations measure iron content in drinking water and other tap waters in every sector including schools, hospitals, industries, etc.[5]

[1] Gunnar Nordberg; Bruce Fowler; Monica Nordberg. *Handbook on the toxicology of metals*, 4th ed.; Amsterdam, Elsevier, 2014, Chapter 41, Iron. pp 879-902.Website http://dx.doi.org/10.1016/B978-0-444-59453-2.00041 (accessed October 20, 2017)

[2] Ibrahim A.Q.; Onyenekwe P.C.; Nwaedozic I.M. An Efficiency Assessment of Lower Usuma Water Treatment Plant in Abuja Metropolis, Nigeria. *Environ. Sci. Toxicol. Food Technol*. [Online] **2014**, 8, Ver.II., pp 46-53 http://www.scribd.com/document/250619321/ (accessed October 16, 2017).

[3] U.S. EPA. *Secondary Drinking Water Standards: Guidance for Nuisance Chemicals*, website https://www.epa.gov/dwstandardsregulations/secondary-drinking-water-standards-guidance-nuisance-chemicals (accessed October 7, 2017).

[4] Fawell, J.K; Land.U; Mintz, B. *Iron in Drinking water*. Back ground document for development of WHO Guidelines for Drinking Water Quality. (online); Geneva, 2003. http://www.who.int/water_sanitation_health/dwq/chemicals/iron.pdf (accessed on October 9 , 2017)

[5] International Organization for Standardization, *Water quality—determination of iron*. (ISO 6332:1988)1988. Website .iso.org http://www.iso.org/standards/12630.htm. (accessed on October 9, 2017)

"In the drinking water supply, iron (II) salts are unstable and are precipitated as insoluble iron (III) hydroxide which forms as a rust colored sediment".[4] When water is directly pumped from the well, the water may contain iron (II) at concentrations of up to several milligrams per liter without any color or turbidity.[6] "When the iron levels are more than 0.05-0.1 mg/L turbidity and color develops in the pipe system. If the concentration is more than 0.3 mg/L staining of laundry and water systems may be damaged".[7] Iron also promotes undesirable bacteria growth within a water works and distribution system because of large deposition of iron minerals on piping.

"The iron concentration in rivers has been reported as 0.7 mg/L, and in groundwater, which is anaerobic, iron is in the form of iron (II), with the concentration being usually 0.5-10 mg/L; and sometimes, the concentration is found as high as 50 mg/L".[8] "The concentration of iron in water should be less than 0.3 ppm (0.3 mg/L); however, it may be higher in countries where various iron salts are used as coagulating agents in water-treatment plants and where cast iron, steel, and galvanized iron pipes are used for water distribution".[8]

"According to WHO and U.S. Federal guidelines, the limit for iron is less than 0.3 ppm (0.3 mg/L) in municipal drinking water".[9] Although iron is only toxic at very high concentrations, it acts as a useful surrogate for other heavy metals. An experiment that mainly focuses on measuring iron content in tap water and determines whether the water meets the standards and may also suggest the presence of other contaminants. Solutions containing iron are colorless at low concentration so the iron solutions are tested by adding a complexing agent that absorbs at a specific wavelength and is analyzed using a spectrophotometer. Iron is used as a constructional material for drinking water pipes and for structural support in automobiles, buildings and bridges. It is also used as pigments in paints. It is also use for treatment of iron deficiency in humans.[10] Various iron salts are used as coagulants in water treatment.

2. Method used for the experiment

[6] *Iron and water: reaction mechanisms, environmental impact and health effects*, website https://www.lenntech.com/periodic/water/iron/iron-and-water.htm#ixzz50JXZPunT (accessed on October 9 2017).

[7] . Weaver LC, Comparative toxicology of iron compounds 1961. *Am J Med Sci*,1961 241,296-302.

[8] World Health Organization. *Iron in drinking-water*. Background document for preparation of WHO Guidelines for drinking-water quality, 2008. Geneva, World Health Organization (WHO/SDE/WSH/03.04/8) 390. Website http://www.who.int/water_sanitation_health/dwq/chemicals/iron.pdf (accessed December 4, 2017)

[9] U.S. EPA. *Ground Water and Drinking Water: National Primary Drinking Water Regulations*. Website, https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations (accessed on October 12, 2017).

[10] Joint FAO/WHO Expert Committee on Food Additives, *Toxicological evaluation of certain food additives and food contaminants*, (WHO Food Additives,Cambridge University Press No. 18,1983).

For the determination of iron in the samples that were provided, Iron Cell Test Kit from Spectroquant in the Test Kit all the iron ions present in the samples was reduced to Fe^{2+} ions by ascorbic acid. In the presence of the medium thioglycolate, a purple complex was formed because of Fe^{2+} reacts with a trizine derivative.[11] The complex was determined photometrically by using UV-Vis spectrophotometer.

A UV-Vis spectrometer is an instrument used to measure the amount of ultraviolet and visible light absorbed by a solution. The light used in UV-Vis spectroscopy, is a very narrow portion of electromagnetic spectrum. The instrument is designed so that the sample is placed between a light source and detector. Depending on the sample, light may be absorbed causing electrons to be promoted from one energy level to another. Since different metal ions, have different absorption patterns, UV-VIS spectroscopy can be used to identify metal ions in solutions.

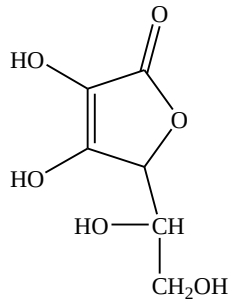
2.1 Materials

Iron metal, concentrated nitric acid (HNO_3) was purchased from Fisher Scientific, 6.0 M sodium hydroxide (NaOH) was purchased from wards science, 6.0 M hydrochloric acid (HCl) was purchased from Fisher Scientific, ascorbic acid was purchased from Acros and triazine present in the Iron Cell Test kit.

2.2 Apparatus

Iron Cell Test kit, Hanna pH meter pH range: pH 0.00 to 14.00 operated at room temperature (~ 20 ^{o}C). A Perkin Elmer Lambda 35 UV-Visible spectrometer was used to perform qualitative analysis. The UV visible spectrometer was operated with Perkin Elmer UV Win lab data processor and viewer vision 1.00.00 with wavelengths ranging from 700 nm to 300 nm using polystyrene cuvettes. HPLC filters 0.45 µm from Thermo Fisher Scientific with a polyethylene or Teflon filter material were used to remove interfering materials and fine particles where stated, micropipettes with differing levels of precision (20-200 µL, 2-20 µL, 100-1000 µL), glass volumetric flasks of 10, 20, 50, 100...500 mL, and plastic transfer pipettes. Iron Cell Test Kit from spectroquant.

[11] Spectroquant *Iron Cell Test* 1.14549.0001 Merck KGaA; website www.analytical-test-kits.com (accessed on 27th October 2017)

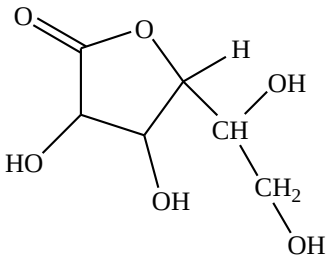


3,4-dihydroxy-5-(1,2-dihydroxyethyl)furan-2(5*H*)-one

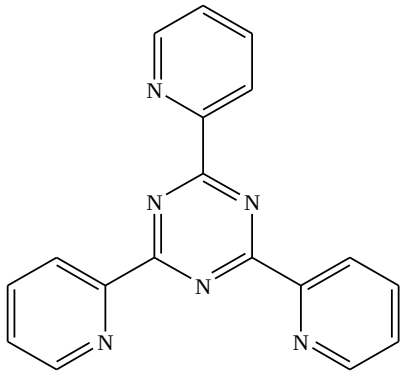
Ascorbic acid

+ Fe(III)

Thioglycolic acid



dihydro-3,4-dihydroxy-5-(1,2-dihydroxyethyl)furan-2(3*H*)-one



2,4,6-tri(pyridin-2-yl)-1,3,5-triazine

Figure 1. Steps involved in the principle chemical reaction[12]

Mechanism:[13] Ascorbic acid contains OH group, the hydrogen present in this OH group is taken up by the Fe^{3+} resulting in the formation of Fe^{2+}. The mechanism involved is reduction reaction. The thioglycolate present herein the reaction is acting as a buffer, when the reduction reaction is taking place i.e. conversion of from Fe^{3+} to Fe^{2+} this is indicated by the color change conforming that reduction reaction.

3. Procedure

The following is a narrative of the procedure that we developed. It is written in a style may be used for the future (see below) researchers or students who wish to perform this procedure.

3.1 Preparation of calibration curve

1. From a 1000 ppm stock solution of Fe3+ in 1% HNO3[14], solution of concentrations of 0.5, 1.0, 2.0, 3.0, 4.0 ppm was prepared

2. The standards were treated according to Iron Cell Test kit instructions and the absorbance was measured for each sample at 565 nm.

Absorbance vs. concentrations was plotted and the y-intercept was obtained slope and correlation were measured for each sample at 565 nm.

3.2 Preparation of samples for measurement

1. Water samples were collected from the tap water or other sources using (a clean 100 mL polyethylene containers provided by the instructor). For tap water, the water was collected after the tap was run for 2 minutes. For another source of water, such as a pond or river, the water was

[12] Klepo, L.; Copra-Janicijevic, A.; Kukoc-Modun, L., A New Indirect Spectrofluorimetric Method for Determination of Ascorbic Acid with 2,4,6-Tripyridyl-S-Triazine in Pharmaceutical Samples. *Molecules* **2016,** *21,101-113*. Web site www.researchgate.net/publication/291365592_A_New (accessed on December 1, 2017)

[13] Drits, V. A.; Manceau, A., A model for the mechanism of Fe3+ to Fe2+ reduction in dioctahedral smectites. Clays Clay Miner. **2000,** *48*, 185-195. Environmental Geochemistry website http://www.clays.org/journal/archive/volume%2048/48-2-185.htm (accessed on December 1, 2017)

[14] **Preparation of Fe^{3+} standard stock solution:** Fe wire (1.000 g) is weighed and dissolved in conc. HNO_3 (10 mL). If necessary, gently heat the solution until the wire is completely dissolved, cool to room temperature and quantitatively transfer the solution to a 1000 mL volumetric flask. Dilute to 1000 mL with purified water.

collected using nitrile glove to protect the sample from contaminated with dirt and bacteria from our hands.

2. The container was labeled with our name, date, and location of the sample.

3. From the water samples, 10 mL was dispensed into a small beaker and the pH was measured. The pH (must be within the range 1-10).

If necessary the pH was adjusted with sodium hydroxide (6.0 M) solution or (6.0M) optimally, the pH was adjusted to 7.

4. Further work was performed on a 25 mL aliquot of the sample.

5. If there were suspended solids, the 25mL aliquot was filtered using a 0.45 µm polyethylene or Teflon filter.

6. The 25 mL aliquot was treated with 0.1 mL of HNO3 (0.1%v/v).7. Then, 5.00 mL was pipette into a pre-prepared test tube containing the buffer ammonium thioglycolate and thioglycolic acid. (Note: this buffer stabilizes the pH to 7.0.)

8. The test tube was tightly capped and mixed well until the reagent and sample were completely combined.

9. The samples were left for 3 min. If the iron was present we will observe the formation of a purple solution.

10. The sample was measured in the UV- Visible spectrophotometer with absorbance at 565 nm.

11. The dissolved iron concentration was calculated from the above calibration curve.

12. For reproducibility check, the procedure was repeated with a 25mL aliquot of the sample solution.

3.3 Limit of detection (LOD) and limit of quantification (LOQ).[15]

The LOD is defined as the lowest amount of the analyte in a sample that can be detected but not necessarily quantified. The detection limit is determined by the analysis of sample with known concentration of analyte and by establishing the minimum level at which the analyte can be reliably detected.

A signal to noise (S/N) ratio analysis is performed by comparing measured signals from samples with known low concentrations of the analyte and with the blank samples. By establishing the

minimum concentration at which the analyte can be reliably detected, a S/N ratio of 3:1 was used in this study.

Where S = height of the signal, and, N = height of the noise.[2]

The LOQ was determined as the minimum concentration of analyte in a sample that can be quantified with acceptable precision and accuracy under the stated operational conditions of the method. The quantitation limit was determined by the analysis of sample with known concentration of analyte and by establishing the minimum level at which the analyte can be reliably quantitated.

The S/N ratio is performed by comparing measured signals from samples with known low concentration of analyte with those of blank samples and by establishing the minimum concentration at which the analyte can be quantitated; an S/N ratio of 10:1 was used in this study. After measuring the signal noise ratio which was 0.0002 mAu and then the concentrations were calculate by using standard calibration curve and the absorbance of the signal noise ratio the LOD and LOQ values are 0.05 ppm and 0.50ppm

[15.] Shrivastava, A., Gupta, V. Methods for the determination of limit of detection and limit of quantitation of the analytical methods. *Chron. Young Sci.* website www.scribd.com/document/325963935/Methods-for, **2011**. *2*, pp 21(accessed on November 26, 2017).

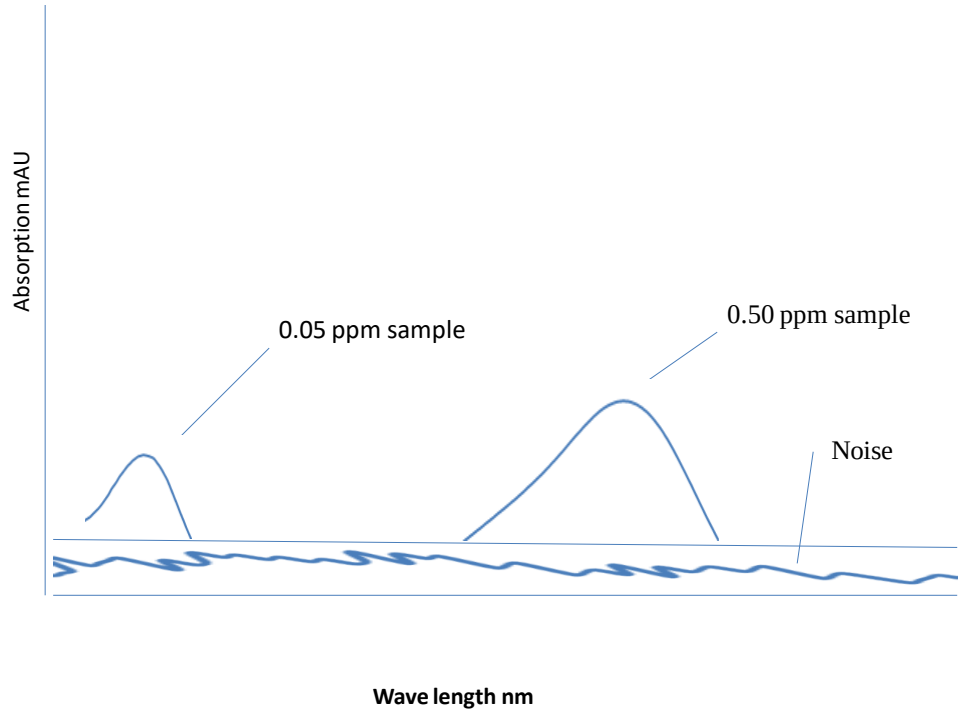


Figure 2. Signals for LOD and LOQ

4. Results:

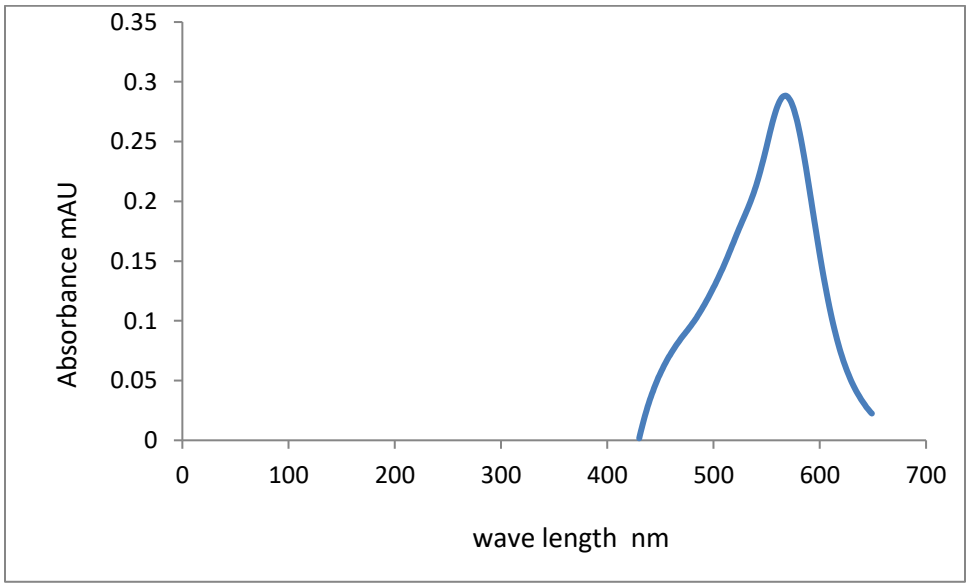


Figure 3. Peak of the real sample concentration 0.75 ppm

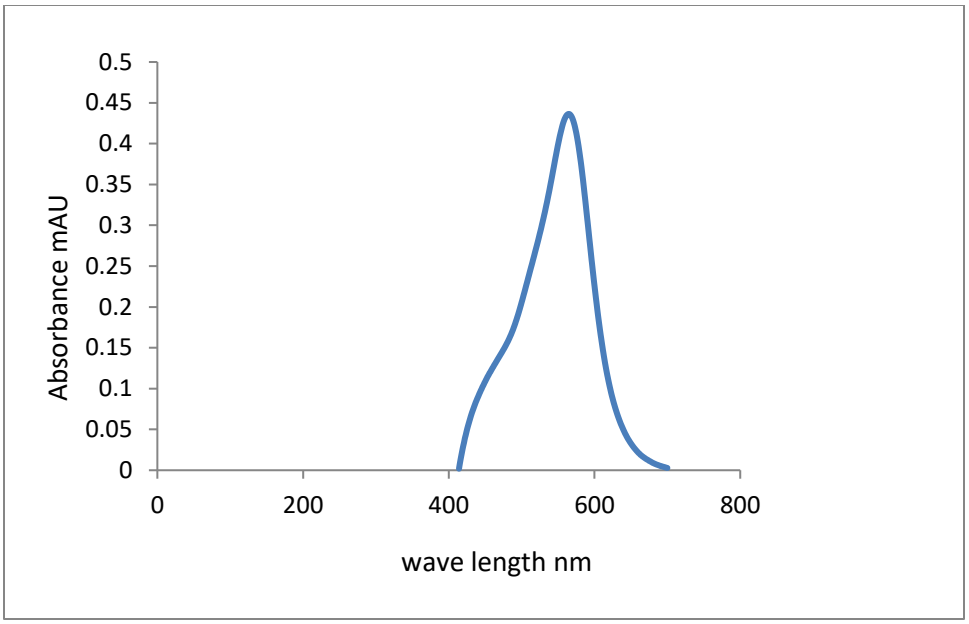


Figure 4. Peak of the standard concentration 1.0 ppm

The results of sample set I that we measured in this study are shown in Table 2.

Table 1. Absorbance and concentration of standards samples

Absorbance (mAU)	Concentration (ppm)
0.20	0.50
0.436	1.00
0.905	2.00
1.36	3.00
1.74	4.00

Table 2. Absorbance and concentration of sample 1 to 7 (set I)

Sample no.	Absorbance (mAU)	Concentration (ppm)
1. Standard	0.436	1.00
2. S-1	0.092	0.45
3.S-2	0.0201	0.13
4.S-3	0.842	3.84
5.S-4	1.24	5.64
6.S-5	1.48	6.72
7.S-6	1.27	5.78

The results of the samples set II that we measured in this study are shown in Table 3.

Table 3. Absorbance and concentration of samples 1 to 5 (set II).

Sample no.[b]	Absorbance (mAU)	Concentration (ppm)
1. S-1	0.0073	0.11
2. S-3	0.0058	0.11
3. S-4	0.0194	0.14[c]
4[a].S-5	0.0056	0.12
5. S-6	0.0074	0.11

Note

4[a]. Initially sample 4 showed a high reading of .097 ppm of iron, but on observing the sample solution which showed lot of sediments in it so, the sample was allowed to settle and then the concentration of settled (unmixed) sample had dropped to 0.14 ppm. Then after mixing the sediment in the sample the concentration of iron again raised to 0.97 ppm so, the sample was filtered in order to get rid of the sediment which upon gave a concentration of 0.12 ppm.

[b] Samples1.00 ppm and S-2 in the table 1 were not tested in table 2.

[c]. we were surprised by this ppm result because of the higher absorbance. However, this results is mathematically consistent with the associative properties of the y= mx+b equation especially since the y- intercept term is Non- zero.

Table 4. Error analysis for samples (set II)

Conc	Abs 1	Abs 2	Abs 3	Error
0.11	0.0061	0.0061	0.0073	±0.0007
0.11	0.0053	0.0059	0.0058	±0.0003
0.14	0.0127	0.0127	0.0194	±0.0039
0.12	0.0048	0.0059	0.0056	±0.0006
0.11	0.0074	0.0074	0.0074	±0.0000

Table 4 describes the error analysis for given set of samples that were analyzed.

Error was calculated using the following formula

$$\frac{\text{Highest observed absorbance} - \text{Lowest observed absorbance}}{\text{Square root of the no of measurements}}$$

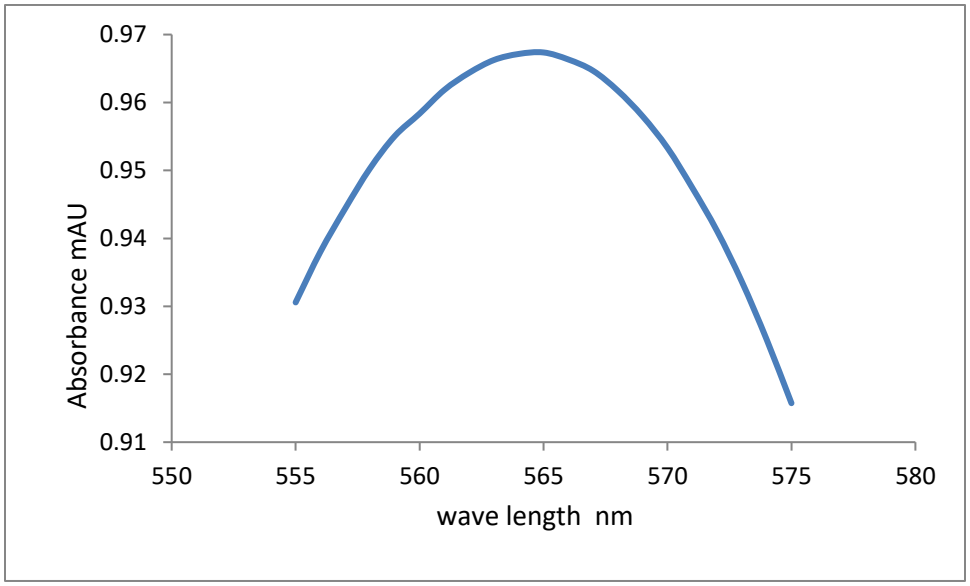


Figure 5. Peak for sample 4 (set II) concentration 0.97 ppm

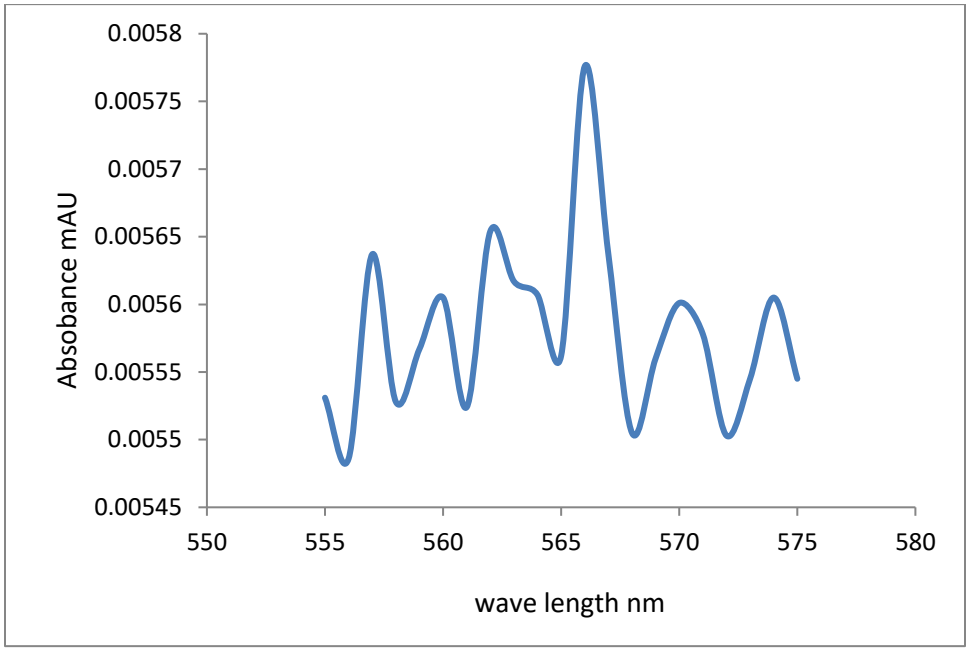


Figure 6. Peak for sample 4 (set II) after filtration

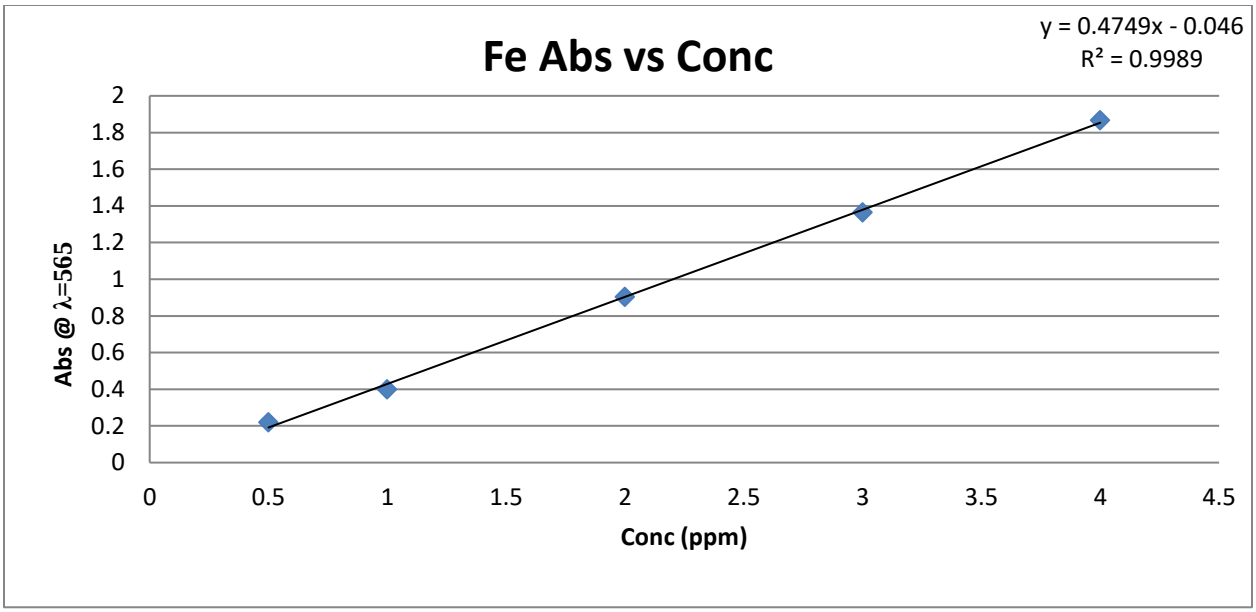


Figure 5. Calibration curve of iron standards samples.

5. Discussion:

Initially two sets of samples were given to us by Environmental Safety Officer at Governors State University. Set I samples, were given to us at the time pipe line failure. Set II samples were given to us after the pipe lines were fixed.

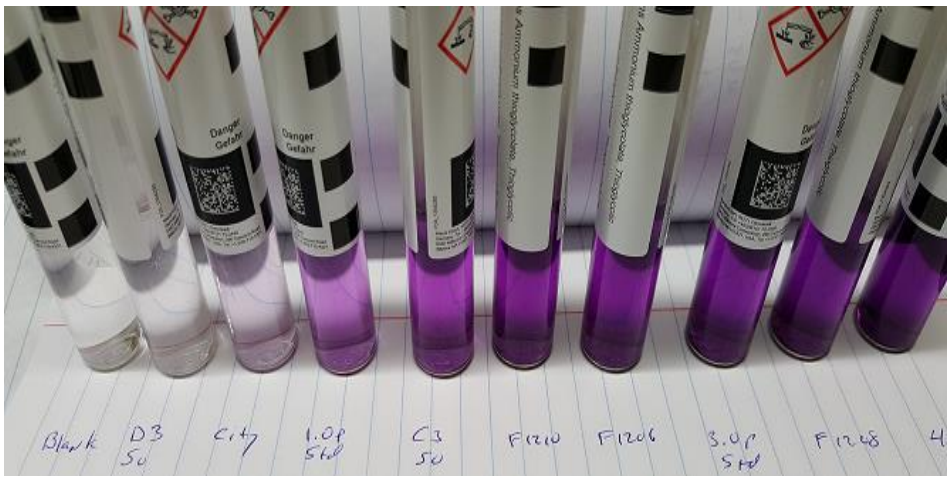


Figure 6. Photograph of set I blank, standards and samples.

Figure 6 Photograph of the set I blank, standards and samples treated according to the procedure in this study. Arranged left to right: a) blank which contains purified water and the chelating agent, b) sample 1 (D3 So), c) sample 2 (city) taken directly from the city of University Park (city), d) the 1.0 ppm standard solution, e) sample 2 (C3 So), f) sample 3 (F1210), g) sample 4 (F1206), h) the 3.0 ppm standard, i) sample 5 (F1208), j) the 4.0 ppm of standard.

From the result set I samples except for the sample 1 (D3 So) and sample 2 (city) contain high concentration of iron. Figure 6 shows 10 solutions that were analyzed in set I. The samples are arranged according to the color and the trends shows the sample that contain more iron it appears progressively deeper blue in color.

Here set II, five water samples were given to us by Environmental Safety Officer at Governors State University, labeled as S-1, S-3, S-4, S-5 and S-6. All samples were acidified to a concentration of 0.1 % (v/v) with HNO_3 and then treated with a chelating agent to yield a violet colored solution. Detection of all samples were performed on UV-Visible spectrometer at 565 nm. Once the water samples concentrations were determined, the results were compared with EPA guidelines for recommended amount of dissolved iron (< 0.3 ppm). For samples1-3 and 5, the water was considered below the EPA guideline. However, sample 4 had a concentration of 0.97 ppm. In this sample, visible sediments were found. We took additional action in determining the cause of the high concentration. We decided to further test sample 4. We filtered the sample and this was analyzed to 0.11 ppm. Additionally, we obtained a sample in which the water was run from the tap for 2 min. This we measured to give a concentration of 0.14 ppm.

6. Conclusion

"According to US EPA, the recommended limit for dissolved iron in drinking water is 0.30 ppm".[8] As per our result, all the samples concentrations in set I are much higher than the limit of iron in drinking water except sample 2 and all samples concentrations in set II are in range of 0.10 to 0.15 ppm. There was one exception in which sample 4 contained suspended solids that initially gave a high dissolved iron concentration (0.97 ppm). However, after filtration and also after having water run from the tap for 2 min, the iron concentration decreased to acceptable

levels. This means all samples pass the EPA recommendation for dissolved iron drinking water. In addition, our analysis indicates that water containing suspended solids may result in high dissolved iron concentrations. Overall, we are able to come to the conclusion that if the water is left running for two minutes, the sediments are able to be flushed and an sample that is within the EPA concentration guideline is obtained.

Finally the water samples that we tested were found to have dissolved iron concentration below the EPA limit of 0.3 ppm

7. Glossary:

L = Liter

M = Molarity

mg = Milligram

mL = Milliliter

nm = Nanometer

pH = Potential hydrogen

ppm = Parts per million

mAU = Milli-Absorbance-Units

µL = Micro liter

λ = Wave length

8. Acknowledgement of collaborators

I would like to say sincerely thanks to Dr. John Sowa (professor in the Department of Chemistry), Rameshbabu Ambari (graduate student, Department of Chemistry) and Enas EI-Khatib (graduate student, Department of Chemistry).