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"ESTIMATION OF IRON METAL CONTENTS IN NATURAL SAMPLES BY UV-VISIBLE SPECTROPHOTOMETER METHOD"

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ABSTRACT

Like other transition metal ions, Fe²⁺ forms many coordination compounds in which the iron ion is surrounded by various Lewis bases, called ligands. Many of these coordination compounds are colored, unlike aqueous ferrous ion itself. If the colored complex is stable and forms quantitatively, it can be used to measure of the amount of iron present. 1,10-Phenanthroline or O-Phenanthroline is a bidentate chelating agent, which forms a stable red complex with ferrous iron. This procedure is selective for Fe²⁺, even in the presence of other metals, for several reasons. Sodium acetate buffer was used to maintain the pH for optimum complex formation while Hydroxylamine

Hydrochloride was added as a reducing agent to intercept oxygen and prevent oxidation of ferrous iron to ferric iron. Fe(II) + 3 phen \Leftrightarrow Fe(phen)₃²⁺ (Red) Determination of Iron in Natural and synthetic samples was carried out using the UV Visible Spectophotometer CL-320 (Chemi line). Concentration of iron in test solution was calculated from the Standared Curve prepared. For each natural and synthetic samples at least two readings were obtained and the average calculated. Result of the study suggests theta method can be used satisfactory for the analysis of iron.

KEYWORDS: UV-Visible Spectrophotometer, Natural sample, Iron test and Analysis.

INTRODUCTION

Iron plays an important role in the body. One of the main roles of iron is to help our red blood cells transport oxygen to all parts of the body.

Iron also plays an important role in specific processes within the cell that produce the energy for our body. It is for this reason that one of the first symptoms of low body iron stores is tiredness and fatigue.

There are two types of iron

Haem Iron: This type of iron is found in animal-based foods, like red meat, poultry and fish. Haem iron is easily absorbed by the body.

Non-Haem Iron: This type of iron is found in plant-based foods like cereals, vegetables and legumes. In contrast to haem iron, our body doesn't absorb non-haem iron as easily. However, because it is present in the diet in much larger quantities than is haem iron, it is an important source of this mineral. We generally obtain around 65% of our iron requirements from non-haem iron.

About 25 percent of the iron in the body is stored as ferritin, found in cells and circulates in the blood. The average adult male has about 1,000 mg of stored iron (enough for about three years), whereas women on average have only about 300 mg (enough for about six months). When iron intake is chronically low, stores can become depleted, decreasing hemoglobin levels.

When iron stores are exhausted, the condition is called iron depletion. Further decreases may be called iron-deficient erythropoiesis and still further decreases produce iron deficiency anemia.

Blood loss is the most common cause of iron deficiency. In men and postmenopausal women, iron deficiency is almost always the result of gastrointestinal blood loss. In menstruating women, genitourinary blood loss often accounts for increased iron requirements. Oral contraceptives tend to decrease menstrual blood loss, whereas intrauterine devices tend to increase menstrual bleeding. Other causes of genitourinary bleeding and respiratory tract bleeding also increase iron requirements.

Iron is also involved in the conversion of blood sugar to energy. Metabolic energy is crucial for athletes since it allows muscles to work at their optimum during exercise or when competing.

The production of enzymes (which play a vital role in the production of new cells, amino acids, hormones and neurotransmitters) also depends on iron. This is important if you are competing professionally or following serious exercise so you can perform at your best.

The immune system is dependent on iron for its normal functioning. Iron also contributes to normal cognitive function in children. Iron is lost by the body through a variety of ways including urination, defecation, sweating, and exfoliating of old skin cells. Women needs more iron than men as they suffer from Bleeding during menstrual cycle.

If iron stores are low, normal haemoglobin production slows down, which means the transport of oxygen is diminished, resulting in symptoms such as fatigue and tiredness.

Iron plays an important role in biology, forming complexes with molecular oxygen in hemoglobin and myoglobin; these two compounds are commonoxygen transport proteins in vertebrates. Iron is also the metal at the active site of important redox enzymes dealing with cellular respiration and oxidation and reduction in plants and animals. A human male of average height has about 4 grams of iron in his body, a female about 3.5 grams.

Iron deficiency causes Anemia. Anemia is a condition characterized by inadequate red blood cells (erythrocytes) or hemoglobin. Iron deficiency anemia occurs when the body lacks sufficient amounts of iron, resulting in reduced production of the protein hemoglobin. Hemoglobin binds to oxygen, thus enabling red blood cells to supply oxygenated blood throughout the body.

The ferric-ferrous ratio of natural silicate liquids equilibrated in air was studied by Kilinc et al.^[1] Determination of Ferrous Iron in the Presence of Ferric Iron With Bathophenanthroline was done by Lee et al.^[2] Determination of arsenic, boron, carbon, phosphorus, selenium and silicon in natural waters by direct current plasma atomic emission spectrometry was observed by Urasa.^[3] Colorimetric flow-injection analysis of dissolved iron in high DOC waters was studied by Pullin et al.^[4] The new method utilizes two selective ligands to stabilize Fe(III) and Fe(II), thereby preventing changes in Fe reduction—oxidation distribution. Complexed Fe(II) is cleanly removed using a silica-based, reversed-phase adsorbent, yielding excellent isolation of the Fe(III) complex. Iron(III) concentration is

measured colorimetrically or by graphite furnace atomic absorption spectrometry (GFAAS).^[5]

Solid Phase Colorimetry of Trace Metal Ions Based on a Tristimulus Chromaticity Diagram and Simultaneous Determination of Iron(II) andIron(III) were observed by Yokota et al.^[6] A procedure for the removal of free iron from soils and clays by a single extraction at 50 °C. with sodium hydrosulphite in a citrate buffer at pH 4.75 is developed by D. E. Coffin.^[7] The effect of the iron ore tailings on the coastal environment of Tolo Harbour, Hong Kong was studied by Wong et al.^[8] Development of a dispersive liquid–liquid microextraction method for iron speciation and determination in different water samples was done by Tabrizi⁹. Visual estimation of iron in saprolite was done by Hurst.^[10]

Solvent extraction and fluorometric determination of fluoride ion at ppb level in the presence of large excess of aluminum(III) and iron(III) by using an expanded porphyrin, sapphyrin were determined by Nishimoto et al.^[11]

Determination of iron(II) in natural waters by capillary zone electrophoresis using on-capillary complexation with 2,4,6-tri(2'-pyridyl)-1,3,5-triazine was observed by Dahlen et al.^[12]

Al(III) and Fe(III) binding by humic substances in freshwaters, and implications for trace metal speciation was studied by Tipping et al.^[13]

MATERIAL AND METHOD

The instrument used for the study was UV Visible Spectophotometer CL-320 (Chemi line).

All Solutions were prepared in deionized water because we were working with dilute solutions of Iron in this experiment and because tap water contains significant concentrations of iron, we have to be very careful to avoid contamination of our solutions with tap water. In other words, all glassware should be rinsed thoroughly with deionized water before use.

Diluted sulfuric acid 0.7 M: This solution was prepared by adding 40 ml of concentrated sulfuric acid to approximately 1 L of deionized water in a beaker, mixing thoroughly and allowing to cool to room temperature.

Hydroxylamine Hydrochloride 10 g/100 ml: This solution was prepared by dissolving approximately 2.5 g of hydroxylamine hydrochloride (H₂NOH·HCl) in approximately 25 ml of water in a small beaker.

1,10-phenanthroline, 0.1 g/ 100 ml: This solution was prepared by dissolving approximately 0.2 g of orthophenanthroline monohydrate in 200 ml of water. If necessary, the mixture is warmed gently and stirred to ensure complete dissolution. The solution stored in the dark until it is used. If the solution darkens at any step in the process, discard it and prepare another solution.

Sodium acetate, 1.22 M: This solution was prepared by dissolving approximately 10 g of sodium acetate in 100 ml of water in a small beaker. Obtain about 100 ml of this solution in a small beaker.

Preparation of Standard ferrous ammonium sulfate solutions:

Stock 1

Weigh approximately 0.210 grams of reagent grade Ferrous Ammonium Sulfate Hexahydrate $(FeSO_4(NH_4)_2SO_4\cdot 6H_2O, MM=392.14)$ to the nearest fourth of a milligram, transfer the salt carefully to a small beaker and dissolve the salt in 12.5 ml (graduated cylinder) of the diluted sulfuric acid solution. When the salt was completely dissolved, transfer the solution quantitatively to a 500-ml volumetric flask using at least five rinses with small volumes of deionized water to ensure that all the solution is transferred to the volumetric flask. Dilute the solution in the volumetric flask to the calibrated mark and mix thoroughly.

Stock 2

Pipet 25.00 ml of the Stock 1 solution into a clean 250-ml volumetric flask, add 5 ml of the diluted sulfuric acid solution to the flask, dilute to volume and mix thoroughly. Natural samples of various groups were purchased from local markets and retail stores for analysis. In Natural sample the edible portions was blended and aliquots taken for analysis.

Preparation of Natural Sample

An amount of 5-15 g of the homogenized sample was dried in an air oven at 105°C for 3 hours. The dried sample then charred until it ceased to smoke. The charred sample was kept in a muffle furnace until a whitish or grayish ash was obtained. The ash was treated with dilute sulfuric acid, transferred to a volumetric flask and make up to 50 ml. For each food to

be studied, two ash solutions were prepared, i.e. duplicate analysis was carried out. Each ash solution was used for the determination of iron by the Atomic absorption spectrophotometer method.

Reaction step (Colour development)

Standard and unknown solutions prepared were used to develop the Iron/Phenanthroline complex ion in a blank, calibration standards and an aliquot of our unknown solution. prepare one blank and one set of standard solutions and prepare unknown sample. UV Visible Spectophotometer CL-320 (Chemi line) was used. Wave length was set at 505 nm. A standard curve was prepared using Ferrous Ammonium Sulphate and used for calculation of iron in the test (unknown) solution.

UV Visible Spectophotometer is used to get a calibration curve with at least 4 concentration of Iron was prepared.

Concentration of iron in test solution was calculated from the Standard Curve prepared. For each natural and synthetic samples at least two readings were obtained and the average calculated.

RESULT AND DISCUSSION

The Absorption Spectrophotometric method was found to give satisfactory result. In the method several steps for the observations recordings. The red colour complex formed is stable for a number of hours. The procedure is relatively economically cheaper, thats requires only a low cost instrument for the measurements.

The Atomic Absorption Spectrophotometer requires the purchase of a high cost instrument which is also expensive to operate and maintain. The ash solution can be used directly in AAS and sample can be destroy. AAS method is more sensitive method than Colorimetric method. We have worked on AAS at Govt. Pollution Control Board, Vijay Nagar, Indore.

The describe method are very simple, rapid, sensitive and selective for estimation of Iron. Like other transition metal ions, Fe²⁺ forms many coordination compounds in which the Iron ion is surrounded by various Lewis bases, called ligands. Many of these coordination compounds are coloured, unlike aqueous ferrous ion itself. If the coloured complex is stable and forms quantitatively, it can be used to measure of the amount of Iron present.1,10-

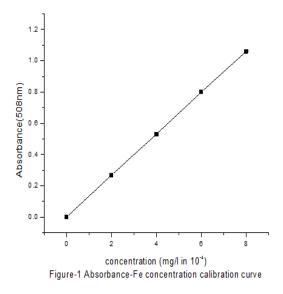
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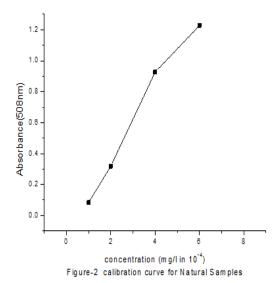
Table 1: Calibration Curve Data for Synthetic Sample by UV-Visible spectrophotometer.

Concentration(mg/ml)	Absorbance
Blank	0.000
2.0×10 ⁻⁴	0.270
4.0×10 ⁻⁴	0.534
6.0×10 ⁻⁴	0.807
8.0×10 ⁻⁴	1.065

Table 2: Curve Data For Synthetic Sample by UV- Visible Spectrophotometer.

S.no.	Sample No.	Absorbance
1.	Sample 1	1.084
2.	Sample 2	0.319
3.	Sample 3	0.926
4.	Sample 4	1.028
5.	Sample 5	0.545
6.	Sample 6	1.026
7.	Sample 7	0.748





CONCLUSION

In the Atomic Absorption spectrophotometric methods, the red colour complex is formed which is stable for number of hours. The procedure is also relatively much cheaper, requiring only a low cost Instrument.

The deficiency of iron cause anemia. Most of the people of our country are poor. So they cannot buy rich food, The person, who suffer from iron deficiency can get rid of their diseases by selecting the iron rich vegetables, fruits, pulses and multivitamins tablets or iron tablets including as source of vitamin C in our meal too. Vitamin C helps Iron absorption. In case of unavailability of Iron rich vegitables, fruits the people can grow such vegetable, fruits and pulses in their gardens or fields.

From this research it is concluded that vegetables are nutritious foods that provide sufficient amount of nutrients needed for normal body function, maintenance and reproduction. It was found that nutrients composition in all the selected vegetables was different. Some vegetables contained high amount of starch while other contained maximum amount of protein. Some vegetables were rich in some minerals such as Fe, P, Na, K etc. but their concentration in other vegetables were poor. Vitamins concentration was also different in all the vegetables. Moisture content was found maximum in all the selected vegetables. Vegetables are poor sources of fat that make them good food for obese people. They are good source of fiber and can decrease the concentration of high cholesterol level in body. From this trial we found that vegetables intake in different combination is essential for the maintenance of healthy life and normal body functioning. However, further investigations are required to notice the effect of cooking and storage conditions on these valuable nutrients.

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