

CHAPTER- 3

Theoretical analysis

Organic reagents shows good analytical properties with various metal ions forming soluble metal complexes (M:L). Literature survey shows that very few benzoyl hydrazones and isonicotinoylhydrazones are employed for the direct and derivative spectrophotometric determination of Pb(II), Cd(II), V(V) and Cu(II) metals.

3.1. Aims and Plan of Research Work

In this context the present investigations are as follows.

- i. Synthesis of new Ligand as Chromogenic reagent
 - a) Diacetylmonoxime-3-amino-4-hydroxy benzoyl hydrazone (DMAHBH)
 - b) 3,5-Dimethoxy-4-hydroxy benzaldehydeisonicotinoylhydrazone(DMHBIH)
- ii. Characterization of the ligand and their analytical properties.
- iii. To develop direct and derivative spectrophotometric determination of Cadmium (II), Lead (II), Vanadium (V) and Copper (II) by using DMAHBH and Pb (II), Cd (II) by DMHBIH.
- iv. Applications-The developed method is applied for the quantitative analysis of Cadmium (II) in soil, tobacco samples, Lead (II) in food and soil samples, Vanadium (V) in alloys, water, Plantmaterials and Copper (II) in Food Samples.

3.2. Preparation of solutions

Preparation of reagent solutions

Stock solutions (1×10^{-2} M) of reagents were prepared in dimethyl formamide (DMF) by dissolving appropriate quantity of each reagent substance separately.

i) 3,5-Dimethoxy-4-hydroxybenzaldehydeisonicotinoylhydrazone(DMHBIH)

0.30103 g of the compound (DMHBIH) was transferred into a 100-ml volumetric flask. It was dissolved and diluted up to the mark with DMF to get 1×10^{-2} M concentration of DMHBIH solution.

(ii)Diacetylmonoxime-3-amino-4-hydroxybenzoyl hydrazone (DMAHBH)

0.2503 g of the compound (DMAHBH) was transferred into a 100-ml volumetric flask. It was dissolved and diluted up to the mark with DMF to get 1×10^{-2} M concentration of DMAHBH solution. These reagent stock solutions were suitably diluted to get the required concentrations wherever necessary.

Preparation of Inorganic stock solutions

Preparation of Lead (II) Solution: Requisite quantity of Lead acetate (AR, BDH) was dissolved in double distilled water in a 250ml Volumetric flask to get 1×10^{-2} M Solution and standardised [175].

Preparation of Cadmium (II) Solution: Stock solution of Cd (II) (1×10^{-2} M) was prepared from CdSO_4 with double distilled water containing few drops of Concentrated H_2SO_4 and made upto 250ml. The stock solution was standardized [175].

Preparation of Vanadium (V) Solution: Vanadium (V) stock solution was prepared by dissolving Sodium metavanadate (S.D.Fine) in double distilled water and then standardized. Working solutions of above metals were prepared by diluting the stock solutions.

Preparation of Copper (II) Solution : Copper (II) Stock Solution was prepared by dissolving Copper Sulphate (S.D.Fine , A.R.Grade) in double distilled water and then standardized [175] .

Preparation of Foreign Ions:

The solutions of various cations and anions were prepared for interference studies [175].

3.3. Preparation of Buffer solutions

Buffer solutions (Phosphate buffer) were prepared by adopting the standard procedures reported in the literature[176].

3.4. Instruments employed:

UV-Visible recording spectrophotometer (UV-160A)

Shimadzu Corporation, Spectrometric instrument plant, Analytical instruments division, Kyoto, Japan. UV-160A is a double beam microprocessor based spectrophotometer designed for the quantitative analysis.

Digital p^H meter

The p^H of the buffer solutions was monitored by using ELICO digital p^H meter (LI-120) manufactured by M/s. ELICO Pvt. Ltd. India.

Digital electronic balance

Sartorius BS/BT 2245 model (Germany make) electronic analytical balance.

3.5. General experimental procedures

Spectrophotometric determination of Lead (II), Cadmium (II) Vanadium (V) and Copper (II) was carried out using synthesized hydrazone reagents. They were namely:

- 1) Diacetylmonoxime-3-amino-4-hydroxybenzoylhydrazone (DMAHBH)
- 2) 3,5-Dimethoxy-4-hydroxybenzaldehydeisonicotinoylhydrazone (DMHBIH).

The reagent Diacetylmonoxime-3-amino-4-hydroxybenzoylhydrazone (DMAHBH) reacts with Pb(II), Cd(II), V(V) and Cu (II) in Phosphate buffer (pH 3.0 – 10.0) medium to give colored complexes in presence of surfactant except Copper (II). 3,5-Dimethoxy-4-hydroxybenzaldehydeisonicotinoylhydrazone (DMHBIH) reacts with Cd (II) and Pb(II) in phosphate buffer solutions gives yellow colored complex in presence of surfactant.

3.5.1. Absorption spectra of reagent solutions and metal complexes

An aliquot of reagent (usually 0.5 ml of 1×10^{-2} M) solution was taken in a 10-ml volumetric flask containing 3-ml of buffer solution and suitable volume of surfactant was added and the volume was made up to the mark with distilled water. The absorbance of the reagent solution was measured against buffer blank. A plot between absorbance and the wavelength was made.

The following procedure was adopted for measuring the absorption spectra of complex (metal + reagent) in aqueous medium. In a 10-ml standard flask, the metal complex was prepared by taking 3-ml of buffer, suitable volume of surfactant, suitable concentration of metal ion and reagent (usually 10-fold molar excess to metal ion) solutions. The contents were diluted up to the mark with distilled water and the absorbance of the complex was measured against the reagent blank prepared identically. A plot between absorbance and the wavelength was plotted from which the analytical wavelength was selected.

3.5.2. Effect of pH on the absorbance of the metal complexes

To examine the effect of p^H on the intensity of the color, the metal complex in solution was prepared using buffer (Phosphate) solutions having different p^H values.

In a set of 10-ml standard flasks, 3-ml of buffer (different p^H values 1.0 to 11.0) solution, constant amount of metal ion, reagent (usually 10-fold molar excess to metal ion) solution and required quantity of surfactant were taken, made up to the mark with distilled water. The absorbance of each solution (metal complex) was measured at a selected wavelength (λ_{max}) against corresponding reagent blank prepared accordingly. A plot was made between absorbance and p^H from which the working p^H was selected.

3.5.3. Effect of reagent concentration on the absorbance

The following procedure was employed to find optimum amount of reagent required for full color development. Varying and known aliquot of reagent solution was taken in a set of 10 (10-ml) calibrated flasks, each containing 3-ml of buffer solutions, fixed amount of metal ion and adequate volume of surfactant. The contents were diluted up to the mark with distilled water and absorbance of the solution in each flask was measured at a selected wavelength against suitable reagent blank prepared identically. From this experiment, required number of folds of the reagent necessary for full color development was ascertained.

3.5.4. Effect of time on the absorbance of reaction mixture and stability

In a 10-ml calibrated flask, 3-ml of buffer solution, 0.5ml of metal ion (usually in Beer's law range) and 0.5ml of reagent (usually 10-fold molar excess) and required quantity of surfactant were taken and diluted up to the mark with distilled water. The absorbance of the colored complex solution was measured at a selected wavelength in different time intervals against reagent blank prepared similarly. From this experiment, the time stability of the complex and time interval required for full color development can be ascertained.

3.5.5: Effect of surfactants on the metal complexes

Surfactant micelles can enhance sensitivity and can bring about changes in solubility, pK_a , chemical equilibrium, stability, reaction rates and mechanisms of some chemical process. Hence, the effect of surfactants (CTAB, Triton X-100 and SDBS) on the solubility, stability and absorbance of metal complexes was studied and suitable surfactant was selected. In order to

arrive at the optimum volume of surfactant required to retain the metal complexes stable in solution state, the following method was adopted.

To different 10-ml volumetric flasks each containing 3 ml of buffer solution, various known aliquots of surfactants, appropriate amount of metal ion and required aliquots of reagent solution were added. The contents of flasks were made up to the mark with distilled water and the absorbance of the solution in each flask was measured at the selected wavelength against reagent blank prepared similarly.

The surfactants such as Triton X-100 (neutral), Cetyltrimethyl ammonium bromide (CTAB) (cationic) and Sodiumdodecylbenzene sulphonate (SDBS) (anionic) were used in the present study of metal complexes in solution state. From this experiment the required quantity of surfactant was ascertained.

3.5.6. Applicability of Beer's law

To ascertain the sensitivity of the color reactions and to explore the possibility of determining micro amounts of metal ions, the following procedure was used in the present study.

In a set of twelve (10-ml) volumetric flasks, 3-ml of buffer solution, varying and known aliquots of metal solution of appropriate concentration, suitable aliquot of surfactant and excess amount of reagent solution were taken and diluted to the mark with distilled water. Then the absorbance of all solutions was measured at a selected wavelength (λ_{\max}) against blank solution.

A plot between absorbance and amount of metal ion ($\mu\text{g/ml}$) was constructed. The slope and intercept of the plot was computed. Various analytical parameters are calculated using the slope of calibration plot.

3.5.7. Effect of foreign ions on the absorbance of reaction mixture

In order to assess the applicability of the proposed methods for the analysis of real or synthetic samples containing the metal ions, the effect of the presence of various foreign ions which were generally found associated with the test metal ions on the absorbance of the complex solution was studied by adopting the following procedure.

Fixed amount of metal ion solution was taken in 10-ml volumetric flasks containing 3-ml of buffer solutions and required quantity of surfactant. Then appropriate amount of foreign ions are added to all the flasks except one. The reagent solution was added to all at the end. The

contents were made up to the mark with distilled water. The absorbance of the complex in each flask was measured at a selected wavelength (λ_{max}) against reagent blank. From this absorbance the tolerance limit of the foreign ion was determined. The amount of foreign ion which brings about a change in absorbance by $\pm 2\%$ was taken as its tolerance limit.

Cations were added mostly as chlorides, sulphates or nitrates and the anions were added as sodium or potassium salts. Some of the cations interfering were masked by using adequate quantity of suitable masking agents.

3.5.8 Applications

The present methods were applied for the determination of Lead, Cadmium, and vanadium in soil samples, tobacco (cigarette), plant material[33,34], alloys[35-37], food stuff samples, real water samples, biological samples and also for Copper in food samples. The above sample solutions were prepared as per literature [33-37]. Wherever original samples are not available, synthetic samples were prepared accordingly and used.

3.5.9 Job's continuous variation method and Molar ratio Method

Composition of the complex and stability of the complex were calculated by Job's and Molar ratio method. The stability constant of the complex were calculated from the experimental data obtained from Job's method.

The stability constant of a complex is defined by the equation

$$\beta = \frac{[\text{Complex}]}{[\text{Metal Ion}] [\text{Ligand}]^n}$$

Applications

The methods developed in the present investigation will be employed for the estimation of metals in various samples like [239-243]cigarette, soil, bulked food, biological, water and plant material, alloy, tobacco, synthetic alloy samples, as per the procedures reported earlier.

3.6. Introduction to Cadmium

The availability of reliable analytical data is an important consideration for a better understanding of the environmental and clinical role of Cadmium.

The two natural radioactive isotopes were ^{109}Cd with half-life 7.7×10^{15} and ^{115}Cd with half-life 2.9×10^{19} . Greenockite (CdS), the only Cadmium mineral of importance, was nearly always associated with sphalerite (ZnS). The main source of Cadmium was from Mining, Metallurgical operations, Electroplating Industries, Plastic Industries, Metallic Plastic Pipes, Superphosphates, Sewage sludge and automotive tyres. Cadmium gets accumulated in the Kidneys as a toxic metal. It is associated with induction of Hypertension, Cirrhosis of the Liver and also leads to pulmonary embolism. Cadmium accumulation in the body causes softening of bones, Fractures, deformities and pain. The maximum permissible limit of Cadmium in drinking water is $0.01 \mu\text{g/L}$.

Cadmium may be separated by precipitation (as Sulphide), extraction (CHCl_3 , or CCl_4 , as diethyl dithio carbamate) and ion-exchange methods. It can also be isolated with caproic acid.

Analytical chemist specifies an extraction spectrophotometric method for Cadmium, based on its reaction with dithizone. The method has a low limit of detection but is not specific. Cadmium ion reacts with dithizone in neutral to strongly alkaline media to give a Pink Colored Complex, which is moderately soluble in carbon tetrachloride and more readily soluble in chloroform. The stability of the complex in strongly alkaline media allows cadmium to be extracted from Pb (II),

Bi (III), Sn (II) and Zn (II), the dithizonate of which cannot exist under such conditions. Dimethyl glyoxime is added to mask Nickel (II) and Cobalt (II). Tartarates prevents the precipitation of metals as hydroxides.

3.7. Introduction to Lead

Lead is a ubiquitous pollutant in the ecosystem. Lead mining and smelting works and automobile exhausts, lead batteries, lead paints, manufacture industries of lead base alloys and automobile exhausts are the main causes of lead pollutants. The leaded dust settles on the plants and food stuff intended for human and animal consumption. Lead causes poisoning of living

organism. It is a cumulative poison causing loss of appetite, constipation, abdominal pain, mental retardation, a blue line at the junction of teeth and gums and finally nervous disorders and brain damage. It also has effects on the haem-bio synthesis, blood pressure on kidney function.

Spectrophotometric methods for the determination of lead (II) are very limited. Even for an experienced worker in the field, it is mandatory to know different spectrophotometric methods for the determination of an element, especially lead. Hence it is considered worthwhile to review spectrophotometric methods for the determination of lead in aqueous medium.

Though a number of reagents are available for the spectrophotometric determination of lead, the well-known dithiozone method, which involves extraction, is often recommended because of its relative high sensitivity. Dithiozone method, however, has some well-known drawbacks. The optimum pH range for extraction of lead is from 7-10 the lead dithiozone complex solution are unstable in sun light several modifications of the dithiozone methods for the determination of lead have been made and applied for its petroleum products, the food stuffs, urine, biological substances, plant materials, and water.

3.8 Introduction to Vanadium

Vanadium is a very rare metal and is available as silvery white solid. In soils Vanadium is present in varying concentrations and is utilized by the living organisms for cell growth. In Plants it is useful for Nitrogen assimilation.

By its oxidation state vanadium may become reasonably toxic and also leads to nervous depression, coughing, vomiting, diarrhoea, anaemia and increased risk of lung cancer.

Professional exposure to airborne vanadium causes bronchitis and bronchopneumonia as well as irritation of eyes and skin. Vanadium complexes are proven to be promising candidates in diabetes management and also in the inhibition of proliferating human tumors but are hazardous when present at high concentrations.

Vanadium is employed in the nuclear reactors due to its inherent ability to withstand the high temperatures as well due to its low neutron adsorption abilities. In the present era Vanadium also finds its applicability in the manufacturing of batteries. In combination with aluminum, vanadium is utilized in jet engines, gears and other critical components

3.9 Introduction to Copper

Copper is an important element in industry and biological chemistry. The human adult requirement is 2 mg per day, and adult human body contains 100-150mg of copper. The greatest concentration exists in liver and bones.

Blood contains a number of copper proteins and copper is known to be necessary for the synthesis of hemoglobin, although there is no copper in it. It is known that some drugs have increased activity when administered as metal complexes and number of metal chelates inhibit tumor growth. It is directly responsible for maintenance of myelin within the nervous system. Copper is essential for mammal, they become hazardous when present in excess. As a result there are metabolic disorders connected with both deficiencies and excess amount of the metal ion. An excessive accumulation of copper in liver, kidney and brain, leads to liver and kidney failure and various neurological abnormalities causing Wilson's disease.

The main source of copper come from mining and metallurgical operations contribute to contamination of aquatic environments. Copper salts are used as algicides and fungicides. Bordeaux mixture , a formulation of copper sulphate and calcium carbonate is still used as fungicide. Copper is used in anti fouling paints for protection of ships. Copper is toxic to many osmotic plants at low concentration. The toxicity of copper to aquatic organism varies with the chemical species present in water and other environmental parameters like temperature, pH, turbidity, Hardness etc., main dissolved species of Cu in aquatic environment are $\text{Cu}(\text{OH})^+$, Cu^{++} and CuCO_3 . The maximum permissible limit of copper in drinking water is 0.05mg/lit.