CHAPTER II

SPECTROPHOTOMETRIC DETERMINATION OF COPPER(II) WITH 2,6-DIACETYLPYRIDINE BIS-4-PHENYL-3-THIOSEMI-CARBAZONE(2,6-DAPBPTSC)

2.1. Introduction:

Copper is both micro-nutrient as well as toxic element for living beings, depending up on the concentration level[1]. Inhalation of dusts, fumes and mists of Cu-salts results in congestion of nasal mucous membranes. Ulceration with perforation of the nasal septum on occasion and some times, pharyngeal congestion[2]. It is also a gastrointestinal tract irritant[3]. The study of copper in food items is of great concern, since it plays a definitive role in the intrinsic mechanisms regulating vital biological processes. Copper is also an essential element and its deficiency causes the ischemic heart disease, anemia, abnormal wool growth and bone disorders[4, 5]. Interest in copper complexes as anti-inflammatory drugs and anti-arthritis is evidenced by the large number of reviews and symposia proceedings published in recent years[6].

Cu(II) is used to control fungal diseases in vineyard plants in France, South Africa and orange orchard in Thailand[7]. High concentrations of copper were detected in some aquatic ecosystems collecting vineyard runoff water[8]. Copper is also a widely used metal industrially[9]. In addition to this, it is an important pollutant in the environment resulting from the industrial discharge in the form of particulate (or) soluble copper waste from electroplating, chemical and textile industries. In view of this, separation and determination of copper from associated elements is indispensable.

2.2. Review of known methods

As per the review of literature some standard, sensitive and/or selective reagents for the spectrophotometric and extractive spectrophotometric determination of copper(II) are discussed here.

Bizil-α-monoximethiosemicarbazone(BMOT) and 1,2 propanedione-2-oxime-thiosemicarbazone(PPDOT)[10] are used for the spectrophotometric determination of copper(II). The copper complexes have the maximum absorbances at 355 nm for BMOT and 465 nm for PPDOT. The complexes show molar absorptivity values as 2.2×10^4 lit $mol^{-1}cm^{-1}$ for Cu(II)-BMOT and 0.58×10^4 lit $mol^{-1}cm^{-1}$ for Cu(II)-PPDOT. The method is successfully applied for the determination of copper(II) in edible oils and seeds.

Diphenylcarbazide[11] is a highly sensitive reagent for the extractive spectrophotometric determination of copper(II). The complex formed by this reagent is easily extractable into benzene. The complex shows the molar absorptivity as 8.0×10^4 lit $mol^{-1}cm^{-1}$ at 545 nm.

2-Hydroxy-4-methoxybenzophenone oxime[12] is used for the spectrophotometric determination of copper(II) at 400 nm. The molar absorptivity of the complex is found to be 0.07×10^4 lit $mol^{-1}cm^{-1}$ and the Beer's law is obeyed up to 31.75 ppm of copper(II).

Thipyapang et al[13] have reported meso-hexamethyl propylene amine oxime (meso-HMPAO) for the determination of copper(II). The molar absorptivity of the complex is found to be 0.338×10^4 lit $mol^{-1}cm^{-1}$ at 497 nm.

Carbonyl derivatives like oximes, hydrazones, thio-and phenylthio-semicarbazones were also used for the spectrophotometric as well as extractive spectrophotometric determi- nation of copper(II). Some of the sensitive and/or selective methods are discussed below.

α-Benzoinoxime[14] was also used for the spectrophotometric determination of copper(II) in the presence of tartrate and at pH 12.5. The complex shows a maximum absorbance at 440 nm.

3-{2-[2-(2-Hydroxyimino-1-methyl-propylideneamino)-ethylamino]-ethylimino}-butan-2-one oxime[15] reacts with copper(II) to form complex. The molar absorptivity of the complex is found to be 0.16×10^4 lit $mol^{-1}cm^{-1}$ at 570 nm. Moreover, this method is easy to perform the determination of copper in pharmaceutical and biological samples.

Benerjea et al[16] have reported Methyl-2-pyridylketoxime for the simultaneous spectrophotometric determination of iron(II) and copper(II). The copper complex has the maximum absorbance at 410 nm. The complex obeys the Beer's law in the range of 0.5-3.7 ppm of copper(II).

Other oximes used for the spectrophotometric and extractive spectrophotometric determination of copper(II) are acetylacetonedioxime[17], α-benzildioxime[18], 2,6-diacetoxime[19], dimethylglyoxime[20], furfuloxime[21], 2-hydroxyacetonaphthoneoxime[22], 6-methylpyridyl-2-aldoxime[23], n-amyl-2-pyridylketoxime[24], N-(2,6-dimethylphenyl)aminoglyoxime[25], and phenyl-2-(6-methylpyridyl)ketoxime[26].

Katsunori et al[27] have reported bis(D-glucose)oxalyldihydrazone (BGOH) for the spectrophotometric determination of copper(II) at 608 nm. The molar absorptivity of the complex is found to be 1.55×10^4 lit $mol^{-1}cm^{-1}$.

Chandrasekhar et al[28] have reported diacetylmonoximeisonicotionylhydrazone (DMIH) for the determination of copper(II) and nickel(II). The molar absorptivity of the copper complex is found to be 1.12×10^4 lit $mol^{-1}cm^{-1}$, and the Beer's law is obeyed in the range 0.25-2.54 ppm of copper(II).

3-hydroxyacetanilide with 3-methyl-2-benzothiazolinone hydrazone[29] is also used for the spectrophotometric determination of copper(II). The complex shows a maximum absorbance at 530 nm. The molar absorptivity of the complex is found to be 2.5×10^5 lit $mol^{-1}cm^{-1}$ and the Beer's law is obeyed in the range 0.8-1.6 ppm of copper(II).

Di-2-pyridylketonebenzoylhydrazone[30] is a highly sensitive reagent for the spectrophotometric determination of copper(II) at 370 nm. The complex shows the molar absorptivity as 3.92×10^4 lit $mol^{-1}cm^{-1}$. This method is successfully applied for the determination of copper(II) in water and alloy samples.

However, not enough work has been reported on the use of thiosemicarbazones as extractants for copper(II), even though a number of thiosemicarbazones have been reported for the spectrophotometric and extractive spectrophotometric determination of copper(II)[31].

2-Acetylthiophenethiosemicarbazone has been used for the determination of copper(II) by Siyaji Rao et al[32]. The molar absorptivity of the complex is found to be 1.8×10^4 lit $mol^{-1}cm^{-1}$ at 370 nm in the pH range of 5.0-7.0. The reagent forms 1:2(M:L) complex with

the metal ion. The method has been successfully applied for the determination of copper(II) in alloys, edible oils and seeds.

Reddy et al[33] have reported Benzildithiosemicarbazone for the extractive spectrophotometric determination of copper(II). The complex has maximum absorbance at 380 nm in the pH range of 3.5-4.5. The reagent forms 1:1 (M:L) complex with metal ion. The molar absorptivity of the complex is found to be 1.63×10^4 lit $mol^{-1}cm^{-1}$ and Beer's law is obeyed in the range 0.5-4.0 ppm of copper(II).

Benzyloxybenzaldehydethiosemicarbazone(BBTSC)[34] is used for the extractive spectrophotometric determination of copper(II). The complex is extracted into n-butanol at 370 nm in the pH 5.0.The molar absorptivity of the complex is found to be 1.5×10^4 lit mol^{-1} cm^{-1}. The complex obeys the Beer's law in the range 0.6-5.2 ppm of copper(II).

Babu et al [35] have reported 3-hydroxybenzaldehydethiosemicarbazone for the simultaneous spectrophotometric determination of copper(II) and palladium(II). The copper complex has the maximum absorbance at 420 nm. The molar absorptivity of the complex is found to be 8.75×10^4 lit $mol^{-1}cm^{-1}$.

Reddy et al[36] have reported N-ethyl-3-carbazolecarboxaldehyde-3-thiosemicarbazone (ECCT) for an extractive spectrophotometric determination of copper(II). The ECCT forms a greenish-yellow coloured 1:1(M:L) complex with copper(II) at pH 3.0, which is well extractable into n-butanol and shows maximum absorbance at 380 nm. The molar absorptivity and Sandell's sensitivity of the complex are 2.243×10^4 lit mol^{-1} cm^{-1} and 0.00283 μg cm^{-2}, respectively. The complex obeys Beer's law in the range 0.4-3.6 ppm of copper(II).

Acetophenone-*p*-chlorophenylthiosemicarbazone has been used for the determination of copper(II) by Ghazy et al[37]. The molar absorptivity of the complex is found to be 5.5×10^3 lit $mol^{-1}cm^{-1}$ at 600 nm in the pH range of 4.0-9.0. The reagent forms 1:2 (M: L) complex with the metal ion. The complex obeys Beer's law in the range 2.25-6.35 ppm of copper(II).

Takeshi and Akira et al[38] have reported 3,5-dibromosalicylaldehyde-4-phenyl-3-thiosemicarbazone(DBPS) for the extractive spectrophotometric determination of copper(II). The complex has maximum absorbance at 400 nm in the pH range of 2.8-7.5. The complex is easily extractable into chloroform. The molar absorptivity of the complex is 2.06×10^4 lit $mol^{-1}cm^{-1}$.

Phenanthraquinone monophenylthiosemicarbazone has been used for the spectrophotometric determination of copper(II) by Khalifa et al[39]. The complex has maximum absorbance at 545 nm in the pH range of 3.0-6.5. The molar absorptivity of the complex is found to be 2.3×10^4 lit $mol^{-1}cm^{-1}$. The complex obeys Beer's law in the range 3.0-40.0 ppm of copper(II).

Pyridoxal-4-phenyl-3-thiosemicarbazone(PPT) has been used for the extractive spectrophotometric determination of copper(II) by Sarma et al[40]. The reagent forms 1:1(M:L) complex with copper(II) at 440 nm in the pH 4.5, which is well extractable into n-butanol. The molar absorptivity of the complex is found to be 2.16×10^4 lit $mol^{-1}cm^{-1}$. The complex obeys Beer's law in the range 0.2-5.0 ppm of copper(II).

Kazuyo and Kiyoharu et al[41] have reported 2-pyridinealdehyde-4,4'-diphenylsemicarbazone for the spectrophotometric determination of copper(II). The complex has maximum absorbance at 380 nm in the pH range of 4.0-6.5. The molar absorptivity of the complex is found to be 1.84×10^4 lit $mol^{-1}cm^{-1}$. The complex obeys the Beer's law in the range 0.14-2.88 ppm of copper(II).

A thorough literary survey has revealed that a number of thiosemicarbazones are available for the spectrophotometric and extractive spectrophotometric determination of copper(II). But many of the thiosemicarbazones are either less sensitive or non-selective. Hence, a new reagent namely 2,6-diacetylpyridinebis-4-phenyl-3-thiosemicarbazone (2,6-DAPBPTSC) is synthesized for the development of a simple, selective and sensitive spectrophotometric method in the determination of copper(II). In the present investigation, the researcher has studied the reactivity of 2,6-DAPBPTSC with copper(II) and the results are reported in terms of its maximum absorbance, pH, molar absorptivity and stoichiometry. which provides the basis for judging the potential utility of 2,6-

DAPBPTSC as an analytical reagent for copper(II). Finally, the developed method is successfully applied for the determination of copper(II) in food and water samples.

2.3. Results and Discussion

2,6-Diacetylpyridinebis-4-phenyl-3-thiosemicarbazone (2,6-DAPBPTSC) forms 1:1(*M: L*) complex with copper(II), in sodium acetate-acetic acid (pH 3.0) buffer. The yellowish orange Cu(II)-2,6-DAPBPTSC complex has a maximum absorbance at 370 nm and is stable for 48 hours. The conditions for effective determination are established after studying the effects of various factors such as pH, reagent concentration, metal ion concentration and influence of diverse ions, in order to develop a sensitive, selective and rapid spectrophotometric method for the determination of copper(II) at micro gram levels.

2.3.1. Absorption spectra of reagent and Cu(II)- 2,6-DAPBPTSC complex

The absorption spectrum of Cu(II)-2,6-DAPBPTSC complex was recorded against the reagent blank. Similarly the absorption spectrum of the reagent (2,6-DAPBPTSC) was recorded against the solvent blank. The absorption spectra of both the complex and reagent are shown in Fig. 2.1. From the absorption spectra it is clear that the complex and reagent have shown maximum absorptions at 370 nm and 360 nm, respectively. The reagent has minimum absorbance at the maximum absorbance of the complex, and the reagent does not interfere with the determination of copper(II). Hence, further absorbance measurements of the complex were recorded at 370 nm.

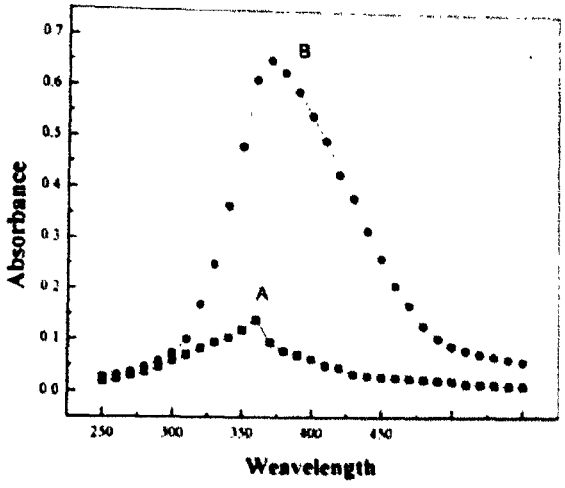

Fig.2.1 (A) Absorption spectrum of 2,6-DAPBPTSC; (B) Absorption spectrum of Cu(II)-2,6-DAPBPTSC complex: Cu(II)= 1.0 mL of 1.0×10^{-4} M; 2,6-DAPBPTSC= 1.0 mL of 1.0×10^{-3} M and pH= 1.0 mL of pH 3.0.

2.3.2. Effect of pH

To arrive at the optimum pH required for maximum colour development, the influence of pH on the colour intensity was studied by using different buffers in the pH range 1.0 to 6.5. The absorbance of the Cu(II)-2,6-DAPBPTSC complex increases as the pH increases from 1.0 to 2.5 and remains constant in the pH range 2.5-3.5. However, it is decreased beyond 3.5. The absorbance of all these solutions was measured at 370 nm against their corresponding reagent blanks and the values were noted in Table 2.1. Hence, sodium acetate-acetic acid buffer was used for further studies, considering 3.0 as the optimum pH. The plot is shown in Fig.2.2.

Table 2.1 Effect of pH on Cu(II)- 2,6-DAPBPTSC complex

S.No.	pH	Absorbance
1	1.0	0.210
2	1.5	0.380
3	2.0	0.525
4	2.5	0.648
5	3.0	0.656
6	3.5	0.635
7	4.0	0.592
8	4.5	0.530
9	5.0	0.453
10	5.5	0.376
11	6.0	0.270
12	6.5	0.160

Cu(II) = 1.0 mL of 1.0×10^{-4} M; 2,6-DAPBPTSC = 1.0 mL of 1.0×10^{-3} M and λ_{max} = 370 nm.

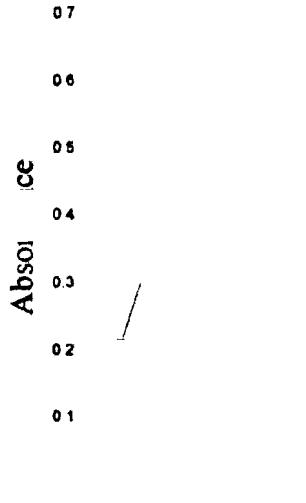

Fig. 2.2 Effect of pH on Cu(II)- 2,6-DAPBPTSC complex: Cu(II) = 1.0 mL of 1.0×10^{-4} M; 2,6-DAPBPTSC = 1.0 mL of 1.0×10^{-3} M and λ_{max} = 370 nm.

2.3.3. Effect of reagent concentration

The absorbances of the complex solutions obtained from the solutions of pH 3.0, containing constant amount of copper(II) and varying amounts of reagent were measured at 370 nm by adopting the following procedure. Different aliquots containing 1.0 mL of 1.0×10^{-4} M

copper(II) solution, 2.0 mL of pH 3.0 buffer solution and the reagent solution containing different concentrations ranging from 1.0×10^{-4} to 20.0×10^{-4} M were taken into a set of 10.0 mL standard flasks. These coloured solutions were made up to the mark with double distilled water. The absorbences of these solutions were measured at 370 nm against their corresponding blanks and the values are noted in Table 2.2. The results clearly indicate that a fifteen fold molar (15.0×10^{-4} M) excess of reagent to that of the metal ion is sufficient for maximum colour development of the Cu(II)-2,6-DAPBPTSC complex. Hence a fifteen fold molar excess of the reagent was maintained for maximum colour formation. The observed values are presented in the form of a plot as shown in Fig.2.3.

Table 2.2 Effect of reagent concentration on Cu(II)- 2,6-DAPBPTSC complex

Concentration of reagent, $\times10^{-4}$ M	No. of folds of reagent concentration with respect to metal	Absorbance
1.0	1	0.102
2.0	2	0.160
3.0	3	0.233
4.0	4	0.286
5.0	5	0.348
6.0	6	0.410
7.0	7	0.468
8.0	8	0.530
9.0	9	0.592
10.0	10	0.657
11.0	11	0.715
12.0	12	0.780
13.0	13	0.842
14.0	14	0.860
15.0	15	0.872
16.0	16	0.881
17.0	17	0.892
18.0	18	0.902
19.0	19	0.910
20.0	20	0.914

Cu(II)= 1.0 mL of 1.0×10^{-4} M, 2,6-DAPBPTSC = 1.0 mL of 1.0×10^{-4} to 20.0×10^{-4} M, pH=3.0 and λ_{max} = 370 nm.

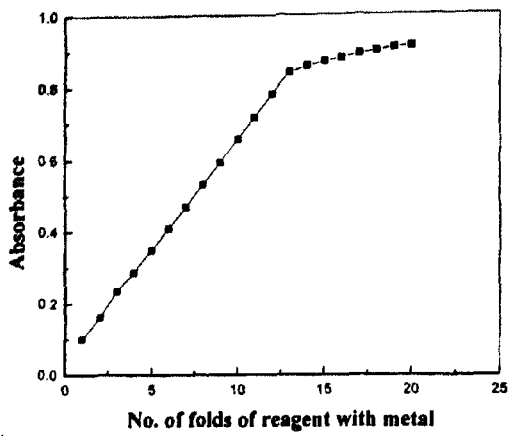

Fig. 2.3 Effect of reagent concentration on Cu(II)- 2,6-DAPBPTSC complex: Cu(II)=1.0 mL of 1.0×10^{-4} M; 2,6-DAPBPTSC = 1.0 mL of 1.0×10^{-4} to 20.0×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

2.3.4. Applicability of Beer's law

Various aliquots containing different amounts of copper(II)(1.0×10^{-5} to 20.0×10^{-5} M), 2.0 mL of pH 3.0 buffer and 1.0 mL of 2,6-DAPBPTSC (15.0×10^{-4} M) were taken into 10.0 mL standard flasks and made up to the mark with double distilled water. The absorbances of all the solutions were recorded at 370 nm, against their corresponding reagent blanks. The obtained results are noted in Table 2.3. A graph plotted between the amount of copper(II) and its absorbance is shown in Fig.2.4. It can be observed from the graph that a linear plot passing through the origin obeys Beer's law in the concentration range 0.63-6.30 ppm of copper(II). The molar absorptivity of the complex was calculated and noted as 0.847×10^{4} L $mol^{-1}cm^{-1}$ and the Sandell's sensitivity of the complex was 0.0075 $\mu g\ cm^{-2}$. The correlation coefficient value of the Cu(II)-2,6-DAPBPTSC complex, with an independent variable as concentration in $\mu g\ mL^{-1}$ and a dependent variable as absorbance, was found to be 0.942. This indicates a satisfactory linearity between the two variables.

Table 2.3 Applicability of Beer's law to Cu(II)- 2,6-DAPBPTSC complex

Concentration of metal, $\times 10^{-5}$M	Amount of metal, ppm	Absorbance
1	0.63	0.095
2	1.26	0.180
3	1.89	0.270
4	2.52	0.355
5	3.15	0.448
6	3.78	0.536
7	4.41	0.612
8	5.04	0.700
9	5.67	0.785
10	6.30	0.875
11	6.93	0.910
12	7.56	0.950
13	8.19	0.990
14	8.82	1.012
15	9.45	1.025
16	10.08	1.032
17	10.71	1.037
18	11.34	1.041
19	11.97	1.044
20	12.60	1.046

Cu(II)= 1.0 mL of 1.0×10^{-5} to 20.0×10^{-5} M; 2,6-DAPBPTSC= 1.0 mL of 15×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

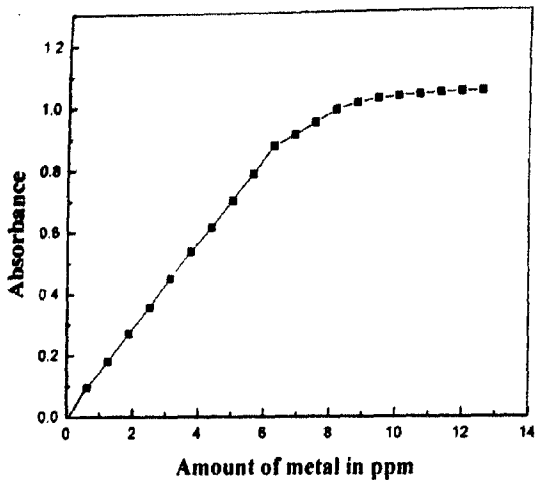

Fig. 2.4 Applicability of Beer's law to Cu(II)- 2,6-DAPBPTSC complex: Cu(II)= 1.0 mL of 1.0×10^{-5} to 20.0×10^{-5} M; 2,6-DAPBPTSC=1.0 mL of 15×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

2.3.5. Ringbom's plot for Cu(II)- 2,6-DAPBPTSC complex

Ringbom's plot is the standard adopted to know the optimum range of the concentration for a system, which emaciates Beer's law. The values are noted in Table 2.4. The plot is drawn between log C of Cu(II) and (1-T) [where T is the transmittance]. The plot has a sigmoid shape with a linear segment at intermediate concentration values ranging from 3.10-3.87 μg L^{-1}, which indicates that copper(II) is precisely determined in the range 3.10-3.75 μg L^{-1}. The slope of the plot obtained from Fig.2.5 is 0.635. Based on this value at 1% photometric error, the ratio between the relative error in concentration and the photometric error is 3.626. Hence, the relative error in concentration is 0.0362.

Table 2.4 Ringbom's plot for Cu(II)- 2,6-DAPBPTSC complex

Amount of copper(II), $\mu g\ L^{-1}$, C	Log C	Absorbance	Transmittance	(1-T)
630	2.799	0.095	0.9.9	0.091
1260	3.100	0.180	0.835	0.165
1890	3.276	0.270	0.763	0.237
2520	3.401	0.355	0.701	0.299
3150	3.498	0.448	0.638	0.362
3780	3.577	0.536	0.585	0.415
4410	3.644	0.612	0.542	0.458
5040	3.702	0.700	0.496	0.504
5670	3.753	0.785	0.456	0.544
6300	3.799	0.875	0.416	0.584
6930	3.840	0.910	0.402	0.598
7560	3.878	0.950	0.386	0.614
8190	3.913	0.990	0.371	0.629
8820	3.945	1.012	0.363	0.637
9450	3.975	1.025	0.358	0.642
10080	4.003	1.032	0.356	0.644
10710	4.029	1.037	0.354	0.646
11340	4.054	1.041	0.353	0.647
11970	4.078	1.044	0.352	0.648
12600	4.100	1.046	0.351	0.649

Cu(II)= 630-12600 $\mu g\ L^{-1}$; 2,6-DAPBPTSC=1.0 mL of 1.5×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

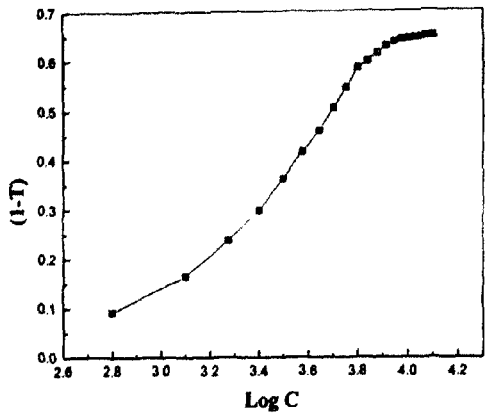

Fig. 2.5 Ringbom's plot for Cu(II)- 2,6-DAPBPTSC complex: Cu(II)= 630-12600 μg L^{-1}; 2,6-DAPBPTSC=1.0 mL of 15×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

2.3.6. Precision, accuracy and detection limit of the method

To assess the precision and accuracy of the method, determinations were carried out for a set of five measurements, with different concentrations of Cu(II), under optimum conditions. Calculations have revealed that the standard deviation of method was not more than 0.0012 and the relative standard deviation was less than 0.777%. From the results it is evident that the method is precise, besides being accurate. The detection limit, C_{min} is determined as the amount of Cu(II) corresponding to three times the standard deviation of the blank values and a value of 0.0056 μg L^{-1} is obtained.

2.3.7. Determination of the composition of Cu(II)- 2,6-DAPBPTSC complex

Job's method of continuous variation, molar ratio method and Asmus' method were employed to elucidate the composition of the complex.

2.3.7.1. Job's method of continuous variation

Equimolar solutions of copper(II) and 2, 6-DAPBPTSC (15×10^{-4} M) were used to determine the metal to ligand ratio by job's method of continuous variation. The absorbance values were recorded at 370 nm against the reagent blank. The valves are reported in Table 2.5 and the corresponding graph drawn between absorbance and

V_M / V_L+V_M (where V_L and V_M are the volumes of the reagent and the metal, respectively) is shown in Fig.2.6. From the graph, it is observed that one mole of copper(II) reacts with one mole of ligand and hence, the composition of metal to ligand complex is 1:1.

Table 2.5 Job's method of continuous variation

Volume of Cu(II), V_M, mL	Volume of 2,6-DAPBPTSC, V_L, mL	V_M/V_M+V_L	Absorbance
0.2	1.8	0.1	0.359
0.4	1.6	0.2	0.600
0.6	1.4	0.3	0.785
0.8	1.2	0.4	0.932
1.0	1.0	0.5	1.030
1.2	0.8	0.6	0.912
1.4	0.6	0.7	0.768
1.6	0.4	0.8	0.695
1.8	0.2	0.9	0.413

[Cu(II)= 2,6-DAPBPTSC]= 1.5×10^{-4} M; pH=3.0 and λ_{max} = 370 nm

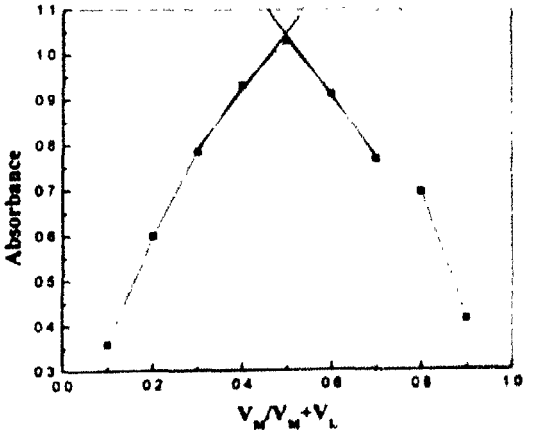

Fig.2.6 Job's method of continuous variation: [Cu(II)]= [2, 6-DAPBPTSC]= 1.5×10^{-4} M; pH=3.0 and λ_{max} = 370 nm.

2.3.7.2. Molar ratio method

In molar ratio method, different aliquots containing 1.0 mL of 1.0×10^{-3} M copper(II), 2.0 mL of sodium acetate-acetic acid buffer (pH 3.0) and varying concentrations (0.25×10^{-3} to 2.0×10^{-3} M) of 2,6-DAPBPTSC were used to determine the metal ligand ratio. The absorbances of the solutions were recorded at 370 nm against their respective reagent blanks. The values are noted in Table 2.6. A plot (Fig.2.7.) is drawn between the absorbance and the concentration of the reagent. From the obtained curve, it is confirmed that one mole of copper(II) complexes with one mole of 2,6-DAPBPTSC.

Table 2.6 Molar ratio method

No. of moles of 2,6-DAPBPTSC per one mole of Cu(II)	Absorbance
0.25	0.326
0.50	0.540
0.75	0.749
1.00	0.950
1.25	1.025
1.50	1.102
1.75	1.168
2.00	1.235

Cu(II)= 1.0 mL of 1.0×10^{-3} M; 2,6-DAPBPTSC= 1.0 mL of 0.25×10^{-3} to 2.0×10^{-3} M; pH=3.0 and λ_{max} = 370 nm.

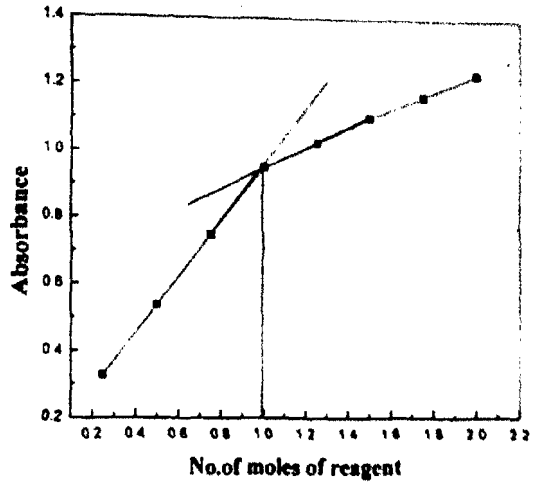

Fig.2.7 Molar ratio method: Cu(II):1.0 mL of 1.0×10^{-3} M; 2,6-DAPBPTSC=1.0 mL of 0.25×10^{-3} to 2.0×10^{-3} M; pH:3.0 and λ_{max} ~ 370 nm.

2.3.7.3. Asmus' method

For Asmus' method the data obtained from molar ratio method was used. 1/m values (where 'm' is extinction modulus) were calculated by dividing the optical density with the cell width, along with 1/V, $1/V^2$ and $1/V^3$ and are given in Table 2.7. The plots between 1/m and 1/V, $1/V^2$ and $1/V^3$ are indicated in Fig.2.8. Among the plots between 1/m and 1/V, $1/V^2$ and $1/V^3$, only the plot between 1/m and 1/V is linear indicating that the composition of the complex is 1:1 (M: L).

Table 2.7 Asmus' method

Volume of 2,6-DAPBPTSC, (V)	Absorbance (m)	1/m	1/V	$1/V^2$	$1/V^3$
0.25	0.326	3.06	4.00	16.00	64.00
0.50	0.540	1.85	2.00	4.00	8.00
0.75	0.749	1.33	1.33	1.77	2.37
1.00	0.950	1.05	1.00	1.00	1.00
1.25	1.025	0.97	0.80	0.64	0.51
1.50	1.102	0.90	0.66	0.44	0.29
1.75	1.168	0.85	0.57	0.32	0.18
2.00	1.235	0.80	0.50	0.25	0.12

Cu(II)= 1.0 mL of 1.0×10^{-3} M; 2,6-DAPBPTSC= 1.0 mL of 0.25×10^{-3} to 2.0×10^{-3} M; pH=3.0 and λ_{max} = 370 nm.

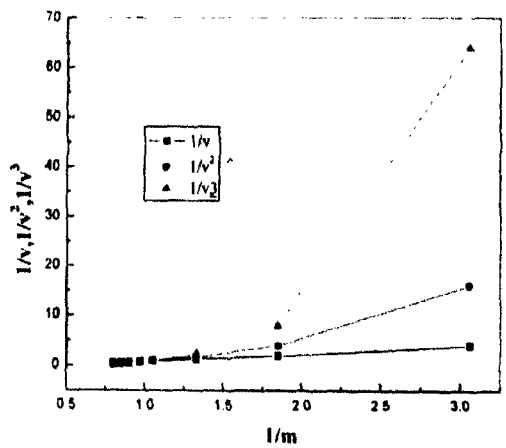

Fig.2.8 Asmus' method: Cu(II)= 1.0 mL of 1.0×10^{-3} M; 2,6-DAPBPTSC=1.0 mL of 0.25×10^{-3} to 2.0×10^{-3} M; pH=3.0 and λ_{max} =370nm.

2.3.8. Calculation of instability constant of Cu(II)-2,6-DAPBPTSC complex

The instability constant of Cu(II)-2,6-DAPBPTSC complex was calculated by using Asmus' method. The absorbance values were obtained at 370 nm for the solutions containing fixed volumes of copper(II) (1.0 mL of 1.0×10^{-3} M) and 2.0 mL of pH 3.0 buffer with different known volumes of 0.25-2.0 mL of 1.0×10^{-3} M of 2,6-DAPBPTSC. The instability constant of Cu(II)-2,6-DAPBPTSC complex was calculated to be 1.415×10^{-4} at room temperature.

2.3.9. Effect of foreign ions on the extraction of Cu(II)- 2,6-DAPBPTSC complex

Interference of a number of cations and anions was studied in the color absorbance of the Cu(II)-2,6-DAPBPTSC complex. A change in absorbance of ± 0.0025 was taken as the tolerance limit for interference. Cations like Ca(II), Mg(II), Mn(II), Bi(III), Pb(II) and anions like bromide, iodide, chloride, nitrate, sulphate, thiosulphate, citrate, acetate and tartate do not interfere, even when present upto 5000 $\mu g\ mL^{-1}$. Interference due to Al(III), Cr(III), Ag(I) and Sb(II) can be tolerated upto 2500 $\mu g\ mL^{-1}$. Determination of copper(II) was not possible in the presence of Co(II), Ni(II), Fe(II), Fe(III), Zn(II), Pd(II), Cd(II), Mo(VI), Se(IV), thiocyanate, oxalate and EDTA due to their interference, even when present in trace amounts. However, Interference of Fe(II) and Fe(III) was suppressed with 1.0 mL of 0.2 percent fluoride as a masking agent and of Co(II), Ni(II), Zn(II), Pd(II), Mo(VI), Se(IV) and Cd(II) is suppressed, by adding 1.0 mL of 0.2 percent citrate solution. Increasing the amounts of their corresponding masking agents proportionately can mask higher amounts of interfering ions. These results are given in Table 2.8.

Table 2.8 Effect of foreign ions on extraction of Cu(II)- 2,6-DAPBPTSC complex

Foreign ions	Tolerance limit, μg mL^{-1}	Foreign ions	Tolerance limit, μg mL^{-1}
Ca(II)	5000	Cd(II)	1000
Mg(II)	5000	Mo(VI)	1000
Mn(II)	5000	Se(IV)	1000
Bi(III)	5000	Bromide	5000
Pb(II)	5000	Chloride	5000
Al(III)	2500	Iodide	5000
Cr(III)	2500	Nitrate	5000
Ag(I)	2500	Citrate	5000
Sb(II)	2500	Sulphate	5000
Co(II)	1000	Thiosulphate	5000
Ni(II)	1000	Acetate	5000
Fe(II)	1000	Tartrate	5000
Fe(III)	1000		
Zn(II)	1000		
Pd(II)	1000		

2.4. Applications of the developed method

The developed sensitive spectrophotometric method for copper(II) was successfully applied for its determination in food and water samples.

2.4.1. Determination of Cu(II) in food samples

The foods like kakara (Momordica charantia), leafy vegetables and milk samples were analyzed for copper(II) using the proposed method. The content of copper(II) present in the solution was determined by using a calibrated plot and the results obtained were conformed by direct atomic absorption spectrophotometer. The data obtained in the analysis of medicinal leaves and leafy vegetables and milk samples are given in Table 2.9.

Table 2.9 Determination of Cu(II) in food samples

Samples[b]	Amount of copper(II) found[a]					
	AAS method	Present method	Present method			
			SD	RSD (%)	F-Test	T-Test
Kakara(Momordica charantia) Sample location						
Srikalahasthi	25.3	25.2	0.205	0.820	1.866	1.542
Madanapalli	18.5	18.3	0.195	1.071	1.397	1.916
Tirumala	21.2	20.9	0.175	0.829	1.741	1.806
Srinivasamanga puram	24.6	24.3	0.172	0.702	2.95	1.581
Pakala	17.3	17.0	0.225	1.30	2.4	3.258
Mangalam	22.5	22.4	0.210	0.948	1.621	3.832
Nagari	22.2	21.9	0.198	0.904	2.969	5.475
Leafy vegetables[b] Name of the samples						
Chama(Colocasia esculenta)	14.28	14.15	0.20	1.428	1.562	1.185
Avalu(Brassica nigra)	16.14	16.10	0.245	1.529	1.185	0.878
Pala teege (Ichnocarpus frutescens)	23.25	22.95	0.250	1.101	1.644	1.702
Drumstick(Moringa oleifera)	20.50	20.32	0.192	0.955	1.491	1.987
Thotakura(Amaranthus gangeticus)	15.45	14.90	0.197	1.362	1.471	1.454
Chilakamukkaku (Impotiens balsamina)	34.80	34.0	0.232	0.685	1.219	1.490
Milk samples[c]						
Raw milk	4.00	3.98	0.022	0.600	1.234	1.054
Choco milk	3.50	3.48	0.026	0.764	1.145	1.288
Cow	5.00	4.92	0.148	0.314	1.056	1.119
Dairy	4.25	4.20	0.017	0.442	1.408	1.739

[a] Average of five determinations, [b] Concentration in µg/g

2.4.2. Determination of Cu(II)in Water samples

The present method was also applied for the determination of copper(II)in water samples. The concentration of copper(II) was determined by adopting the procedure

described in general procedure. The data obtained in the analysis of water samples were given in Table 2.10.

Table 2.10 Determination of Cu(II) in water samples

Amount of copper(II) found [a]						
Name of the area [b]	**AAS method**	**Present method**	**Present method**			
			SD	**RSD (%)**	**F-Test**	**T-Test**
River water (Penna)						
Sample 1	1.02	1.00	0.014	1.41	1.13	2.25
Sample 2	0.98	0.97	0.015	1.54	1.33	3.63
Sample 3	1.04	1.02	0.017	1.73	1.20	3.99
Lake water(Kollate)						
Sample 1	0.92	0.90	0.015	1.68	1.32	3.16
Sample 2	0.95	0.94	0.158	1.64	1.49	1.99
Sample 3	0.97	0.95	0.019	2.03	1.65	3.16
Polluted water						
Sample 1	0.83	0.82	0.014	1.71	1.37	3.38
Sample 2	0.87	0.87	0.016	1.83	1.20	3.16
Sample 3	0.85	0.84	0.015	1.78	1.32	5.26

[a] Average of five determinations, [b] Concentration in µg/mL.

2.5. Conclusions

The present investigations had proved that 2,6-DAPBPTSC is a promising complexing agent for Cu(II) and its subsequent determination by spectrophotometry is rapid and precise. This method is equally sensitivity when compared to other existing spectrophotometric determination methods. The selectivity of this method is improved by using masking agents for Co(II), Ni(II), Fe(II), Fe(III), Zn(II), Pd(II), Cd(II), Mo(VI) and Se(IV). It has been successfully applied for the determination of copper(II) in food, leafy vegetables, milk samples and water samples.

REFERENCES

1. Sharma, B. K., *Environmental chemistry*, Goel publishing House, Meerut, India, 1997.

2. Raymond, W. M. and Donahue, R. L., *Soils in our Environment*, 7th ed. Prentice-Hall of India, New Delhi, 1997.

3. Judith, T. Z., Peter, T. and Thomas, T., *Immunotoxicology of Environment occupational metals, School of medicine*, New York, 1998.

4. Underwood, E. J., *Trace Elements in human and animal nutrition 4th Ed*, New York, Academic Press, 1979.

5. Eichhorn, G. L., *Inorganic biochemistry*. NewYork, Elsevier Scientific Publishing Company, 1975.

6. Sorenson, R.J.R., *Prog. Med. Chem.*, **26**, 437, 1989.

7. Schlotfeldt, H. J., *Current practices of Chemotherapy in fish culture*. In: Michel, C., Alderman, D.J. editors. *Chemotherapy in Aquaculture*: From Theory to Reality, Paris, OIE, 25, 1992.

8. GERBE, A., *programme de recherche Toxicologie, ecotoxicologie des pesticides et des metaux lourds*. Reims, Francce: Europol' Agro universite de Reims Champagne- Ardenne, 39, 1996.

9. Purnima, L. M. and Vijay, M. S., Ion- pair extraction and determination of copper(II) and zinc(II) in environmental and pharmaceutical samples. *Analyst*, **116**, 1081, 1991.

10. Reddy, K.H. and Prasad, N.B.L., *Indian J. Chem.*, **43**, 111, 2004.

11. Turkington, R.W. and Tracy, F.M., *Anal Chem.*, **30**, 1699, 1958.

12. Purohit, M.K. and Desai, K.K., *E-Journal of Chemistry*, **2**, 48, 2005.

13. Thipyapang, K. and Suksai, C., *Bull. Korean Chem. Soc.*, **24**, 1767, 2003.

14. Mandera, J., *Anal. Chem.*, **27**, 1955, 2003.

15. Dalman, O. and Karabocek, S., *Turk. J. Chem.*, **27**, 649, 2003.

16. Benerjea, D.K. and Tripathi, K.K., *Ana. Chem.*, **32**, 1196, 1960.

17. Ben-Bassat, A.H., Saat, Y. and Sarel, S., *Bull. Soc. Chim., France*, 948, 1960.

18. Uzumasa, Y. and Washizuka, T., *Bull. Chem. Soc., Japan*, **29**,403, 1956.

19. Gagliardi, E. and Prestinger, P., *Microchim. Acta*, 1047, 1965.

20. Peshkova, V.M., Levontin, M.E. and Litvin, K.I., *Zh. Analit. Khim.*, **3**, 161, 1948.

21. Rodriguez, J.J., Hernandez, G., Siva, J.A., Chacon, E. and Mocelo, R., *Rev. Cubana Quim.*, **1**, 89, 1985.

22. Reddy, D.V. and Reddy, K.H., *Curr. Sci.*, **53**, 798, 1984.

23. Hartkamp, H., *Z. Anal. Chem.*, **176**, 185, 1960.

24. Trussel, F. and Leiberman, K., *Anal. Chim. Acta*, **30**, 269, 1964.

25. Macit, M., Bati, B. and Ozdemir, M., *Turkish J. Chem.*, **19**, 127, 1995.

26. Pemberton, J.R. and Diehl, H., *Talanta*, **16**, 393, 1969.

27. Katsunori, K., Tsugikatsu, O. and Hajime, I., *Anal. Sci.*, **2**, 417, 1986.

28. Chandrasekhar, K.B. and Reddy, K.H., *Indian J. Chem.*, **41**, 1643, 2002.

29. Kumar, M.S., Nagaraja, P. and Yathirajan, H.S., *Anal. Bioanal. Chem.*, **375**, 315, 2003.

30. Pinto, J.J., Moreno, C. and Garcia-Vargas, M., *idem.*, **373**, 844, 2002.

31. Singh, R.B. and Ishii, H., *Critical Reviews in Analytical Chemistry*, **22**, 381, 1991.

32. Siyaji Rao, M., Prasad, N.B.L. and Reddy, K.H., *Indian J. Chem.*, **45**, 1659, 2006.

33. Reddy, B.K., Kumar, J.R., Reddy, K.J., Sarma, S.L. and Reddy, A.V., Anal. Sci., **19**, 423, 2003.

34. Narayana, S.L., Reddy, K.J., Reddy, S.A., Sarala, Y. and Reddy, A.V., *Food analytical methods*, **1**, 293, 2008.

35. Babu, S.V. and Reddy, K.H., *J. Indian Chem. Soc.*, **83**, 20, 2006.

36. Reddy, K.J., Kumar, J.R., Narayana, S.L., Chandraiah, C.R., Thriveni, T. and Reddy, A.V., *Environ. Monit. Asses.*, **124**, 309, 2007.

37. Ghazya S.E., El-Shazlya, R.M., El-Shahawib, M.S., Al-Hazmia, G.A.A. and El-Asmya, A.A., *J. Iranian Chem. Soc.*, **3**, 140, 2006.

38. Takeshi, K. and Akira, H., *Japan Soc. Anal. Chem.*, **31**, 579, 1982.

39. Khalifa, M.E., Akl, M.A. and Ghazy, S.E., *Chem. Pharm. Bull.*, **49**, 664, 2001.

40. Sarma, L.S, Kumar, J.R., Reddy, K.J. and Reddy, A.V., *J. Agri. and Food Chem.*, **53**, 5492, 2005.

41. Kazuyo, I and Kiyoharu, I., *Japan Soc. Anal. Chem.*, **31**, 565, 1982.