DETERMINATION OF COPPER (II)

Determination of Copper (II)

Copper is probably most technically important metal after Iron. It is used in many fields either as metal or its salts, such as in industries, laboratories or medicines, food and beverage. Traces of copper estimation is very essential In chemicals, Pharmaceuticals and metals industries. Traces amounts of copper and its salt is highly toxic to lower organism, much more than man. It is, however, an essential constituent of certain proteins. In biochemical systems copper counter acts the toxicity of zinc by Zn – Cu antagonism[1]. Recent data suggest that test is impaired with copper deficiency is induced[2]. Its toxic effects are the main cause of Wilson's desease[3].

The presence of copper in the plants and animal tissue are recognized more than 150 years ago[4]. In plant physiology it is essential compound[5]. It is harmful impurities in semi-conductor materials[6].

It is one of the nine elements essential to mammalian life. Many organic compounds containing oxygen, nitrogen & sulpher reacts with copper (II) forming chelae complexes, which are widely used for separation and / or extractive spectrophotometric determination of copper. Coulorimetric reagents used for extraxtion & determination of copper are numerous[7-21]. Several workers have reviewed[22-23] the spectrophotometric methods for its determination. Some more reagents are also reported in the literatures.

Procedure for Extraction of copper

An aqueous solution containing 1 mg of copper was taken in a beaker, and mixed with 2 ml of 0.4M sodium acetate solution and 1 ml of 6% isonitrosothiocamphor in alcohol. The pH of the solution was adjusted up to the desire value, with dilute solution of hydrochloric acid or sodium hydroxide, keeping total volume of 10 ml with purified water. The mixture was transferred

in a separating funnel and equilibrated for one minute with an equal volume of chloroform. The two phases are allowed to separated out.

The compound was analyzed as anal. Calcd. For C_{20} H_{28} $N_2O_2S_2Cu$:

C52.66%; H 6.4%; N 6.14%; S 14.64%., and Cu 13.04%,

Found C 52.6; H 6.64%; N 6.06%; S 14.85%. and Cu 13.0%

Procedure for Spectrophotometer determination of copper

To an aqueous solution 4 ml, containing 1 to 50 mcg of copper, is taken in a beaker. 2 ml of 0.4 M sodium acetate solution and 0.5 ml of 0.5% Isonitrosothiocamphor in alcohol was added. The pH of solution was adjusted to 4 with the help of dilute solution of hydrochloric acid or sodium hydroxide solution. The volume of solution maintained up to 10 ml & transfer this mixture into a separatory funnel. Mixed the content for one minute with 10 ml of chloroform. Filter the chloroform layer through anhydrous sodium sulphate. The chloroform layer was collected in a test tube and absorbance of same was taken at 430nm against reagent blank.

The amount of copper is determined with a calibration curve, which was prepared by a series of solution containing known amount of copper were treated by method as mentioned above & the graph of absorbance against concentration of copper were plotted (Refer Table I)

Procedure for mole ratio curve:

A series of mixture containing 50 mcg of copper & increasing amount of reagent in 1 ml of alcohol were treated as described in procedure of spectrophotometer determination of Copper. Absorbance of such extract was measured at 400 nm using chloroform as a blank (Refer Table 2)

Stability of complex:

The extracted colour remain constant for more than 90 minutes. (Refer. Table 3)

Effect of pH:

It was observed that in pH range 2.0 to 6.0, gives better results and at this pH range colour extraction in solvent was stable up to 90 minutes. (Refer. Table 4)

Results and Observation:

The results of extraction studies (Table 5), shows that copper can be quantitavely extracted into chloroform from an aquous solution containing isonitrosothiocamphor at pH 3.0 to 6.0. Bellow and above pH extraction of copper is decreases.

The value of extraction of copper gives following order for the organic solvent used (Refer Table 6)

Chloroform = Benzene = Carbontetrachloride > Toluene > Butanol > ether > chlorobenzene.

Benzene, carbon tetrachloride also can be used for the extraction of copper but the colour is in chloroform is longer time stable as compaired with others.

Validation of Analytical Technique [26-28]

Parameter: The analytical procedure is prepared for the following parameter.

1. **Specificity:-** Specificity is carried out to check the interference due to diluents in analysis.

   Acceptance Criteria: The absorbance proves that there is no interference due to diluents indicating method is specific.

2. **Linearity :** Linearity is established by demonstrating that the results obtained, are directly proportional to that concentration. The Solution were prepared at different concentrations level and absorbance was measured.
   A graph of concentration v/s absorbance was plotted which is Linear.

3. **Accuracy:** Accuracy of the method was established by recovery experiment known amount of Copper was added at there different concentration and analysis was performed.

Sr.No.	Qty. added in ppm	Absorbance	Quantity recovered in ppm	% Recovery
1	0	0	0	--
2	40	0.242	49.59 ppm	99.1 %
3	50	0.488	100.00 ppm	100.00 %
4	60	0.731	149.79 ppm	99.86 %
Mean recovery.				99.52%

Mean recovery of 3 level is calculated, which is 99.52 and is in acceptable limit, indicating that method is accurate under the prescribed conditions.

4. Precision: Precision of the method was established by performing assay 3 time by same analyst.

Sr.No.	Conc. of Copper in ppm	Absorbance	% of Copper
1	100 ppm	0.485	99.38 %
2	100 ppm	0.487	99.79 %
3	100 ppm	0.490	100.40 %
Mean			99.85 %

Mean of 3 analysis is 99.85 % and is in acceptance limit, indicated that method is validated.

5. Robustness: The robustness is proceeded by obtaining the analytical results on the portion of same sample by different analyst.

Sr.No.	Conc. of Copper in ppm	Analyst	Absorbance	% of Copper
1	100 ppm	Analyst I	0.491	100.51 %
2	100 ppm	Analyst II	0.486	99.59 %
3	100 ppm	Analyst III	0.489	100.20 %
Mean				100.13 %

Mean of results of all three analysts is 100.13 % which is in acceptable limit, and indicated that the method is validated.

Table 1

Absorbance measurement of Copper in chloroform with the help of Isonitrosothiocamphor at different wavelength.

Concentration of Copper : 100 ppm

Volume of chloroform : 10 ml

Time of equilibrium : 1 min.

Sr. No.	Wavelength	Absorbance
1	350	0.442
2	355	0.432
3	360	0.427
4	365	0.425
5	370	0.418
6	375	0.424
7	380	0.424
8	385	0.438
9	390	0.433
10	395	0.441
11	400	0.455
12	405	0.462
13	410	0.468
14	415	0.460
15	420	0.472
16	425	0.483
17	426	0.484
18	427	0.480
19	428	0.487
20	429	0.487
21	430	0.488
22	431	0.487

Cont.

23	432	0.485
24	433	0.482
25	434	0.477
26	435	0.470
27	440	0.469
28	445	0.458
29	450	0.459
30	455	0.444
31	460	0.432
32	465	0.424
33	470	0.428
34	475	0.406
35	480	0.392
36	485	0.390
37	490	0.387
38	495	0.361
39	500	0.347
40	505	0.322
41	510	0.306
42	515	0.290
43	520	0.253
44	525	0.228
45	530	0.208
46	535	0.200
47	540	0.188
48	545	0.172
49	550	0.155
50	555	0.136

Table 2

Effect of Reagent concentration on the absorbance of Copper(II) and Isonitrosothiocamphor.

Concentration of Copper : 100 ppm
Organic phase : 10ml Chloroform
Time of equilibrium : 1 min.
Wavelength : 430 nm

Sr.No.	Volume of Isonitrosothiocamphor	Absorbance
1	0.1	0.425
2	0.2	0.460
3	0.3	0.477
4	0.4	0.480
5	0.5	0.488
6	0.6	0.488
7	0.7	0.488
8	0.8	0.488
9	0.9	0.488
10	1.0	0.488
11	1.5	0.488
12	2.0	0.488

Table 3

Absorbance of Copper (II) with the help of Isonitrosothiocamphor in chloroform at different pH

Concentration of Copper : 100 ppm
Organic phase : 10 ml Chloroform
Wavelength : 430 nm.

Sr.No	pH	Absorbance	% extraction
1	1.0	0.478	97.7%
2	2.0	0.482	98.7%
3	3.0	0.486	99.5%
4	4.0	0.488	100.0%
5	5.0	0.488	100.0%
6	6.0	0.485	99.3%
7	7.0	0.481	98.5%
8	8.0	0.456	93.4%
9	9.0	0.430	88.1%
10	10.0	0.406	83.1%

Table 4

Absorbance of Copper(II) in chloroform with Isonitrosothiocamphor as function of time.

100 ppm of Copper(II) in 10 ml of chloroform at pH 4 equilibrated with respect to different time at 430nm.

Sr.No.	Time in minute	Absorbance
1	1	0.488
2	5	0.488
3	10	0.488
4	20	0.488
5	40	0.488
6	60	0.488
7	90	0.488
8	120	0.482
9	150	0.477
10	180	0.460

Table 5

Absorption measurement of copper extracted with different concentration of chloroform at pH 4

Concentration of copper : 100ppm

Wavelength : 430 nm

Time of equilibrium : 1 min.

Sr.NO.	Volume of Chloroform in ml	Absorbance
1	5 ml	0.962
2	10 ml	0.488
3	15 ml	0.357
4	20 ml	0.241
5	25 ml	0.113

Table 6

Extraction of Copper (II) with Isonitrosothiocamphor using different solvent

Organic phase : 10 ml

Time of equilibrium : 1 min.

Sr.No.	Solvent	Absorbance	% extraction
1	Chloroform	0.488	100.0%
2	Benzene	0.488	100.0%
3	Carbon tetra chloride	0.478	97.95%
4	Toluene	0.462	94.67%
5	Butanol	0.450	92.2%
6	Ether	0.404	82.7%
7	Chlorobenzene	0.360	73.7%

Table 7

Calibration Curve for Copper (II)

Organic phase : 10 ml chloroform
Time of equilibrium : 1 min.
Wavelength : 430 nm

Sr.No.	Conc. Of Copper in ppm	Absorbance
1	25	0.122
2	50	0.242
3	75	0.368
4	100	0.488
5	125	0.606
6	150	0.731
7	175	0.452
8	200	0.980

Infrared studies of Copper(II) Complex with isonitrosothiocamphor

Infrared spectrpphotometry involves vibrational energy level and again organic functional groups exhibits characteristics absorption peaks. Infrared spectrophotometry obeys the Lambert's Beer's law. Quantitative measurements are also made more difficult because transparent in the infrared as water is in the visible region.

Infrared radiation promotes transition in a molecule between rotational and vibrational energy level of lowest electronic energy state, and the energy that is absorbed from a radiation beam as a result of such a transition and the corresponding frequencies or wavelength of an absorption band, is directly associated with the atomic mosses, bond forces and special geometry of the molecules. The infrared absorption spectrum thus has a high degree of specificity and no two compounds have identical spectra. Small difference in structure frequently results in significant difference in spectra. Early workers used this technique for material identification and functional group analysis. Improved instrumentation and development are more efficient source units, monochromaters. Detectors and measuring systems are used in quantitative organic analysis.

The determination of Functional groups determined with the help of infrared are as bellow.

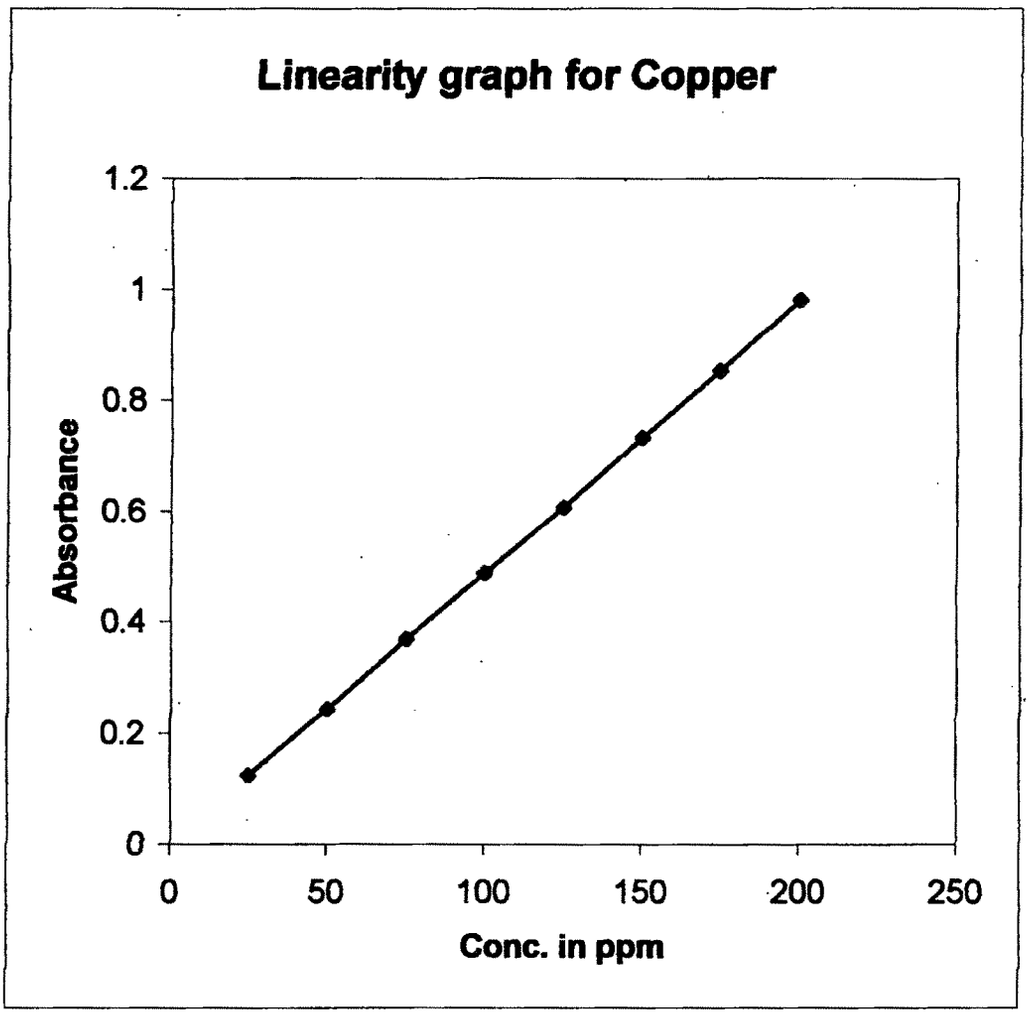
1. **Nitroso compound** : N=O bond is stable in nitroso compounds is characterized by a stretching vibration in the range [29] 1621 -1488 cm^{-1}

Nitroso compound tend to dimerise, secondary primary nitroso compound readily rearrange to oxime. In monomeric state absorbs in the 1621-1488 cm^{-1} [30] region, but in the solution they exist preferably as dime rise and then absorbs near 1290 cm^{-1}.

Linearity graph for Copper

Calibration curve for Nickel at 430 nm in Chloroform

Conc. of Copper in ppm	Absorbance
25	0.122
50	0.242
75	0.368
100	0.488
125	0.606
150	0.731
175	0.852
200	0.98



2. **→C– CH_3 group**: The mode of → C– CH_3 gives an observation at about [30] 2872 cm^{-1}

3. **>C=S group** : The range of >C=S group occurs in the range[31] of 1000-1400 cm^{-1}

4. **>C<** $^{CH3}_{CH3}$ **group** : The frequency of >C< $^{CH3}_{CH3}$ is at about [32] 1340 - 1460 cm^{-1}

Two new bands around 470 and 350 cm^{-1} appeared in the spectra of all the complexes, which could be assigned to complete vibrations having contribution from υ MN υ MS.

υ – OH in free ligend disappear in the complex, and a new weak new band at 280 cm^{-1} appears. This indicates the deprotonation of the ligend molecule during the complexation. Further, the intensity of υ = C – decreases and it shifts to downward direction in the complex indicating thereby complaxation of oxygen side of the ligend.

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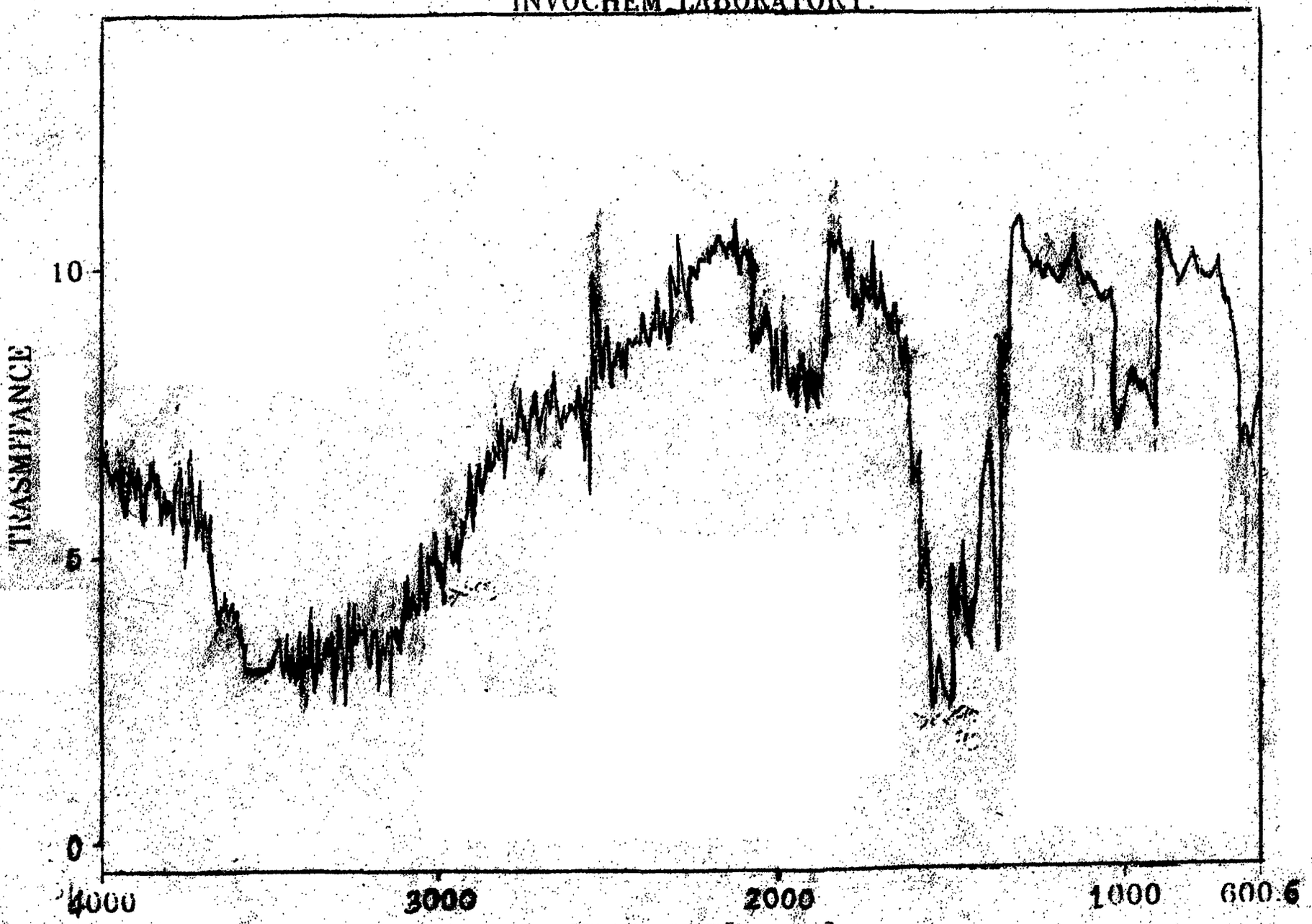

ARTY : SANJAY KUMAR SINGH B.NO. :

R.NO. :

PLE :

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