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BSTOICHIOMETRIC DETERMINATION OF COPPER BY ISOTOPIC DILUTION ANALYSIS IN BIOLOGICAL AND ENVIRONMENTAL SAMPLES

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ABSTRACT

A rapid and selective method has been used for the determination of copper by isotopic dilution employing substoichiometric extraction with dithizone in carbon tetrachloride. The appropriate pH range for the substoichiometric extraction was 2-7. In the analysis, even a large excess of elements which form extractable complexes with dithizone does not interfere. The accuracy and precision of the method was evaluated. The method has been applied in SRMs, wheat flour, wine and beer.

INDEX ENTRIES: Copper, isotopic dilution, substoichiometry, copper dithizonate, application of tracers, environmental samples.

INTRODUCTION

Copper is one of the elements that has biological interest at trace and ultra-trace levels. Analytical dificulties often seem to increase drastically when analyzing materials with very low contents of trace elements(1).Thus the availability of sensitive and selective methods is of great importance for biological and environmental samples. Neutron activation analysis with inherent sensivity is the obvious method of choice. However ^{64}Cu isotope can also be produced by the reaction $^{64}Zn(n,p)^{64}Cu$, which has a cross section of 390 mbarn and ^{64}Zn has 48,9% natural abundance. Since most biological samples contain amounts of Zn comparable with those of copper, this reaction may cause some interference in the determination of copper. In this context, substoichiometric isotopic dilution analysis proposed by Suzuki(2) and Ruzicka and Stary(3) is a very attractive analytical method to determine copper in these kinds of samples.

The method offers an accurate and precise determination of trace and ultra-trace amounts of elements by measurements of radioactivity alone, without corrections for chemical yield. The advantages of substoichiometric separation are that it is highly selective, quick and reproducible. The principles of the method have been described in several papers(4-7).

This paper describes a simple procedure for the determination of copper in biological and environmental samples by isotope dilution analysis involving the substoichiometric extraction technique with dithizone into carbon tetrachloride.

MATERIALS AND METHODS

Countig apparatus

A Nuclear Chicago single channel analyzer coupled to a 3.7 x 6.2 cm well-type NaI(Tl) crystal was used for the measurements of the activity.

Reagents

All the chemicals, reagents and solvents used were of A.R. grade.The acids and ammonium hydroxide were purified by distillation.

Radio-copper solution

^{64}Cu was produced by the neutron irradiation of metallic copper for 30 min or 8 h at a neutron flux of 10^{12} ncm^{-2}s^{-1}. Irradiated target was dissolved in nitric acid, and acid excess was eliminated by evaporation. The radioactive solution was transferred to give a concentration of 1 mg/mL.

Dithizone solution

The solution was prepared dissolving dithizone in CCl_4 to give a concentration of 10^{-3} M. The solution was transferred into an amber flask and kept in the refrigerator because it decomposes rapidly with the formation of carbadiazone if exposed to strong light and subjected to relatively high temperatures.

Buffer solution

Prepared by the dissolution of 15 g of potassium iodide, 13 g of ammonium acetate in 100 mL of water and the addition of glacial acetic acid to attain a pH value of 4.8(8). Purified by shaking with a concentrated solution of dithizone in CCl_4.

Interfering elements solutions

Appropriate amount of the elements: salts, oxides or metals were dissolved in acid or deionized distilled water. The excess of acid was eliminated by evaporation. The solutions were transferred to give a concentration of 1 mg/mL. The procedure was applied to prepare As(II), Au(III), Bi(III), Cd(II), Co(II), Hg(II), In(III), Mn (II), Ni(II), Pb(II), Pd(II), Pt(II), Sb(III), Se(II), Sn(IV) and Zn(II).

Sodium thiossulfate solution 2%

An amount of sodium thiossulfate was dissolved in distilled water.

COPPER EXTRACTION

The high selectivity and simplicity of the method which give optimum conditions for the determination of copper were studied. The influence of hydrochloric, sulfuric and nitric acids were examinated due to the possibility of using these acids in the digestion process of samples.

EQUILIBRIUM TIME

A series of solutions, pH 5, with the same amounts of ^{64}Cu were shaken with dithizone in CCl_4. The solution was shaken during different times. The gamma activity of the extracted copper was measured and plotted against shaking time. The equilibrium was reached with a shaking time of 2 min.

EFFECT OF ACIDS

The influence was verified by the addition of an aqueous solution to the tracer dissolved in HCl, HNO_3 or H_2SO_4 separately. The excess of acid was eliminated by evaporation and the pH was adjusted to 4.8 with buffer solution. The solution was shaken for 2 min with dithizone solution. The phases were separated and the activity of the organic aliquot was measured.

The results showed that copper is quantitatively extracted independent of the acid used.

SUBSTOICHIOMETRIC EXTRACTION

A known amount of ^{64}Cu and a substoichiometric amount of dithizone in CCl_4 solution were added to an aqueous solution. A series of preliminary experiments were carried out to ascertain the best conditions of pH range and influence of masking agent for finding the reproducibility of the substoichiometric extraction.

pH-RANGE

A series of solutions with known amounts of radio-copper with a constant but substochiometric amount of dithizone in CCl_4 was prepared and investigated at different pH. The dependence of the substoichiometric extraction of Cu with dithizone on pH can be seen in Table 1. Copper could be extracted substoichiometrically from pH 2-7 which is in agreement with the predicted value(9).

REPRODUCIBILITY OF SUBSTOICHIOMETRIC EXTRACTION

The reproducibility of the substoichiometric extraction of copper with dithizone into CCl_4 was examined in solutions containing different amounts of radioactive Cu(II). A graph was plotted of the activity of an aliquot of the organic phase vs the amount of Cu(II) taken. As shown in Table 2, the activity of the organic phase increased with increase in the amount of Cu(II),after this point which corresponds to a copper-dithizone ratio of 1:2, the amount of copper in the organic phase remains constant.The aqueous solution containing excess of labelled copper was treated repeatedly with dithizone in CCl_4. In each case the activity of the organic phase was noted to be the same as that of the other extracts. This shows that the same amount of copper was extracted in each extraction.

INTERFERENCES OF OTHER ELEMENTS IN THE SUBSTOICHIOMETRIC EXTRACTION OF Cu(II).

The study of the selectivity of substoichiometric separation of Cu(II) was carried out by means *of a series of solutions* containing known amounts of non-active copper and active copper and substoichiometrically extracted with dithizone in CCl_4, in presence of As(III), Au(III), Bi(III), Co(II), Cd(II), Hg(II), In(III), Mn(II), Ni(II), Pb(II), Pd(II), Pt(II), Sb(III), Se(II), Sn(IV) and Zn(II). As shown in Table 3 even a large excess of metals which form extractable complexes with dithizone when iodide is present in the aqueous phase will not inhibit the extraction. It can be seen that only Au, Pd and Pt interfere.

APPLICATIONS

The method described has been applied to the determination of copper in SRMs:Citrus Leaves:NBS-SRM-1472; Wheat Flour:NBS-SRM-

1567a; Rice Flour:NBS-SRM-1568; Fish Homogenate:MA-A-2 621, wine, beer and wheat flour.

Sample Preparation

SRMs: An amount of 200 mg of sample was added to 2-3 mL of HNO_3 and a known amount of ^{64}Cu in a microwave acid digestion bomb. The sample was digested in a microwave furnace at medium power for 2 min and cooled for 1 h. The sample solution was evaporated to eliminate the excess acid.

WINE and BEER: The procedure used was described by Kumar et al(10). An aliquot of 50 mL of wine or beer was evaporated until dryness, the residue was dissolved in 100 mL of HNO_3.

Substoichiometric determination of copper

The appropriate amount of HNO_3 dil or NH_4OH dil was added to the sample and standard solutions to reach the 4.0-5.0 pH. After adjusting the acidity, 2 mL of buffer solution and 0,5 mL of sodium thiossulfate solution 2% was added. The solutions were simultaneously extracted with equal substoichioetric portions of dithizone in CCl_4. The activity of the organic layer was measured. Table 4 shows the results obtained.

DISCUSSION

As showed above, Cu(II) could be extracted with a substoichiometric amount of dithizone in CCl_4 solution between pH 2 to 7. Above and below this pH range the extraction is not quantitative. To examine the influences of other elements, the substoichiometric extraction with $1x10^{-5}M$ dithizone was applied to the synthetic samples solutions containing a small amount of copper and large amounts of other elements. As presented in Table 3, the substoichiometric determination of copper is possible together with other elements that have higher extraction constants than copper. The extraction constant of mercury with dithizone is log26,85, however, the results showed that mercury does not interfere in the extraction of copper. This occurs because a buffer solution containing iodide was added to the aqueous solution which formed a complex species with mercury being most stable in the aqueous phase. The thiossulfate present in the aqueous phase prevents oxidation of the dithizone by free iodine.

This method is found to be highly selective for Cu(II) and was observed that the elements Au, Pt and Pd are extracted by solution with dithizone in these conditions. However, these elements may be left out of consideration because of their rarity in these kinds of samples.

The precision and accuracy of the method was tested with Standard Reference Materials. From Table 4 the good precision of the method is shown. Even for materials with the lowest copper contents, relative standard deviation does not exceed 8%. The precision of substoichiometric analysis has mainly two components: one associated with substoichiometric separation and the other associated with radioactivity measurements. As the latter component can be improved by use of a radioisotope with a high specific activity, the precision depends primarily on the former component.

The sensivity limit was found to be less than 13 ng which was obtained by irradiation of Cu(II) with thermal neutron flux of $10^{12}ncm^{-2}s^{-1}$ for 8 h and then subjected through the radiochemical separation procedure as mentioned.

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REFERENCES

1. R. Dybczynski, M. Wasek, H. Maleszewska, *J. Radioanal. Nuclear Chem. Articles* **130**, 365 (1989)
2. N. Suzuki, *Proceedings of 2nd Conf. Radioisotopes;* Japan Atomic Industrial Forum, Tokyo, 1958, pp. 151.
3. J. Ruzicka, J. Stary, *Substoichiometry in Radiochemical Analysis*, Pergamon Press, London, 1968.
4. K. Kudo, N. Suzuki, *J. Radioanal. Chem.* **26**,327 (1975)
5. P. C. Reddy, B. Polalah, R. Rengamanner, *Radioisotopes* **39**, 496 (1990).
6. S. S. Shetty, Z. R. Turel, *J. Radioanal. Nuclear Chem. Articles* **145**,223 (1990).
7. N. Suzuki, *J. Radioanal. Nuclear Chem. Articles* **124**,197 (1988).
8. J. Ruzicka, J. Stary. *Talanta* **9**,617 (1962)
9. K. Kudo, K. Kobayashi. *J. Radioanal. Chem.* **53**,163 (1979)
10. B. Kumar, H. B. Singh, M. Katyal, R. L. Sharma. *Mikrochim. Acta* **III**,79 (1991).

TABLE 1 Effect of pH on the substoichiometric extraction of copper
$10^{-3}M\ H_2Dz$, $6.3x10^{-3}M\ ^{64}Cu$
Shaking time: 3 min

pH	Percentage of extraction(%)
0.5	67.8
1.0	84.5
1.5	87.1
2.0	89.4
3.0	89.0
4.0	90.9
5.0	93.3
6.0	95.4
7.0	95.3
8.0	64.9

TABLE 2 Substoichiometric extraction of Cu(II) with dithizone into CCl_4
10-4M H2Dz, shaking time:3 min

^{64}Cu(II)(ug)	Activity of 1mL aliquot of organic phase (cpm)
8.09	4003
12.13	5967
16.18	7720
24.27	8391
32.36	8550
36.40	8835
40.45	8841
50.56	9016

TABLE 3 Interference with extraction of copper caused by addition of non-active elements

Copper taken (ug)	Active copper (ug)	Other cations present (ug)	Copper found (ug)*
1.0	1.0	–	1.17±0.02
		25.0ug Hg	1.01±0.02
1.0	1.0	–	1.25±0.03
		1.01ug Au	1.44±0.04
		0.95ug Pd	3.12±0.08
		1.04ug Pt	1.33±0.03
1.0	1.0	–	1.22±0.03
		93.2ug Bi	1.10±0.03
		98.5ug In	1.40±0.03
		100.8ug Mn	1.31±0.03
		98.7ug Se	1.38±0.03
1.0	1.0	–	1.01±0.03
		100.0ug As	1.12±0.04
		100.0ug Cd	1.05±0.03
		100.0ug Co	1.01±0.03
		100.0ug Ni	1.05±0.03
		100.0ug Pb	1.00±0.03
		100.0ug Sb	1.10±0.03
		100.0ug Sn	1.08±0.03
		100.0ug Zn	1.09±0.03

* Arithmetic mean ,n=4

TABLE 4 Results of copper determination

Material	Certified value (ug/g)	Experimental value (ug/g)*	RSD	Deviation from the certified value %
SRM 1567a	2.1±0.2	1.79±0.16	0.057	-14.6
SRM 1568	2.2±0.3	2.04±0.22	0.043	-7.4
SRM MA-A-2 621	4.0±0.1	4.83±0.6	0.078	20.8
SRM 1572	16.5±1.0	17.0±0.6	0.022	3.0
Wheat flour		1.00±0.09	0.053	
Wine (ug/mL)		0.17±0.01	0.051	
Beer (ug/mL)		0.28±0.02	0.063	

* Arithmetic mean (n=4) and its confidence limits($\bar{x} \pm t_{0.05} s n^{-1/2}$)