Spectrophotometric Determination of Aliphatic Thiols Based on Redox Color Reaction with Copper(II) Complex of 1,10-Phenanthroline in Dimethyl Sulfoxide

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A novel spectrophotometric method was developed for the determination of aliphatic thiols. The method was based on the stoichiometric redox reaction between aliphatic thiol and a copper(II) complex with 1,10-phenanthroline in dimethyl sulfoxide to form an intensely colored copper(I) complex, which had a molar absorptivity of 7000 $dm^3 mol^{-1} cm^{-1}$ at 444 nm. The calibration curves for five aliphatic thiols of carbon numbers 1 - 8 had the same slope over the range from the detection limit of 5.0×10^{-6} to 1.5×10^{-4} mol dm^{-3}. The relative standard deviation of five successive experiments was 2.0% at 8.6×10^{-5} mol dm^{-3} of 1-butanethiol. The method was applied to the determination of methanethiol in gas samples.

Keywords Spectrophotometric method, thiol, copper-1,10-phenanthroline complex

Volatile aliphatic thiols are typical air pollutants, because of their characteristic offensive odor. Gas chromatography (GC) is widely used to determine traces of aliphatic thiols.[1,2] Recently, high performance liquid chromatography (HPLC) with fluorescence detection has been applied to the determination of aliphatic thiols, which were derivatized with fluorescence agents prior to the measurement by HPLC.[3] On the other hand, it is necessary to develop a simple and selective screening method employed for field testing in order to rapidly determine aliphatic thiols contained in a sample gas. Spectrophotometric methods provide substantial advantages with respect to simplicity of instrumentation, and speed of analysis. However, no spectrophotometric method for aliphatic thiols has been established, though many spectrophotometric methods have been widely used for such water-soluble thiols as cysteine and glutathione.[4,5]

We have previously shown that a copper(II)-1,10-phenanthroline(phen) complex ion in dimethyl sulfoxide is quantitatively reduced by aliphatic thiols to the intensely colored copper(I)-phen complex ion.[6] Based on this finding, we proposed a spectrophotometric method for the determination of aliphatic thiols. In this paper we describe the effects of aprotic solvents and the concentrations of phen and copper(II) on the stoichiometry of the redox reaction between the aliphatic thiols and $[Cu(II)(phen)_n]^{2+}$ ion. The proposed method was applied to the determination of methanethiol in gas samples.

Experimental

Reagents

Dimethyl sulfoxide (DMSO), dimethylformamide (DMF) and acetonitrile (AN) were used as received with no further purification. All other chemicals were guaranteed grade reagents and were used without further purification.

Standard solutions of 1-propanethiol, 1-butanethiol, 1-hexanethiol and 1-octanethiol were made by dissolving 0.10 cm^3 of the respective thiols in hexane and diluting to 25 cm^3. A standard solution of methanethiol was purchased from Wako Pure Chemical Ind. Working solutions of the thiols from 5.0×10^{-6} to 1.5×10^{-4} mol dm^{-3} were prepared by subsequent dilution of the standard solutions with hexane.

Apparatus

The absorption spectra of the copper complex ion were measured with a Hitachi U-2000-type spectrophotometer equipped with 1-cm quartz cells. Disulfide and unreacted thiols in hexane were measured with a Hitachi 663-30-type gas chromatograph equipped with a hydrogen flame ionization detector. Measurements of sulfur compounds in gas samples were made on a Hitachi 163-type gas chromatograph equipped with a flame photometric detector (FPD).

Procedure

$[Cu(II)(phen)_n]^{2+}$ ion solutions were prepared by dissolving copper(II) nitrate (final concentration: 1.0×10^{-2}

mol dm⁻³) and phen (4.0×10⁻² mol dm⁻³) in DMSO, DMF and AN or a mixture of these aprotic solvents and distilled deionized water (water), except for experiments for the optimization of the concentration of copper(II) and phen.

The recommended optimized procedure for the determination of the aliphatic thiols is as follows: equal volumes (10 cm³) of DMSO containing [Cu(II)(phen)$_n$]²⁺ ion and hexane containing thiol are placed, together in a 50-cm³ separatory funnel, and the mixture is shaken for 15 min at room temperature.   After phase separation, the absorption spectra of [Cu(I)(phen)$_n$]⁺ ion in the DMSO phase are measured against a reagent blank.

Determination of methanethiol in gas samples

A volume of 20 dm³ of gas at the inlet and outlet of a dryer in a fish meal factory was bubbled at 1 dm³/min through octane (10 cm³) placed in two fritted bubblers (50 cm³), connected in series.   Then, the methanethiol absorbed in the octane was analyzed by the proposed method and gas chromatography with FPD.

Results and Discussion

Redox reaction between 1-butanethiol and [Cu(II)(phen)$_n$]²⁺ ion in aprotic solvents

In order to clarify the effect of aprotic solvents on the redox reaction, the oxidation of 1-butanethiol (8.6×10⁻⁵ mol dm⁻³) with [Cu(II)(phen)$_n$]²⁺ ion was examined in a DMSO, DMF or AN medium.   The intensely colored [Cu(I)(phen)$_n$]⁺ ion was formed in all aprotic solvents without any formation of the copper thiolato complex. The redox color reaction was completed within 10 min, independent of the aprotic solvents used.   Then, the absorption spectra of the [Cu(I)(phen)$_n$]⁺ ion formed were measured after 15 min from initiation of the reaction.   The copper(I)-phen complex ion exhibited an absorption maximum at 444 nm, as shown in Fig. 1. However, the maximum absorbance was dependent on the aprotic solvents, indicating that the extent of reduction of [Cu(II)(phen)$_n$]²⁺ ion to the [Cu(I)(phen)$_n$]⁺ ion was affected by aprotic solvent used.

Next, we examined the effect of the volume percent of aprotic solvent in the final solution on the molar fraction of 1-butanethiol oxidized to 1-butyldisulfide.   The oxidation of 1-butanethiol (2.0×10⁻² mol dm⁻³) was carried out using [Cu(II)(phen)$_n$]²⁺ ions (1.0×10⁻² mol dm⁻³) prepared in DMSO, DMF and AN or a mixture of these aprotic solvents and water in the volume percent range 60 – 100%.   The concentrations of 1-butyldisulfide and unreacted 1-butanethiol in hexane were determined after 15 min from initiation of the reaction.   Figure 2 shows the molar fraction of 1-butanethiol oxidized to 1-butyldisulfide.

When [Cu(II)(phen)$_n$]²⁺ ions and 1-butanethiol (RSH) in equi-molar amounts react quantitatively to form [Cu(I)(phen)$_n$]⁺ ions and 1-butyldisulfide (RSSR), the reaction yield (mol%) of the thiol oxidized to the disulfide
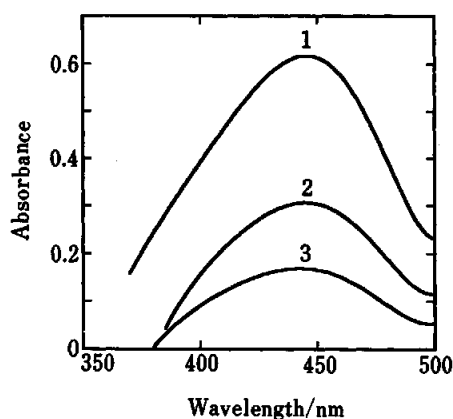

Fig. 1   Absorption spectra of copper(I) complex ions in aprotic solvents.   Cu(II), 1.0×10⁻² mol dm⁻³; phen, 4.0×10⁻² mol dm⁻³; 1-butanethiol, 8.6×10⁻⁵ mol dm⁻³.   1, DMSO; 2, DMF; 3, AN.
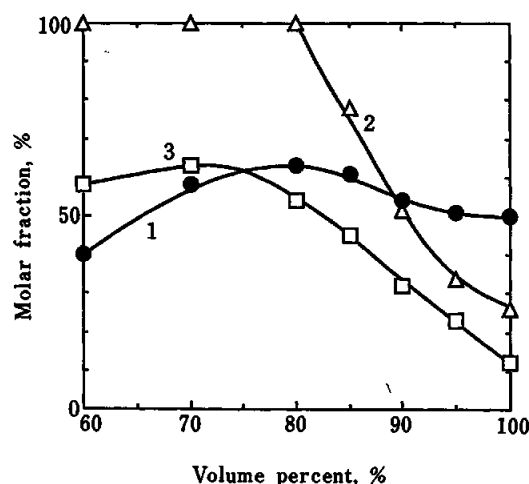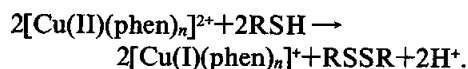

Fig. 2   Molar fraction of 1-butanethiol oxidized to 1-butyldisulfide in aprotic solvents.   Cu(II), 1.0×10⁻² mol dm⁻³; phen, 4.0×10⁻² mol dm⁻³; 1-butanethiol, 2.0×10⁻² mol dm⁻³. 1, DMSO; 2, DMF; 3, AN.

should be 50%, as expressed by the following equation:

$$2[Cu(II)(phen)_n]^{2+}+2RSH \longrightarrow$$
$$2[Cu(I)(phen)_n]^++RSSR+2H^+.$$

The results shown in Fig. 2 indicate that equimolar amounts of 1-butanethiol and [Cu(II)(phen)$_n$]²⁺ ions reacted to form [Cu(I)(phen)$_n$]⁺ ions and 1-butyldisulfide (RSSR) when the volume percent of DMSO was above 95% in aqueous-DMSO.   In aqueous-AN and DMF solutions, [Cu(II)(phen)$_n$]²⁺ ions and 1-butanethiol (RSH) react in equi-molar amounts at 80 and 90% in DMF and AN, respectively.   It is therefore possible to determine aliphatic thiols by measuring the absorbance of [Cu(I)(phen)$_n$]⁺ ions under these conditions.

The degree of oxidation of 1-butanethiol to its disulfide decreased when the volume percent of DMF and AN in their aqueous solutions was above 80 and 90%, respectively. Along with a decrease in the degree of oxidation of 1-butanethiol in DMSO, DMF and AN, the absorbance of $[Cu(I)(phen)_n]^+$ ions decreased in the same order. On the other hand, the degree of oxidation was greater than 50% when the volume percent of DMSO, AN and DMF in their aqueous solutions was less than 90, 90 and 80%, respectively. This was attributable to the. oxidation of $[Cu(I)(phen)_n]^+$ ions with dissolved oxygen to $[Cu(II)(phen)_n]^{2+}$ ions. Cu(II) complex ions reacted again with the thiol. Under these conditions, the redox reaction proceeded nonstoichiometrically and the color of the solution faded rapidly. Thus, DMSO was chosen as a solvent for the reaction media. In aqueous-AN and DMF solutions the redox reaction proceeded quantitatively in a very limited range of volume percent of AN or DMF, thus precluding their use.

Optimum concentrations of phen and copper(II) in DMSO

We examined the optimum concentrations of phen required for a quantitative redox reaction. The oxidation of 1-butanethiol $(2.0\times10^{-2}$ mol dm$^{-3})$ in DMSO was carried out by varying the molar ratio of phen to Cu(II) $(1.0\times10^{-2}$ mol dm$^{-3})$ up to 6. The concentrations of disulfide and unreacted 1-butanethiol in hexane were determined after 15 min from initiation of the reaction. The concentration of 1-butanethiol as present in the copper thiolato complex was determined by subtracting the concentrations of 1-butanethiol oxidized to 1-butyldisulfide and unreacted 1-butanethiol from the initial 1-butanethiol concentration. The result of an analysis of the reaction products is shown in Fig. 3.
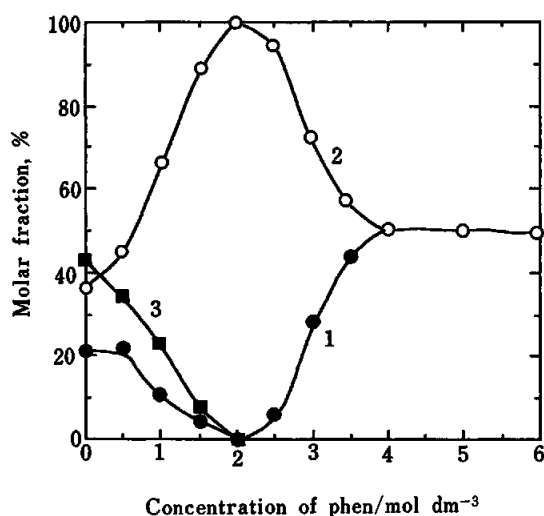
When the mole ratio of phen to Cu(II) was greater than 4, the redox reaction proceeded stoichiometrically. However, when the mole ratio was below 4, excess amounts of 1-butanethiol were oxidized to 1-butyldisulfide by catalytic oxidation. In molar ratios up to 2, copper(II)-phen complex ions react with 1-butanethiol to form a copper thiolato complex and 1-butyldisulfide. The optimum concentration of phen was thus determined to be 4.0×10^{-2} mol dm^{-3} in the recommended procedure.

Next, the Cu(II) concentration must be optimized. The oxidation of 1-butanethiol $(1.0\times10^{-4}$ mol dm$^{-3})$ was achieved by using $[Cu(II)(phen)_n]^{2+}$ ions, the concentration of which was within the range 1.0×10^{-4} – 2.5×10^{-2} mol dm^{-3}. The absorbance of $[Cu(I)(phen)_n]^+$ was measured after 15 min from the initiation of the reaction. Figure 4 shows the effect of the Cu(II) concentration on the absorbance. The reduction of $[Cu(II)(phen)_n]^{2+}$ ions to $[Cu(I)(phen)_n]^+$ ions was markedly influenced by the initial concentration of Cu(II). The absorbance was constant when the concentration of Cu(II) was greater than 1.0×10^{-2} mol dm^{-3}. Thus, the most appropriate concentration of Cu(II) was found to be 1.0×10^{-2} mol dm^{-3} by taking into account of the solubility of $[Cu(II)(phen)_n]^{2+}$ ions in DMSO.

The effect of the shaking time on the absorption spectra in DMSO was examined during periods from 5 to 30 min. The oxidation of 1-butanethiol $(5.0\times10^{-5}$ mol dm$^{-3})$ was carried out under the optimum conditions for the respective agents. The oxidation was completed within 10 min. The reoxidation of $[Cu(I)(phen)_n]^+$ ions in DMSO was so slow that the absorption spectra remained unchanged for two hours. Thus, 15 min was selected for the procedure.

Analytical results and parameters

Analytical calibration curves were prepared for five aliphatic thiols under the optimized conditions. The calibration curves at 444 nm obeyed Beer's law and had the same slope over the range from a detection limit of
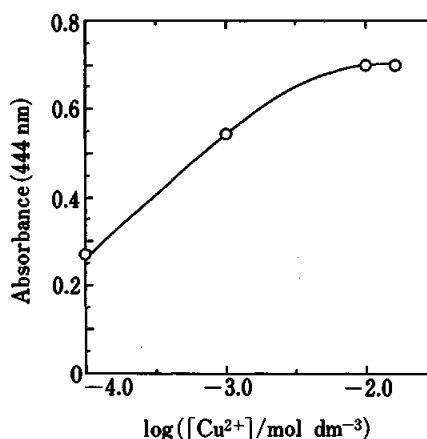

Fig. 3 Variation of the molar fraction of 1-butanethiol with the phen concentration after a redox reaction in DMSO. Cu(II), 1.0×10^{-2} mol dm^{-3}; 1-butanethiol, 2.0×10^{-2} mol dm^{-3}. 1, 1-butanethiol in hexane; 2, 1-butanethiol oxized to 1-butyldisulfide; 3, 1-butanethiol formed copper thiolato complex.

Fig. 4 Variation of the absorbance with the copper(II) concentration in DMSO. 1-Butanethiol, 1.0×10^{-4} mol dm^{-3}; [phen]/[Cu(II)]=4.

Table 1 Determination of sulfur compounds in gas samples

Sulfur compound	Concentration, ppm(v/v)	
	Inlet	Outlet
Dimethyl sulfide	0.16[a]	0.05[a]
Dimethyl disulfide	0.03[a]	0.27[a]
Methanethiol	0.21[a]	0.98[a]
Aliphatic thiol	0.22[b]	0.93[b]

a. Gas chromatographic method with FPD.
b. Present method.

5.0×10^{-6} to 1.5×10^{-4} mol dm^{-3}, independent of the carbon numbers of the thiols. The detection limit for the thiols was defined as the concentration yielding an absorbance equal to three times the standard deviation of the reagent blank. The molar absorptivity of [Cu(I)-(phen)$_n$]$^+$ ion was 7000 dm^3 mol^{-1} cm^{-1} at 444 nm. The relative standard deviation of five successive experiments was 2.0% at 8.6×10^{-6} mol dm^{-3} of 1-butanethiol.

Determination of methanethiol in gas samples

In order to investigate interference from sulfur compounds, the determination for 8.6×10^{-6} mol dm^{-3} of 1-butanethiol was examined by adding such sulfur compounds as dimethyl sulfide, dimethyl disulfide, tetrahydrothiophene and thiophene into hexane. The tolerance ratio of each foreign compounds was taken as being the largest amount yielding an error of less than 3% in absorbance. No interference from dimethyl disulfide was observed, even at 1000-times greater than the concentration of 1-butanethiol. The tolerable amount of tetrahydrothiophene was two-hundred times greater than that of 1-butanethiol. One-hundred times of dimethyl sulfide and thiophene was tolerated.

The present method was applied to a determination of aliphatic thiols in gas at the inlet and outlet of a dryer in a certain fish meal factory. In order to prevent organic solvent losses during sampling, octane was used in the place of hexane for dissolving the thiols in sample gas. Table 1 presents the analytical results of methanethiol and other sulfur compounds by the gas chromatographic method. Though dimethyl sulfide and dimethyl disulfide were contained in the gas samples, they had little influence on the oxidation of methanethiol with [Cu(II)(phen)$_n$]$^{2+}$ ions. We next determined the concentration of aliphatic thiol by the proposed methods. As shown in Table 1, the concentration of aliphatic thiol is very close to the value obtained by the gas chromatographic method.

In conclusion, a new indicator reaction has been developed for the spectrophotometric determination of aliphatic thiol by using DMSO as the reaction media of the redox reaction between aliphatic thiols and [Cu(II)(phen)$_n$]$^{2+}$ ions. In the present method, aliphatic thiol is determined by measuring the absorbance of intensely colored [Cu(I)(phen)$_n$]$^+$ ions formed in the redox reaction. The proposed method can also be used for the determination of the total amount of aliphatic thiols, whose carbon numbers are less than 8, since their calibration curves have the same slope. The sensitivity of the proposed method is similar to that of Ellman method[4], which is widely used to spectrophotometrically determine water-soluble thiols, such as cysteine. The present spectrophotometric method is useful for the determination of aliphatic thiols in view of the simple operation and high precision.

References

1. V. B. Stein and R. S. Narang, *Anal. Chem.*, **54**, 991 (1982).
2. K. J. Rygle, G. P. Feulmer and R. F. Scheideman, *J. Chromatogr. Sci.*, **22**, 514 (1984).
3. Y. Nishikawa and K. Kuwata, *Anal. Chem.*, **57**, 1864 (1985).
4. G. L. Ellman, *Arch. Biochem. Biophys.*, **82**, 70 (1959).
5. A. Swatdiat and C. C. Tsen, *Anal. Biochem.*, **45**, 349 (1972).
6. T. Kamidate, A. Katayama and H. Watanabe, *Anal. Sci.*, **4**, 329 (1988).