

CHROM. 5805

## Oxidation of thiosulphate during chromatography in the presence of copper or gold ions

Thiocyanate formation from thiosulphate and cyanide is catalyzed by copper(II) or gold(III) ions<sup>1-3</sup>. With gold and with limiting concentrations of copper, only partial conversion of thiosulphate to thiocyanate occurs<sup>1-3</sup> and attempts were made to identify the other products of these cyanolytic reactions. This note reports the hazards encountered in attempting these analyses and shows that both copper and gold ions catalyze thiosulphate oxidation during paper chromatography.

### Experimental

Reaction mixtures (1-ml or 2-ml volumes) were prepared at 20° in small test-tubes and are described in detail in Tables I and II. Thiocyanate was determined colorimetrically by mixing samples (up to 5  $\mu$ moles of thiocyanate) with 3 ml of 1.5 *M* ferric nitrate in 4 *N* perchloric acid in 25-ml volumetric flasks, diluting to 25 ml with water and measuring the absorbance of the ferric thiocyanate at 460 nm in a Unicam SP600 spectrophotometer. Samples (0.01-0.02 ml) of the reaction mixtures were analysed by chromatography and electrophoresis. Paper chromatography was carried out at 20° on Whatman No. 1 paper with: (A), *n*-butanol-acetic acid-pyridine-water (30:6:20:24); or (B), *n*-butanol-acetone-water (2:2:1).  $R_F$  values obtained in descending separations with these solvents were: thiosulphate, 0.18 and 0.02; trithionate, 0.42 and 0.13; tetrathionate, 0.55 and 0.21; and thiocyanate, 0.81 and 0.71. Electrophoresis in Gelman electrophoresis equipment was run in 0.1 *M* citrate buffer, pH 4.9, at 500 V (30-50 mA) at 2°. The supporting media used were: (C), Whatman anion-exchange paper DE20; (D), Whatman anion-exchange paper ET20; (E), Gelman glass microfibre sheets; and (F), Gelman Sepraphore III strips. With the last medium, thiosulphate and tetrathionate migrated distances of 86 and 77 mm, respectively, in 30 min. Distances migrated (mm) by thiosulphate, trithionate and tetrathionate respectively, were 81, 76, 69 for glass microfibre (E) in 45 min; 62, 29, 19 on ET20 (D) in 60 min; and 73, 43, 32 on DE20 (C) in 45 min; thiocyanate migrated 57 mm on DE20. Sulphur compounds were detected by spraying with 8% (w/v) silver nitrate in 90% acetone. <sup>35</sup>S-regions were detected by scanning or by radioautography and counted with a Beckman Lowbeta II gas-flow counter or a Packard scintillation spectrometer using 0.8% (w/v) 2,5-bis(5'*tert.*-butyl benzoxalolyl-[2'])thiophene BBOT (CIBA) in toluene as scintillator.

Thiosulphate was in some cases estimated by titration with 0.01 *N* iodine solution<sup>2</sup>; formaldehyde solution was added to mixtures also containing cyanide to complex any free sulphite.

### Results and discussion

When reaction mixtures containing thiosulphate, cyanide and gold or copper ions were analysed chromatographically in system A, excellent agreement was obtained between the recovery of <sup>35</sup>S-labelled thiocyanate and the colorimetric estimation of thiocyanate (Table I). No loss or further cyanolysis thus occurred during chromatography. In mixtures in which partial cyanolysis had occurred with limiting cata-

TABLE I

APPARENT PRODUCTS OF REACTIONS OF THIOSULPHATE CATALYSED BY COPPER(II) AND GOLD(III)

(a) Final concentrations of  $\text{Na}_2^{35}\text{SSO}_3$ , 2.68 mM; KCN, 25 mM.  $\text{CuSO}_4$  and  $\text{NaAuCl}_4$  as indicated were added separately, mixed or added in succession with intermediate mixing. Chromatographic analysis with system A. Analysed 30 min after mixing.

Metal added (mM)		Products (as mM initial $^{35}\text{SSO}_3^{2-}$ )			
1st addition	2nd addition	Thiocyanate		Thiosulphate	Tetrathionate
		Colorimetric	$^{35}\text{SCN}$	$^{35}\text{S}$	$^{35}\text{S}$
—	—	0	0	2.68	0
Au (1)	—	0.26	0.27	0.53	1.87
Au (2)	—	0.58	0.51	0.28	1.89
Au (4)	—	1.20	1.10	0.22	1.36
Au (8)	—	ppt	0.10	0.80	1.74
Cu (1)	—	0.30	0.27	0.24	2.16
Cu (2)	—	0.62	0.57	0.31	1.80
Cu (4)	—	1.31	1.34	0.08	1.26
Cu (8)	—	2.69	2.58	0.00	0.09
Au (2); Cu (2)	—	1.20	1.12	0.17	1.39
Au (4); Cu (4)	—	1.74	1.68	0.07	0.93
Au (2)	Cu (4)	1.90	1.80	0.14	0.74
Au (4)	Cu (4)	1.74	1.71	0.10	0.87
Cu (4)	Au (4)	1.78	1.85	0.06	0.77

(b) 2.67 mM  $\text{Na}_2^{35}\text{SSO}_3$ , mixed with  $\text{CuSO}_4$  or  $\text{NaAuCl}_4$ , and chromatographed in system A after 10 min.

Metal added (mM)	Products (% of initial $^{35}\text{S}$ )		
	Thiosulphate	Tetrathionate	Others (origin)
—	100	0	0
Cu (0.5)	40.3	59.7	0
Cu (1)	6.4	93.6	0
Au (2)	16.3	67.9	15.8
Au (4)	35.2	36.4	28.4
Au (6)	41.0	27.9	31.1

lytic amounts of copper or gold ions, or when cyanide was absent from such mixtures, the predominant product (as well as or instead of thiocyanate) was apparently tetrathionate, when mixtures were analysed in system A (Table I). "Tetrathionate" was isolated by preparative-scale chromatography in system A from mixtures containing  $^{35}\text{S}$ -thiosulphate after reaction with copper or gold both with and without cyanide. Co-chromatography with authentic tetrathionate showed no difference in chromatographic mobility in systems A or B. The  $^{35}\text{S}$  in the "tetrathionates" underwent stoichiometric exchange with unlabelled thiosulphate, as would be expected from tetrathionate<sup>4,5</sup>; and treatment of them with cyanide converted them to thiosulphate and thiocyanate containing equal amounts of  $^{35}\text{S}$ , again as expected for tetrathionate. The apparent presence of tetrathionate in the cyanide-containing reaction mixtures was surprising as tetrathionate and cyanide should undergo spontaneous reaction to form thiocyanate<sup>2,6,7</sup>. Even in the presence of copper at relatively low pH, some cyanolysis of tetrathionate would have been expected. Failure to obtain progressive thiocyanate formation suggested that tetrathionate might only be formed during

chromatography. Moreover, copper and thiosulphate ions are known to form complexes in aqueous solution<sup>7</sup>, rather than to give rise to tetrathionate. These possibilities were tested in three ways. Firstly, it was established that the separation procedure determined the amount of each product (other than thiocyanate) recovered (Table II).

TABLE II

EFFECT OF SEPARATION METHOD ON APPARENT PRODUCTS OF REACTION OF  $\text{Na}_2^{35}\text{SSO}_3$  WITH COPPER AND CYANIDE

2.68 mM  $\text{Na}_2^{35}\text{SSO}_3$  was analysed by paper chromatography and electrophoresis 10 min after mixing with cyanide or copper as indicated. For details of separation methods (A-E), see text.

Additions (mM)	Products (% of total $^{35}\text{S}$ )								
	Thiosulphate			Tetrathionate			Thiocyanate		
	A	B	C-E	A	B	C-E	A	B	C-E
—	92	93	99	8	7	1	0	0	0
KCN(25)	93	92	99	7	8	1	0	0	0
KCN(25); Cu (1)	28	70	89	64	22	0	8	8	11
KCN(25); Cu (4)	0	42	55	54	21	0	46	37	45
Cu (1)	7	37	61	93	63	39	0	0	0
Cu (4)	5	16	48	95	84	52	0	0	0

The two paper-chromatographic systems differed markedly in the amount of tetrathionate indicated after reaction with copper, with or without cyanide. When cyanide was present, no tetrathionate was found after analysis by any of the electrophoretic systems: thiosulphate and thiocyanate accounted for all the initial  $^{35}\text{S}$ -thiosulphate. Without cyanide, tetrathionate occurred in all separation systems, although much less was produced during electrophoresis (Table II). Secondly, iodometric titration of mixtures of thiosulphate with gold or copper, with or without cyanide, indicated titratable thiosulphate to be present in mixtures yielding predominantly tetrathionate (and thiocyanate) on chromatography in system A. Thirdly, addition of excess copper to a mixture (*e.g.*, 10 mM of cupric sulphate to a mixture of 2.5 mM thiosulphate and 25 mM potassium cyanide) in which partial cyanolysis had been produced 30 min previously by limiting amounts (*e.g.* 1, 2 or 4 mM) of copper or gold resulted in immediate and complete cyanolysis of all the thiosulphate initially present. Such treatment would not effect primary cyanolysis of tetrathionate, and indicates that all the initial thiosulphate was available for cyanolysis but existed in some complexed form (with cyanide and the previously added copper or gold) in solution. Stepwise additions of small amounts of copper or gold similarly produced additive cyanolysis not compatible with reaction by tetrathionate (Table Ia).

In summary, it is the aim of this note to point out that chromatography can reliably be used to isolate thiocyanate from cyanide- and metal-containing mixtures, but that thiosulphate in such mixtures undergoes oxidation to tetrathionate during chromatography. Electrophoresis is apparently suitable for mixtures containing thiosulphate and gold or copper with cyanide, but not for mixtures lacking cyanide. Thiosulphate apparently exists in complexed form in copper and gold solutions and is

available for oxidation by iodine at low metal concentrations and for further reaction with cyanide, but the complexes oxidize during some separation procedures.

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