

While at pH 7, the initial, rate-determining, reduction reaction shows dependence on both the nitro and thio species, indicating that Eq. 4 may be simplified to

$$-d(\text{PNBA})/dt = k_4 (\text{PNBA}) (\text{SO}_3^{2-}) \quad (\text{Eq. 6})$$

to describe the S_N2 initial reaction which dominates at higher pH values.

The apparent heat of activation of the initial reaction between stoichiometric amounts of reactants (3 bisulfite:1 nitro compound) determined at pH 7 where S_N2 characteristics dominate gave the value 22 Kcal. mole⁻¹ (Fig. 7).

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Thioacetamide II

Substitute for Hydrogen Sulfide in the Quantitative Analysis of Medicinal Chemicals

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Assay procedures utilizing thioacetamide and hydrogen sulfide gas have been compared. The advantages of using thioacetamide are discussed.

THE PURPOSE of this study is to investigate the use of thioacetamide as a substitute for hydrogen sulfide in the analysis of medicinal chemicals. A survey of the literature concerning the use of thioacetamide as a substitute for hydrogen sulfide has been made by Jue and Huyck (1).

Swift and Butler (2) reported that since the rate of acid-catalyzed hydrolysis follows the equation

$$\frac{d\text{TA}}{dt} = -K \text{H}^+ \text{TA}$$

(where TA = thioacetamide), the value of K is 0.21 L. mole⁻¹ min.⁻¹ at 90° and 0.019 L. mole⁻¹ min.⁻¹ at 60°. The activation energy is 19.1 Kcal. per mole. These figures mean that in a solution which is 1 M in hydronium ion, thioacetamide would be half hydrolyzed at 100° in 100 seconds, at 60° in 36 minutes, and at 25° in 21 hours. In solutions of pH 10

or more, hydrolysis is quite rapid and yields both thioacetate ions and acetamide.

The relation of hydronium ion concentration to the time of precipitation of arsenic in 0.1 M thioacetamide was studied by Butler and Swift (3). Two kinds of reactions with thioacetamide in sulfide precipitations were proposed: (a) hydrolysis controlled reaction in which hydrogen sulfide or hydrosulfide ions are formed as intermediates; (b) a direct reaction of metal ions with thioacetamide itself. Thioacetamide is a more rapidly acting reducing agent than hydrogen sulfide since arsenic acid is reduced much faster with thioacetamide than with hydrogen sulfide. The rate of precipitation of arsenic with valence of three by thioacetamide was measured in solutions from pH 1.0 to 3.8 and was found to follow quantitatively the calculated rate of hydrolysis of thioacetamide to hydrogen sulfide.

Molybdenum in titanium alloys was determined by precipitation as sulfide with thioacetamide by McNerney and Wagner (4). Stoner and Finston (5) clearly separated uranium from 200 times its weight of bismuth by precipitating bismuth sulfide from solution. Such a separation is impossible with gaseous hydrogen sulfide without a long digestion period.

The precipitation of metal sulfides is governed by the solubility product relation as shown by

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Butler, *et al.* (6). If the solubility product is high, precipitation with thioacetamide may be incomplete due to insufficient hydrogen sulfide in the solution.

In volumetric analysis titrations, thioacetamide was used as a reagent for determination of silver in photographic fixing solutions by Bush, *et al.* (7). The solutions were made 0.1 to 1.0 *N* in sodium hydroxide and titrated with 0.01 *N* thioacetamide to an end point determined potentiometrically, using the silver sulfide electrode. This method made it possible to overcome the interference of thiosulfate and halide ions in the usual Volhard titration of silver

Interference of other divalent ions such as Cu⁺⁺, Cd⁺⁺, Zn⁺⁺, Co⁺⁺, and Ni⁺⁺ was prevented by adding ethylenediaminetetraacetic acid tetrasodium salt.

Using hydrogen sulfide, zinc can be precipitated completely at pH 3.0, or even less, but thioacetamide precipitated only a very little zinc at this pH (8). One remedy is to use a large excess of thioacetamide and perform the precipitation under pressure in a closed vessel. Hydrogen sulfide gas escapes faster than it is generated in the hydrolysis of thioacetamide solutions when they are heated. The solution of thioacetamide should not be boiled during

TABLE I.—ASSAY OF MERBROMIN BY PRECIPITATION WITH HYDROGEN SULFIDE GAS

No.	Weight of Sample, Gm.	Weight of Precipitate, Gm.	% Weight of Mercury
1	0.2611	0.0780	25.76
2	0.2690	0.0785	25.16
3	0.3391	0.1020	25.93
4	0.3201	0.0986	26.57
5	0.2987	0.0891	26.60
6	0.3467	0.1032	25.66
7	0.3303	0.0989	25.82
8	0.3316	0.1001	26.02
9	0.2850	0.0844	25.53
10	0.3110	0.0927	25.70
11	0.3288	0.0954	25.01
12	0.2706	0.0785	25.02
			308.78
			Mean = 25.73

$$S.D. = \sqrt{\frac{\sum(X_1 - \bar{X})^2}{n - 1}}$$

$$S.D. = \sqrt{\frac{2.9888}{n - 1}} = \sqrt{\frac{2.9888}{11}} = \sqrt{0.2717} = 0.5214$$

TABLE II.—ASSAY OF MERBROMIN BY PRECIPITATION WITH THIOACETAMIDE

No.	Weight of Sample, Gm.	Weight of Precipitate, Gm.	% Weight of Mercury
1	0.2711	0.0807	25.66
2	0.2990	0.0899	25.92
3	0.3226	0.0970	25.92
4	0.3642	0.1086	25.72
5	0.2615	0.0775	25.56
6	0.2908	0.0861	25.53
7	0.3122	0.0957	26.42
8	0.3364	0.1010	25.89
9	0.2846	0.0852	25.22
10	0.2916	0.0876	25.90
11	0.2778	0.0834	25.88
12	0.3289	0.0984	25.79
			309.41
			Mean = 25.78

$$S.D. = \sqrt{\frac{\sum(X_1 - \bar{X})^2}{n - 1}} = \sqrt{\frac{0.9277}{11}} = \sqrt{0.0833} = 0.2886$$

TABLE III.—ASSAY OF MERCURIC CHLORIDE BY PRECIPITATION WITH HYDROGEN SULFIDE GAS

No.	Weight of Sample, Gm.	Weight of Precipitate, Gm.	% Weight of Mercury
1	0.5122	0.4375	99.62
2	0.4968	0.4261	100.01
3	0.4664	0.3979	99.56
4	0.4912	0.4215	100.01
5	0.5244	0.4475	99.60
6	0.5218	0.4449	99.52
7	0.5190	0.4420	99.39
8	0.4758	0.4068	99.59
9	0.5324	0.4552	99.80
10	0.5010	0.4274	99.56
11	0.4883	0.4178	99.84
12	0.4439	0.3778	99.32
13	0.5103	0.4357	99.62
			1295.44
			Mean = 99.64

$$S.D. = \sqrt{\frac{\sum(X_1 - \bar{X})^2}{n - 1}} = \sqrt{\frac{0.5353}{12}} = \sqrt{0.04460} = 0.2112$$

TABLE IV.—ASSAY OF MERCURIC CHLORIDE BY PRECIPITATION WITH THIOACETAMIDE

No.	Weight of Sample, Gm.	Weight of Precipitate, Gm.	% Weight of Mercuric Chloride
1	0.4822	0.4110	99.49
2	0.4618	0.3940	99.59
3	0.4929	0.4198	99.39
4	0.5446	0.4645	99.54
5	0.5003	0.4268	99.54
6	0.5118	0.4361	99.44
7	0.4432	0.3789	100.00
8	0.4681	0.3995	99.60
9	0.4924	0.4198	99.49
10	0.5100	0.4348	99.50
11	0.4223	0.3610	99.76
12	0.3886	0.3315	99.80
13	0.4216	0.3590	99.39
14	0.4108	0.3508	99.69
			1394.22
			Mean = 99.58

$$S.D. = \sqrt{\frac{\sum(X_1 - \bar{X})^2}{n - 1}} = \sqrt{\frac{0.3868}{13}} = \sqrt{0.02975} = 0.1725$$

precipitation of metallic sulfides but should be as hot as possible without actually boiling.

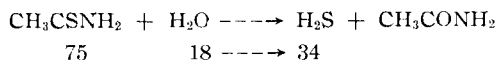
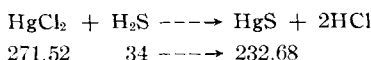
EXPERIMENTAL

A comparison was made of the methods and results of the quantitative analysis of merbromin N.F. (9) and mercury bichloride U.S.P. (10) for mercury content, using hydrogen sulfide gas and thioacetamide solution.

The hydrogen sulfide gas was generated by the conventional method of adding dilute acid to iron sulfide and then charging the generators after each precipitation. Sufficient commercial, reagent grade thioacetamide was dissolved in distilled water to make a 1 M or 7.5% w/v solution, and it was stored in a dark amber-colored bottle. The solution was heated to approximately 80° before use to hasten hydrolysis of thioacetamide. Otherwise, the analysis of mercuric chloride was carried out in accordance with the United States Pharmacopeia (10); the analysis of merbromin was carried out in accordance with the National Formulary (9, 11).

Identical analyses were conducted for each of the two chemicals described above (see Tables I-IV), using thioacetamide wherever hydrogen sulfide was called for. The amount of thioacetamide required for the reaction was calculated on the basis of its hydrolysis product, hydrogen sulfide:

For mercuric chloride



75 Gm. thioacetamide is needed to convert 271.52 Gm. of HgCl_2 to HgS , giving a gravimetric factor of $75/271.52 = 0.276$.

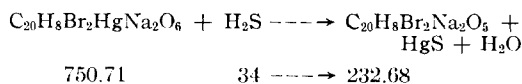
COMPARISON BETWEEN TABLES I AND II^a

	Table I	Table II
No. of analyses	12	12
Mean	25.73	25.78
S.D.	0.5214	0.2886
S.D.D.	$\sqrt{\frac{\sigma_1^2 + \sigma_2^2}{n_1 + n_2 - 2}} = \frac{0.5214 + 0.2886}{\sqrt{0.22}} = \frac{0.8100}{4.69} = 0.1727$	
Computing "t" from	$\frac{\bar{X}_1 - \bar{X}_2}{\text{S.D.D.}} = \frac{0.05}{0.1727} = 0.2895$	

$t_{0.01} 22 \text{ d.f.} = 2.819$

^a S.D. = standard deviation, S.D.D. = standard deviation of the difference, σ_1 and σ_2 = standard deviation of the arithmetic mean, \bar{X}_1 = mean of first group, \bar{X}_2 = mean of second group, and n = No. of analyses.

For merbromin



75 Gm. thioacetamide is required for 750.71 Gm. of merbromin, giving a gravimetric factor of $75/750.71 = 0.0999$.

A statistical evaluation (12) for the confidence limits for the two methods was made for both chemicals assayed. These tests indicate no significant difference of results between the use of hydrogen sulfide and thioacetamide in the analysis of mercuric chloride and merbromin.

COMPARISON BETWEEN TABLES III AND IV

	Table III	Table IV
No. of analyses	13	14
Mean	99.64	99.58
S.D.	0.2112	0.1725
S.D.D.	$\sqrt{\frac{\sigma_1^2 + \sigma_2^2}{n_1 + n_2 - 2}} = \frac{0.2112 + 0.1725}{\sqrt{25}} = \frac{0.3837}{5} = 0.0767$	
Computing "t" from	$\frac{\bar{X}_1 - \bar{X}_2}{\text{S.D.D.}} = \frac{0.06}{0.0767} = 0.3395$	
$t_{0.01} 25 \text{ d.f.} = 2.787$		

DISCUSSION

In the preceding analyses, it was found that thioacetamide could be used in place of hydrogen sulfide gas in the assay of merbromin and mercuric chloride. Results between the two methods agree fairly well. Thioacetamide solution is easier to use than hydrogen sulfide gas. Since, in the use of thioacetamide a moderate excess yielding, in turn, a moderate excess of hydrogen sulfide can be calculated quite accurately, there is very little danger of toxicity and hardly any unpleasant odor.

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