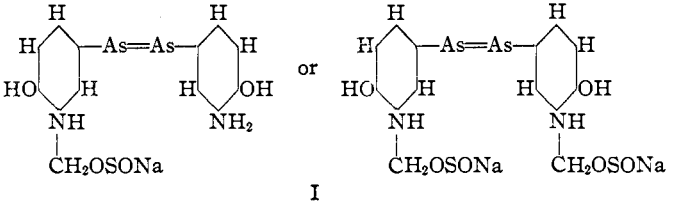
[CONTRIBUTION FROM RESEARCH LABORATORIES, E. R. SQUIBB & SONS]

STUDIES ON THE ANALYSIS AND CHEMISTRY OF NEOARSPHENAMINE[1]

BY ALFRED E. JURIST AND WALTER G. CHRISTIANSEN

RECEIVED SEPTEMBER 15, 1927 PUBLISHED JANUARY 5, 1928

Neoarsphenamine is officially described as a condensation product of 3,3′-diamino-4,4′-dihydroxyarsenobenzene (I)



I

and sodium formaldehyde sulfoxylate. Since it is not a pure crystalline substance but a complex mixture of varying composition, an accurate method of analysis is important in making a synthetic study. Investigation of the available methods showed that of Elvove[1a] most satisfactory. However, it was found necessary to make two modifications, and as a result of these two modifications of Elvove's method it is possible to gain a clearer insight into the composition and chemistry of neoarsphenamine than has been possible hitherto.

The Elvove method of analysis is based on a differential sulfur determination, chiefly on the assumption that the combined and free sodium formaldehyde sulfoxylate differ in their reaction to iodine in neutral[2] solution in that the free sulfoxylate only reduces iodine in neutral solution whereas both the free and combined sulfoxylate reduce iodine in alkaline solution. Freedman[3] calls attention to the fact that this is not correct and we have found that the combined sulfoxylate as well as the free sulfoxylate reduces iodine in neutral solution. This can be demonstrated by oxidizing neoarsphenamine with an excess of iodine, reducing the excess iodine with sodium arsenite, acidifying the solution and then precipitating the sulfur as barium sulfate instead of titrating the excess iodine with sodium thiosulfate as in the Elvove method. The sulfur thus determined directly as barium sulfate is usually greater than that calculated by the Elvove method.

Free sodium formaldehyde sulfoxylate reduces 4 atoms of iodine, whereas combined sodium formaldehyde sulfoxylate reduces only 2 atoms of iodine.

[1] This paper was presented before the American Chemical Society, Section on Chemistry of Mechanical Products, at Detroit, in September, 1927.

[1a] Elvove, *U. S. Pub. Health S. Repts.*, **40**, 1235 (1925).

[2] The P_H of neoarsphenamine solutions varies from 5.8 to 9.0.

[3] Freedman, *J. Lab. Clin. Med.*, **11**, 6 (1926).

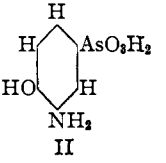
By applying these facts and the results obtained by the method just described, which we shall call the arsenite method to distinguish it from the Elvove direct titration, along with the Elvove titration two simultaneous equations may be set up, as follows

$$x + y = A; \frac{x}{0.8} + \frac{y}{1.6} = B$$

where x is the free sulfoxylate sulfur, y the combined sulfoxylate sulfur, A the sulfur found by the arsenite method less the free sulfate sulfur and B the iodine titration in neutral solution corrected for the arseno group iodine equivalent. These equations give directly both the free and combined sulfoxylate sulfur.

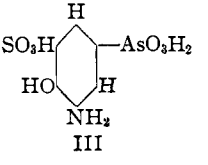
In addition to the free and combined sulfoxylate sulfur determinations, Elvove uses a free sulfate sulfur determination and a total sulfur determination. The difference between the total sulfur and the sulfur oxidizable by iodine in alkaline solution, the total sulfoxylate sulfur, is then considered to be sulfur present as sulfarsphenamine. It has been found that this sulfur difference represents not sulfarsphenamine but nuclear sulfur introduced in the preparation of the arsphenamine base from which the neoarsphenamine was prepared.

In order to demonstrate this, two preparations of arsphenamine base were made in which great care was taken to avoid the introduction of nuclear sulfur by starting with 3-amino-4-hydroxyphenylarsonic acid (II),



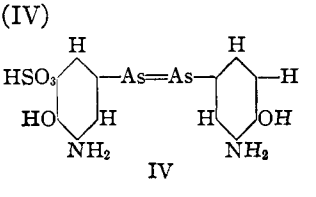
II

which has been shown by Christiansen[4] to yield a base low in nuclear sulfur, but in one of these preparations 10% of 3-amino-4-hydroxy-5-sulfophenylarsonic acid (III)



III

was substituted for the "amino" acid, yielding a base containing 10% of the sulfonic acid derivative (IV)



IV

[4] (a) Christiansen, THIS JOURNAL, 43, 2202 (1921); 44, 2334 (1922).

The two bases so obtained were analyzed for sulfur and arsenic and then converted into neoarsphenamines, using formaldehyde sulfoxylate which was known to be free from formaldehyde bisulfite. The neoarsphenamines were than also analyzed for total sulfur and sulfur oxidizable by iodine in alkaline solution. The results of the sulfur analyses of the neoarsphenamines and the bases from which they were made are as follows:

Sulfur content of base made free from sulfonic acid	1.51%
Sulfur content of base with 10% of sulfonic acid	2.31%
Calculated sulfur due to sulfonic acid	0.77%
Sulfonic acid sulfur found	.80%
Nuclear sulfur content of neoarsphenamine made from base free from sulfonic acid	.06%
Calculated sulfur content of neoarsphenamine made from base with 10% sulfonic acid	.53%
Found sulfur content of neoarsphenamine made from base with 10% sulfonic acid	.57%

These results serve to demonstrate conclusively the fact that the "sulfarsphenamine sulfur" of Elvove is actually nuclear sulfur.

A further addition to the methods given here permits the determination of the percentage composition of neoarsphenamine by utilizing the results of the differential sulfur determinations, the arsenic content and several factors as follows:

The per cent. of sulfoxylate sulfur × 3.69 = % of free sulfoxylate

The per cent. of combined sulfoxylate sulfur as mono-substituted arsenical ÷ 6.87 = % of mono-substituted arsenical

The per cent. of combined sulfoxylate sulfur as di-substituted arsenical ÷ 11.31 = % of di-substituted arsenical

(The per cent. of arsenic—the per cent. of arsenic as substituted arsenical) × 2.44 = % of free base

The per cent. of free sulfate sulfur × 4.44 = % of sodium sulfate

A large number of neoarsphenamines have been analyzed by the foregoing methods and the results so obtained are given in the following table, showing examples of some of the different types observed.

The results given here show a great difference between the results given by the Elvove method and by the arsenite method for determining free and combined sulfoxylate sulfur. In types A and B, the free sulfoxylate sulfur is always less by the arsenite method than by the Elvove method and the combined sulfoxylate sulfur always greater. In certain cases, namely, type C, the amount of combined sulfoxylate sulfur is small by the Elvove method, but still less by the arsenite method, there being none in two instances. This latter result is not a fault, however, in the method but rather an indication of the existence of a type of combination between arsphenamine base and sulfoxylate differing from that usually described, a probability which must be assumed since the Elvove and arsenite methods both

TABLE I

RESULTS OF ANALYSES

Number[a]	% Arsenic	% Total sulfur	% Sulfur oxidizable by iodine in alkaline solution (Elvove)	% Nuclear sulfur	% Free sulfate sulfur	% Free sulfoxylate sulfur (Elvove)	% Free sulfoxylate sulfur (arsenite)	% Combined sulfoxylate sulfur (Elvove)	% Combined sulfoxylate sulfur (arsenite)	% Non-sulfoxylate sulfur [3 − (7 + 9)]	% Sulfur required for mono-substitution calcd. on % of arsenic	% Free sulfoxylate	% Mono-substituted arsenical	% Di-substituted arsenical	% Free arsphenamine base	% Sodium sulfate	% Inert matter
Column no.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
A-1-P	21.53	6.14	6.03	0.11	0.48	3.42	1.49	2.13	3.87	0.19	4.59	5.37	56.32	0.00	8.26	2.13	27.92
A-2-P	20.84	6.80	5.81	.99	.64	3.73	1.96	1.44	3.54	.31	4.45	7.23	51.52	.00	10.37	2.84	28.04
A-3-P	22.06	6.76	6.50	.26	.77	3.54	1.57	2.19	3.93	.23	4.71	5.79	57.20	.00	8.88	3.42	24.71
A-4-P	19.53	6.88	6.61	.27	1.14	4.20	3.36	1.27	1.68	.43	4.17	12.40	24.45	.00	28.46	5.06	29.63
B-1-E	24.39	9.13	9.08	.05	.66	4.18	.46	4.24	7.43	.53	5.20	1.70	43.23	39.46	0.00	2.93	12.68
B-2-E	27.82	8.64	7.83	.81	.38	3.90	.34	3.55	7.11	.00	5.93	1.25	69.13	20.88	.00	1.69	7.05
B-3-P	19.53	8.88	8.57	.31	1.22	2.92	.00	4.43	6.43	.92	4.17	.00	27.80	39.99	.00	5.42	26.79
B-4-E	27.25	8.92	8.09	.83	1.19	2.98	.05	3.92	5.85	1.00	5.81	.19	83.97	0.70	.00	5.28	19.86
C-1-P	19.35	7.66	7.37	.29	.87	5.77	5.41	0.73	0.72	0.37	4.13	19.96	10.48	.00	38.78	3.86	26.92
C-2-P	20.24	8.07	7.92	.15	1.06	5.38	5.18	1.48	.41	1.27	4.32	19.12	5.97	.00	44.71	4.71	25.49
C-3-P	19.70	9.30	8.25	1.05	1.10	6.62	6.62	0.53	.00	0.53	4.20	24.43	0.00	.00	48.08	4.88	22.61
C-4-P	19.49	8.42	8.61	.00	0.78	7.05	6.79	.78	.00	1.04	4.16	25.06	.00	.00	47.56	3.46	23.92

[a] P = commercial product, E = experimental product.

sometimes give results for the combined sulfoxylate which are almost impossibly low.

The directly detectable difference between the two types of combined sulfoxylate sulfur is their reaction to iodine in neutral solution in which the usual or N-methylene type of combined sulfoxylate reduces but two atoms of iodine whereas the second type of combined sulfoxylate reacts in the same manner as free sulfoxylate and reduces four atoms of iodine. This latter type of combined sulfoxylate can be explained on the basis of a double salt of arsphenamine base and sulfoxylate similar to the metallic compounds of arsphenamine, such as the silver compound, due, perhaps, to reactions of secondary valences of the arseno linkage or the amino group. While this possibility has previously been expressed verbally to one of us, this is the first evidence presented to support this view and the results also show the possibility of the existence of both types of combined sulfoxylate in one product.

In addition to the differences between the amounts of free and combined sulfoxylate sulfur given by the Elvove method and the arsenite method, the latter shows the presence in neoarsphenamine of a type of sulfur hitherto unrecorded. This is here called the non-sulfoxylate sulfur and varies in these analyses from 0.19 to 1.27%. No information is available as to the nature of this sulfur but it probably has its origin in decomposition or reaction with the benzene ring of the sulfoxylate used in the preparation of neoarsphenamine. However, several preparations which were exposed to air contained large amounts of non-sulfoxylate sulfur, indicating that it may be an oxidation product. While the nature of this sulfur is not clearly indicated, it can be oxidized to sulfate by iodine in alkaline solution but not in neutral solution so that the amount can be determined by subtracting the total arsenite sulfur from the sulfur oxidized to sulfate in alkaline iodine solution.

In the foregoing data it will be noted that there is a wide range of variation in the nuclear sulfur from zero to 1.05%. This indicates clearly to what extent the variation in nuclear sulfur can occur with slight variations in the base, but in only four instances is this nuclear sulfur present in amounts greater than 0.31%.

The data showing the compositions of the neoarsphenamines illustrated disclose a wide range of variation in composition from products which have no sulfoxylate directly condensed on the amino group to products which contain more di-substituted arsenical than mono-substituted arsenical. The free sulfoxylate varies from zero to 25.00% and many of the products examined contain at least small amounts of arsphenamine base. All neoarsphenamines examined contain a small amount of sodium sulfate introduced as an impurity in the sulfoxylate used in their preparation.

Summary

The method of analysis given here leads to a fairly complete knowledge of the composition of neoarsphenamine and it has been found:

1. That the arsenite method of analysis gives a more accurate distribution of the sulfur in neoarsphenamine than has been previously possible.
2. That the so-called "sulfarsphenamine sulfur" is, in fact, nuclear sulfur.
3. That there are probably two types of combination between arsphenamine base and sodium formaldehyde sulfoxylate, one being of the N-methylene type, and the other resembling a double salt formation.
4. That there is a type of sulfur present in neoarsphenamine which owes its origin to some reaction or decomposition of sodium formaldehyde sulfoxylate but it is no longer present as sulfoxylate.

BROOKLYN, NEW YORK

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF BARNARD COLLEGE]

THE DIBROMIDES OF METHYLCOUMARIC AND METHYLCOUMARINIC ACIDS

BY MARIE REIMER AND MARION HOWARD

RECEIVED SEPTEMBER 28, 1927 PUBLISHED JANUARY 5, 1928

In connection with a study of *o*-methoxybenzalpyruvic acid it became necessary, for purposes of identification, to prepare α-bromo-*o*-methoxycinnamic acid (169–171°). As reactions for the preparation of this substance by the action of a 50% aqueous solution of potassium hydroxide on methylcoumaric acid dibromide, according to the directions of Perkin,[1] failed to give the substance he described, we have made a study of the action of bromine on methylcoumaric and methylcoumarinic acids and of the elimination of hydrogen bromide from the resulting compounds. Perkin[1] described two isomeric dibromo addition products, neither of which, however, he prepared in the pure condition. Fittig and Ebert[2] questioned Perkin's results, as they were able to obtain but one dibromo compound. Later investigators[3,4,5] were also unable to identify two products of the reaction, although Stoermer mentions an "impure methylcourmarinic dibromide" left in the filtrates. We find that the product of addition of bromine to methylcoumaric and methylcoumarinic acids is in each case a mixture of isomers which we have succeeded in separating and purifying; a dibromo acid melting, with vigorous decomposition, at 177° and, in smaller quantity, an isomeric acid melting, with very slight de-

[1] Perkin, *J. Chem. Soc.*, **39**, 418 (1881).
[2] Fittig and Ebert, *Ann.*, **216**, 146 (1883).
[3] Werner, *Ber.*, **39**, 27 (1906).
[4] Stoermer and Friemel, *Ber.*, **44**, 1838 (1911).
[5] Biilmann and Lund, *Ann. chim.*, [9] **18**, 263 (1922).