acid; reduction was apparent in a few seconds, a white crystalline precipitate forming in the course of a minute. After two or three minutes 5 cc. more of vanadous chloride and 2 cc. more of hydrochloric acid were added and after ten minutes 50 cc. of boiled water was added. The precipitate was filtered off, washed and dried. It weighed 1.2 g.; recrystallized as described above, it yielded 0.7 g. of pure product.

Dixanthyl.—Half a gram of xanthydrol was dissolved in 15 cc. of acetone; 3 cc. of *N* vanadous chloride and a few drops of hydrochloric acid were added. Reduction was rapid and after a few minutes a further addition of 3 cc. of vanadous chloride and 1 cc. of hydrochloric acid was made and then the mixture was diluted with boiled water, the crystalline precipitate filtered off and dried. There was thus obtained 0.38 g. of dixanthyl; m. p., 204–205°.

We wish to express our indebtedness to the Wolcott Gibbs Fund of the National Academy for a grant in aid of this investigation.

Summary

1. Mixtures of triphenylmethyl (hexaphenylethane) and triphenylmethyl sulfate in glacial acetic acid give reproducible potentials which accord with the usual electrochemical equations.
2. The single electrode potentials of triphenylmethyl, phenylxanthyl and phenyldimethoxyxanthyl have been measured by a titration method, in acetic acid and acetone solutions of varying acidity.
3. A method has been developed of estimating the activity of the hydrogen ion in non-aqueous solvents by means of oxidation-reduction cells involving tetrachloroquinone (chloranil).
4. The changes of the single electrode potentials of the free radicals with changes in hydrogen-ion activity of the solutions are in accord with an electrochemical equation which has been derived.
5. A new convenient method of preparing dissociated and undissociated ethanes directly from the carbinols is described.

CAMBRIDGE, MASSACHUSETTS

[CONTRIBUTION FROM THE DERMATOLOGICAL RESEARCH LABORATORIES]

MERCURY DERIVATIVES OF AZO DYES

BY A. PROSKOURIAKOFF AND GEORGE W. RAIZISS

RECEIVED MARCH 2, 1925 PUBLISHED JULY 3, 1925

Dimroth, who of early workers contributed most to the subject of organic mercury compounds, suggested the use of mercuric acetate for the mercuration of organic compounds. He demonstrated that mercuric acetate reacts readily with aromatic amines and phenols, one or more acetoxy-mercury groups entering the *ortho* or *para* position with respect to the amino or hydroxyl groups. He further found that mercury does not enter in the *meta* position to the amino or hydroxyl groups. Raiziss and Proskouriakoff[1] also showed that mercury takes a position either

---

[1] Raiziss and Proskouriakoff, THIS JOURNAL, **44**, 787 (1922).

para or *ortho* to an hydroxyl or a carboxyl group because in several instances, when the *para* and one or both of the *ortho* positions were already occupied by a substituent, mercury did not enter the ring. The assumption of Raiziss, Kolmer and Gavron[2] that the position of mercury in the molecule of the 3-nitro-4-hydroxyphenylarsinic acid, prepared by them, was *ortho* to the hydroxyl group, has been corroborated by Stieglitz, Kharasch and Hanke[3] who prepared the same substance and actually determined the position of mercury.

Attempts to prepare mercury derivatives of azophenols and azonaphthols by coupling mercurated naphthols and phenols with diazonium salts were successful only when the *para* position to the phenolic hydroxyl was unoccupied. In such cases the mercury in the *ortho* position remains intact and the formation of mercurated azophenols takes place. This method was used in preparing mercury derivatives of benzene-azophenol and benzene-azo-*p*-cresol.[4] By diazotizing *o*-mercuri-bis-*p*-aminobenzoic acid and *p*-acetoxymercuri aniline and by allowing them to interact with phenol and resorcinol, corresponding mercurated azo compounds were obtained.

The direct mercuration of azophenols and azonaphthols was studied but little. In the course of investigating the therapeutic value of organic mercury compounds, we obtained mercury derivatives of various dyes by direct mercuration.

When the sodium salt of hydroxy-azobenzenesulfonic acid (Tropaeoline Y) was treated with mercuric acetate, and the resulting product dissolved in sodium hydroxide and reprecipitated with methyl alcohol, a dihydroxymercury compound was obtained, probably because both *ortho* positions to the phenolic hydroxyl were open. $NaSO_3.C_6H_4.N{=}N.C_6H_2.ONa(4)(HgOH)_2(3,5)$.

Two hydroxymercury groups could also be introduced into a potassium salt of dihydroxy-azobenzenesulfonic acid (Chrysoine) by treating it with mercuric acetate in water or alcoholic solution. $KSO_3.C_6H_4.N{=}N.C_6H.(OH)_2(4,6)\ (HgOH)_2(3,5)$.

The sodium salt of β-naphthol-azobenzenesulfonic acid (Orange II) on being treated with mercuric acetate in aqueous solution and prolonged boiling also yields a dihydroxymercury compound. β-Naphthol-azobenzenesulfonic acid itself, however, on being treated in the cold with mercuric acetate in alcoholic solution yields a mercury compound of a different type. In this case an hydroxymercury group is evidently attached to one of the rings while the other mercury atom stands between the two salts forming sulfonic acid groups, thus forming mercuri-bis-(hydroxy-

---

[2] Raiziss, Kolmer and Gavron, THIS JOURNAL, **43**, 582 (1921).

[3] Stieglitz, Kharasch and Hanke, *ibid.*, **43**, 1185 (1921).

[4] Dimroth, *Ber.*, **35**, 2859 (1902).

mercury-β-naphthol-azobenzenesulfonic acid). $C_{10}H_5.OH.(HgOH).N{=}N.C_6H_4.SO_3{-}Hg{-}SO_3.C_6H_4.N{=}N.(HgOH).OH.C_{10}H_5$.

Another example of the formation of a compound of the above type is that obtained by mercuration of Alizarine Yellow R, which is nitrobenzene-azosalicylic acid.

This azo dye also contains a strong salt-forming group. When this dye is treated with mercuric acetate in water or alcoholic solution it yields a mercuri-bis(nitrobenzene-azosalicylic acid) with mercury standing between the two salt-forming carboxyl groups.

The azo dyes used for mercuration were prepared in our Laboratory and thoroughly purified until the analytical results corresponded to the calculated values.

Mercury analyses were made simultaneously with those for carbon and hydrogen by a method of Falkov and Raiziss.[5]

Toxicity tests were carried out by injecting white rats intravenously according to the method standardized by the Hygiene Laboratories at Washington, D. C. The bactericidal effect was determined by exposing bacteria to the chemical for one hour according to the method described by Raiziss and Severac.[6] Mercuric chloride and Mercurochrome, the biological properties of which are well known, were used for the purpose of comparison with our preparations.

TABLE I

TOXICITY WITH WHITE RATS AND BACTERICIDAL EFFECTS OF MERCURY DYES

Chemical compound	Mercury content %	Maximum tolerated dose	Lethal dose	Bactericidal dilution *Staphylococcus Aureus*	Bactericidal dilution *Bacillus Typhosus*
Mercuric chloride	73.89	0.002	0.003	1:114,000	1:118,000
Hydroxymercuridibromofluorescein (mercurochrome 220)	26.00	.032	.040	1:4,000	1:3,000
Dihydroxymercuri-azonaphtholbenzene-sulfonic acid	51.63	.030	.040	1:5,000	1:5,000
Mercuri-bis(nitrobenzene-azosalicylic acid)	26.07	..	.015	1:40,000	1:20,000
Dihydroxymercuri-dihydroxy-azobenzene-sulfonic acid	51.80	..	.008	....	....

The perusal of Table I indicated that the mercury dyes studied are in general considerably less toxic than mercuric chloride. There is no relationship between mercury content and toxicity. Mercuri-bis(nitrobenzene-azosalicylic acid) contains 26.07% of mercury and is more toxic than dihydroxymercuri-azonaphtholbenzenesulfonic acid which contains

---

[5] Falkov and Raiziss, THIS JOURNAL, **45**, 1000 (1923).

[6] Raiziss and Severac, *J. Lab. Clin. Med.*, **9**, No. 2 (1923).

51.63%. This indicates that mercury can be considerably detoxified by modifying the organic compound to which it is attached. It seems that the dye constituent, sulfanilic acid, contributes materially to the lowering of the toxicity, probably due to the presence of the sulfonic acid group which is known in chemotherapy as the group that reduces toxicity. While the naphthol constituent gives a low toxicity, resorcinol greatly increases the toxicity. In mercuri-bis(nitrobenzene-azosalicylic acid) the nitro groups and the salicylic acid component did not decrease the toxicity of mercury as much as the sulfanilic acid and the naphthol in the other compounds. The values indicating the bactericidal effect show that the mercury dyes studied are less powerful than mercuric chloride and more powerful than Mercurochrome. The results confirm the statements of other investigators that mercury, when bound to the nuclear carbon, is less toxic but is also less destructive in its action on bacteria. This finding is brought out particularly well with mercury dyes.

Experimental Part

Dihydroxymercuri-dihydroxy-azobenzenesulfonic Acid (Mercury Compound of Chrysoine).—Chrysoine (monopotassium salt) was prepared by coupling *p*-diazobenzenesulfonic acid with resorcinol dissolved in potassium hydroxide. On adding an excess of acetic acid to the alkaline solution of the formed dye the potassium salt was precipitated. This was purified by repeated crystallization from water until the values obtained in analysis corresponded to that calculated.

Anal. Calcd. for $C_{12}H_9O_5N_2SK$: N, 8.43. Found: 8.45.

Four g. of the compound described above was dissolved in 600 cc. of hot water and treated with 7.6 g. (2 molecular equivalents) of mercuric acetate dissolved in 300 cc. of water and acidified with 0.5 cc. of acetic acid. The solution was warmed on the water-bath for one hour, evaporated to a very small volume, the residue treated with alcohol and the substance reprecipitated with ether. The product obtained was thoroughly washed with boiling alcohol, dissolved in a cold 5% solution of potassium hydroxide and reprecipitated by treating with alcohol. The orange-brown compound was filtered off, washed with alcohol and ether and dried in a vacuum over calcium chloride. The same compound may be prepared by using alcoholic solutions of Chrysoine and mercuric acetate. In this case the mercury compound is precipitated at once.

Anal. Calcd. for $C_{12}H_9O_7N_2Hg_2SK$: C, 18.82; H, 1.12; N, 3.66; Hg, 52.41. Found: C, 18.91; H, 1.39; N, 3.61; Hg, 51.80.

It is an amorphous powder insoluble in common organic solvents but soluble in dil. sodium hydroxide solution.

Dihydroxymercuri-azonaphtholbenzenesulfonic Acid (Mercury Compound of Orange II).—The dye (sodium salt) was prepared by coupling *p*-diazobenzenesulfonic acid with β-naphthol, and thoroughly purified until a chemically pure product was obtained.

To prepare the acid dye, 5 g. of the sodium salt was dissolved in 50 cc. of hot water, the solution neutralized with 30 cc. of 10% hydrochloric acid, the precipitate filtered and washed with dil. hydrochloric acid, then dried at 80°, pulverized, washed with ether and dried again at 100°. It is soluble in water and also in alcohol.

Anal. Calcd. for $C_{16}H_{12}O_4N_2S$: N, 8.52. Found: 8.33.

Twenty g. of Orange II (sodium salt) was dissolved in 150 cc. of hot water. To this was added 36 g. (2 molecular equivalents) of mercuric acetate dissolved in 300 cc. of water and acidified with 10 cc. of glacial acetic acid.

The mixture was boiled under the reflux condenser for two hours, and evaporated in a vacuum to about 200 cc. Upon cooling, 1 liter of methyl alcohol was added and the compound precipitated with 2 liters of ether. Fifteen g. of crude compound was boiled for five minutes with 300 cc. of water, the water decanted, the precipitate washed with methyl alcohol by decantation, then boiled twice with 100 cc. of alcohol, filtered off, washed with ether and dried in a desiccator.

The mercurial compound is not soluble in common organic solvents but very slightly soluble in water and also in dil. hydrochloric acid when warmed. On being cooled the solution forms a thick, gelatinous mass soluble in dil. sodium hydroxide solution. More concentrated alkaline solutions, prepared by warming, become gelatinous when cooled.

Anal. Calcd. for $C_{16}H_{12}O_6N_2SHg_2$: C, 25.23; H, 1.58; N, 3.68; Hg, 52.74. Found: C, 25.35; H, 1.80; N, 3.85; Hg, 51.63.

Mercuri-bis(hydroxymercuri-β-naphtholazobenzenesulfonic Acid).—Two g. of Orange II was dissolved in 100 cc. of methyl alcohol, the solution treated with 3.6 g. (2 molecular equivalents) of mercuric acetate dissolved in 150 cc. of methyl alcohol acidified with a few drops of acetic acid. The bright red precipitate which appeared at once was filtered off, placed in 200 cc. of water which was boiled for 15 minutes, the water changed and the boiling repeated four times. Thus purified, the compound was washed with alcohol and ether and dried in the desiccator. It appears as a red, amorphous powder insoluble in common organic solvents but soluble in dil. sodium hydroxide solution.

Anal. Calcd. for $C_{32}H_{22}O_{10}N_4S_2Hg_3$: C, 29.80; H, 1.71; N, 4.34; Hg, 46.71. Found: C, 28.56; H, 2.00; N, 4.36; Hg, 46.56.

Dihydroxymercuri-hydroxy-azobenzenesulfonic Acid (Mercury Compound of Tropaeoline Y).—The dye was purified by recrystallization from water and gave analytical results corresponding to the chemically pure compound. Two g. of the dye was dissolved in 150 cc. of water. To this was added 4.24 g. (2 molecular equivalents) of mercuric acetate dissolved in 100 cc. of water acidified with 2 cc. of acetic acid. The solution was boiled for one-half hour. A fine precipitate appeared gradually, forming a suspension. To the cooled mixture, 500 cc. of ethyl alcohol was added which immediately changed the appearance of the precipitate. It then settled to the bottom of the beaker. It was thoroughly washed by decantation with ethyl alcohol, filtered off and dried in a vacuum. It appears as a brownish-red powder insoluble in common organic solvents but soluble in a dil. solution of sodium hydroxide.

Anal. Calcd. for $C_{12}H_{10}O_6SN_2Hg_2$: N, 3.93; Hg, 56.40. Found: N, 3.66; Hg, 57.46.

DISODIUM SALT.—The disodium salt of mercurated Tropaeoline Y is prepared by dissolving the mother compound in dil. sodium hydroxide solution, precipitating it with methyl alcohol, purifying it by boiling several times with alcohol and drying to constant weight in a vacuum over calcium chloride. The compound is insoluble in common organic solvents but soluble in water.

Anal. Calcd. for $C_{12}H_8O_6N_2SHg_2Na_2$: C, 19.07; H, 1.06; N, 3.71; Hg, 53.11. Found: C, 18.87; H, 1.46; N, 3.76; Hg, 53.64.

Mercuri-bis(nitrobenzene-azosalicylic Acid) (Mercury Compound of Alizarine Yellow R).—The dye was purified until the data of analyses corresponded to those for the sodium salt. To obtain the acid dye, 7 g. of the sodium salt was dissolved in hot water, the solution treated with 20 cc. of 10% hydrochloric acid, the precipitate filtered off, thoroughly washed with water and dried in the oven at 100°. It is soluble in hot ethyl alcohol.

Anal. Calcd. for $C_{13}H_9O_5N_3$: C, 54.35; H, 3.13; N, 14.63. Found: C, 53.89; H, 2.97; N, 14.26.

Two g. of Alizarin Yellow R (sodium salt) was dissolved in 150 cc. of hot water to which was added 2 g. of mercuric acetate dissolved in 75 cc. of water acidified with 3 cc. of glacial acetic acid. A voluminous precipitate appeared at once. The mixture was boiled for two hours, the precipitate filtered off and washed successively with water, alcohol and ether; the crude product was then boiled twice with 100 cc. of ethyl alcohol, washed with ether and dried in a vacuum over calcium chloride. The compound appears as a light brown powder readily soluble in dilute solutions of sodium hydroxide and sodium carbonate.

Anal. Calcd. for $C_{26}H_{16}O_{10}N_6Hg$: N, 10.87; Hg, 25.96. Found: N, 10.65; Hg, 26.07.

Mercuration of the dye in form of its acid was accomplished in the following manner. Three g. of nitrobenzene-azosalicylic acid was dissolved in 250 cc. of boiling ethyl alcohol to which was added 1.6 g. (0.5 molecular equivalent) of mercuric acetate dissolved in 100 cc. of ethyl alcohol acidified with 2 cc. of glacial acetic acid (temperature of mixture, 40°). A precipitate formed at once. It was allowed to stand for two hours. The compound was then filtered off, washed with alcohol and ether, purified by boiling for 25 minutes with 200 cc. of ethyl alcohol and dried in a vacuum over calcium chloride.

Analysis showed that the compound obtained had the same composition as that prepared from the sodium salt in water solution.

Summary

We have studied the preparation and some of the chemical properties of the following compounds.

Dihydroxymercuri-hydroxy-azobenzenesulfonic acid and its disodium salt, dihydroxymercuri-dihydroxy-azobenzene sulfonic acid (potassium salt), dihydroxymercury-β-naphtholazobenzenesulfonic acid, mercuri-bis-(hydroxymercury-β-naphtholazobenzenesulfonic acid), mercuri-bis-(nitrobenzene-azosalicylic acid).

We have pointed out the possibility of obtaining two types of compounds by direct mercuration of azo dyes containing salt-forming groups.

Instances are recorded confirming the theory of formation of dimercury derivatives of azophenols when both *ortho* positions to the phenolic hydroxyl are unoccupied.

The chemotherapeutic investigation of the above-mentioned mercury dyes shows that their toxicity is lower than that of inorganic mercury compounds; but the bactericidal effect is also lower.

PHILADELPHIA, PENNSYLVANIA