for an hour to ensure complete reaction. During this time the mass in the reaction flask became black and gummy.

The flask was allowed to cool to room temperature and then the excess of pentoxide was hydrolyzed by adding 30–60 cc. of water through the dropping funnel (air excluded). When hydrolysis was completed, the gaseous contents of the flask were swept into the carboy with 2 liters of dry nitrogen and the total gas analyzed for carbon dioxide and carbon monoxide.

The results are given in tabular form. The data for *n*-butyric, isobutyric, trimethylacetic and methyl-*t*-butylneopentylacetic acids, determined previously,[1,4] were included for purposes of comparison.

Acid	% CO	% CO_2
Primary		
n-Butyric	1.1	5.5
n-Caproic	0.9	6.3
Isocaproic	.8	.77
t-Butylacetic	.5	5.3
n-Caprylic	.14	9.9
Stearic	.0	13.0
Secondary		
Isobutyric	8.8	1.7
Diethylacetic	26.0	1.6
Methyl-*n*-propylacetic	21.7	1.6
Di-*n*-propylacetic	27.5	0.6
Diallylacetic	17.6	.5
Di-*n*-butylacetic	24.6	.5
Methylneopentylacetic	26.6	.8
Dineopentylacetic	30.0	.5
Tertiary		
Trimethylacetic	55.0	0.2
Ethyldimethylacetic	68.0	.0
Diethylmethylacetic	84.5	.6
Triethylacetic	77.5	.0
Methyl-*t*-butylneopentylacetic	90.0	.2

Summary

Fifteen aliphatic acids have been treated with phosphorus pentoxide at 150–160° and the yields of carbon monoxide and carbon dioxide recorded.

On the basis of the yield and composition of the gaseous products of this reaction a method of discriminating between acids having a primary, secondary or a tertiary alkyl group attached to the carboxyl carbon is proposed.

STATE COLLEGE, PENNA. RECEIVED JUNE 27, 1938

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[4] Whitmore and Laughlin, THIS JOURNAL, **56**, 1128 (1934).

[CONTRIBUTION NO. 384 FROM THE FOOD RESEARCH DIVISION, BUREAU OF CHEMISTRY AND SOILS, UNITED STATES DEPARTMENT OF AGRICULTURE, AT THE DEPARTMENT OF PHARMACOLOGY, STANFORD UNIVERSITY SCHOOL OF MEDICINE]

Studies on Phenothiazine. IV. Potentiometric Characterization of Thionol

BY FLOYD DEEDS AND C. W. EDDY

In a previous report[1] we have described a simple method for the conversion of phenothiazine to thionol (hydroxyleucophenthiazone). We have pointed out that thionol is a reversible oxidation–reduction system and reported the potential of the system under conditions when the ratio of the concentration of the oxidant to the concentration of the reductant was unity. This paper is concerned with details of the potentiometric studies of the system thionol–leuco thionol.

All oxidation–reduction potentials were measured with bare platinum electrodes, the saturated calomel half cell being used as the working standard of reference. Two platinum electrodes were used in each measurement and no potential reading was accepted as final until the two electrodes agreed within 0.1 mv. when the system was well poised. When the concentration of one component was low relative to that of the other, a discrepancy of not more than 0.5 mv. was considered indicative of equilibrium. Before using the electrodes they were cleaned in aqua regia, washed thoroughly, and their behavior checked in 0.001 *M* ferri–ferrocyanide solution in which the concentration of oxidant and reductant was equal. All potentials were measured with a Leeds and Northrup type K potentiometer placed in the grid circuit of a General Electric FP 54 tube as shown in the accompanying diagram.

Measurements were made in an air-bath at a temperature of 21–22°. Potentials were measured in an atmosphere of pure nitrogen which was passed over heated copper gauze to remove all traces of oxygen. The leuco base of thionol was prepared by reduction of thionol with platinized asbestos and a stream of hydrogen freed of oxygen by passage over heated platinized asbestos. When reduction was completed the solution of leuco base was freed of hydrogen with a stream of oxygen-free nitrogen. The stream of nitrogen also served to force the solution of leuco thionol through a sintered glass filter to remove the asbestos and to transfer the solution to an oxygen-free buret. Since the measured amount of solution of oxidant (10 cc.) placed in the titration vessel and the solution treated for reduction to the leuco base were aliquots of the same thionol solution, the ratio of oxidant to reductant after each addition of reductant from the buret was determined readily.

For the determination of the curve relating the potential to the ratio of reductant to oxidant a solution of thionol

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[1] F. DeEds and C. W. Eddy, THIS JOURNAL, **60**, 1446 (1938).

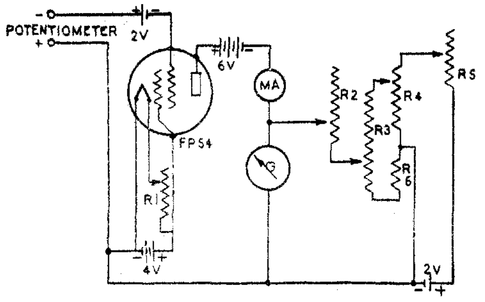

Fig. 1.—Diagram of static vacuum tube voltmeter: G, L. & N. galvanometer 0.025 m. a. per mm. scale division; R_1, 50-ohm rheostat; R_2, 200-ohm variable resistance; R_3, 700-ohm potentiometer; R_4, 400,000-ohm potentiometer; R_5, 500,000-ohm rheostat; R_6, 10,000-ohm resistance; MA. microammeter.

in a phosphate buffer of pH 6.57 was prepared by volatilizing a chloroform solution of thionol from the heated buffer during constant mechanical stirring. The concentration of thionol was approximately $M/4600$. The sigmoid curve showing the variation in potential from 0–75% reductant is given in Fig. 2, and the data obtained on the two platinum electrodes are given in Table I.

TABLE I

TITRATION OF 10 CC. OF BUFFERED THIONOL WITH BUFFERED LEUCO THIONOL [pH 6.57]

Leuco thionol, cc.	Reduction, %	E_h, obsd., v.	E_h, calcd., v.	Diff., v.
0.00		0.2462		
.50	4.76	.2277	0.2269	+0.0008
1.00	9.09	.2193	.2180	+ .0013
2.00	16.66	.2110	.2090	+ .0020
3.00	23.07	.2057	.2037	+ .0020
4.00	28.57	.2018	.2000	+ .0018
5.00	33.33	.1987	.1971	+ .0016
7.00	41.17	.1940	.1928	+ .0012
8.00	44.44	.1917	.1911	+ .0006
10.00	50.00	.1882		.0000
11.00	52.38	.1867	.1870	− .0003
13.00	56.52	.1839	.1843	− .0004
15.00	60.00	.1818	.1830	− .0012
20.00	66.66	.1762	.1793	− .0031
25.00	71.42	.1719	.1764	− .0045
30.00	75.00	.1677	.1740	− .0063
40.00	80.00	.1605	.1703	− .0098

It has been shown by Clark and Cohen[2] that at constant pH the relation of potential to the ratio of reductant to oxidant is given by the equation

$$E_h = E_0' - \frac{RT}{nF}\ln\frac{[Sr]}{[S_0]}$$

where E_h is the observed potential referred to the normal hydrogen electrode.

$$E' = E_h \text{ when } \frac{[Sr], \text{ the concentration of reductant}}{[S_0], \text{ the concentration of oxidant}} = 1$$

R is the gas constant.
T is the absolute temperature.
F is one faraday.
n is the number of electrons exchanged when one molecule of oxidant is changed to one molecule of reductant, or *vice versa*.

At a temperature of 21° this electrode equation may be given numerical form

$$E_h = E_0' - \frac{0.05833}{n}\log\frac{[Sr]}{[S_0]}$$

In the reports of Clark, including that on methylene blue, the value of n was found to be 2. Since thionol is very closely related to methylene blue, we have assumed $n = 2$ in our studies. Using this value we have solved the electrode equation for E_h for all values of the ratio $[Sr]/[S_0]$. Comparison of the calculated values with the observed is shown in Table I.

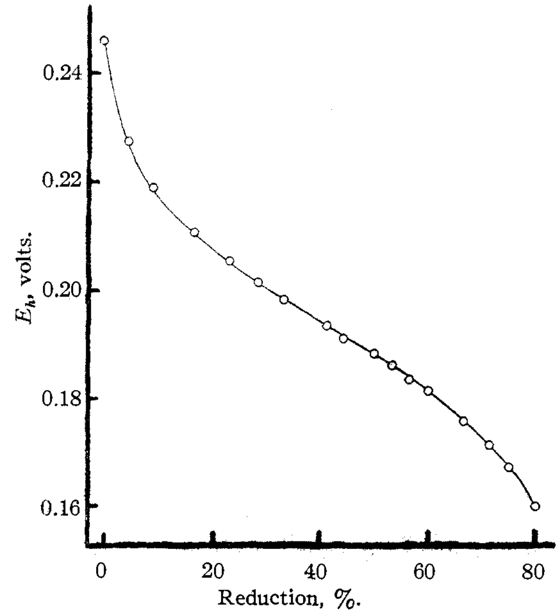

Fig. 2.—Relation between electrode potential and percentage reduction when pH is constant at 6.57: titration of thionol with leuco thionol at pH 6.57.

Comparison of the observed and calculated values for E_h shows that the difference is appreciable as the ratio $[Sr]/[S_0]$ becomes smaller or greater than unity. We do not believe that this error is due to the assumption that $n = 2$, but rather to the fact that leuco thionol is photosensitive. In other experiments to be reported in another paper, the agreement between observed and calculated E_h values was much closer, the maximum difference being 0.0011 v. in a single instance. The deviation reported does not invalidate the conclusion that thionol–leuco thionol is a reversible oxidation–reduction system nor does

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[2] W. M. Clark and B. Cohen, *U. S. Pub. Health Repts.*, **38**, 666 (1923).

it detract from the significance of the potential when $[Sr]/[S_0] = 1$ and the system is well poised.

The photosensitivity of the leuco thionol was demonstrated by the following experiment. A solution of thionol was reduced to the leuco base with platinized asbestos and hydrogen. A portion of the colorless solution was transferred to each of two oxygen-free Pyrex vessels. One of these was kept in the dark as a control for six days, and the other subjected to irradiation under a sun lamp. A pronounced red color developed in the irradiated vessel. At the end of six days the solution in the control vessel was colorless, but after two hours of exposure under the skylight in the laboratory a definite red coloration was noticeable. The rapidity with which the leuco thionol was converted to the red thionol under the influence of irradiation with the appropriate wave length of light suggested the feasibility of following the photosensitive action potentiometrically. At the same time advantage was taken of the opportunity to observe the effect of conversion of leuco thionol to thionol upon hydrogen ion concentration under strict anaerobic conditions.

A solution of thionol in distilled water was prepared and reduced to leuco thionol as previously described. The reduction vessel was then swept free of hydrogen with a stream of oxygen-free nitrogen which also served to force the solution of leuco thionol through a sintered glass filter into an oxygen-free vessel provided with a glass electrode for detecting potential changes due to alteration in *p*H and a bare platinum electrode for measuring of oxidation–reduction potential changes. The vessel was wrapped with black paper provided with a flap which could be raised for purposes of irradiation. The source of light for irradiation was an argon-filled glow lamp, which emits a wave length of light fairly efficient in converting leuco thionol to thionol. Throughout the course of the potential measurements the solution was stirred by a stream of oxygen-free nitrogen.

The time–potential curves of two such experiments are presented in Fig. 3. Since small increments of thionol induced by irradiation of leuco thionol produce large changes in the oxidation–reduction ratio, the irradiation in the early part of the experiment was limited to five-minute periods. Later when the rate of change of the ratio was small continuous irradiation was resorted to.

The change in potential registered by the bare platinum electrode is in harmony with the observation that as irradiation was prolonged the intensity of red color, due to increasing concentration of thionol, increased. The potential changes are due chiefly to a change in oxidation–reduction potential but not entirely so, for the solution was intentionally unbuffered to permit detection of *p*H changes. In the experiment at a temperature of 26° where the data are designated by circles the glass electrode registered a change of 64 mv., or somewhat more than one *p*H unit in the direction of increased acidity. At the same time the platinum electrode registered a change of 288.2 mv. The quantitative aspects of the experiment cannot be emphasized in the absence of exact knowledge of the molar concentration of the leuco thionol solution. The important points are the demonstration of the photosensitivity of leuco thionol, and the fact that conversion to thionol is accompanied by an increase in hydrogen ion concentration in unbuffered solutions. Clark, Cohen and Gibbs[3] have called attention to the photosensitivity of methylene white. These authors state: "We are not sure that the absence of visible coloration can be regarded as proof that there did not take place changes too small for positive identification but large enough to have a significant part in cumulative errors. Indeed, our titration curves often have the form which would result from the presence of very small percentages of oxidant in a solution treated as if it were completely reduced." Similarly, in the present study of thionol–leuco thionol inspection of Table I shows that the difference between the observed and calculated values of E_h might well be accounted for by the presence of small amounts of oxidant in the solution of reductant as the result of photodynamic action.

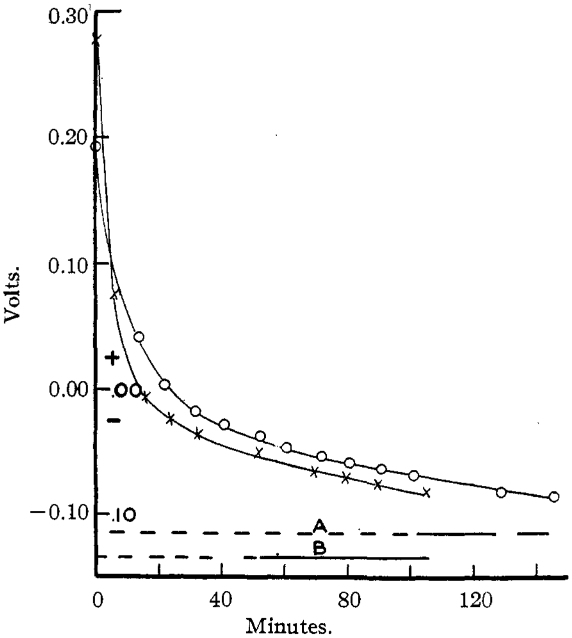

Fig. 3.—Time–potential curve showing change in oxidation–reduction potential due to photosensitivity of leuco thionol: A, length of dash indicates time of irradiation for experiment plotted with circles; B, length of dash indicates time of irradiation for experiment plotted with crosses.

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[3] W. M. Clark, B. Cohen and H. D. Gibbs, *U. S. Pub. Health Repts.*, **40**, 1131 (1925).

Because of the presence in the system thionol-leuco thionol of OH and NH groups, which may be capable of ionization, the system may be characterized further by a determination of the curve relating *p*H to the potential when $[Sr]/[S_0] = 1$. The curve was determined in the following manner. An approximately 0.2 *M* solution of phosphoric acid was titrated with an approximately 0.2 *M* solution of sodium hydroxide, and the change in *p*H followed with glass electrode measurements. The resulting curve relating *p*H to cc. of sodium hydroxide added was plotted on a large scale. From this curve the amount of sodium hydroxide required to bring a given amount of phosphoric acid to a given *p*H was determined. On the basis of such information a series of buffers was prepared and their exact *p*H values then determined with the hydrogen electrode. A chloroform solution of thionol was prepared by dissolving 0.1 g. in 100 cc. Five cc. of this chloroform solution was added slowly to 75 cc. of each buffer heated and mechanically stirred until all the chloroform was volatilized. The buffer solution of thionol was cooled and made up to 100 cc. with additional buffer. This procedure did not modify the *p*H of the buffers except in the case of the two most alkaline buffers used. The exact *p*H of these two buffer solutions of thionol was determined by duplicating the procedure in the absence of thionol and measuring their values with the hydrogen electrode. These buffers originally had values of 8.89 and 10.69 which were shifted to 8.70 and 9.90, respectively. A portion of each thionol solution was reduced as previously stated, and, under anaerobic conditions, 10 cc. of the reductant added to 10 cc. of the oxidant. The potentials were measured as before using two bare platinum electrodes. Final readings were recorded when the two electrodes were in substantial agreement. The data are given in Table II and presented in the form of a curve in Fig. 4.

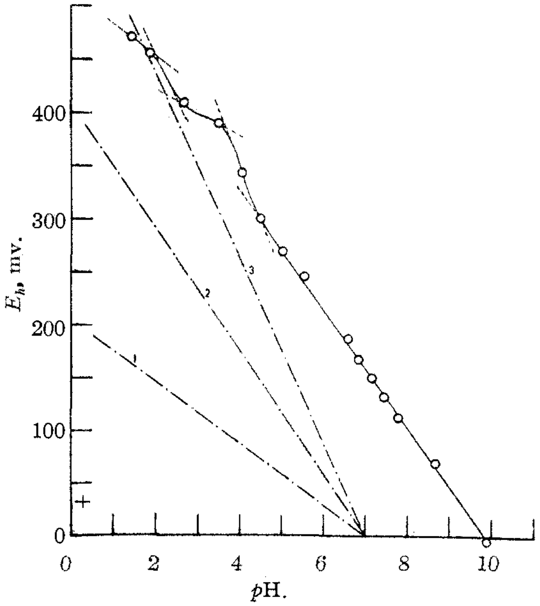


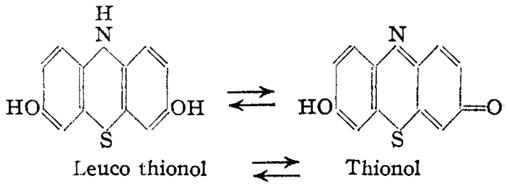
Fig. 4.—Relation between electrode potential and *p*H when total oxidant and total reductant are equimolecular: thionol, E_0' *vs.* *p*H.

Table II

Variation of E_0' with *p*H, $T = 21°$

*p*H	$E_h = E_0'$, v. Electrode 1	Electrode 2
1.42	+0.4712	+0.4725
1.85	+ .4573	+ .4579
2.65	+ .4105	+ .4105
3.47	+ .3917	+ .3917
4.03	+ .3450	+ .3450
4.47	+ .3021	+ .3023
4.99	+ .2710	+ .2710
5.53	+ .2482	+ .2477
6.57	+ .1882	+ .1882
6.84	+ .1687	+ .1687
7.14	+ .1515	+ .1515
7.44	+ .1333	+ .1333
7.78	+ .1132	+ .1132
8.70	+ .0700	+ .0700
9.90	− .0041	− .0038

The deflections of the curve in Fig. 4, in terms of ionizable groups, may be interpreted as follows. Each point on the curve designated by a circle represents the potential of a mixture of oxidant and reductant in equal concentrations at a definite *p*H value. As will be seen from the formula for leuco thionol there are two hydroxyl groups and an NH (3) capable of ionization. The formula for the oxidant shows that there is one hydroxyl group capable of ionization and a double bonded oxygen which may ionize if hydration occurs. For convenience of interpretation the broken lines 1, 2 and 3 radiating from a *p*H value of 7 in Fig. 4 represent slopes or −d*E*/d*p*H values of 29.2, 58.3 and 87.5 mv. per unit of *p*H, respectively, at 21°. It will be seen that between *p*H



values of 4.47 and 9.90 the observed potentials fall on a line, the slope of which approximates that of line 2. Between *p*H values 4.47 and 3.47 the potentials fall on a line approximately parallel to line 3. The potentials at *p*H values of 3.47 and 2.65 fall on a line approximately parallel to line 1. From *p*H 2.65 to 1.85 the slope of the curve once more approximates that of line 3. The dotted line extensions of the straight line portions of the curve give intersections corresponding to *pK* values of 3.16×10^{-3}, 2.24×10^{-4}, and 3.39×10^{-5}.

As has been shown in the papers by Clark and co-workers, deflections in the E'_0:*p*H curve denote the presence of detectable dissociations either in the oxidant or reductant, or in both. To determine whether a particular dissociation occurs in the oxidant or reductant these authors made use of a principle which Hall, Preisler and Cohen[4] stated as follows: "In the E'_0:*p*H curve, the bend with a convexity in the direction of negative e. m. f. always corresponds to a dissociation in the reductant; and, vice versa, a deflection with a concavity toward negative e. m. f. denotes a dissociation in the oxidant. That is, when the change in slope (defining slope as $-\mathrm{d}E/\mathrm{d}p\mathrm{H}$) is negative, the dissociation causing the change is assignable to the reductant; when the change is positive, the corresponding dissociation belongs to the oxidant." Therefore at *p*H 4.47 the dissociation is assignable to the reductant and likely due to the NH group. At *p*H 3.65 the change in slope involves two ionizable groups in the oxidant. Since the change in slope occurs suddenly both groups have the same or very nearly the same dissociation constant. At *p*H 2.5 the change in slope involves the two ionizable groups in the reductant. Here again the change in slope occurs suddenly, suggesting that both groups have the same dissociation constant, as might be expected from the symmetrical placement of the hydroxyl groups in the molecule.

Development of an equation relating the potential of the fixed ratio of thionol to leuco thionol with change in hydrogen ion concentration failed to yield a set of calculated E'_0 values showing satisfactory agreement with the observed values. This failure to correlate the physical chemical findings with the structural formulas for thionol and leuco thionol might be attributed to inaccurate *pK* values traceable to errors caused by the photosensitivity of the leuco thionol. However, the attempt to explain the deflections of the E'_0:*p*H curve requires the assumption that both hydroxyl groups of the reductant have the same *pK* value, and that the double bonded oxygen of the oxidant undergoes hydration and subsequent ionization. Therefore an explanation for the deflections of the E'_0:*p*H curve was sought on some basis other than ionizable groups.

The close relationship of thionol to α-phenazine and pyocyanine studied by Michaelis[5] and Friedheim and Michaelis[6] suggested the possibility that the system thionol–leuco thionol might be characterized by the existence of a semiquinone in the *p*H range from 0 to 4. Evidence for the existence of a semiquinone should be obtainable both colorimetrically and electrometrically.

Colorimetric tests for the presence of a semiquinone were made at *p*H values of 1.00, 1.21, 1.47, 1.85, 3.06, and higher. At *p*H values of 3.06 and higher reduction of thionol with stannous chloride, titanium trichloride, or platinized asbestos and hydrogen gas gave a change of color which proceeded directly from red to colorless, and was reversed directly to red by the addition of hydrogen peroxide. At the lower *p*H values of 1.00, 1.21, 1.47, and 1.85 reduction by the above methods gave a color change from red through purple to blue, light green and finally colorless. Reoxidation with hydrogen peroxide reversed the order of color change. The more acid the reaction the more pronounced the intermediate colors and the greater the ease with which partial reduction characterized by the intermediate colors could be maintained over a period of time. These observations are in conformity with the behavior to be expected in the presence of a semiquinone.

Attempts to confirm this colorimetric evidence with potential measurements have failed, a major difficulty being the instability of the potentials in the extreme acid range.

Nevertheless, the difficulties encountered in seeking an explanation of the deflections in the E'_0:*p*H curve do not invalidate the curve, which was found to be reproducible, nor do they detract from the usefulness of the curve as discussed in a succeeding paper.[7]

It is of interest to compare the potential of this

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[4] W. L. Hall, P. W. Preisler and B. Cohen, *ibid.*, Supplement No. 71 (1928).

[5] L. Michaelis, *J. Biol. Chem.*, **92**, 211 (1931).

[6] E. Friedheim and L. Michaelis, *ibid.*, **91**, 355 (1931).

[7] F. DeEds, C. W. Eddy and J. O. Thomas, *J. Pharmacol. Exptl. Therapeutics*, in press.

thiazine, thionol, with the values given by Clark[3] for two other thiazines, Lauth's violet and methylene blue. At *p*H 7 the E'_0 for thionol is 0.158, for methylene blue 0.011, and for Lauth's violet 0.062 at *p*H 6.967, and 0.045 at *p*H 7.517, which gives 0.061 at *p*H 7 by interpolation. These values place thionol appreciably closer to the oxygen electrode than the other two thiazines, being about midway between 1-naphthol-2-sulfonate-indophenol and 2,6-dichlorophenol-indo-*o*-cresol. This is in agreement with the observation that spontaneous oxidation of leuco thionol exposed to air occurs more slowly than is the case with methylene white at the same *p*H. Likewise, the reducing power of a given sample of urine is more readily demonstrated with thionol than with methylene blue.

Summary and Conclusions

1. The potentiometric characteristics of thionol have been determined.
2. Colorimetric evidence for the existence of a semiquinone has been submitted.
3. The three thiazines, methylene blue, Lauth's violet and thionol have been compared with regard to their position on the oxidation–reduction scale.

SAN FRANCISCO, CALIF. RECEIVED JUNE 15, 1938

[COMMUNICATION NO. 675 FROM THE KODAK RESEARCH LABORATORIES]

Oxidation Processes. XII.[1] The Autoxidation of Hydroquinone and of the Mono-, Di- and Trimethylhydroquinones

BY T. H. JAMES, J. M. SNELL AND A. WEISSBERGER

A study of the primary reaction in the autoxidation of hydroquinone is severely complicated by the presence of secondary reactions involving nuclear hydrogens. In order to avoid such complications, we investigated the autoxidation of durohydroquinone.[1] In this compound, the four nuclear hydrogens of hydroquinone are replaced by methyl groups. This prevents secondary reactions, which complicate the hydroquinone autoxidation. Hydrogen peroxide and duroquinone could be shown to be the reaction products, and it was further observed that duroquinone exerts a marked catalytic effect upon the reaction. In order to apply these results to the autoxidation of hydroquinone, we have investigated the autoxidation of the intermediate members of the homologous series, namely, toluhydroquinone, the three xylohydroquinones, and ψ-cumohydroquinone (trimethylhydroquinone) and we have extended previous observations on the oxidation of hydroquinone itself.

Materials[2]

***p*-Benzoquinone**, twice sublimed; m. p. 116°.

Hydroquinone, recrystallized from benzene, or three times from water slightly acidified with hydrochloric acid; m. p. 172°. No kinetical difference between these samples and the original material was detected.

Toluquinone, recrystallized from ligroin (b. p. 70–90°), and sublimed at 100°.

Toluhydroquinone, recrystallized from toluene; m. p. 127–128°.

Dimethylquinones were made from the corresponding xylidines, as follows.

The xylidine (61 g., 0.5 mole) was dissolved in 200 ml. of concentrated sulfuric acid and 1000 ml. of water, and a solution of 60 g. of sodium bichromate in 150 ml. of water was slowly run in, with stirring, at a temperature below 10°. The mixture was left standing in a cool place for twenty-four hours, and then 80 g. of bichromate in 240 ml. of water was added in the same manner. After another twenty-four hours, the precipitate was collected, washed with water, and the filtrate extracted with ether. The combined precipitate and ether solution was steam distilled. The distillate was half saturated with sodium chloride, cooled, the quinone collected, and the filtrate extracted with ether. The combined precipitate and ether extract residue was recrystallized from petroleum ether or ligroin and sublimed.

2,5-Dimethylquinone.—Yield 40%; m. p. 124–124.5°.

2,5-Dimethylhydroquinone was prepared according to Conant and Fieser[3]; recrystallized from ethanol, yield 46%; m. p. 210–212°.

2,6-Dimethylquinone.—Yield 10%; m. p. 72–73°.

2,6-Dimethylhydroquinone was prepared analogously to the 2,5-dimethylhydroquinone;[3] recrystallized from water (Norite), yield 60%; m. p. 153–154°.

2,3-Dimethylquinone.—From 3-amino-1,2-dimethylbenzene; yield 8%; m. p. 59–60°.

2,3-Dimethylhydroquinone.—The solution of the quinone (0.6 g.) in 25 ml. of 25% ethanol was reduced at 0° with sulfur dioxide for one hour; yield 17%; m. p. 224–225°.

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(1) Part XI: T. H. James and A. Weissberger, THIS JOURNAL, **60**, 98 (1938).

(2) All materials the origin of which is not stated are Eastman grades.

(3) Conant and Fieser, THIS JOURNAL, **45**, 2199 (1923).