155. *Polarographic Reduction of Some Heterocyclic Molecules. Part I. The Reduction of Cinchoninic Acid.*

By D. J. Casimir and L. E. Lyons.

Cinchoninic acid is reduced at the dropping-mercury cathode over the pH range 1—13. The mechanism differs with the pH but in every case a total of two hydrogen ions and two electrons is used for each molecule. Waves with $n = 2$ occur in both extremes of pH, but around the neutral point the reduction proceeds in two separate stages, each involving one electron. As the slopes of the waves on logarithmic analysis do not give simple values the theory of van Rysselberghe has been tested but found to fail. Acid conditions are most suitable for the determination of cinchoninic acid.

The polarographic reduction of quinoline-2- and -8-carboxylic acids has been studied by Stock (*J.*, 1944, 427; 1949, 586). The detailed mechanism still remains in some doubt. The behaviour of quinoline-4-carboxylic acid is now reported and discussed. All work was done on well-buffered aqueous solutions, at 25·0° generally, from pH 1 to pH 13; potentials are referred to the saturated calomel electrode at 25°. The concentration used was, unless otherwise specified, $2{\cdot}00 \times 10^{-3}$ M. The present work is part of a wider study which should complement that of Stock.

The reduction of cinchoninic acid gives waves over the entire pH range: at pH 1—8 a single wave, *A*, which at pH 8—11 is replaced by two waves, *B* and *C*, each half the height of *A*. At still higher pH's the curve is once more a single wave, *D*, similar to *A* in height. As the pH rises the reduction becomes more difficult and the curves are displaced to more negative potentials. Typical waves are shown in Fig. 1. The lowering of hydrogen overvoltage was observed, as is usual with quinoline derivatives (cf. Stock, *J.*, 1949, 763), but in this case was not sufficient to interfere with the reduction curve even at pH 1·3. In addition to the main waves pre-steps to *A* and *D* were observed (see below). Maxima were eliminated by the use of gelatin. These pre-steps occurred around pH 7 but only to a much less extent than with quinaldinic acid. Some minor aberrations from a smooth form were observed near the neutral point.

The half-wave potential, $E_{\frac{1}{2}}$, of wave *A* was a linear function of pH, and for pH <8 is given by $E_{\frac{1}{2}} = -(0{\cdot}550 + 0{\cdot}068\text{ pH})$ volts. This same equation applies to $E_{\frac{1}{2}}$ of wave *B* when pH is >8. Fig. 2 shows $E_{\frac{1}{2}}$-pH relationships for all waves. $E_{\frac{1}{2}}$ of *C* and *D* appeared independent of pH.

The wave-heights when plotted against pH showed that waves A and D were of approximately constant height (11·1 μa.) for pH 1—8 and pH >11 respectively, while B and C were constant at 5 3 μa. for pH 8—11, for the given capillary characteristics. In order to compare one compound with another examined on a different set-up it is necessary to eliminate the latter factor; consequently, a plot of pH against the wave-height divided by $m^{2/3}t^{1/6}$ is shown in Fig. 3, the wave-height having been corrected for variations in residual current and in drop-time variation with electrode potential. It is suggested that such a plot should become standard in future polarographic studies rather than a plot of pH against wave-height in μa.

FIG. 1.

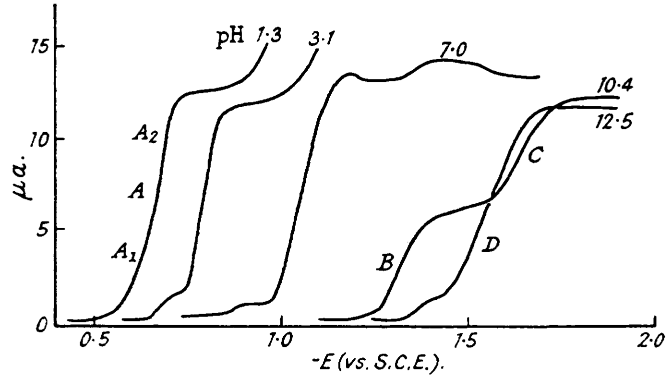


FIG. 2.

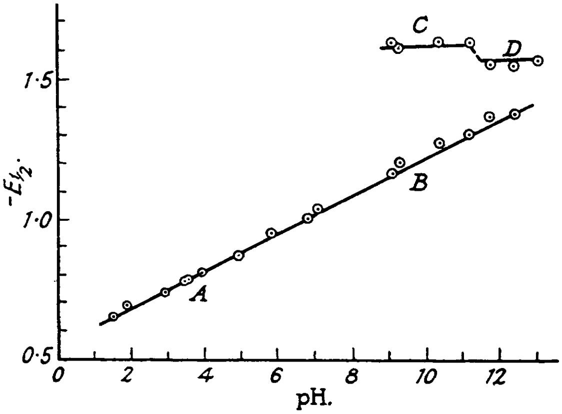
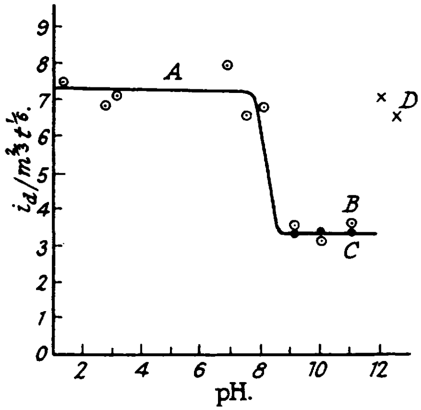


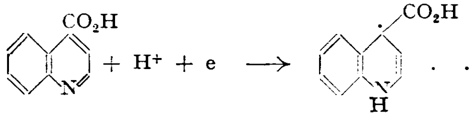
FIG. 3.



The number, n, of electrons per molecule concerned in the reduction process at each diffusion-controlled wave may be estimated from Ilkovic's equation $n = i_d/605D^{\frac{1}{2}}Cm^{2/3}t^{1/6}$, where i_d is the diffusion current in μa., D the diffusion coefficient in cm.^{2} sec.$^{-1}$, C the concentration of the reducible substance in millimoles/l., m the mg. of mercury dropping per second, and t the drop-time in seconds. Taking the value of D to be the same as that of quinaldinic acid, $0·80 \times 10^{-5}$ cm.^{2} sec.$^{-1}$ (Stock, *loc. cit.*), the following (nearest integer) values were obtained: A, $n = 2$; B, $n = 1$; C, $n = 1$; D, $n = 2$. With the reverse procedure, assuming the value of n to be exactly either 2 or 1, the mean value of the diffusion coefficient was $0·88$ ($\pm 0·04$) $\times 10^{-5}$ cm.^{2} sec.$^{-1}$, to be compared with quinoline-2-carboxylic acid $0·80 \times 10^{-5}$ (from conductivity data), quinoline-8-carboxylic acid $0·92 \times 10^{-5}$, and 8-hydroxyquinoline $0·85 \times 10^{-5}$ (both from polarography).

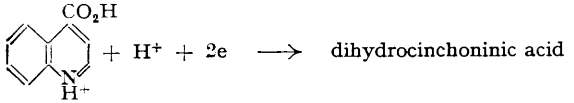
The plots of $\log i/(i_d - i)$ against E were generally linear for E near $E_{\frac{1}{2}}$, but the slopes varied from curve to curve. Thus for wave A slopes of 0·062, 0·070, 0·079, 0·082, 0·084, and 0·086 were obtained. Exclusion of a section of wave A which seemed possibly to overlie a pre-step lowered these values, *e.g.*, from 0·062 to 0·051. For a two-electron process the corresponding slope for a "reversible" reaction equals 0·030, and for a one-electron process 0·059. The discrepancy between 0·030 and the observed values might, it was thought, be explicable by the theory of van Rysselberghe (*J. Amer. Chem. Soc.*, 1946, **68**, 2047). This theory attempts to explain the departure of certain polarographic curves from the reversible reaction slope in terms of a Freundlich adsorption isotherm for the product of the electrode reaction. The theory is not applicable if the half-wave potential varies markedly with either temperature or concentration of the reducible substance. For cinchoninic acid, experimentally $E_{\frac{1}{2}} = 0{\cdot}795$ v. at 25°, 0·787 v. at 35°, and 0·793 v. at 45° (all compared to S.C.E. at 25°; $c = 2{\cdot}00 \times 10^{-3}$ M.; pH 3·55). Also $E_{\frac{1}{2}} = 0{\cdot}795$ v. for $2{\cdot}00 \times 10^{-3}$ M., and 0·792 v. for $1{\cdot}00 \times 10^{-3}$ M. (25°; pH 3·55). The value of $E_{\frac{1}{2}}$ in every case was obtained from a curve which had been corrected for residual current (cf. Delahay, *Rec. Trav. chim.*, 1948, **67**, 159) and for the iR drop across the cell (R being measured each time using A.C.). Thus the van Rysselberghe theory might be thought applicable. Under our conditions the theory gives rise to the relation $dE/di = 0{\cdot}051x$, where x = the number of molecules which react with two hydrogen ions and two electrons. The number of electrons reacting with each molecule reduced is thus $2/x$. Measurement of dE/di for wave A gave x values in eleven different cases ranging from 1·8 to 3·6 with an average of 2·6. It follows that n obtained from the Ilkovic equation differs from that obtained from the van Rysselberghe theory by a factor of more than two. Thus the theory seems not to be of general applicability.

The variation in ultra-violet absorption spectrum with pH has been studied for quinaldinic acid (Lyons, *Proc. Roy. Soc. N.S.W.*, in the press), the polarography of which resembles that of cinchoninic acid. From this and other evidence there is no doubt that at pH $\leqslant$ 4 the latter is present in the form containing H^+ on the nitrogen atom. Further evidence may be obtained from the polarography of 8-hydroxyquinoline (Stock, *loc. cit.*), where the $E_{\frac{1}{2}}$-pH plot of (Stock's) wave B shows an inflection at pH 5·3, although his larger wave A, which may be considered analogous to A of the present paper, shows no such inflection. Casimir and Lyons (unpublished) obtained such an inflection for quinoline itself, but here wave A seems to correspond to a one-electron process (resembling 8-hydroxyquinoline but not the 2- or 4-carboxylic acids). All this, and the fact that the $E_{\frac{1}{2}}$-pH plot (Fig. 2) for cinchoninic acid shows no inflection about pH 5, means that the first half (A_1) of A may *not* be identified in origin with B which is attributable to

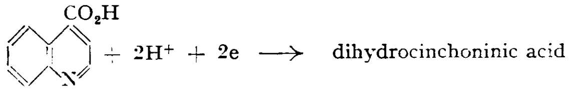


. . . . (1)

But to connect A_1 with the addition of only a single electron to $\equiv\mathrm{NH}^+$ as the potential-determining step is not valid as the half-wave potential of A_1 varies with pH. All that can be done at the present stage of theory is to associate the whole wave A with reaction (2*a*) when pH < p*K*, and with reaction (2*b*) when pH > p*K* < 8 ($K \equiv$ dissociation constant of the base).

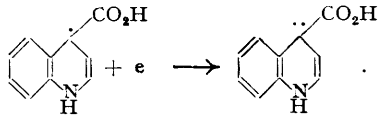


. . . . (2*a*)



. . . . (2*b*)

For pH > 8 < 11 reaction (2) is replaced by reaction (1), followed presumably by



. . . . (3)

which may then react with water to give the dihydro-compound. It may be possible to verify reaction (3) by theoretical calculations after the method of Maccoll (*Nature*, 1949, **163**, 178). Reaction (3) may be associated with wave C. The fact that $E_{\frac{1}{2}}$ of C is independent of pH supports the mechanism.

For pH >11 the potential-determining step must consist solely of the addition of two electrons, in order that $E_{\frac{1}{2}}$ of D be independent of pH, the product ion reacting to give the dihydro-compound. In each instance formation of the latter would be followed by di- or tri-merisation.

Comparative data (see table) for the first main wave of certain quinoline derivatives of quinoline show that a similar mechanism to the above may apply to quinoline-2- (Stock) and -4-carboxylic acid and 8-hydroxyquinoline (Stock), but not to quinoline-8-carboxylic acid (Stock) or to quinaldine or quinoline itself (Casimir and Lyons, unpublished).

Data for the first main wave of quinoline derivatives.

Derivative :	$2\text{-}CH_3$.	$2\text{-}CO_2H$.	$4\text{-}CO_2H$.	$8\text{-}CO_2H$.	8-OH.
n.	1.	2.	2.	1.	2.
$E_{\frac{1}{2}}$ at pH 0	0·52	0·52	0·59	0·78	0·95
$E_{\frac{1}{2}}$–pH slope	0·066	0·060	0·061	0·060	0·042

It is suggested that $E_{\frac{1}{2}}$ at pH 0 values, together with *n* values, might serve to distinguish the various compounds analytically. The best conditions for straightforward polarographic analysis exist in solutions of pH <3 where the wave is single and $E_{\frac{1}{2}}$ least negative.

Pre-steps were observed with both the waves *A* and *D* and in both cases occurred at potentials mostly independent of pH. [That to *A* occurred at 0·67 v. (pH 1·3 and 3·1); that to *D* at 1·36 v. (pH 11·8), 1·35 v. (pH 12·5), but 1·21 v. (pH 10·4).] The pre-step to *A* indicates adsorption of the ion, and that to *D* adsorption of the free base on the surface of the mercury drop (cf. Brolička, *Z. Elektrochem.*, 1942, **48**, 278), with the energy of adsorption utilised in the reduction process.

EXPERIMENTAL.

Buffer solutions used were made from the following solutions : pH 1·3 and 2·1 : 0·2N-KCl–0·2N-HCl; pH 3·1 and 4·0 : 0·05M-borax–0·05M-succinic acid; pH 4·9 : 0·1N-NaOH–0·1M-potassium hydrogen phthalate; pH 5·9 and 7·0 : 0·1N-NaOH–0·1M-KH_2PO_4; pH 7·2 : 0·05M-borax–0·2M-H_3BO_3; pH 8·2, 10·3, 10·6, 11·1, 11·7, and 12·4 : 0·1N-NaOH–0·1N-NaCl–0·1M-glycine; pH 8·8 and 9·0 : 0·1N-NaOH–0·1M-H_3BO_3. The cinchoninic acid (m. p. 252—253°) was prepared by H. C. Freeman, whom the authors thank. The apparatus of conventional design was manually controlled. The cell was connected by an agar–saturated potassium chloride bridge to a saturated calomel electrode, all in a thermostat keeping the temperature within ±0·01°. Purified hydrogen was used to remove dissolved oxygen. The characteristics of the capillary used for most of the work were $m = 1{\cdot}24$ mg./sec.; $t = 5{\cdot}66$ sec. (closed circuit; pH 3·08); ($m^{2/3}t^{1/6} = 1{\cdot}55$).

The 50-Ω coil resistance galvanometer was shunted and calibrated with a standard meter in series.

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[*Received, November 17th*, 1949.]