325. *The Polarographic Reduction of Some Heterocyclic Molecules. Part II. Coumarin and Certain Derivatives thereof.*

By A. J. Harle and L. E. Lyons.

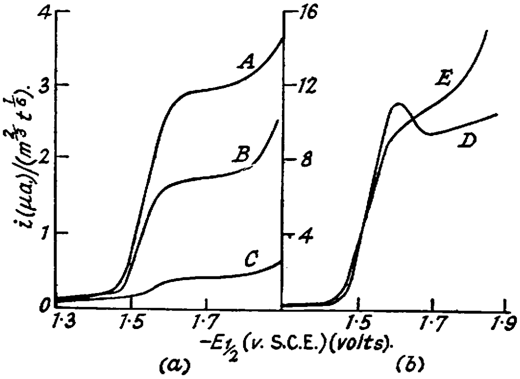
The polarographic behaviour and the absorption spectra of coumarin indicate that at pH $>11{\cdot}2$ it exists entirely in the lactone form and at pH $<6{\cdot}8$ entirely as coumarinic acid, both forms being present at intermediate pH. The reduction leads to *meso-* and racemic forms of tetrahydrodi-4-coumarinyl.

Coumarin is reported to be reduced in several ways. Zinc dust in alkaline solution gives two dicoumarins (Fries and Fickewirth, *Annalen*, 1908, **362**, 30); sodium amalgam in water gives melilotic acid (Zwenger, *Annalen*, Suppl., 1872, **8**, 32), and in alcohol gives hydrocoumaric acid. Dyson (*J.*, 1887, **51**, 61) studied various reductions but both his and Fries and Fickewirth's conclusions have been questioned by Assahini and Fujita (*J. Pharm. Soc. Japan*, 1919, **444**, 97). The present paper reports the first study of the polarographic reduction of the coumarin nucleus and gives a method which can be used to determine small amounts of coumarins.

The observed polarographic behaviour was simple in that for any pH where reduction occurred there was a single wave, the half-wave potential, $E_{\frac{1}{2}}$, of which was independent of pH. Maxima, which generally were not observed, appeared at a concentration of 0·005M. (see Fig. 1*b*) and could be removed by gelatin. The wave-height for a given concentration varied with pH as shown in Fig. 1*a*. Below pH 4 no wave was obtained because of the overlying hydrogen discharge.

FIG. 1.

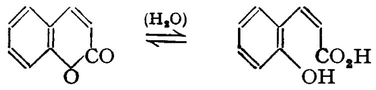
Polarographic reduction of coumarin.



Curve A : *pH* 6·82; $c = 1{\cdot}5 \times 10^{-3}$M.
B : *pH* 9·46; $c = 1{\cdot}5 \times 10^{-3}$M.
C : *pH* 10·20; $c = 1{\cdot}5 \times 10^{-3}$M.
D : *without gelatin* ; *pH* 6·45; $c = 5 \times 10^{-3}$M.
E : *with* 3 *drops of* 1% *gelatin* ; *pH* 6·45; $c = 5 \times 10^{-3}$M.

The ultra-violet absorption spectra of coumarin in aqueous buffers at a strength of $1{\cdot}5 \times 10^{-4}$ M. are shown in Fig. 2. It is seen that there is a gradual change in the character of the absorption from that at pH 7·56 [$\lambda_{\text{max.}}$ = (i) 310, (ii) 278 mμ.; $\varepsilon_{\text{max.}}$ (i) 6000, (ii) 10,500] to that at pH 12·18 [$\lambda_{\text{max.}}$ (i) 330, (ii) 226 mμ.; $\varepsilon_{\text{max.}}$ (i) 4900, (ii) 13,400]. Both the polarographic and the spectroscopic results are explicable on the assumption of a pH-variable interchange between the lactone (I) and the coumarinic acid (II) form of coumarin.

(I.)



(II.)

If the optical densities at the two maxima are plotted against pH (Fig. 3), a marked change in the character of the absorption is seen around pH 9·8. This pH agrees with that (9·3—10·3) at which the polarographic wave-height decreases markedly. Hence it is concluded that the lactone (I), which is from chemical evidence the stable form in acid media, is reducible over the range of potentials studied but that the acid (II) is not reducible over that range; and also that (I) is present to a negligible extent above pH 11·2 and (II) to a negligible extent below pH 6·8. For pH between 6·8 and 11·2 the polarographically estimated proportions of (I) and (II) are shown in Fig. 4 for 25° and 35°. Qualitatively similar figures may be estimated spectroscopically but these are in error owing to the impossibility of finding wave-lengths where the absorption is due exclusively either to (I) or to (II). By use of 240 mμ. to measure (II) and 280 mμ. to measure (I), the curve marked *Sp.* in Fig. 4 was obtained, on the assumption that no lactone is present at pH 11.

The effect of temperature on the relative amounts of (I) and (II) is not great. The pH at which most of the molecules change from (I) to (II) is similar at 35° and 25°, the slight difference observed being of the order of the experimental error, but it is seen that the percentage of (I) is reduced more rapidly with rising pH at 35° than at 25°.

The current at the top of the wave at pH 6·45 (at 25°) is controlled by the diffusion to the surface of the mercury drop, as i_d is proportional to the concentration in the bulk (Fig. 5). It is therefore possible to use these conditions for the quantitative determination of coumarin in small amounts, except in solutions containing a great excess of more easily reducible substances. The effect of temperature on the diffusion current is shown by the following

FIG. 2.

Absorption spectra of coumarin at pH stated.

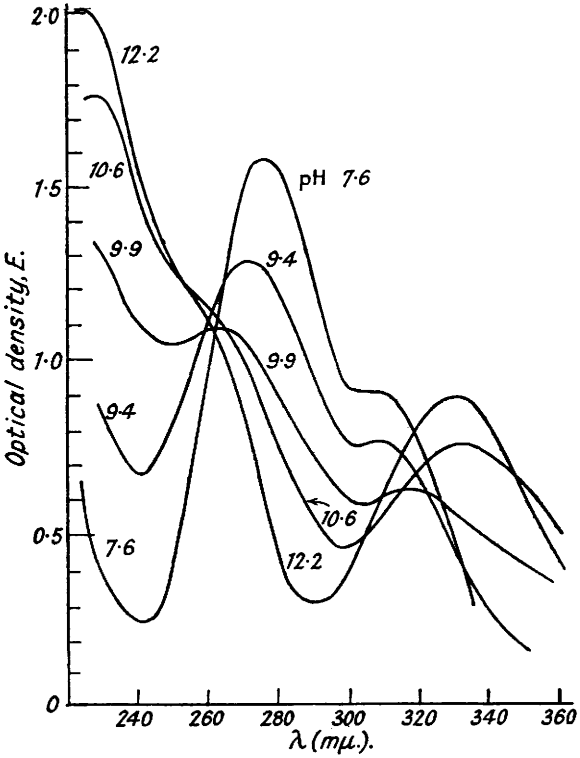


FIG. 3.

Dependence of the optical densities at absorption maxima (for coumarin) on pH.

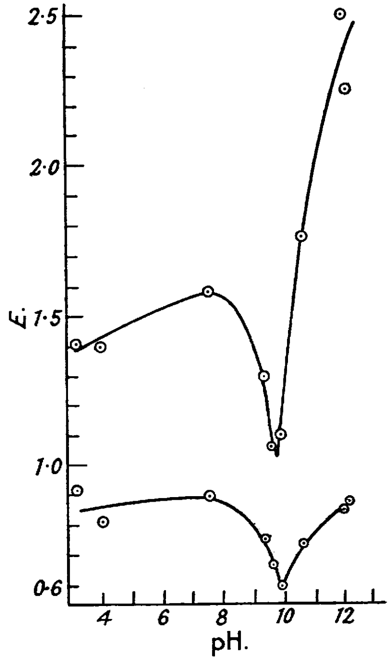


FIG. 4.

Percentage of lactone form (I) *present in coumarin solutions.*

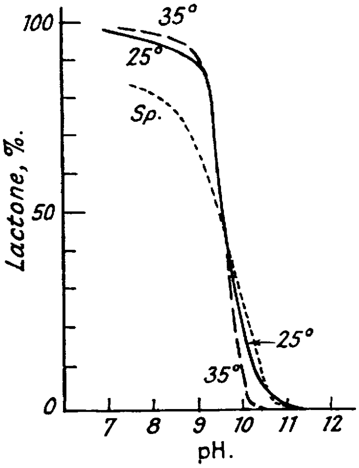
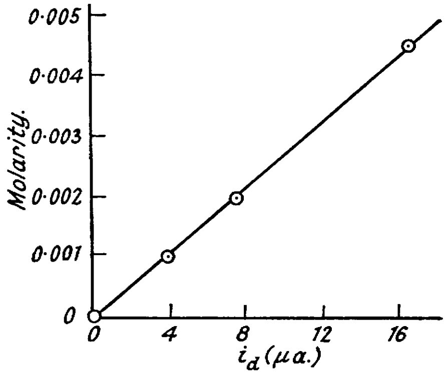


FIG. 5.

Relation of i_d *to molarity, for coumarin.*



values (μa.) at pH 6·65 : 25°, 4·92; 35°, 5·48; 45°, 6·58; these results are attributable to the increase in diffusion coefficient, D, with temperature, i_d varying as $D^{\frac{1}{2}}$ according to the Ilkovic equation, $i_d = 605\, ncD^{\frac{1}{2}}m^{\frac{2}{3}}t^{\frac{1}{6}}$, the symbols having the usual meaning quoted in Part I.

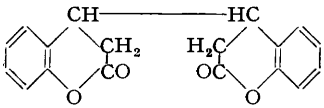
The number, n, of electrons concerned in the reduction process may be calculated, assuming a value of $0{\cdot}8 \times 10^{-5}$ cm.^{2} sec.$^{-1}$ for D (by taking the value for quinaldic acid in the absence of other information). A value of 1·02, *viz.*, 1, was obtained for n.

Evidence to support this value of n was obtained from the reciprocal slopes of the plots of log $i/(i_d - i)$ against E. For several polarograms the mean value of this reciprocal slope was 0·062 ($\pm$0·010), in agreement with the value of 0·059 expected for a reversible one-electron step (cf. 0·030 and 0·020, expected for 2- and 3-electron steps respectively).

$E_{\frac{1}{2}}$ was independent within experimental error of both pH and temperature. The value of $-E_{\frac{1}{2}}$ was 1·529, 1·551, 1·528, 1·523, 1·526, and 1·534 at pH 6·82, 7·63, 9·31, 9·46, 9·66, and 9·89 respectively, and for pH 6·6 at 35° was 1·526 and at 45° was 1·530 (versus S.C.E. at 25°). Corrections were made for residual current at every point and for the iR drop across the cell due to its resistance R.

The independence of $E_{\frac{1}{2}}$ of pH means that at least the potential-determining step in the reduction is free of H^+ participation, and so the step must consist in the addition of a single electron to the neutral molecule. If the correlation of the half-wave potential with the coefficient, a, of the molecular orbital resonance integral in the expression for the energy of the lowest unoccupied orbital be made (Maccoll, *Nature*, 1949, **163**, 178), the experimental value for a is 0·22, indicating that the first unoccupied level in the coumarin molecule lies below the corresponding level in the naphthalene molecule.

The product of the reduction was obtained in appreciable quantity by using an apparatus similar to that of Lingane (*J. Amer. Chem. Soc.*, 1943, **65**, 1348). The cathode potential was maintained at $-1{\cdot}7$ v., sufficient alcohol being added to a pH 6·8 buffer to dissolve 1 g. of coumarin in about 200 ml. of solution. The white product was precipitated (0·75 g.) as the reaction proceeded and a polarographic determination of coumarin in the resultant cathode solution showed that 99·8% of coumarin had been reduced. The amount of product unaccounted for was possibly still in solution, but was not detectable polarographically below $-$ 2·0 v. Several recrystallisations from glacial acetic acid revealed two difficultly separable components : A, less soluble, m. p. 280° (darkening above 235°), and B, m. p. 247° (darkening above 220°). Both A and B were insoluble in sodium carbonate solution but soluble in hot concentrated sodium hydroxide, and acidification of this solution gave a precipitate, thus indicating a lactone rather than an acid structure. Neither A nor B was optically active. Comparison of solubilities in alcohol, benzene, ether, acetone, and chloroform and of melting points of the separated compounds with those mentioned in the literature suggested that A and B were isomers $C_{18}H_{14}O_4$, *tetrahydrodi-4-coumarinyl* (Found, A : C, 72·4; H, 4·7; B : C, 72·9; H, 4·9. $C_{18}H_{14}O_4$ requires C, 73·5; H, 4·8%).



According to Assahini and Fujita (*loc. cit.*) two coumarin nuclei are united at position 4, although there is some doubt about this. A and B are assigned the annexed formula (*meso*- and racemic forms).

The mechanism of the polarographic reduction suggested by the results is :

$$R + e \longrightarrow R\cdot^- \qquad (1)$$

$$R\cdot^- \ (+H^+) \longrightarrow RH\cdot \qquad (2)$$

$$2RH\cdot \longrightarrow R_2H_2 \qquad (3)$$

where R represents the lactone coumarin. Step 1 is proved by the pH-invariance of $E_{\frac{1}{2}}$, and step 3 is supported by the formula of the product and the fact that isomers are obtained. Step 2 is the necessary intermediate. Whether $R\cdot^-$ reacts with H_3O^+ or H_2O is indefinite.

As well as coumarin, four naturally occurring derivatives were examined at pH's where the lactone form predominated. A single wave was given as with coumarin. The polarographic results for a concentration of 2×10^{-3}M. are :

	$-E_{\frac{1}{2}}$ (corr.).	$i_d/m^{\frac{2}{3}}t^{\frac{1}{6}}$.
7-Hydroxy-8-methoxycoumarin	1·594	2·74
7 : 8-Dimethoxycoumarin	1·590	2·60
7-Methoxy-6-dimethylallylcoumarin	1·668	1·84
8-Methoxy-7-geranyloxycoumarin (collinin)	1·577	1·85

EXPERIMENTAL.

The apparatus and buffer solutions used were similar to those described in Part I. Capillary characteristics were (a) $m = 1{\cdot}808$ mg. sec.$^{-1}$, (b) $m = 2{\cdot}583$ mg. sec.$^{-1}$. The coumarin was recrystallised four times from aqueous alcohol (m. p. 67·5°). The resistance, R, of the cell was measured by a " Philoscope." The coumarin derivatives were kindly supplied by Mrs. J. Ewing and Mr. F. Anet.

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