497. *Melanin and its Precursors. Part III.* New Syntheses of 5 : 6-Dihydroxyindole and its Derivatives.*

By JOHN D. BU'LOCK and JOHN HARLEY-MASON.

As previously reported briefly (*Nature*, 1950, **166**, 1036), β-3 : 4-dihydroxyphenyl-*N*-methylalanine and 2-3′ : 4′-dihydroxyphenylethylmethylamine have been converted directly into 5 : 6-dihydroxy-1-methylindole by oxidation with potassium ferricyanide and isomerisation of its resulting quinones with zinc acetate. Similar treatment of β-3 : 4-dihydroxyphenylalanine leads to the formation of 5 : 6-dihydroxyindole.

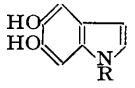
Oxidation of β-3 : 4-dihydroxyphenylalanine ethyl ester with potassium iodate gives a red crystalline iodo-quinone, and a similar iodo-quinone is obtained from 2-3′ : 4′-dihydroxyphenylethylmethylamine. Both products are isomerised by zinc acetate.

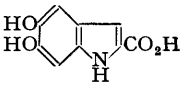
The mechanism of the zinc-catalysed isomerisation is discussed.

THE formation of the melanin intermediates 5 : 6-dihydroxyindole (I; R = H) and its 2-carboxylic acid (II) by enzymic oxidation of tyrosine has been established (Raper, *Biochem. J.*, 1927, **21**, 89; Dulière and Raper, *ibid.*, 1930, **24**, 239); the products were isolated as their *O*-methyl derivatives, which have not been successfully demethylated. (I) and (II) have since been synthesised by another and longer route (Beer, McGrath, Robertson, and Woodier, *J.*, 1949, 2061).

* Part II, *J.*, 1951, 703.

It appeared probable that these compounds should be obtainable from the primary oxidation product of tyrosine, 3 : 4-dihydroxyphenylalanine, by oxidation to the red quinone * (III)


(I.)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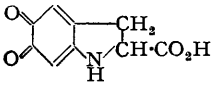
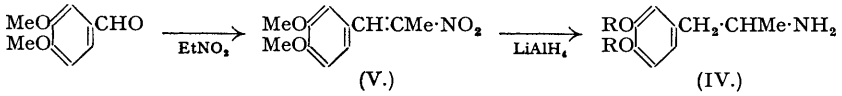

(III.)

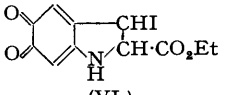
followed by isomerisation. Oxidation by means of silver oxide had already been examined by Dulière and Raper (*loc. cit.*) who isolated the two *O*-methyl derivatives mentioned above. We have now found that oxidation is best effected by potassium ferricyanide, and that the quinone thus obtained is isomerised and simultaneously decarboxylated to give (I; R = H) by the action of zinc acetate. This use of zinc acetate was prompted by Fischer, Dérouaux, Lambot, and Lecomte's observation (*Bull. Soc. chim. Belg.*, 1950, **59**, 72) that adrenochrome is converted by this reagent into 3 : 5 : 6-trihydroxy-1-methylindole. This catalysed isomerisation of 2 : 3-dihydroindole-5 : 6-quinones is, in fact, general and of considerable preparative value : its mechanism is discussed below. The yields of (I; R = H) obtained by this process were, however, variable and rather low, apparently owing to oxidation or polymerisation of the product in the dilute solutions used. Much better yields were obtained by application of a similar procedure to β-3 : 4-dihydroxyphenyl-*N*-methylalanine, leading to the formation of 5 : 6-dihydroxy-1-methylindole (I; R = Me). Similar treatment of 2-3′ : 4′-dihydroxyphenylethylmethylamine (" epinine ") also gave (I; R = Me) in good yield.

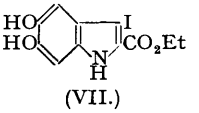
Oxidation of 2-3′ : 4′-dihydroxyphenylethylamine and 2-3′ : 4′-dihydroxyphenyl*iso*propylamine (IV; R = H) was next examined in the hope of obtaining 5 : 6-dihydroxyindole and its 2-methyl derivative. In both cases, however, only very small amounts of the indoles were formed, and the method was of no preparative value. (IV; R = H) was first obtained by Mannich and Jacobsohn (*Ber.*, 1910, **43**, 194). We adopted a more convenient synthesis in which veratraldehyde was condensed with nitroethane to give 1-(3 : 4-dimethoxyphenyl)-2-nitropropene (V), which was then reduced with lithium aluminium hydride to the amine (IV; R = Me). Demethylation with hydrobromic acid then gave the dihydroxy-amine (IV; R = H).

Although its structure is hardly in doubt, the quinone (III) has in fact never been isolated, since it is very soluble in water and the solutions decompose on concentration, and moreover it is not extracted from water by organic solvents. It seemed probable that a derivative of (III) containing a heavy substituent might be less soluble and thus more readily isolated. By the action of potassium iodate on β-3 : 4-dihydroxyphenyl-L-alanine ethyl ester we have obtained crystalline, sparingly soluble ethyl 2 : 3-dihydro-3-iodoindole-5 : 6-quinone-2-carboxylate (VI). On treatment with zinc acetate or alkali, isomerisation to ethyl 5 : 6-dihydroxy-


(V.) (IV.)

3-iodoindole-2-carboxylate (VII) occurred readily, indicating that the iodine atom did not occupy the 2-position in the quinone. This isomerisation also took place on storage of the solid material; the deep red crystals slowly became brown and recrystallisation then gave (VII). Reduction of (VI) gave a colourless leuco-compound which readily re-oxidised to the


(VI.)


(VII.)

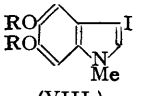
quinone. The preparation of (VI) starting from the racemic rather than the L-ester was much less satisfactory. The entrance of the iodine atom creates a new asymmetric centre, so that from the L-ester two diastereoisomers could be formed. The material isolated in this case appeared to be homogeneous, so that it is probable that in fact an asymmetric synthesis under

* This compound is frequently referred to as " hallachrome " on the basis of its supposed identity with the red pigment isolated from the polychæte worm *Halla parthenopoea* (Mazza and Stolfi, *Arch. Sci. biol.*, 1931, **16**, 182). This identity has now been disproved (Bu'Lock, Harley-Mason, and Mason, *Biochem. J.*, 1950, **47**, xxxii) and the alternative trivial name " dopachrome " is proposed for (III).

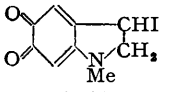
the influence of the neighbouring asymmetric atom takes place in the iodination, leading to a single product. In the case of the racemic ester, two products must necessarily be formed and this would make crystallisation more difficult.

Treatment of epinine with potassium iodate also led to the formation of a crystalline iodoquinone, which on treatment with acetic anhydride and pyridine gave a colourless diacetoxyiodo-1-methylindole, m. p. 145—146°, differing from 5 : 6-diacetoxy-2-iodo-1-methylindole, m. p. 153—155° (Bergel and Morrison, *J.*, 1943, 48). Our product was therefore the corresponding 3-iodo-compound (VIII; R = Ac), and the iodo-quinone was 2 : 3-dihydro-3-iodo-1-methylindole-5 : 6-quinone (IX). The action of zinc acetate on (IX) gave 5 : 6-dihydroxy-3-iodo-1-methylindole (VIII; R = H).

The results obtained from the oxidation of ten 2-3′ : 4′-dihydroxyphenylethylamine derivatives, including four from our earlier work, indicate the scope of the reaction and are collected in the Table. Inspection reveals that, in general, better results were obtained with secondary amines; with the primary amines examined the failures or poor yields appeared



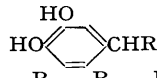
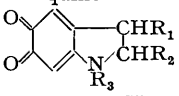
(VIII.)



(IX.)

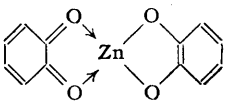
in most cases to be due to unsatisfactory cyclisation in the formation of the dihydroindole-quinone. This may be correlated with the basicity of the nitrogen atom which will be higher in the secondary amines, thus facilitating the cyclisation which involves anionoid attack on the *o*-benzoquinone nucleus formed as the first stage in the oxidation (cf. Robinson and Sugasawa, *J.*, 1932, 789; Bu'Lock and Harley-Mason, *J.*, 1951, 712).

The action of zinc ions in catalysing the rearrangement of dihydroindole-quinones is apparently unique, and no analogies could be found : its mechanism was therefore of particular interest. Oxidation of catechols in aqueous solution in the presence of zinc acetate led to the formation of insoluble, dark green complexes derived from two molecules of the catechol and one atom of zinc. These complexes are decomposed by acids and are clearly similar to those obtained by Criegee and Klonk (*Annalen*, 1949, **564**, 1) by the oxidation of " ene-diol " compounds in the presence of alkaline-earth metal hydroxides, and those obtained by Michaelis and Granick (*J. Amer. Chem. Soc.*, 1948, **70**, 624) by the alkaline reduction of 9 : 10-phenanthraquinone in the presence of the same metal ions. They are therefore formulated as in (X); as shown, one ring is benzenoid and the other *o*-quinonoid, although in fact each ring is at an

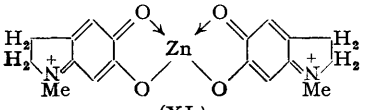
β-3 : 4-Dihydroxyphenylethylamine derivative 	Oxidising agent.	2 : 3-Dihydroindole-5 : 6-quinone 	Yield (quinone or isomerisation product).
$R_1 = R_2 = R_3 = H$ (Hydroxytyramine)	Ferricyanide	$R_1 = R_2 = R_3 = H$	Trace
	Iodate		Nil
$R_1 = R_3 = H$; $R_2 = Me$	Ferricyanide	$R_1 = R_3 = H$; $R_2 = Me$	Trace
	Iodate		Nil
$R_1 = R_2 = H$; $R_3 = Me$ (Epinine)	Ferricyanide	$R_1 = R_2 = H$; $R_3 = Me$	60%
	Iodate	$R_1 = I$, $R_2 = H$, $R_3 = Me$	50%
$R_1 = R_3 = H$, $R_2 = CO_2H$ (β-3 : 4-Dihydroxyphenylalanine)	Ferricyanide	$R_1 = R_3 = H$, $R_2 = CO_2H$	5—30%
	Iodate		Nil
$R_1 = R_3 = H$, $R_2 = CO_2Et$ (β-3 : 4-Dihydroxyphenylalanine ethyl ester)	Iodate	$R_1 = I$, $R_2 = CO_2Et$, $R_3 = H$	30%
$R_1 = H$, $R_2 = CO_2H$, $R_3 = Me$ β-(3 : 4-Dihydroxyphenyl-*N*-methylalanine)	Ferricyanide	$R_1 = H$, $R_2 = CO_2H$, $R_3 = Me$	40%
$R_1 = OH$, $R_2 = R_3 = H$ (Noradrenaline)	Ferricyanide	$R_1 = OH$, $R_2 = R_3 = H$	Nil
	Iodate	$R_1 = OH$, $R_2 = I$, $R_3 = H$	16%
$R_1 = OH$, $R_2 = H$, $R_3 = Me$ (Adrenaline)	Ferricyanide	$R_1 = OH$, $R_2 = H$, $R_3 = Me$	62%
	Iodate	$R_1 = OH$, $R_2 = I$, $R_3 = Me$	65%
$R_1 = OH$, $R_2 = H$, $R_3 = Pr^i$ (Aludrine)	Ferricyanide	$R_1 = OH$, $R_2 = H$, $R_3 = Pr^i$	40%
	Iodate	$R_1 = OH$, $R_2 = I$, $R_3 = Pr^i$	20%
$R_1 = OH$, $R_2 = Me$, $R_3 = H$ (Corbasil)	Ferricyanide	$R_1 = OH$, $R_2 = Me$, $R_3 = H$	20% *
	Iodate		44%

* As quinone semicarbazone.

equivalent intermediate state of oxidation and the complexes should properly be regarded as derived from two molecules of the semiquinone.



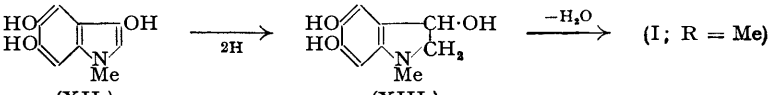
(X.)



(XI.)

Complexes of this kind could be formed from 2 : 3-dihydroindole-5 : 6-quinones and zinc ions as in the example (XI); in these cases the presence of the positive charges would render the complex water-soluble. Spontaneous decomposition could occur by release of protons from the 2- and 3-positions of a dihydroindole nucleus, resulting in isomerisation to a 5 : 6-dihydroxyindole.

Another method for the preparation of 5 : 6-dihydroxy-1-methylindole was investigated. This consisted of the reduction of 3 : 5 : 6-trihydroxy-1-methylindole (XII), easily obtained from adrenaline *via* adrenochrome, with Raney nickel–aluminium alloy and aqueous sodium hydroxide. Since it was found that the same reagents reduce indole to 2 : 3-dihydroindole,



(XII.) (XIII.) (I; R = Me)

the reaction must involve reduction to leuco-adrenochrome (XIII) followed by dehydration. That this dehydration occurs with remarkable ease has already been shown by Harley-Mason (*J.*, 1950, 1276).

Experimental.

Oxidation of β-3 : 4-Dihydroxyphenyl-N-methylalanine.—To a solution of the amino-acid (0·2 g.) and sodium hydrogen carbonate (0·1 g.) in water (5 c.c.), a solution of potassium ferricyanide (1·3 g.) and sodium hydrogen carbonate (0·3 g.) in water (15 c.c.) was added with stirring. After 10 minutes the deep red solution thus obtained was treated with 20% zinc acetate solution (8 c.c.), and the mixture shaken vigorously. During 5 minutes the red colour changed to grey, and a little sodium dithionite was added to prevent oxidation. The precipitated zinc ferrocyanide was filtered off and washed. The filtrate was extracted with peroxide-free ether (4 × 10 c.c.), the ethereal solution separated and dried over alkali-free sodium sulphate, and the solvent removed. Recrystallisation of the residual gum (benzene–light petroleum) gave 5 : 6-dihydroxy-1-methylindole (60 mg., 40%), m. p. 136°.

Similar oxidations were performed with β-3 : 4-dihydroxyphenylalanine in place of the *N*-methyl compound. In one experiment a yield of 30% of 5 : 6-dihydroxyindole, m. p. 140°, was obtained, but this could not be repeated and it appeared that the solid light brown residue obtained after removal of the ether was polymeric, and only very small amounts of the required product could be obtained from it by high-vacuum sublimation or solvent extraction.

Oxidation of L-β-3 : 4-*Dihydroxyphenylalanine Ethyl Ester.*—L-β-3 : 4-Dihydroxyphenylalanine (2·2 g.) was suspended in ethanol (50 c.c.) and saturated with dry hydrogen chloride without cooling. The solvent was removed under reduced pressure, and the residue dried in a desiccator over phosphoric oxide and potassium hydroxide, giving the ethyl ester hydrochloride as a glass which could not be crystallised. To a solution of the ester (2·2 g.) in water (10 c.c.), a saturated aqueous solution of potassium iodate (2·5 g.), followed by *n*-butanol (20 c.c.), was added. The mixture was gently agitated for about 40 minutes, during which the product crystallised out in the butanol layer, which was then separated and filtered, and the residue washed with water, methanol, and ether. *Ethyl 2 : 3-dihydro-3-iodoindole-5 : 6-quinone-2-carboxylate* (VI) (1·0 g.) formed red needles, m. p. 127° (decomp.), very sparingly soluble in water and slightly soluble in ethanol, giving a deep red solution. Light absorption in ethanol: $\lambda_{max.}$ = 2980 and 4900 A.; $\varepsilon_{max.}$ = 8330 and 1710 (Found: C, 38·5; H, 2·9; I, 36·7. $C_{11}H_{10}O_4NI$ requires C, 38·0; H, 2·9; I, 36·7%).

Isomerisation of the Iodo-quinone (VI).—(a) *Spontaneously.* A specimen of (VI) which had been kept for 6 months had become light brown. Fractional crystallisation from ethyl acetate–light petroleum gave *ethyl 5 : 6-dihydroxy-3-iodoindole-2-carboxylate* (VII) as small colourless plates, m. p. *ca.* 140° (decomp. and varying with rate of heating, with softening at 100°) (Found: C, 36·4, 36·9; H, 3·9, 3·2. $C_{11}H_{10}O_4NI,H_2O$ requires C, 36·2; H, 3·3%).

(b) *With alkali.* Addition of one drop of 2N-sodium hydroxide to a solution of (VI) in aqueous ethanol caused immediate decolorisation. The solution was at once acidified with acetic acid and extracted with ether. Removal of the solvent left a gum which gave (VII) on recrystallisation from ethyl acetate–light petroleum.

(c) *With zinc acetate.* When the quinone (VI) was stirred with 10% zinc acetate solution, the suspension of red crystals slowly became grey. Extraction with ether, filtration from dark undissolved material, and removal of the ether left a gum from which a small amount of (VII) was isolated by recrystallisation as above.

Reduction of (VI).—A solution of (VI) in aqueous ethanol was rapidly decolorised by addition of sodium dithionite. Extraction with ether and removal of the solvent left a gum which was recrystallised from ethyl acetate–light petroleum, giving *ethyl* 2 : 3-*dihydro*-5 : 6-*dihydroxy*-3-*iodoindole*-2-*carboxylate*, colourless needles, m. p. 103° (decomp.) (Found : C, 37·7; H, 3·4. $C_{11}H_{12}O_4NI$ requires C, 37·8; H, 3·4%); its ethanolic solution rapidly became red on exposure to air and the absorption spectrum was then identical with that of (VI).

1-(3 : 4-*Dihydroxyphenyl*)*prop*-2-*ylamine*.—A solution of veratraldehyde (8 g.), nitroethane (4 c.c.), and ammonium acetate (2 g.) in acetic acid (30 c.c.) was refluxed for 2 hours and then poured into water. The precipitated 1-(3 : 4-dimethoxyphenyl)-2-nitropropene formed yellow plates from ethanol, m. p. 73°.

The nitro-compound (6 g.) was placed in a small Soxhlet thimble and extracted under reflux into a solution of lithium aluminium hydride (4·1 g.) in ether (200 c.c.), boiling being continued for 6 hours. The resulting ethereal suspension was decomposed with a concentrated solution of sodium potassium tartrate, the ethereal layer separated and dried, and the solvent removed. The residual oil, 1-(3 : 4-dimethoxyphenyl)prop-2-ylamine (IV; R = Me), distilled at 95—97°/0·05 mm.

The amine (3 g.) was treated cautiously with hydrobromic acid (*d* 1·49; 10 c.c.) and refluxed 2 hours. The excess of acid was removed under reduced pressure and the residual gum crystallised on storage in a desiccator, giving the hydrobromide, which was washed with acetone.

Oxidation of Epinine.—(i) *With Potassium Iodate*. To a solution of epinine hydrochloride (0·5 g.) in water (30 c.c.) a saturated aqueous solution of potassium iodate (0·55 g.) was added. The solution rapidly became red and some brownish-red amorphous solid separated. After 10 minutes, this solid was filtered off, and the filtrate kept at 0°. After 6 hours, the crystalline deposit was collected and washed with a little water. 2 : 3-*Dihydro*-3-*iodo*-1-*methylindole*-5 : 6-*quinone* (IX) (0·4 g.) formed deep red needles, m. p. 85—87° (decomp.), sparingly soluble in water and ethanol, giving deep red solutions which decomposed rapidly on being warmed. The solid rapidly decomposed on storage (Found : C, 38·0; H, 3·0; I, 43·6. $C_9H_8O_2NI$ requires C, 37·4; H, 2·8; I, 44·0%).

The iodo-quinone (0·2 g.) was kept with a mixture of acetic anhydride (5 c.c.) and pyridine (5 c.c.) for 24 hours at room temperature, then poured into water; the precipitate was collected and twice recrystallised from aqueous methanol. 5 : 6-*Diacetoxy*-3-*iodo*-1-*methylindole* (VIII; R = Ac) formed colourless plates, m. p. 146—147° (Found : C, 42·3; H, 3·3. $C_{13}H_{12}O_4NI$ requires C, 41·8; H, 3·2%).

The iodo-quinone (0·2 g.) was stirred with 10% zinc acetate solution (5 c.c.); the red crystals were rapidly converted into a dark sticky mass, which was extracted with ether, the extract filtered, and the ether removed, leaving a yellow gum. Recrystallisation from benzene–light petroleum gave 5 : 6-*dihydroxy*-3-*iodo*-1-*methylindole* as yellow needles, m. p. 88—90° (decomp.) (Found : C, 37·1; H, 2·9. $C_9H_8O_2NI$ requires C, 37·4; H, 2·8%). The iodo-indole was unstable and rapidly decomposed in boiling solvents and in the solid state. It gave an intense blue colour with aqueous alkali.

(ii) *With potassium ferricyanide*. To a solution of epinine hydrochloride (0·8 g.) in water (50 c.c.) a solution of potassium ferricyanide (5·2 g.) and sodium hydrogen carbonate (1·7 g.) in water (50 c.c.) was added with stirring. After 10 minutes, 30 c.c. of a 20% solution of zinc acetate were added and the mixture was shaken vigorously. After 10 minutes the red colour had changed to grey. A little sodium dithionite was added to prevent oxidation, and the precipitated zinc ferrocyanide filtered off. The pale yellow filtrate was extracted with peroxide-free ether (4 × 30 c.c.), the ethereal extract dried (Na_2SO_4), and the ether removed. The residual gum was recrystallised from benzene–light petroleum, giving 5 : 6-dihydroxy-1-methylindole (0·45 g.), m. p. 136°.

Similar treatment of 2-(3 : 4-dihydroxyphenyl)ethylamine and 1-(3 : 4-dihydroxyphenyl)prop-2-ylamine gave brownish-red rather than red solutions on oxidation, and the ethereal extracts, although giving a positive Ehrlich reaction and showing a bluish-violet fluorescence indicating the presence of dihydroxyindoles, contained insufficient material for working up.

Reduction of 3 : 5 : 6-*Trihydroxy*-1-*methylindole*.—The trihydroxy-indole (1·0 g.) was dissolved in 10% sodium hydroxide solution (50 c.c.) and Raney nickel–aluminium alloy (5 g.) was added in small portions without cooling. The yellow colour slowly faded and the green fluorescence disappeared as the reaction proceeded. The supernatant liquor was decanted from undissolved metal and acidified with dilute sulphuric acid. The inorganic precipitate was filtered off, and the filtrate extracted with peroxide-free ether (4 × 20 c.c.). Removal of the solvent after drying (Na_2SO_4) gave a gum from which 5 : 6-dihydroxy-1-methylindole (0·4 g.), m. p. 135°, was isolated by recrystallisation from benzene–light petroleum.

Formation of Zinc Complexes from Oxidised Catechols.—To a filtered solution of catechol (1·1 g.) and zinc acetate (10 g.) in water (200 c.c.) a filtered aqueous solution of sodium persulphate (2·5 g.) was added. The solution rapidly became green and a very dark green precipitate was deposited. After 2 hours the precipitate was collected, well washed with water, and dried (P_2O_5). The *zinc* complex (Found : C, 52·2; H, 3·0; ZnO, 29·5. $C_{12}H_8O_4Zn$ requires C, 51·1; H, 2·8; ZnO, 28·8%) formed a very dark green powder insoluble in the common organic solvents with the exception of acetic acid, which gave an orange-red solution with apparent decomposition. It also dissolved with decomposition in aqueous mineral acids.

Similar treatment of 4-methylcatechol (1·2 g.) gave a similar *zinc* complex (Found : C, 56·1; H, 4·9; ZnO, 23·1. $C_{14}H_{12}O_4Zn$ requires C, 57·0; H, 4·75; ZnO, 24·0%).

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