106. *The Action of Alkali on Rotenone and Related Substances.*

By R. S. Cahn, R. F. Phipers, and J. J. Boam.

The precursor of toxicarol in *Derris* extract is purified and shown to be the *l*-form of the known toxicarol.

Very mild alkali, *e.g.*, potassium carbonate or a trace of alkali hydroxide in acetone, alkali acetate in alcohol, alkali hydroxide in benzene–methyl alcohol, or " activated " alumina in non-hydroxylic solvents, racemises the two central tertiary carbon atoms of rotenone, toxicarol, and related substances by enolisation, followed by fission of the dihydropyranone ring. The same conditions lead to equilibration of toxicarol with a structural isomeride, and proof of the structure of this isomeride also proves the mechanism postulated for the racemisation. Changes in rotation and absorption spectra in the presence of alkali indicate that enolisation is more facile in the toxicarol than in the rotenone series. In all cases one form of ring-junction is greatly favoured.

The " resin " or " extract " removed by organic solvents such as ether, benzene, etc., from the roots of *Derris elliptica* has long been known to contain rotenone. From so-called " Sumatra-type " *Derris* roots Cahn and Boam (*J. Soc. Chem. Ind.*, 1935, **54**, 42T) isolated sumatrol. By freeing the extract as far as possible from rotenone and treating

* We are indebted to Messrs. Howards and Sons, Ltd., for the preparation of this ketone.

the residual gum with alkali, three substances have been isolated, namely, toxicarol, deguelin, and tephrosin [Clark, *J. Amer. Chem. Soc.*, (*a*) 1930, **52**, 2461; (*b*) 1931, **53**, 313; (*c*) 1931, **53**, 729. Haller and LaForge, (*a*) *ibid.*, 1934, **56**, 2415. Cahn and Boam, *loc. cit.* Buckley, *J. Soc. Chem. Ind.*, 1936, **55**, 285T. Back-reference to papers by Clark, Haller, LaForge, and their collaborators will be made by the letters *a*, *b*, etc. Papers by Haller or LaForge and their collaborators form one series]. Buckley (*loc. cit.*) reports a fourth substance, m. p. 183°, obtained by treatment with alkali, but we have never encountered it and consider its individuality as yet insufficiently established. It is now generally recognised that toxicarol, deguelin, and tephrosin do not exist as such in the extract; tephrosin is formed by oxidation of deguelin during its isolation, and deguelin and toxicarol result from some change in the original components caused by the alkali used for their isolation. Rotenone and sumatrol are lævorotatory. Toxicarol, deguelin, and tephrosin, as isolated, are optically inactive. Haller and LaForge's work made it probable, but not certain, that deguelin existed in the extract at least partly as the *l*-isomeride.

Failing, as did also other workers, to isolate other ingredients from the extract without the use of alkali, we investigated the effect of alkali on rotenone and derived substances of known structure in the hope, ultimately realised, that knowledge of this action would throw light on the nature of the deguelin and toxicarol precursors. In this paper we describe (*A*) the action of alkali on rotenone and similar substances and (*B*) the action of alkali on toxicarol.

When this work had been completed, Tattersfield and Martin (*Ann. Appl. Biol.*, 1936, **23**, 899; *J. Soc. Chem. Ind.*, 1937, **56**, 77T, 88T) isolated from *Derris* a substance, m. p. 99°, $[\alpha]_D$ −70° in benzene, which yielded inactive toxicarol when treated with alkali and was correctly considered by them to be the precursor, or a mixture of precursors, of toxicarol. In section (*C*) we show that their product was not quite pure and we establish its constitution.

The bearing of our results on the nature of the components of *Derris* extract and the quantitative determination of 90% of its total ingredients are discussed by us elsewhere (*J. Soc. Chem. Ind.*, in the press).

Unusual difficulties are caused in this field by dimorphism, variability of melting points, and, occasionally though not often, absence of mixed melting point depressions. It is thus essential that claims to individuality or identity should be thoroughly established; preparation of a series of derivatives, optical rotation (values in the series are large and differ widely), crystallo-optical data, and, occasionally, absorption spectra are suitable for this purpose. We have been at pains in our work to indicate clearly the means of identification in all cases in which it seemed to us at all doubtful.

A. *Action of Alkali on Rotenone and Derived Substances.*

Tattersfield and Martin (*loc. cit.*) showed that addition of large amounts of methyl-alcoholic potassium hydroxide to rotenone in benzene caused a gradual fall in optical rotation, the rate depending largely on the amount of alcohol added. At the time of this publication we had already carried out rather similar experiments, in which, however, we used other solvents and very much milder alkaline conditions, and we had also fully elucidated the changes occurring.

We shall need a nomenclature for stereoisomerides of the series. Rotenone (I) has three asymmetric carbon atoms, C7, C8, and C20 (the numbering is designed to retain C7 and C8 as they are in the customary system of partial numbering). The junction of rings *B* and *C* may be *cis* or *trans*. Although, by analogy, the *trans*-structure appears more probable for rotenone, we prefer not to dogmatise. We shall call that mode of ring junction present in rotenone the normal form and the other the *allo*-form; we shall designate the latter by the prefix *allo* to the names of substances containing it, whilst substances without this prefix will be understood to have the normal ring junction as present in rotenone. It is apparent that change in the configuration of only one of the atoms C7 and C8 involves also a change from the normal to the *allo*-form or *vice versa*. When once we have decided whether a substance belongs to the normal or the *allo*-series, it suffices for our initial purpose to consider C7 and C8 together as a single asymmetric

centre. We shall define the stereochemistry of the third asymmetric atom, C20, by the absence or presence of the prefix *epi*; when this prefix is absent, the hydrogen atoms

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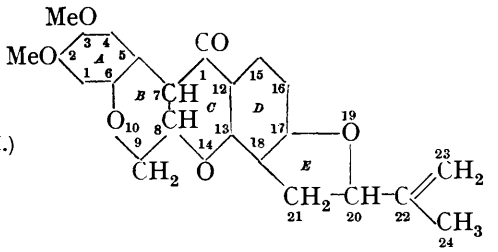

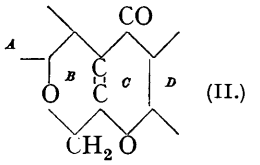

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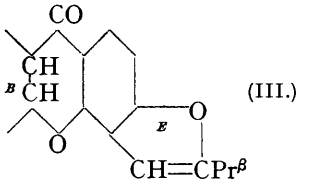


(III.)

attached to C20 and C8 bear the same steric relation to each other as in rotenone. It is inadvisable to relate C20 to C7, since the asymmetry of C7 is destroyed by enolisation.

Rotenone, which is lævorotatory in benzene, is oxidised by iodine, ferricyanide, etc., to *l*-dehydrorotenone (II); in this reaction the asymmetry of C7 and C8 is destroyed and the lævorotation of the dehydro-compound is due solely to C20. It follows that C20 in rotenone is lævorotatory. When rotenone is treated with sulphuric acid, best in acetic acid (Wright, *J. Amer. Chem. Soc.*, 1928, **50**, 3355), it gives *iso*rotenone (III), which is lævorotatory in benzene; in this reaction the asymmetry of C20 has been destroyed, but there is no reason to suppose that C7 and C8 have been affected. It follows that in rotenone C7 and C8, considered as a single asymmetric centre, are lævorotatory. We thus arrive at the nomenclature set out in Table I.

TABLE I.

Nomenclature of the Stereoisomerides of Rotenone.

Prefix.	Asymmetry at C7—C8, normal series.	C20,	Racemic forms.	Prefix.	Asymmetry at C7—C8, *allo*-series.	C20,	Racemic forms.
(nil or *l*-)	*l*	*l*	*dl*-	*l*-*allo*-	*l*	*l*	*dl*-*allo*-
d-	*d*	*d*		*d*-*allo*-	*d*	*d*	
l-*epi*-	*l*	*d*	*dl*-*epi*-	*l*-*epiallo*-	*l*	*d*	*dl*-*epiallo*-
d-*epi*-	*d*	*l*		*d*-*epiallo*-	*d*	*l*	

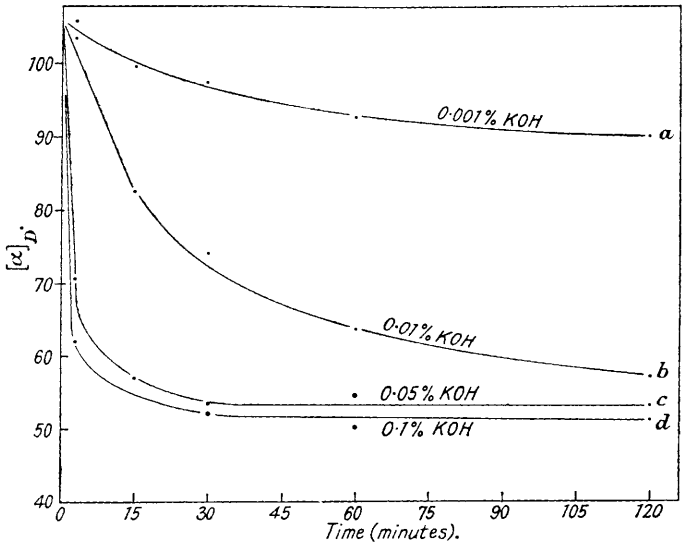
For *iso*rotenone, deguelin, and other substances in which there is no asymmetry in ring *E*, we have the simpler case of *l*-, *d*-, and *dl*-normal and *l*-, *d*-, and *dl*-*allo*-forms. We note also that dehydro-compounds cannot exist in normal and *allo*-forms, so that there can exist only inactive dehydro*iso*rotenone and *l*-, *d*-, and *dl*-dehydrorotenone.

In this scheme we follow the method used in the sterol series. We refer all rotations to benzene solutions, not for any theoretical reason, but because we find it convenient; the necessity for specifying the solvent will appear later. The prefix *l*- or *d*- for the four *epi*-compounds is fixed by the rotation at the C7—C8 centre and may not coincide with the sign of the total rotation. It should be noted that the rules of simple optical superposition do not apply in this series; for instance, *l*-dehydrorotenone has $[\alpha]_D$ −77° and *l*-*iso*rotenone has $[\alpha]_D$ −73°, whereas rotenone, containing only the asymmetric centres of these two substances, has $[\alpha]_D$ −226° (all in benzene). Finally we have neglected isomerism which would arise if rings *B* and *C* were not planar; LaForge and Haller [(*b*) *J. Amer. Chem. Soc.*, 1934, **56**, 1620] have envisaged this possibility, but we consider it no longer necessary (*vide infra*).

When 1 c.c. of 1—10% ethyl-alcoholic potassium hydroxide is added to 4 g. of rotenone in 100 c.c. of acetone, the rotation falls rapidly; the rate depends on the amount of alkali

used, as shown in Fig. 1, but, except in very weak solutions, an equilibrium is rapidly reached, the final value remaining constant for many hours. The reaction mixture contains small amounts of oxidation products, but the bulk of the product is a new substance, m. p. 146°, $[\alpha]_D$ −83° in benzene, which we term *mutarotenone*; analysis shows it to be isomeric with rotenone. The same substance is formed by the Tattersfield–Martin procedure mentioned above and also by heating rotenone with an alkali acetate in absolute or 96% alcohol or with anhydrous potassium carbonate in acetone containing 1—2% (not less or much more) of water. The effect of rather vigorous or exceedingly mild alkaline conditions is thus the same. The product, mutarotenone, is, however, not a single substance. Nearly half its weight of rotenone separates as the crystalline solvate containing one molecule of carbon tetrachloride of crystallisation when mutarotenone is dissolved in this solvent. The mother-liquors yield a gum, which we have been unable to crystallise; but we are able to state its composition with complete certainty.

FIG. 1.



Change of $[\alpha]_D$ *with time when* 1 *c.c. of* (a) 0·1%, (b) 1%, (c) 5%, *and* (d) 10% *ethyl-alcoholic potassium hydroxide solution is added to* 4 *g. of rotenone in* 100 *c.c. of acetone. Percentages stated on the curves are the* % KOH *in the total solution.*

With iodine, mutarotenone gives an almost quantitative yield of *l*-dehydrorotenone (II), showing that all the components of the compound have a lævorotatory C20, whilst isomerisation by sulphuric acid gives *dl-iso*rotenone (III), showing that only normal ring-forms are present and that the C7—C8 centre is present in mutarotenone in equal amounts of *d*- and *l*-forms. Half of mutarotenone is rotenone, in which both C20 and C7—C8 are lævorotatory; the other half must, therefore, be the isomeride of the normal series in which C20 is lævorotatory and C7—C8 dextrorotatory, that is, *d-epi*rotenone. Mutarotenone is then a 1 : 1 molecular compound of rotenone and *d-epi*rotenone, a kind of semi-racemic compound. Rotenone separates from carbon tetrachloride because it forms a solvate, whereas *d-epi*rotenone (which is not the enantiomorph of rotenone) apparently does not, but in other solvents no separation has been observed.

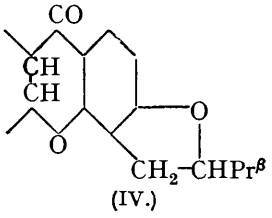
The purity of mutarotenone cannot be judged by its melting point, which varies only slightly on recrystallisation; optical rotation is, however, a satisfactory guide to purity, for material with $[\alpha]_D$ −83° in benzene gives *iso*rotenone which is optically inactive in the crude state, whereas material having about the same melting point but a higher rotation gives slightly active crude *iso*rotenone, from which the activity departs on purification.

Since rotenone has $[\alpha]_D$ −226° in benzene and mutarotenone has $[\alpha]_D$ −83°, *d-epi*-rotenone is expected to have $[\alpha]_D$ +60° in benzene. The highest rotation observed for

our gummy material is +30°. We have also been unable to prepare a crystalline derivative of *d-epi*rotenone, *e.g.*, an oxime; the structure assigned to it is, however, confirmed by hydrogenation, isomerisation by sulphuric acid, and by the fact that racemisation of C7 and C8 is a general reaction. Potassium carbonate in acetone or sodium acetate in alcohol provides, in fact, the readiest method of preparing inactive *iso*rotenone, dihydrodeguelin, and rotenonic acid (*iso*dihydrorotenone) from their active isomerides—in these three compounds only C7 and C8 are asymmetric.

Isomerisation of our gummy *d-epi*rotenone yielded readily and in quantity d-iso-*rotenone*, m. p. 182°, $[\alpha]_D$ +75° in benzene, −10° in acetone. This is obviously the enantiomorph of the known *l-iso*rotenone, m. p. 184°, $[\alpha]_D$ −73° in benzene, +7·5° in acetone; we confirmed this by preparing *dl-iso*rotenone, m. p. 170°, identical (mixed m. p.) with an authentic specimen (m. p. 170°), by crystallising together equal parts of the *d*- and the *l*-form, and observing that 1 : 2 and 2 : 1 mixtures of the active forms melted at about 165°. Clearly, then, the main component of our *d-epi*rotenone has a dextrorotatory C7—C8 centre.

The results of hydrogenation of *d-epi*rotenone would in themselves provide a complete proof of structure, but the yields are poor and we base our proof on the other reactions mentioned, in all of which excellent yields are obtained. Hydrogenation of rotenone under various conditions gives the dihydro-compounds, dihydrorotenone (IV) and rotenonic acid (*iso*dihydrorotenone) (V), or the fully reduced phenolic tetrahydro-compound, according to the conditions [Kariyone, Kimura, and Kondo, *J. Pharm. Soc. Japan*, 1924, **514**, 1049. LaForge and Smith, (*c*) *J. Amer. Chem. Soc.*, 1929, **51**, 2574. Takei, Miyajima, and Ōno, *Ber.*, 1933, **66**, 479; *Bull. Inst. Phys. Chem. Res. Tokyo*, 1933, **12**, 239. Haller and Schaffer, (*d*) *J. Amer. Chem. Soc.*, 1933, **55**, 3494; (*e*) *Ind. Eng. Chem.*, 1933, **25**, 933]. *l*-Rotenonic acid (V) is cyclised by sulphuric acid to *l*-dihydro-deguelin (*l*-β-dihydrorotenone) (VI), m. p. 156°. The phenol (V) is best obtained by the



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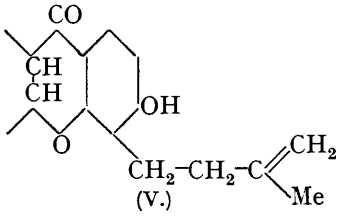

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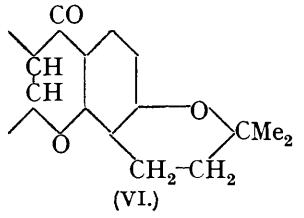
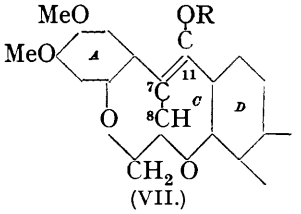


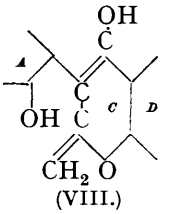
(VI.)

use of platinum in alkaline solution, but, when the amount of alkali is increased sufficiently to give reasonable yields, racemisation also occurs. We needed a method of preparing the active phenol in quantity, and after some experimentation (see p. 527) found it in hydrogenation in dioxan in the presence of palladised barium sulphate and potassium acetate. We would remark, in passing, that racemisation by hot, fairly concentrated alkali has been well established for some time [Haller and LaForge, (*f*) *J. Amer. Chem. Soc.*, 1931, **53**, 3426 and *a*], but that this racemisation by traces of alkali in the cold during hydrogenation (Takei *et al.*, *loc. cit.*, 1933), fully comparable with our conditions of racemisation, has hitherto passed without comment. We hydrogenated, by the methods thus available, the purest *d-epi*rotenone which we could obtain; by using Adams's catalyst in ethyl acetate, we got d-*dihydro*epi*rotenone* (IV), which, as anticipated, is not enantiomorphic with *l*-dihydrorotenone (IV). With palladised barium sulphate and potassium acetate in dioxan we got a phenolic gum, which we believe to have been impure *d*-rotenonic acid (V), since cyclisation yielded d-*dihydrodeguelin* (d-*dihydro*-β-*rotenone*) (VI), m. p. 154°; we had insufficient of this material for accurate determination of the rotation, but a definite dextrorotation was observed, and identification was rendered certain by the facts that the crystallo-optical data were identical with those of the authentic *l*-form and that crystallisation of equal parts of the *l*- and the *d*-form together gave the higher-melting *dl*-form, m. p. 173°, which did not depress the m. p. (176°) of the authentic *dl*-form and was crystallo-optically identical with it.

We turn now to the mechanism of the racemisation. It is natural to assume that the

first step is enolisation, since a number of enolic acetates (VII; R = Ac) have been known for some time [Smith and LaForge, (*g*) *J. Amer. Chem. Soc.*, 1932, **54**, 2996]. Enolisation of rotenone to (VII; R = H), followed by ketonisation, could not, however,


(VII.)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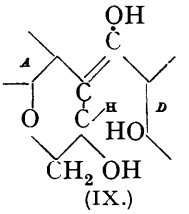
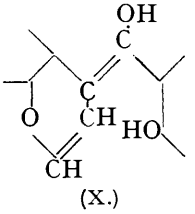
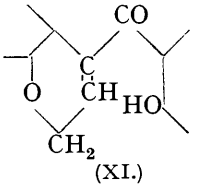
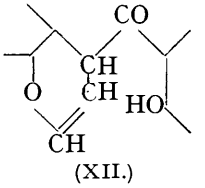

(IX.)

racemise C8, but would merely equilibrate the normal with the *allo*-form. To account for racemisation of C7 and C8 we have considered the three intermediates of types (VIII), (IX), and (X). We can exclude (VIII), because our experiments with toxicarol show that fission of ring *C* occurs. We rule out (IX), because Professor Robertson has told us (private communication) that ring closure and opening of dihydroxy-compounds of this type are not sufficiently facile. We conclude, therefore, that racemisation proceeds by way of (X), ring fission being initiated by enolisation. This ring fission is not a novel


(X.)


(XI.)


(XII.)

conception. Butenandt and Hilgetag (*Annalen*, 1932, **495**, 172) considered a formula of type (XI) for toxicarol, but the case for the ring-closed formula now generally accepted has been clearly stated by George and Robertson (J., 1937, 1535). In a later paper Butenandt and Hilgetag (*Annalen*, 1933, **506**, 158) suggested that a little of the form (XII) exists in alkaline solution in equilibrium with the ordinary closed-ring form; their reasons were, however, not conclusive, for they rested mainly on the formation of minute yields of certain degradation products. We consider the ketonic form (XII) an extremely unlikely product, for fission of ring *C* would give the αβ-unsaturated ketone (XI) rather than (XII); but (XI) is not a suitable intermediate for the degradations discussed by Butenandt and Hilgetag. The enolic form (X) of (XII) is free of all objections and our experiments confirm the validity of Butenandt's degradative mechanisms. If it is agreed that (X), but not (XI) or (XII), is the intermediate, then the necessity for enolisation prior to ring fission is apparent.

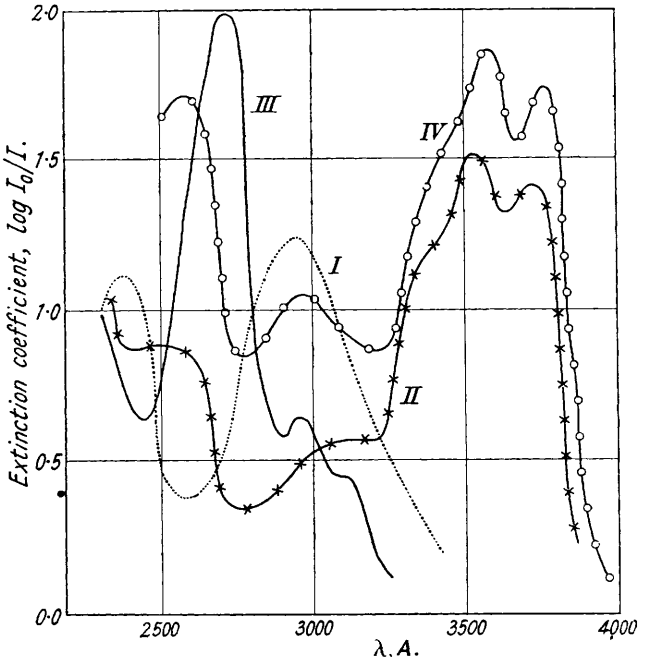
We have discovered the hitherto unsuspected fact that racemisation accompanies formation of the enolic acetyl derivatives. Smith and LaForge (*g*), who prepared acetyl-*iso*rotenone from *l-iso*rotenone, did not determine its rotation. Under suitably modified conditions (milder than those used by Smith and LaForge, so that no objection can be raised on this score), *l-iso*rotenone gives a 75% yield of inactive acetate, which is obtained also from *dl-iso*rotenone and regenerates *dl-iso*rotenone when hydrolysed by dilute acid—dilute acid does not cause racemisation. Haller and LaForge (*a*) report hydrolysis of the acetate to an *iso*rotenone having $[\alpha]_D$ −21·1° in benzene, ordinary *l-iso*rotenone having $[\alpha]_D$ −73°; this was presumably due to their acetate having contained some unacetylated material. Acetylation of rotenone presents a more complicated picture. In acetyl*iso*-rotenone, only C8 is asymmetric and racemisation thus gives a true *dl*-compound. In acetylrotenone, C20 retains its asymmetry; acetylation of rotenone, accompanied by racemisation of C8, must yield a mixture unless acetyl-*l*-rotenone and acetyl-*d-epi*rotenone form a molecular compound analogous to mutarotenone. No such compound appears to be formed. The crystalline product isolated is acetyl-*l*-rotenone and not the *epi*-compound, since it gives on hydrolysis an excellent yield of ordinary rotenone of unchanged

rotation. However, the crystalline compound is accompanied by much gummy material, and, since there is no reason to expect rotenone and *iso*rotenone to behave differently, it is at least possible to connect the gummy by-product with the presence of the *d-epi*-compound.

The mechanism of racemisation during acetylation is obscure; and the position is further complicated by anomalous acetylation, of the isomerides of toxicarol (see pp. 528, 533, 534). If racemisation occurred in this case also by way of the " open " form (X), one would expect to obtain substances in which the phenolic hydroxyl group was acetylated. The enolic acetate structure (VII; R = Ac) was, however, proved by Smith and LaForge (*g*), who hydrogenated acetylrotenone to dihydrodeoxyrotenone in which the >C:C·OAc group has been reduced to >CH·CH_2. The structure (VII) is confirmed by absorption spectra. In Fig. 2 we show the absorption spectra of

FIG. 2.

Absorption spectra. Alcoholic solutions.



I, *Rotenone* 0·00302%, 1 *cm.* III, *Toxicarol* 0·00239%, 1 *cm.*
II, *Acetylrotenone* 0·00222%, 1 *cm.* IV, *Diacetyltoxicarol* 0·00335%, 1 *cm.*

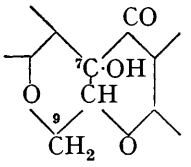
rotenone, acetylrotenone, toxicarol, and diacetyltoxicarol. We shall discuss toxicarol and its acetate later. At this point we would note merely that in the acetates the main absorption maxima are displaced nearly into the visible region; this indicates a series of six or seven conjugated ethylenic linkings, which is provided in (VII) by the aromatic rings *A* and *D* united by the C7—C11 ethylenic linking. In rotenone and toxicarol the system Ar·C:C·Ar is replaced by Ar·CH·CO·Ar and the principal absorption maxima are in regions of much shorter wave-length. Finally, hydrolysis of acetylrotenone gives a 83% yield of rotenone and not mutarotenone; the latter would be produced if the acetate were a derivative of the " open " form (X).

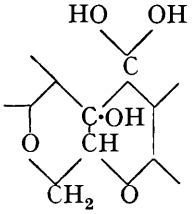
One further aspect calls for comment at this point. Acid hydrolysis of the enolic acetates gives excellent yields of the parent ketones. This regenerates the asymmetry of C7, but no signs of acid conditions affecting C8 have been observed. We should thus expect hydrolysis to produce mixtures of normal and *allo*-forms; actually normal forms are obtained in all cases in excellent yields (as has just been mentioned for acetylrotenone),

and *allo*-forms could not be detected. In alkaline reactions, too, no *allo*-forms were isolated, but in these cases yields are not so good and the conditions are consequently less searching. It is clear that steric relations favour the normal to the complete or nearly complete exclusion of the *allo*-form and that newly formed asymmetry at C7 is determined by any asymmetry already existing at C8. There is a partial analogy, kindly pointed out to us by Dr. S. H. Harper, in that *cis*-α-decalone passes readily and apparently irreversibly into the *trans*-isomeride (Hückel, *Annalen*, 1925, **441**, 1).

This generalisation requires qualification in one respect. Oxidation by air or hydrogen peroxide in the presence of alkali gives a series of hydroxy-derivatives (LaForge and Haller, *b*, who give earlier references). Some, at least, of these are pairs of 7-hydroxy-derivatives (XIII), doubtless obtained by way of (XIV), which arises by 1 : 2 addition of 2OH to

(XIII.)





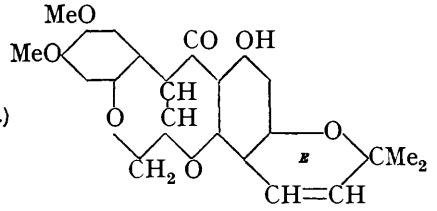
(XIV.)

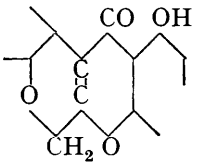
(X); it is suggestive that in these apparently well-authenticated cases of normal and *allo*-forms interconversion by enolisation is impossible. LaForge and Haller considered that non-planar configurations of rings might be necessary to account for other isomerides discovered, but we see now that 1 : 4 addition to (X) might lead to 9-hydroxy-derivatives, two of which could exist by virtue of the new asymmetry at C9. Further work on these interesting substances is desirable.

B. *Action of Alkali on Toxicarol.*

Elegant proof of the structure of toxicarol (XV) has finally been provided by Robertson and his co-workers (J., 1937, 1535 and earlier papers). When toxicarol (which is optically inactive) is heated with potassium carbonate in acetone, it is partly changed into an isomeride, which we term β-*toxicarol*, the two substances being readily separable by reason of the much greater solubility of the new compound in ether. The reaction is reversible,

(XV.)



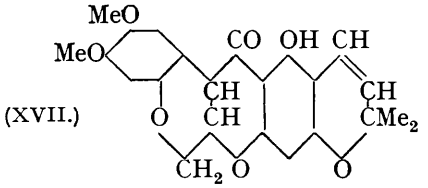


(XVI.)

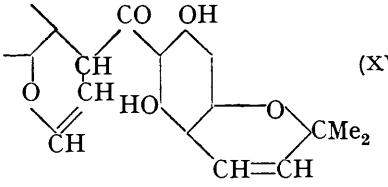
the new compound being partly reconvertible under the conditions of its formation into ordinary toxicarol (we shall term this α-toxicarol when we need to contrast it with the new isomeride, but shall otherwise omit the prefix). β-Toxicarol is equilibrated also by alcoholic alkali, but in this case the much less soluble α-toxicarol crystallises, so that in alcohol the change β → α is almost complete. The α-toxicarol which has separated after the solution has cooled contains, however, some of the β-isomeride and this largely accounts for the various melting points reported for α-toxicarol prepared from *Derris* extracts [213° by Butenandt and Hilgetag, *loc. cit.*, 1932; 219° (corr.) by Clark, *a*; 220° by Cahn and Boam, *loc. cit.*]; if α-toxicarol prepared from the extract is subjected to purification by the ether method, the melting point rises at once to 226° and subsequent crystallisation raises it to 232—233°; repeated crystallisation from acetic acid or benzene is needed if the ether purification is omitted. The removal of β-toxicarol can be followed quantitatively by the Goodhue colour test (*J. Assoc. Off. Agric. Chem.*, 1936, **19**, 118), which is given by β- but not by α-toxicarol; we find that α-toxicarol of m. p. 219° contains 4·5% of the β-isomeride.

We had to consider two possibilities for the structure of β-toxicarol. The first, that it

was the *allo*-form of toxicarol, was readily eliminated, since oxidation of β-toxicarol gives a dehydro-compound (XVI), which is not identical with ordinary dehydrotoxicarol. The alternative was the linear form (XVII). George and Robertson (*loc. cit.*) have pointed out that (XVIII) is an intermediate common to (XV) and (XVII). Professor Robertson had mentioned this in conversation with one of us (R. S. C.) earlier and before the discovery of β-toxicarol, and realisation of the relationship greatly simplified elucidation of the structure

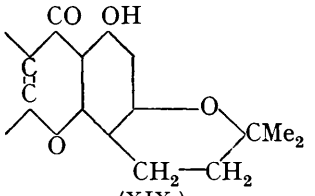


(XVII.)

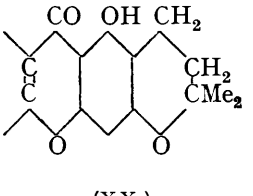


(XVIII.)

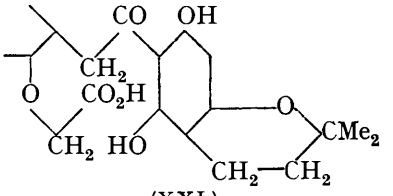
of this compound. Now (XVIII) (or, rather, its enolic equivalent) is the form which, by analogy with rotenone, we should expect toxicarol to assume in the presence of alkali, and equilibration of (XV) and (XVII) by alkali would be in line with our experiences in the rotenone series. This interpretation was readily proved correct. Hydrogenation of toxicarol in acetic acid at 75° [Clark, (*f*) *J. Amer. Chem. Soc.*, 1931, **53**, 2264] or, as we find, more conveniently in dioxan at room temperature, gives dihydrotoxicarol, in which ring *E* is saturated. Oxidation then gives dehydrodihydrotoxicarol (XIX). β-Toxicarol gives similarly the isomeric *dihydro*-β- and *dehydrodihydro*-β-*toxicarol* (XX). When heated with potassium hydroxide, (XIX) gives dihydrotoxicarolic acid (XXI) (Clark, *d*).



(XIX.)



(XX.)



(XXI.)

Dehydrodihydro-β-toxicarol gives the same acid (XXI), identified by the melting points and mixed melting points of the acid, its ester, and ester diacetate. This is explicable only if dehydrodihydro-β-toxicarol has the structure (XX), which necessitates formula (XVII) for β-toxicarol. The interconversion of (XVII) and (XV) proves that fission of ring *C* occurs under the influence of alkali, which in turn is very strong support for the mechanism postulated by us for racemisation.

As was the case with rotenone, toxicarol reacts in the closed form (XV) when acetylated. Mild treatment gives a monoacetate, in which only the phenolic hydroxyl group is acetylated. More drastic treatment gives excellent yields of a diacetate, analogous to acetylrotenone, the structure of which is proved in the same way. First, Clark (*f*) hydrogenated diacetyltoxicarol to acetyldeoxydihydrotoxicarol. Secondly, the absorption spectrum of diacetyltoxicarol shows the three-banded maximum characteristic of acetylrotenone as shown in Fig. 2 (these and other absorption data are discussed further in the experimental portion, p. 535). Finally, hydrolysis of diacetyltoxicarol by dilute acid gives a quantitative yield of α-toxicarol, with no trace of β-toxicarol or of *allo*-forms.

Two further theoretical points follow. First, the quantitative hydrolysis of diacetyltoxicarol to α-toxicarol shows that *allo*-forms are as little favoured in the toxicarol as in the rotenone series, and, secondly, this hydrolysis and that of acetylrotenone are experimental justification for our contention that dilute acid does not affect C8.

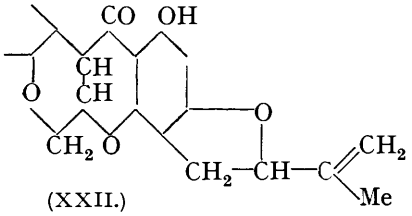
Clark (*d*) obtained acetyldehydrotoxicarol by ring-closure of (XXI) by acetic anhydride. George and Robertson (*loc. cit.*) saw that this reaction should give a mixture of the acetates of (XIX) and (XX) and isolated a little of a compound, m. p. 238—239° (decomp.), which they thought might be (XX) [they termed the then hypothetical (XVII) *iso*toxicarol, but we prefer to use the prefix *iso* only for isomerism of the rotenone–*iso*rotenone type]; however, (XX) has m. p. 226—227°; George and Robertson's compound may have been

impure dehydrodihydro-α-toxicarol (XIX), which has m. p. 260°. Knowing that *acetyldehydrodihydro-β-toxicarol*, readily obtained from (XX), is more soluble than its α-isomeride, we had independently searched for it in the mother-liquors obtained in this reaction; we failed to find it there.

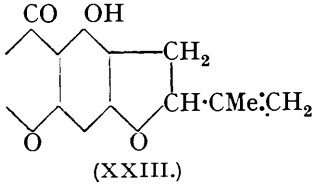
C. *The Toxicarol Precursor.*

Tattersfield and Martin (*locc. cit.*) isolated their toxicarol precursor by crystallising from ethyl acetate either the whole or the alkali-soluble part of a Sumatra-type *Derris* extract (*i.e.*, an extract giving much toxicarol with alkali). Our difficulties connected with this substance were confined to its isolation and purification. At first we could not isolate it at all, but after Dr. Tattersfield had very kindly provided us with samples, we were able to obtain crystals from ethyl acetate–alcohol or ether; ethyl acetate alone could not be used until after preliminary purification. Plain recrystallisation gave material identical with that of Tattersfield and Martin in m. p. and rotation ($[\alpha]_D$ −68° in benzene), but analyses were unintelligible, values for carbon being 0·5—1·4% low for toxicarol, even after thorough drying. Proof that the material was heterogeneous came by acetylation, which gave very poor and erratic results; in one experiment a small amount of a *monoacetate*, m. p. 158°, of the precursor was obtained; in others a little *monoacetylsumatrol*, m. p. 217°, was isolated, which gave sumatrol on hydrolysis by acid and was also obtained (m. p. 218—219°) in 60% yield by gentle acetylation of sumatrol [Cahn and Boam (*loc. cit.*) and Robertson and Rusby (*loc. cit.*) were unable to crystallise acetylsumatrol]. After long trials we found that the sumatrol could be separated by stirring with an insufficiency of pure ether. Our final product had $[\alpha]_D$ −53° in benzene, but had the same m. p., 101—102°, as before removal of the sumatrol, which melts at 189°. It now gave correct analytical results for $C_{23}H_{22}O_7$ (toxicarol). The low analytical figures for carbon obtained for the earlier product cannot have been caused by the sumatrol, which is isomeric with toxicarol and crystallises from alcohol or ethyl acetate without solvent of crystallisation; it seems that a third substance must have been present, in which case the constancy of the m. p. appears still more remarkable. We should add that sumatrol has an asymmetric atom, C20, not present in the precursor and that the sumatrol isolated has its full activity; progressive change of the precursor into sumatrol is thus impossible.

Tattersfield and Martin (*locc. cit.*) found that, when methyl-alcoholic potassium hydroxide was added to a benzene solution of their precursor, $[\alpha]_D$ −70° in benzene, the initial small lævorotation changed immediately to a powerful dextrorotation, +300° to +321° after 2 minutes, and then sank slowly. Our material, $[\alpha]_D$ −68°, behaved similarly, but material having $[\alpha]_D$ −53° gave a dextrorotation of +350°, which sank later to exactly zero. Under Tattersfield–Martin conditions sumatrol, which is almost certainly 15-hydroxyrotenone (XXII) [the only alternative (XXIII) is much less probable (George and Robertson, *loc. cit.*; *cf.* also the evidence of absorption spectra, p. 535)], changes its rotation, −158°, instantaneously to +105°, which falls slowly through zero to −42°; the final rotation is due to C20, for we have seen in the rotenone series that under Tattersfield–Martin conditions only C7 and C8 are racemised—we shall discuss the change of sign later. This indicates that Tattersfield and Martin's material, $[\alpha]_D$ −70°, resembled our material, $[\alpha]_D$ −68°, and so probably contained sumatrol. The final fall of α of our



(XXII.)



(XXIII.)

material to absolutely nil proves its freedom from sumatrol and that the precursor has no asymmetry beyond that of C7 and C8.

Once purity had been attained, we readily proved the precursor to be l-*α-toxicarol*.

With alcoholic alkali it gives excellent yields of ordinary α-toxicarol, as was found for the impure material by Tattersfield and Martin. With methyl-alcoholic potassium hydroxide and benzene, potassium carbonate in acetone, etc., it gives a mixture of inactive α- and β-toxicarol. These reactions, together with the analytical figures, prove the precursor to be one or other active form of α- or β-toxicarol. The next step, proof that it belongs to the α-series, is provided by its oxidation to dehydro-α-toxicarol and its hydrogenation to l-*dihydro-α-toxicarol*, which is oxidised by iodine to dehydrodihydro-α-toxicarol.

This chemical evidence is confirmed by the absorption spectrum of the precursor, which is almost identical with that of *dl*-α-toxicarol.

We must next consider the meaning of the change in sign of rotation under Tattersfield–Martin conditions. If the alkaline, dextrorotatory solution is acidified before the decline to zero commences, *l*-toxicarol is recovered with unchanged rotation, $[\alpha]_D$ −53° in benzene. This disposes of the possibility that, in spite of the analytical figures, the precursor is really some labile compound of an active toxicarol and that the remarkable change in sign of rotation is due to liberation of active toxicarol from such a compound; this possibility is further discounted by the exactly analogous behaviour of sumatrol, the structure of which is beyond doubt. Actually we believe the instantaneous changes in sign are adequately explained by enolisation and this leads to further interesting conclusions. When *l*-toxicarol is enolised by the potassium hydroxide, the asymmetry of C7 is destroyed, but that of C8 remains; the optical behaviour is explained if C7 is lævorotatory and C8 dextrorotatory, the contribution of C7 being numerically greater than that of C8; the final fall to zero is caused by racemisation also of C8, which we believe proceeds by way of the " open " form. We explain the behaviour of sumatrol similarly; the final fall of $[\alpha]_D$ to −42° shows C20 to be lævorotatory; C8 is dextrorotatory and C7 strongly lævorotatory. We notice that with *l*-toxicarol the orientation of C8 determines the orientation of the hydrogen atom returning to C7, since the *l*-toxicarol regenerated from the enol has its full optical rotation, behaviour which is exactly parallel to the hydrolysis of acetylrotenone. We note further that the opposing signs of the rotations due to C7 and C8 afford a rational explanation of *l*-toxicarol being lævorotatory in benzene and dextrorotatory in acetone—for the relative numerical contributions of different asymmetric centres may well vary in different solvents.*

The rotation of rotenone shows no immediate change under Tattersfield–Martin conditions, but only the slow change due to racemisation. This indicates in our opinion that rotenone is less easily enolised than is toxicarol. This is endorsed by the absorption spectra data. Thus, the absorption of rotenone in alcohol is unchanged by the addition of 1% of sodium hydroxide. Alkali immediately changes the colour of toxicarol solutions from yellow to orange-yellow; the displacement of the absorption maximum to longer wave-lengths (cf. data, p. 535) is in agreement with conjugation of the C7—C11 ethylenic linking with the two aromatic rings *A* and *D* consequent upon enolisation. There is also evidence, more convincing than that of Butenandt and Hilgetag (*loc. cit.*), that the ring-fission which follows enolisation is more facile in the toxicarol than in the rotenone series, but we reserve this work for a later communication.

Owing to the incomplete enolisation we cannot allocate definite signs of rotation to C7 and C8 in rotenone. Rotenone is lævorotatory in all solvents tried; but *iso*rotenone is lævorotatory in benzene and dextrorotatory in acetone, dioxan, or chloroform, behaviour which we accounted for in the case of *l*-toxicarol by the opposing contributions of C7 and C8. It is thus probable that in *iso*rotenone and, therefore, in rotenone C7 and C8 have rotations in opposite directions; in rotenone C20 is also lævorotatory, so that the total rotation is negative in all solvents.

One point of the structure of the toxicarol precursor has been left unsettled above, namely, whether it is a normal or *allo*-form. Now, we have seen from the hydrolysis of diacetyltoxicarol that ketonisation of the enolic form gives only the normal keto-form; if we are correct in interpreting the change in sign of rotation in alkaline solution as due to enolisation, then the precursor must be the normal form of *l*-α-toxicarol; for, if it were an *allo*-form, enolisation by alkali, followed by immediate acidification, would lead to a new

* This invalidates the complicated calculations of Jones (*J. Agric. Res.*, 1936, **53**, 831).

compound having the stable normal ring structure, and we have seen that this does not occur.

l-Toxicarol can be isolated by direct crystallisation from Sumatra-type extract or by first extracting the ethereal solution of the extract with aqueous alkali and then crystallising the phenolic portion. These two methods give products which are chemically indistinguishable, and identity was confirmed by crystallo-optical comparison of material isolated by each method and purified to the $[\alpha]_D$ −68° stage. This identification was necessary in view of the known effects of alkali.

The remarkable difference in melting point and solubility between *l*- and *dl*-α-toxicarol, as well as the difficulty of purifying the *l*-form, might be held to indicate that the toxicarol precursor is in fact a mixture; while we cannot exclude definitely the existence of, *e.g.*, other steric forms, there is no valid evidence at all of the occurrence of more than one precursor.

Finally we would point out the dependence of the rate of racemisation on the solvent used. No racemisation occurs when an ethereal solution of *Derris* extract is shaken for a short time with aqueous alkali; in benzene, the rate is greatly accelerated by large additions of methyl alcohol, but even then large amounts of alkali hydroxide appear to be needed. In acetone containing 1% of ethyl alcohol, traces of alkali hydroxide suffice; and when potassium carbonate in acetone is used, the permissible water content is restricted to about 1—2%. Alkali acetates are effective in 96% or absolute ethyl alcohol, even after addition of a little acetic acid. Lastly, Merck's or B.D.H.'s "activated alumina for adsorption," which has been shown to contain unexpectedly large amounts of adsorbed alkali (Cahn and Phipers, *Nature*, 1937, **139**, 717), racemises rotenone derivatives in benzene or chloroform. Thus chromatographic adsorption of *l*-α-toxicarol gave some *dl*-α-toxicarol (the more soluble β-isomeride could not be separated from unchanged *l*-toxicarol), and rotenone was partly converted into a gummy material (this was observed before the discovery of mutarotenone; doubtless *d-epi*rotenone was formed, but we did not confirm this by experiment).

Experimental.

The *Derris* extracts used were commercial extracts; in order to define the materials, Goodhue and ferric chloride values are stated; the meaning of these terms will be explained elsewhere. Owing to the variability of *Derris* extract, workers using other raw material should expect qualitatively, but not quantitatively, similar results. Analyses are by Dr. G. Weiler, Oxford, except methoxyl determinations, which, unless otherwise stated, were done by us by Clark's volumetric semi-micro method (*J. Assoc. Off. Agric. Chem.*, 1932, **15**, 136), using a simplified apparatus. Macro-methods of determining methoxyl are unsatisfactory in this series; micro-methods are better, but, we find, not always as reliable as that used. Optical rotations are only approximate, as unfortunately no accurate polarimeter was available; we, therefore, omit references to temperature and concentrations. In experiments on racemisation the acetone used was B.D.H.'s "Acetone, redistilled," unless otherwise specified; this material usually contained just sufficient water to allow racemisation, but addition of 1% of water was, however, needed for some samples.

Mutarotenone.—(*a*) Rotenone (10 g.) and anhydrous potassium carbonate (20 g.) in acetone (100 c.c.) were heated under reflux for 2 hours. The filtered solution and washings from the carbonate were acidified with acetic acid and evaporated. The residue, crystallised from alcohol, gave 8·3 g., m. p. 137—140°, which on recrystallisation afforded 5·4 g. (*A*), m. p. 141—145°, $[\alpha]_D$ −103° in benzene. Concentration of the filtrates gave 0·1 g. of dehydrorotenone, m. p. 229—233°, identified by a mixed m. p. determination and its failure to give the Durham reaction (a useful diagnostic in this and similar cases); evaporation to dryness gave a gum, which in ether deposited first 0·3 g. of indefinite material and then pale yellow crystals (0·8 g.), m. p. 145—150°. An alcoholic solution of (*A*) deposited first 0·05 g. of dehydrorotenone, later (overnight) 1·8 g. of mostly colourless crystals, and, when concentrated, a further 2·8 g. of similar crystals. Fractional extraction of the two last fractions (bulked) with hot alcohol removed preferentially a lævorotatory component, probably rotenone; the first fraction (1·7 g.) had m. p. 141—142°, $[\alpha]_D$ −120° in benzene, the second (*B*) (1·6 g.) had m. p. 145—146°, $[\alpha]_D$ −100° in benzene, and concentration of the combined mother-liquors gave (*C*) (0·5 g.), m. p. 141—145°. Addition of ether (100 c.c.) to (*B*) and (*C*) in the least amount of benzene caused crystallisation

of pure *mutarotenone* (1·2 g.), m. p. 146—148° (Found : C, 69·6; H, 5·8. $C_{23}H_{22}O_6$ requires C, 70·0; H, 5·6%). Concentration of the ether–benzene mother-liquors gave first rotenone (0·25 g.) (prisms, m. p. 164° alone or mixed with an authentic specimen) and then material (1·5 g.), $[\alpha]_D$ −120°, which by crystallisation from ether–benzene gave a further 1·2 g. of pure mutarotenone. For pure mutarotenone, which separates from ether or alcohol in clusters of fine, colourless needles, m. p. 145—146°, $[\alpha]_D$ was variously determined as −80° to −86° in benzene and −37° in acetone.

In pilot experiments rotenone was recovered unchanged when the acetone contained 0, 0·25, or 0·5% of water and only partly racemised when the water content was 5 or 10%.

(*b*) 1% Alcoholic potassium hydroxide (25 c.c.) was added to rotenone (50 g.) in acetone (1 l.), and the whole kept under coal gas for 6 hours. A slight excess of acetic acid was added, the solvents removed, and the residue stirred with warm alcohol (400 c.c.). After cooling, the pale yellow crystals (41·5 g.), m. p. 140—142°, $[\alpha]_D$ −115° in benzene, were collected and fractionally extracted with two 200 c.c. portions of hot alcohol; the alcohol deposited (i) 4·5 g., m. p. 143—144°, $[\alpha]_D$ −118°, and (ii) 9 g., m. p. 145—146°, $[\alpha]_D$ −100° in benzene. The less soluble residue, crystallised from alcohol–benzene, gave nearly pure mutarotenone (21·5 g.), m. p. 145—146°, $[\alpha]_D$ −87°, which was finally purified (17·5 g.) to $[\alpha]_D$ −80° in benzene by twice adding ether (200 c.c.) to its solution in the minimum amount of benzene. Fractions (i) and (ii) were dissolved in the least amount of acetone and poured into a mixture of ether and water; the ethereal layer was rapidly filtered from dehydrorotenone (2·5 g.), m. p. 229—231° after recrystallisation, identified as above, and then deposited crystals (4·5 g.), m. p. 147—148°, $[\alpha]_D$ −163° in benzene, which by treatment with carbon tetrachloride gave 69% of rotenone and a residual gum having $[\alpha]_D$ +3° in benzene.

(*c*) Rotenone (1 g.) and anhydrous sodium acetate (3 g.) in alcohol (20 c.c.) were heated under reflux for 2 hours. The product which separated on dilution with water had $[\alpha]_D$ −82° in benzene and on recrystallisation gave mutarotenone (0·72 g.), m. p. 143—145°, $[\alpha]_D$ −72°. Similar experiments with potassium acetate in absolute and in 96% alcohol gave substantially similar results. These are much the most convenient methods of preparing mutarotenone, as no dehydro-compounds are formed and purification is thus simpler, but we have not practised them on a larger scale.

Pyridine also racemises the C7—C8 system. When pyridine (10 c.c.) was added to rotenone (2·5 g.) in acetone (50 c.c.), $[\alpha]_D$, originally −106°, fell to −95° in 5 minutes and −80° in 1 hour, but then remained constant overnight. When rotenone (1 g.) was heated in pyridine (5 c.c.) on the water-bath for 1 hour, the product, isolated by pouring the solution into dilute hyrochloric acid, had $[\alpha]_D$ −145° in benzene.

Oxidation of mutarotenone. Iodine (1 g.) in alcohol (10 c.c.) was added gradually to mutarotenone (1 g.) and anhydrous sodium acetate (3 g.) in hot alcohol (40 c.c.) and the whole was heated for a further 2 hours. When cold, the solution had deposited dehydrorotenone (0·82 g.), m. p. 228—230° alone or mixed with an authentic specimen and having $[\alpha]_D$ −70° in benzene (this agrees with the correct figure within our experimental error).

Isomerisation of mutarotenone. Concentrated sulphuric acid (3 c.c.) was added gradually to mutarotenone (0·5 g.; $[\alpha]_D$ −70° in benzene) in warm acetic acid (5 c.c.) and the resulting solution was poured into water. The precipitate, collected and dried, had no rotation in benzene. Crystallisation from a limited amount of alcohol gave *dl-iso*rotenone, m. p. 163—164°, identified by its giving a blue colour in the Durham reaction and not depressing the m. p. of an authentic sample. A small, less soluble residue, crystallised from alcohol, gave needles, m. p. 218—220°, identified as dehydro-compounds by their yellow colour and failure to give the Durham test; they undoubtedly arose from traces of rotenolones present in the not quite pure mutarotenone.

A sample of mutarotenone (2 g.), having $[\alpha]_D$ −110° in benzene, with acetic acid (16 c.c.) and sulphuric acid (12 c.c.) gave a crude product, $[\alpha]_D$ −23° in benzene, which, when crystallised from alcohol containing a little benzene, gave completely inactive *dl-iso*rotenone (1·7 g.), m. p. 168° alone or mixed with an authentic sample.

d-epi*Rotenone.*—Pure mutarotenone (14 g.) in carbon tetrachloride deposited 8·1 g. (41·5%) of rotenone–carbon tetrachloride solvate, identified by its rotation, $[\alpha]_D$ −217° in benzene (calc. as rotenone; pure rotenone has $[\alpha]_D$ −226°), and by conversion by alcohol into rotenone, m. p. 163—164° alone or mixed with an authentic specimen. Evaporation of the carbon tetrachloride mother-liquors gave a gum (8 g.), having $[\alpha]_D$ +8° in benzene; when kept in ether, this slowly deposited small amounts of mutarotenone, m. p. 144°, $[\alpha]_D$ −83°, and finally gave on evaporation impure *d-epi*rotenone, $[\alpha]_D$ +30° in benzene. As usually obtained, the product gave a reddish-brown colour with alcoholic ferric chloride, but removal of the phenolic

decomposition product with alkali did not lead to a crystallisable material. Attempts to prepare an oxime, hydrochloride, and acetyl derivative gave only gums.

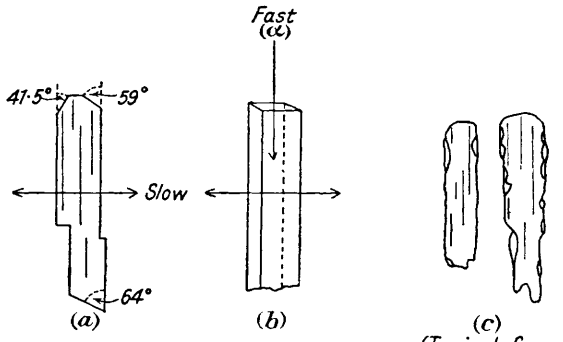
d-iso*Rotenone*.—Crude *d-epi*rotenone [2·8 g., obtained from mutarotenone prepared by method (*b*); $[\alpha]_D$ + 12° in benzene] was treated in acetic acid with sulphuric acid, as described for mutarotenone. The crude product obtained by pouring the reaction mixture into water had $[\alpha]_D$ + 34° in benzene, and, crystallised from alcohol, gave d-iso*rotenone* (1·25 g.) in long, colourless, silky needles, m. p. 182°, $[\alpha]_D$ +75° in benzene, −10° in acetone (Found: C, 70·0; H, 6·1. $C_{23}H_{22}O_6$ requires C, 70·0; H, 5·6%). Mixtures of 1 part of this product with 0·5 and 2 parts of *l-iso*rotenone melted at 166—167° and 165—166°, respectively, but equal weights of the two compounds, crystallised together from alcohol, gave *dl-iso*rotenone, α 0, m. p. 170° alone or mixed with an authentic specimen. *d-iso*Rotenone was similarly obtained from crude *d-epi*rotenone prepared by the potassium carbonate method.

*l-iso*Rotenone has $[\alpha]_D$ about −73° in benzene, +7·5° in acetone, +10° in chloroform, and +12·5° in dioxan.

d-*Dihydro*epi*rotenone* (IV).—Crude *d-epi*rotenone (2·3 g.) in ethyl acetate (25 c.c.) absorbed 130 c.c. of hydrogen in 10 minutes in the presence of a reduced Adams's platinum catalyst (0·25 g.). Hydrogenation was then stopped. The filtered solution was diluted with water, the product was removed by ether and divided into portions soluble and insoluble in aqueous sodium hydroxide. The alkali-soluble part gave only a little gum. The alkali-insoluble part, recovered from the ether, was dissolved in hot alcohol. When cold, the supernatant liquid was decanted from the gum, which had been deposited, and was kept. A small amount of crude d-*dihydro*epi*rotenone*, m. p. 123—125°, slowly separated. Purified by several crystallisations from alcohol, it formed stout prisms, m. p. 127—128°, $[\alpha]_D$ +99° in benzene (Found: C, 69·0; H, 5·9. $C_{23}H_{24}O_6$ requires C, 69·7; H, 6·1%).

FIG. 3.

d- *or* l-*Dihydrodeguelin*.



d-*Dihydrodeguelin* (d-β-*Dihydrorotenone*) (VI).—Crude *d-epi*rotenone (5 g.; $[\alpha]_D$ +25° in benzene) in dioxan (100 c.c.) in the presence of potassium acetate (5 g.) and palladised barium sulphate [5 g.; Houben–Weyl, Vol. 2, p. 270 (1922)] absorbed 260 c.c. of hydrogen in 48 minutes. The solution was filtered, and the inorganic matter thoroughly washed with dioxan. The filtrates were diluted with water and extracted with ether. The ethereal layer, freed from dioxan by several washings with water, was shaken with 5% aqueous potassium hydroxide solution and then retained only uncrystallisable material. The material recovered from the alkaline layer by acidification and extraction with ether, etc., was a gum; test portions decomposed during attempted crystallisation from benzene, benzene–light petroleum, or ether–light petroleum. To one half of the product in acetic acid (5 c.c.) was added concentrated sulphuric acid (20 drops); after 5 minutes the deep red solution was diluted with water and extracted with ether. Uncyclised and other phenolic products were removed with aqueous alkali. The ethereal solution, washed finally with water, dried, and evaporated, gave a gum, from which a pale yellow powder, m. p. 152—153°, was obtained by trituration with a little cold ether. Several crystallisations from methyl alcohol afforded d-*dihydrodeguelin* in colourless prisms (48 mg.), m. p. 154° (Found: C, 69·0; H, 5·6. $C_{23}H_{24}O_6$ requires C, 69·7; H, 6·1%).

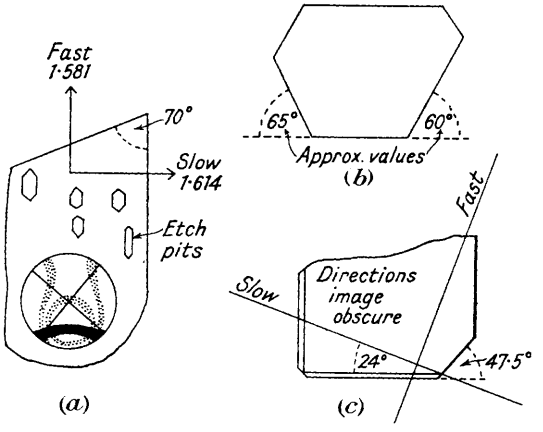
d-Dihydrodeguelin is crystallo-optically identical with the authentic *l*-form, m. p. 153—154°. Both are colourless, elongated prisms or needles, mainly without good crystal form, but occasionally showing terminations as in Fig. 3*a* and *b*. The extinction in the prism zone is straight; the length direction is fast and this is also the direction of the trace of the optic axial plane. $2V$ is large. n_D are: α (along the length) 1·521, β (across the length) 1·600, γ (across the length) 1·714 (all ±0·002). No etch pits were seen.

9·5 Mg. each of the *d*- and the *l*-form, crystallised together, gave *dl*-dihydrodeguelin, melting at 173° alone and at 173—174° when mixed with authentic specimens prepared either from deguelin or from *dl*-rotenonic acid. Our product, m. p. 173°, and an authentic specimen were crystallo-optically identical. They form thin, flat plates, mainly of irregular shape, but some-

times showing pseudohexagonal outlines (Fig. 4*b*). In convergent light a very characteristic directions image is presented, one melatope appearing about two-thirds of the distance from the centre of the field to the margin (Fig. 4*a*). The direction of the trace of the optic axial plane is fast. $2V$ is very large. n_D, measured on this section, which occurred almost to the exclusion of any other, were: in the direction of the optic plane 1·581; normal to the optic plane 1·614 (both ±0·002). Other sections showed two good cleavages and an obscure directions image, and an occasional section as shown in Fig. 4*c* was seen; the common section is, however, so characteristic that these others are not further described. Almost as characteristic were the etch-pits developed in liquids in which the substance is slowly soluble (*e.g.*, *s*-tetrabromoethane) (see Fig. 4*a*). The pseudo-hexagonal pits are elongated in the direction of the optic axial plane, and are symmetrical about this direction and one at right angles to it. They are formed by the coalescing of two groups of hemimorphic pits oriented parallel to the optic axial plane but in opposite directions.

Hydrogenation of Rotenone.—(*a*) l-*Rotenonic acid* (l-iso*dihydrorotenone*) (V). Rotenone (10 g.) in " purified " dioxan (200 c.c.) in the presence of potassium acetate (10 g.) and palladised barium sulphate (5 g.; *vide supra*; best freshly prepared and not dried, but washed with dioxan) absorbed 520 c.c. of hydrogen in 95 minutes. The mixture was then treated with much water and 450 c.c. of ether and filtered. The ethereal layer was washed thrice with 5% aqueous potassium hydroxide solution, whereupon the bulk of the sparingly soluble *l*-dihydrorotenone crystallised and was removed by filtration. Acidification of the alkaline washings precipitated *l*-rotenonic acid, which was collected in ether and, after recovery therefrom, was best crystallised from benzene, in which it is sparingly soluble; it formed colourless needles, m. p. 206—208°, $[\alpha]_D$ +46° in chloroform, +49·5° in acetone. Yield, about 45%. Hydrogenation did not occur in technical dioxan.

FIG. 4.
dl-*Dihydrodeguelin.*



(*b*) Our other experiments were not exhaustive, as our object was achieved by discovery of the above method, but a summary of our experiences is of interest. Using Adams's platinum oxide in ethyl acetate, we got 86% of pure *l*-dihydrorotenone and only a trace of phenols; under the conditions of Takei *et al.* (*loc. cit.*) [rotenone (5 g.), alcohol (500 c.c.), 10% aqueous sodium hydroxide (3 c.c.)] we got a very good yield of *dl*-rotenonic acid, m. p. 184°, α 0; hydrogenation of rotenone (5 g.) in alcohol (100 c.c.) containing 1% alcoholic sodium hydroxide (2 c.c.) was slow and incomplete, probably owing to poor quality of the batch of platinum catalyst, but the phenolic part of the product was almost entirely the *dl*-substance; addition of anhydrous sodium acetate (5 g.) to alcohol (100 c.c.) gave only a trace of phenols. Using palladised barium sulphate, we got only traces of phenols from 2 g. of rotenone by adding potassium carbonate (2 g.) to acetone or dioxan (40 c.c.); potassium acetate in acetone led to 11·5% of the active phenol; addition of water to the mixture as in (*a*) did not alter the yield. Hydrogenation in pyridine containing palladised barium sulphate gave 38% of partly racemised phenol, $[\alpha]_D$ +23·5° in chloroform.

l-Dihydrorotenone gave a Goodhue value of 111, but rotenonic acid gave no colour in the test. Both substances gave a blue colour in the Durham test.

dl-iso*Rotenone.*—After *l-iso*rotenone (10 g.) and potassium carbonate (25 g.) had been heated under reflux in acetone (100 c.c.) for 2 hours, the mixture was poured into water; the precipitate, crystallised from alcohol, gave pure *dl-iso*rotenone (8·7 g.), m. p. 169—171°, α 0. The product gave the known inactive oxime, crystallising from alcohol in leaflets, m. p. 230° (cf. Takei, *Ber.*, 1928, **61**, 1003). *l-iso*Rotenone (1 g.) was partly racemised by heating in pyridine (5 c.c.) for 1 hour on the water-bath; the product had $[\alpha]_D$ −32·5° in benzene.

The oxime of *l-iso*rotenone was obtained by us only as an amorphous powder; it separated as a gel from quite dilute or concentrated solutions in alcohol, benzene, carbon tetrachloride, etc. Rotenoneoxime was readily obtained with m. p. 240—241°, $[\alpha]_D$ +134° in acetone, +37·5° in benzene; the dextrorotation is remarkable.

Other Racemisations.—After *l*-dihydrodeguelin (10 mg.), m. p. 155—156°, and anhydrous sodium acetate (10 mg.) had been boiled in absolute alcohol (3 c.c.) under reflux for ½ hour, the solution, when cold, deposited *dl*-dihydrodeguelin, which, after washing with warm water to remove acetate, melted at 172° alone and at 173—174° when mixed with an authentic specimen, m. p. 175—176°.

A similar experiment with *l*-rotenonic acid (10 mg.), m. p. 208°, gave *dl*-rotenonic acid, which after crystallisation from benzene–ligroin melted at 183° and at 184° when mixed with an authentic specimen.

The feasability of doing such racemisations on a micro-scale is thus demonstrated.

Acetyl-dl-iso*rotenone.*—(*a*) *l-iso*Rotenone (2 g.) and anhydrous sodium acetate (1 g.) were boiled gently under reflux in acetic anhydride (40 c.c.) for 1 hour. The mixture was poured into water and stirred until most of the anhydride was destroyed. The gummy product, separated by decantation of the supernatant liquid and crystallised from alcohol, gave material, m. p. 124—126°, $[\alpha]_D$ about −3° in benzene, which was separated by fractional extraction with hot methyl alcohol into (i) a more soluble fraction (75%), consisting of the *dl*-acetate, needles, m. p. 146°, α 0 in acetone or benzene, and giving no colour in the Durham test, and (ii) a less soluble fraction, needles, m. p. 150—155°, α 0, which gave a blue colour in the Durham test and thus contained unacetylated but racemised *iso*rotenone. Acetylation for only ½ hour gave some of the *dl*-acetate, but after heating for 10 minutes no acetate could be isolated.

(*b*) *dl-iso*Rotenone (2 g.) similarly gave the acetate (1·06 g.), m. p. 146° alone or mixed with the acetate described in (*a*) (Found: C, 68·8; H, 5·5. Calc. for $C_{25}H_{24}O_7$: C, 68·8; H, 5·5%), and a less soluble fraction which gave the Durham test.

Acetyl-*dl-iso*rotenone (1·75 g.) was boiled for 2 hours with alcohol (85 c.c.) and concentrated hydrochloric acid (15 c.c.). The product, isolated by pouring the mixture into water and filtering, was crystallised from the least amount of alcohol; it gave a 72% yield of *dl-iso*rotenone, m. p. 167°; a second crystallisation gave material m. p. 170°, unchanged by admixture with an authentic specimen. This experiment was done with identical results with the acetate prepared on one occasion from *l*- and on another from *dl-iso*rotenone.

Acetylrotenone and its Hydrolysis.—When preparing acetylrotenone (Cahn and Boam, *loc. cit.*), it is essential to dissolve the crude gummy product in at least 20 parts of hot alcohol and to start crystallisation by seeding and scratching; otherwise oils separate. Crystals for seeding are best obtained from more dilute solution. After attempted hydrolysis by very dilute acid, acetylrotenone was recovered in a *form*, m. p. 159—160° (Found: C, 68·8; H, 5·75. $C_{25}H_{24}O_7$ requires C, 68·8; H, 5·5%); thereafter, this form was obtained by acetylation of rotenone. The known form, m. p. 137°, for which we found $[\alpha]_D$ −172·5° in benzene, yields the new form when crystallised from alcohol and seeded with the latter, but the reverse change could not be effected.

Acetylrotenone (10 g.) was hydrolysed by boiling for 1 hour in alcohol (425 c.c.) and concentrated hydrochloric acid (75 c.c.). The precipitate obtained by pouring the reaction mixture into water gave, when crystallised from alcohol, 6·3 g. of rotenone, $[\alpha]_D$ −228° in benzene, m. p. 161—162° alone and 162—163° when mixed with an authentic specimen. Evaporation of the alcoholic mother-liquor and dissolution of the residue in carbon tetrachloride gave 1·75 g. of rotenone–carbon tetrachloride solvate (equivalent to 1·25 g. of rotenone), which was identified by conversion by alcohol into rotenone, the authenticity of which was confirmed in the way just described. Total yield, 83%.

β-*Toxicarol.*—Ordinary (α-)toxicarol (5 g.) and anhydrous potassium carbonate (10 g.) were heated under reflux in acetone (100 c.c.) for 2 hours. The mixture was then treated with water, acidified, and extracted with ether. The ethereal layer, kept overnight, deposited α-toxicarol (1·4 g.), which was removed by filtration and identified by crystallisation from acetic acid (m. p. and mixed m. p. 226°). The residue obtained by evaporating the filtrate was triturated with alcohol; a solid (3 g.), m. p. 155—160°, separated; several crystallisations of this from alcohol yielded pure β-*toxicarol* (2·1 g.) in pale yellow plates, m. p. 165—167° (Found: C, 67·4; H, 5·1 $C_{23}H_{22}O_7$ requires C, 67·3; H, 5·4%). Our product had a Goodhue value of 88; the deficiency below 100 may have been due to a content of α-toxicarol, but we have never obtained a higher value for β-toxicarol and so believe that given to be correct. In this connexion it is noteworthy that *l*-dihydrorotenone gave a Goodhue value over 100. In the Durham test β-toxicarol gives a blue colour, fading through pale green to yellow; this is quite distinct from the deep green colour given immediately by α-toxicarol.

When β-toxicarol (0·25 g.) and anhydrous sodium acetate (0·2 g.) in acetic anhydride (10 c.c.) were gently boiled under reflux for 20 minutes, the product (0·1 g.), isolated by pouring the

reaction mixture into water and crystallising the precipitate from acetic acid, had m. p. 220—223° and did not depress the m. p. of diacetyl-α-toxicarol. This tranformation indicates ring-fission by acetic anhydride. However, in a similar experiment with 2 g. of β-toxicarol we obtained material (1 g.), which melted at 180—190° and seemed from analyses to contain about 1·5 acetyl groups, but was unchanged by further acetylation. Acetylation for 6·5 hours gave a similar product. No definitely homogeneous product could be obtained by acetic anhydride in pyridine. It is possible that acetylation of β-toxicarol is sterically hindered, for that of the α-isomeride is a very smooth reaction, in which apparently no ring fission occurs.

Effect of Alkali on β-Toxicarol.—(*a*) β-Toxicarol (1 g.) and potassium carbonate (2 g.) in acetone (20 c.c.) were heated under reflux for 1 hour. The reaction mixture was poured into water, acidified, and shaken with ether. The ethereal layer, kept overnight, deposited α-toxicarol (0·2 g.), m. p. 220—223° (mixed m. p.). Evaporation of the filtrate gave a gum, from which trituration with alcohol separated β-toxicarol (0·6 g.), m. p. 155—160°, raised to 163—166° (alone or mixed with an authentic specimen) by recrystallisation from alcohol.

(*b*) β-Toxicarol (0·5 g.) in alcohol (10 c.c.) and 10% aqueous sodium hydroxide (0·2 c.c.) was boiled for 10 minutes. The solution, when cold, had deposited crude α-toxicarol (0·3 g.), m. p. 197—203°, just as in the preparation of toxicarol from a *Derris* extract; recrystallisation from acetic acid raised the m. p. to 227—229°, alone or mixed with an authentic specimen. The alcoholic mother-liquors, poured into water and worked up as in (*a*), gave 0·1 g. of red crystals, m. p. 180—185°.

(*c*) The mixture obtained by boiling β-toxicarol (1 g.) and anhydrous sodium acetate (3 g.) in alcohol (30 c.c.) was worked up as in (*a*) and gave 0·75 g. of crude α-toxicarol, m. p. 210—212°.

Dehydro-β-toxicarol.—Iodine (1 g.) in a little alcohol was added gradually to a boiling solution of β-toxicarol (1 g.) and anhydrous sodium acetate (3 g.) in alcohol (30 c.c.). After being boiled for a further 1 hour, the mixture was poured into water. The solid iodo-compound precipitated was collected, dried on porous tile, and reduced with zinc dust (2·5 g.) in boiling acetic acid for 2 hours, further zinc dust (1 g). being added at the end of the first hour. The hot solution was filtered from the zinc, which was washed with hot acetic acid. The filtrate and washings, poured into water, yielded *dehydro-β-toxicarol*, which crystallised from alcohol in minute yellow needles (0·6 g.), m. p. 180—184°, raised to 183—184° by further crystallisation by adding hot methyl alcohol to its solution in a little chloroform. In a similar experiment, in which, however, 2 c.c. of acetic acid were added before the iodine, we obtained 0·6 g. of the dehydro-compound, m. p. 183—188°. Three specimens were analysed, but, although we believe them to have been pure, gave low values for carbon (Found: C, 66·5, 66·9, 66·7; H, 5·0, 4·8, 4·9. $C_{23}H_{20}O_7$ requires C, 67·6; H, 4·9%); similar difficulty was met with dehydrodihydro-β-toxicarol. The structure was confirmed by acetylation.

The *acetyl* derivative, prepared by boiling the dehydro-compound (0·25 g.) and anhydrous sodium acetate (0·2 g.) in acetic anhydride (10 c.c.) for 10 minutes and pouring the resulting mixture into water, was obtained after several crystallisations from alcohol as colourless crystals (0·2 g.), m. p. 196—197° (Found: C, 66·4; H, 4·9. $C_{25}H_{22}O_8$ requires C, 66·5; H, 4·9%).

Dihydro-β- and -α-toxicarol.—β-Toxicarol (2 g.) and an Adams's platinum catalyst (0·3 g.) in " purified " dioxan (50 c.c.) were stirred under hydrogen until absorption was complete. The dioxan also absorbed hydrogen in the presence of this catalyst (but not of palladised barium sulphate), so that absorption was not measured. *Dihydro-β-toxicarol*, isolated from the filtered solution by precipitation by water, crystallised from alcohol–benzene in long, colourless needles (1·8 g.), m. p. 213—215° (Found: C, 66·7; H, 5·4. $C_{23}H_{24}O_7$ requires C, 67·0; H, 5·9%).

Dihydro-α-toxicarol was similarly obtained from α-toxicarol in 80—90% yield as colourless prisms, m. p. 206—207°. α-Toxicarol could not be hydrogenated when palladised barium sulphate was used in dioxan or hot or cold acetic acid.

Both dihydro-compounds give a blue colour in the Durham test.

Dehydrodihydro-β-toxicarol.—This *substance* was prepared from dihydro-β-toxicarol (1 g.), iodine (1·4 g.), and sodium acetate (3 g.) in absolute alcohol (100 c.c.) in the way described for dehydro-β-toxicarol, except that, owing to ready crystallisation, it was more convenient to decant the acetic acid solution from the zinc dust than to filter. Yellow leaflets, m. p. 226—227°, were obtained from chloroform–alcohol. Yield, 70%. Two preparations, believed to be pure, gave erroneous figures for carbon (Found: C, 68·3, 66·3; H, 5·3, 5·0. $C_{23}H_{22}O_7$ requires C, 67·3; H, 5·4%) (cf. dehydro-β-toxicarol). The *acetyl* derivative, prepared in the usual way, formed pale yellow needles, m. p. 175—176°, from alcohol and gave correct analytical figures (Found: C, 66·5; H, 5·6. $C_{25}H_{24}O_8$ requires C, 66·4; H, 5·3%).

Dihydrotoxicarolic Acid.—50% Aqueous potassium hydroxide solution (4·2 c.c.) was added to

dehydrodihydro-β-toxicarol (0·7 g.) and zinc dust (0·7 g.) in alcohol (17·5 c.c.), and the mixture heated for ½ hour under reflux. By working as described by Clark (*J. Amer. Chem. Soc.*, 1932, **54**, 2546), we obtained a 64% yield of dihydrotoxicarolic acid. This separated from aqueous methyl alcohol, as described, in colourless plates, m. p. 123—125° (decomp.), which, contrary to Clark, we found to contain two molecules of water of crystallisation (Found: C, 57·25; H, 6·2. $C_{23}H_{26}O_9,2H_2O$ requires C, 57·25; H, 6·3%. Calc. for $C_{23}H_{26}O_9$: C, 61·9; H, 5·9%). The acid, m. p. 125—128° (decomp.), prepared similarly from dehydrodihydro-α-toxicarol in 70% yield, was also the dihydrate (Found: C, 57·4; H, 6·45%). Neither specimen gave with aqueous or alcoholic ferric chloride the purple colour described by Clark; both gave, however, the methyl ester, colourless leaflets from aqueous methyl alcohol, m. p. 174—176°, and methyl ester diacetate, colourless needles from methyl alcohol, m. p. 143°. (For substances prepared in the β-series: ester: Found: C, 62·3; H, 6·3. Calc. for $C_{24}H_{28}O_9$: C, 62·6; H, 6·1%. Diacetate: Found: C, 61·9; H, 6·2. Calc. for $C_{28}H_{32}O_{11}$: C, 61·7; H, 5·9%.) The acids and their derivatives from the α- and the β-series gave no depressions of the melting points when appropriately mixed.

Hydrolysis of Diacetyltoxicarol.—Diacetyltoxicarol (2 g.), alcohol (100 c.c.), and concentrated hydrochloric acid (5 c.c.) were heated under reflux for 1 hour, poured into water, and shaken with ether. When kept overnight, the ethereal solution deposited *dl*-α-toxicarol, m. p. 225°, in quantitative yield.

Acetylsumatrol.—Sumatrol (0·5 g.) and anhydrous sodium acetate (1 g.) in acetic anhydride (5 c.c.) were gently boiled under reflux for 10 minutes. The mixture was poured into water and stirred until most of the anhydride was destroyed. Two crystallisations of the oily residue from alcohol afforded *acetylsumatrol* in long colourless needles, m. p. 218—219°, $[\alpha]_D$ −57·5° in benzene, −20·5° in acetone (Found: C, 66·3; H, 5·4. $C_{25}H_{24}O_8$ requires C, 66·3; H, 5·35%).

Hydrolysis by 5% alcoholic hydrochloric acid gave sumatrol, $[\alpha]_D$ −180° in benzene, m. p. 195° alone or mixed with an authentic specimen, in excellent yield.

l-α-*Toxicarol.*—(*a*) *Direct isolation from an extract.* In order to obtain *l*-α-toxicarol from a commercial *Derris* extract by ethyl acetate, we found it essential first to remove interfering substances. Addition of light petroleum (b. p. 40—60°) in small portions to a Sumatra-type extract in a little benzene precipitated first very discoloured fractions, which were discarded. When the precipitated gum had only a pale yellow colour, a large volume of light petroleum was added to precipitate the remainder of the insoluble matter. This left the waxy or fatty substances in solution and these too were discarded. The light-coloured precipitates were dissolved in a little ethyl acetate and treated with alcohol until, on seeding liberally and scratching, slow crystallisation began. After being kept overnight, the crystals were collected and recrystallised a few times from ethyl acetate–alcohol or ethyl acetate alone. As thus obtained, crude *l*-α-toxicarol had m. p. 100—101°, $[\alpha]_D$ about −65° to −70° in benzene and +46° in acetone, and was very soluble in cold acetone, ethyl acetate, acetic acid, benzene, chloroform, or carbon tetrachloride, hot ether, ethyl or methyl alcohol, and, which is unusual for this series, moderately soluble in hot light petroleum. Its rotation and melting point were not materially changed by further crystallisation. Further purification is described in (*d*) below.

(*b*) The following method is the counterpart to the above, but using alkali. It is essential for good yields that contact of the alkaline and ethereal solutions should be reduced to a minimum, and, particularly if emulsification occurs, portions of the solution should be sacrificed to speed of working; otherwise oxidation and partial racemisation occur. We shall refer to a solution of 50 g. of potassium hydroxide and 250 g. of potassium chloride in 1 l. of water as the "alkaline solution." A Sumatra-type extract (100 g.) in ether (500 c.c.) was shaken with two 20 c.c. portions of the "alkaline solution" and the precipitated dark brown, oily sludges were discarded (this is essential for ease of crystallisation later). The ethereal solution was then shaken with 150 c.c. of the "alkaline solution" and decanted from the yellowish sludge. This sludge was washed once or twice with ether and then acidified. The precipitated gum was taken up in ether and the whole process was repeated to effect further cleaning and to remove entangled non-phenolic material. The phenols were finally recovered from the ether and crystallised from ethyl acetate–alcohol as described in (*a*); crystallisation was made considerably easier by the use of alkali as just described. Both methods gave substances of similar properties and the following crystallo-optical description applies to both; though not absolutely complete, it is ample to establish identity.

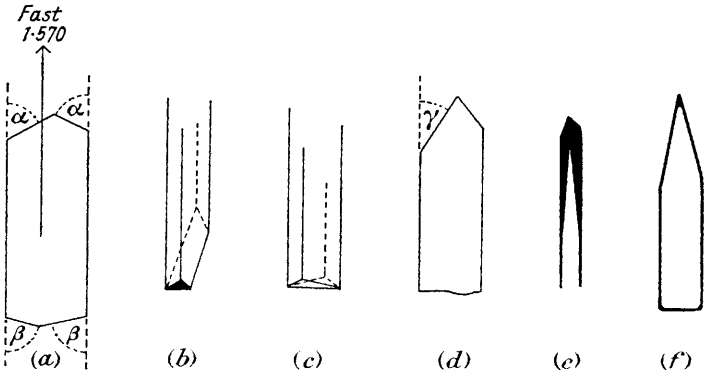
The substance separated from a mixture of equal parts of alcohol and ethyl acetate in pale pistachio green, bladed needles, most often arranged in radiating aggregates. A very characteristic form is shown in Fig. 5*a*. Different terminations, shown in Figs. 5*b*, *c*, and *d*, were common.

The needles almost always showed different terminations at opposite ends (Fig. 5*a*). Even the largest seen (0·03 mm. long) were practically non-pleochroic. Extinction was straight, the length direction fast, and the directions image in specimens as in Fig. 5*a* was either a large obtuse axial angle or else the " flash figure " of the normal to the optic axial plane. The double refraction was moderate and n_D along the length was 1·570 ± 0·003. Forms shown in Figs. 5*e* and *f* were common in some recrystallisations. Angles measured were : α 61 ± 1°, β 79 ± 1°, γ 36 ± 3°.

(*c*) The following method is the simplest and gives the best yield. A Sumatra-type extract (200 g.) in ether (1 l.) was well shaken in a succession of flat-bottomed flasks with successive portions of the " alkaline solution " until no more precipitation of potassum salts occurred (there is no need to discard the early precipitates in this case); the ethereal layer was on each occasion readily decanted from the aqueous alkaline sludge. These sludges were washed individually by shaking with ether, separated by decantation, united, and acidified; the precipitated phenols were removed in a large volume of ether. Solid material (*A*) separated and was removed after 36 hours; this (10—12 g.) was a complex mixture, from which fractional extraction with alcohol removed first crude sumatrol, m. p. 175—180°, and then high-melting products, which were probably dehydro-compounds but may have contained some *dl*-α-toxicarol.

FIG. 5.

Crude l-α-*toxicarol*, $[\alpha]_D$ *about* −68° *in benzene, about* +50° *in acetone ; crystallised from* 1 : 1 *ethyl acetate–alcohol.*



The ethereal filtrate from (*A*) was dried over sodium sulphate, concentrated to about 250 c.c., seeded, and kept for 10 days, during which time a crumpled, twisted mass of compacted, nodular aggregates of small needles separated. This (74 g.) was broken up, collected, and washed with ether, and then had m. p. 101°, $[\alpha]_D$ +50° in acetone. A gum obtained from the ethereal filtrate by evaporation gave a further 16 g. of impure *l*-α-toxicarol by the ethyl acetate–alcohol method. The material recovered from these final mother-liquors was extracted from ether by the " alkaline solution," this treatment being repeated several times; a final crop of 6 g. of crude *l*-α-toxicarol was thus obtained.

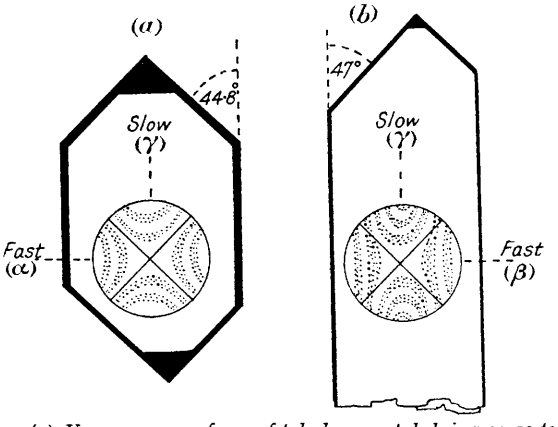
(*d*) *Purification.* Repeated recrystallisation of the products obtained by any of the above methods gave material having constant $[\alpha]_D$ +50° in acetone, +68° in benzene, and a constant m. p. about 101—102°. All the preparations, however, yielded to the following treatment. It is advisable to use batches of not more than 5 g. for the early operations, but to have at least 20 g. in all of material available. The crystals obtained from ethyl acetate were triturated with not too much ether; a small amount of colourless sumatrol remained behind; the amount of ether to be taken is best judged by the fact that the undissolved crystals are pure white. Concentration of the ethereal filtrate gave crystals, which were converted by crystallisation from ethyl acetate into purer material (*A*). The ethyl acetate mother-liquors were diluted with about 10 volumes of alcohol and shaken with water and ether (the alcohol assists removal of the ethyl acetate, which is essential); concentration of this ethereal solution gave crystals, from which trituration with a little fresh ether (*B*) separated a further quantity of sumatrol. Concentration of the ethereal filtrate (*B*) therefrom gave crystals (*A'*). Crystals (*A*) and (*A'*) were united and crystallised alternately from ether and ethyl acetate until they reached constant [α]. Ethyl acetate mother-liquors were treated as just described (alcohol, water, ether, etc.); ethereal mother-liquors were concentrated to give fresh crops. All crystals obtained from

mother-liquors were triturated with ether; when, with the purer specimens, this failed to separate sumatrol, they were crystallised alternately from ethyl acetate and ether. [α] was determined for each crop, and similar crops and similar mother-liquors were combined. In this way the bulk of the material was separated into sumatrol and pure l-α-*toxicarol*, m. p. 101—102°, $[\alpha]_D$ −53° in benzene, +69° in acetone (Found in different preparations: C, 67·3, 67·2; H, 5·9, 5·9. $C_{23}H_{22}O_7$ requires C, 67·3; H, 5·4%). The crude sumatrol obtained had m. p. about 180°; it was very readily purified by recrystallisation, probably best from acetone–alcohol or ethyl acetate, to give material, m. p. 189°. It is known to crystallise without solvent of crystallisation from alcohol and was found to do so also from ethyl acetate (Found: C, 67·65; H, 5·4. Calc. for $C_{23}H_{22}O_7$: C, 67·3; H, 5·4%).

Well-defined crystals are obtained from ethyl acetate solutions of *l*-α-toxicarol, $[\alpha]_D$ +50° in acetone, but from ether or light petroleum only if $[\alpha]_D >$ +60° in acetone. When pure, the substance forms beautiful canary-yellow needles, m. p. 101—102° (capillary tube), from ethyl acetate, ether, or ethyl acetate. From alcohol or light petroleum we got a *form*, m. p. 126—127° (Found: C, 67·3; H, 5·3. $C_{23}H_{22}O_7$ requires C, 67·3; H, 5·4%).

FIG. 6.

Pure l-α-*toxicarol, m. p.* 127°, *crystallised from alcohol.*

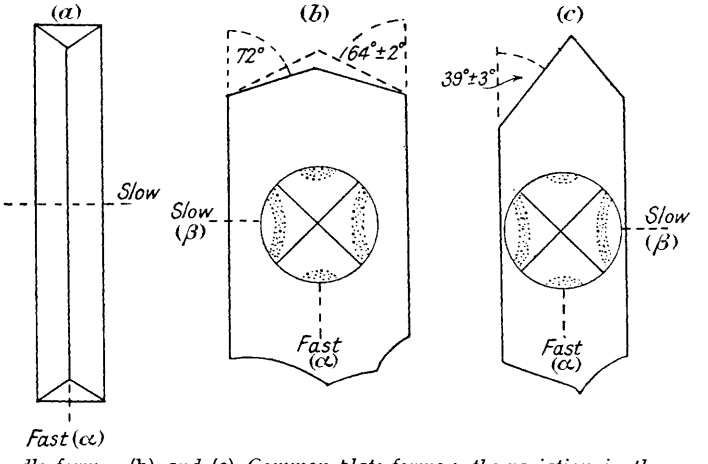


(a) *Very common form of tabular crystal, lying so as to show directions image normal to the optic axial plane.* (b) *Crystal lying on edge, to show the emergence of the acute bisectrix, 2V large, negative.*

As observed under the microscope, *l*-α-toxicarol crystallises from alcohol fairly easily in pale green, tabular crystals, m. p. 127°, having the characteristic forms shown in Fig. 6*a* and *b*.

FIG. 7.

Pure l-α-*toxicarol, m. p.* 102°, *crystallised from ethyl acetate.*



(a) *Common needle form.* (b) *and* (c) *Common plate forms; the variation in the angular values is in part due to the crystals being tilted slightly. The dotted termination in* (b) *shows another common end form. A bisectrix, probably that of the obtuse angle, emerges normal to the plate.*

Straight extinction is always given parallel to the length, which is the direction of slow vibration γ. n_D are α 1·551, β 1·664, γ 1·742 (all ±0·003). The common form (Fig. 7*a*) presents a section normal to the optic axial plane and gives α and γ. Some crystals lie at right angles to this (Fig. 6*b*) and give a large acute bisectrix directions image of negative character; these give β and γ. The optic axial angle is large, 2*V* being about 75°.

The form, m. p. 103—104° (on a warm stage), is quite distinct from that, m. p. 127°. It presents two habits: fine needles of the type shown in Fig. 7*a*, and plates, characteristic forms

of which are shown in Figs. 7*b* and *c*. The colour is very faint yellowish-green, and the fast direction is along the length of the needles and elongated plates. n_D are α 1·570, β 1·645, γ 1·679 (all ±0·003). All sections presented by plates and needles give straight extinction. The characteristic directions image given on the broad face of the plates is shown in Fig. 7*b*. It was difficult to measure γ across the edges of the plates and this value may not be so accurately determined as the others.

(*e*) The experiments described in (*a*)—(*c*) were done with extract containing 4% of rotenone and giving a ferric chloride value of 60. Two other extracts were treated by method (*b*). One, containing 2% of rotenone and giving a ferric chloride value of 68, gave material, which by repeated crystallisation was obtained with m. p. 102°, $[\alpha]_D$ +54° in acetone; treatment by method (*d*) separated sumatrol from these crystals. The other extract, containing 6% of rotenone and giving a ferric chloride value of 53, gave by the same methods a material, m. p. 102—102·5°, $[\alpha]_D$ +59° in acetone, from which also a little sumatrol was obtained by method (*d*). These additional extracts were chosen as representative of the two extremes of Sumatra-type extracts. It is plain that the occurrence of sumatrol and the difficulty of separating it from *l*-α-toxicarol are general, but it seems that simple crystallisation gives finally a material which differs slightly from case to case; the formation of a true molecular compound seems to be excluded.

(*f*) A normal *Derris* extract (100 g.), containing 28% of rotenone and giving Goodhue and ferric chloride values of 49 and 13, respectively, in ether (1 l.) was shaken with an excess of the "alkaline solution." The precipitate formed in the aqueous layer (which assumed a deep brown colour) was collected by filtration and treated with dilute acid and ether. Evaporation of the ethereal layer gave a residue, which was dissolved in ethyl acetate (about 10 c.c.), treated with alcohol (about 100 c.c.), seeded, and kept overnight. The yellowish crystalline sludge (2 g.), which had separated, was collected and triturated with cold ether, whereby a mixed insoluble residue was obtained; concentration of the ethereal filtrate gave, after cooling, crystals, from which by trituration with ether 0·15 g. of sumatrol, m. p. 178—181°, was isolated. The sumatrol, when recrystallised, had $[\alpha]_D$ −175° in benzene and m. p. 188° alone or mixed with an authentic specimen. The final ethereal mother-liquors gave by concentration impure *l*-α-toxicarol, m. p. 100°, $[\alpha]_D$ +60° in acetone. N.B. This experiment has qualitative, but not quantitative, significance.

(*g*) Acetylation of *l*-α-toxicarol was not a smooth process, no matter what the degree of purity of the material, and results were not reproducible. Material, $[\alpha]_D$ +50° in acetone, gave in various experiments small amounts of acetylsumatrol, m. p. 217°, $[\alpha]_D$ −56·5° in benzene, hydrolysed by acid to sumatrol of correct [α] and m. p. (mixed m. p.), and/or more soluble, colourless prisms, m. p. 158°, $[\alpha]_D$ +70° in acetone, separable by fractional extraction with hot alcohol. Neither acetate gave a ferric chloride colour, so that in both it is the phenolic hydroxyl group which is acetylated. The prisms appear to be *monoacetyl-*l-α-*toxicarol* (Found: C, 66·0; H, 5·2; CH_3·CO, 11·7. $C_{23}H_{21}O_7$·CO·CH_3 requires C, 66·3; H, 5·35; CH_3·CO, 9·7%), for the same product was obtained in some experiments in poor yield from pure *l*-α-toxicarol and it gave on hydrolysis by dilute alcoholic hydrochloric acid a substance, m. p. 98—101°, which did not depress the m. p. of pure *l*-α-toxicarol. We are unable to assert categorically—although it seems inevitable—the structure of this acetyl derivative, as we had insufficient material to determine [α] of the regenerated phenol. The difficulty of acetylating *l*-α-toxicarol may be due to racemisation and is paralleled by the behaviour of β-toxicarol, but is in marked contrast to the smooth acetylation of *dl*-α-toxicarol. In this connexion the acetylation of *l*-dihydro-α-toxicarol to *dl*-diacetyldihydro-α-toxicarol, described below, should be noted.

Equilibration of l-α-*Toxicarol.*—(*a*) To *l*-α-toxicarol (0·5 g.; $[\alpha]_D$ +68° in acetone) in benzene (10 c.c.) was added 1·7% methyl-alcoholic potassium hydroxide solution (4 c.c.; 1 equiv.) and thereafter within 10 seconds an excess of acetic acid. The solution was washed with water, dried, and evaporated. The residue, crystallised from alcohol, gave *l*-α-toxicarol (0·27 g.), $[\alpha]_D$ +67° in acetone, m. p. 126—128° alone or mixed with an authentic specimen.

In a similar experiment, in which, however, no acid was added, the following rotations were observed when the solution was kept:

Time, mins.	< 0·5	2	5	10	15	30	120	300	1440
$[\alpha]_D$	+350°	+280°	+224°	+193°	+171°	+140°	+70°	+46°	0°

A similar solution in benzene–methyl alcohol without alkali had $[\alpha]_D$ −46°. When worked up at the end of 24 hours, the inactive solution yielded *dl*-α-toxicarol (0·3 g.), m. p. 225—227° alone or mixed with an authentic specimen.

(*b*) 5% Aqueous potassium hydroxide solution (0·5 c.c.) was added to *l*-α-toxicarol (2 g.) in boiling alcohol (10 c.c.) and the whole was boiled for 10 minutes. The resulting rather

stiff paste was acidified with acetic acid and kept overnight. *dl*-α-Toxicarol (1·95 g.), m. p. 217—219° (mixed m. p.), was then collected in a Gooch crucible.

(*c*) *l*-α-Toxicarol (1 g.) and anhydrous sodium acetate (3 g.) in alcohol (25 c.c.) were heated under reflux for 2 hours The product, obtained by dilution with water and recrystallised, afforded *dl*-α-toxicarol (0·7 g.), m. p. 224° (mixed m. p.). In a similar experiment, in which, however, 2 c.c. of acetic acid were added to the boiling mixture, *dl*-α- (0·55 g.), m. p. 218° (mixed m. p.), and also *dl*-β-toxicarol (0·3 g.), m. p. 155° (mixed m. p.), were isolated. For separation of the two isomerides in this and similar experiments, see the preparation of β-toxicarol.

(*d*) 1% Methyl-alcoholic potassium hydroxide solution (3 c.c.) was added to *l*-α-toxicarol (2·5 g.) in acetone (50 c.c.). After 48 hours *dl*-α-toxicarol (0·9 g.) had separated. The filtrate therefrom yielded *dl*-β-toxicarol (0·85 g.).

(*e*) *l*-α-Toxicarol (1 g.) and anhydrous potassium carbonate (2 g.) in acetone (20 c.c.) were heated under reflux for 2 hours. The mixture, poured into water, gave *dl*-α- (0·4 g.) and *dl*-β-toxicarol (0·45 g.).

(*f*) When *l*-α-toxicarol (3 g.) in ether (50 c.c.) was shaken with an excess of 3% aqueous barium hydroxide, a bright yellow, insoluble barium salt was precipitated. This was soluble in acetone, but sparingly so in alcohol. The salt, washed successively with water and ether, was boiled in suspension in alcohol (150 c.c.) for 10—15 minutes. The mixture, acidified with acetic acid, gave *dl*-α-toxicarol (2 g.).

Dehydrogenation of l-α-*Toxicarol.*—Iodine (1·2 g.) in a little alcohol was added gradually to *l*-α-toxicarol (1 g.) and anhydrous sodium acetate (3 g.) in boiling alcohol (25 c.c.), and boiling was continued for a further 2 hours. Further working proceeded as described for dehydro-β-toxicarol. The product crystallised from chloroform–methyl alcohol in pale yellow needles, α 0, m. p. 228—230° alone or mixed with dehydro-α-toxicarol (m. p. 229—231°).

Dihydro-l-α-*toxicarol.*—*l*-α-Toxicarol (4 g.) and a platinum oxide catalyst (0·25 g.) in " purified " dioxan (50 c.c.) were stirred under hydrogen until absorption ceased. l-*Dihydro*-α-*toxicarol*, isolated by pouring the filtered mixture into water and crystallising the product from alcohol, formed colourless needles (2·6 g.), m. p. 178—180°, $[\alpha]_D$ −57° in benzene (Found : C, 67·5; H, 5·85. $C_{23}H_{24}O_7$ requires C, 67·0; H, 5·9%). Further crystallisation did not affect these constants. This substance crystallises very readily and is easily obtained in good yield from quite crude *l*-α-toxicarol; so obtained, however, it is completely purified only with difficulty.

A similar experiment in acetic acid gave a better yield (85%).

When this substance (0·5 g.) and anhydrous sodium acetate (0·2 g.) in acetic anhydride (5 c.c.) were gently boiled under reflux for 10 minutes and then poured into water, the residue left after destruction of the anhydride crystallised from alcohol in colourless needles (0·3 g.), m. p. 184—186°, $[\alpha]_D$ +64·5° in acetone. This was the *monoacetate* of the active compound (Found : C, 66·1; H, 5·8; $CH_3{\cdot}CO$, 9·7. $C_{23}H_{23}O_7{\cdot}CO{\cdot}CH_3$ requires C, 66·0; H, 5·8; $CH_3{\cdot}CO$, 9·7%); the phenolic group is esterified, as the compound gives no colour with ferric chloride. Hydrolysis by hot 5% alcoholic hydrochloric acid for ½ hour gave the active parent phenol of unchanged rotation and m. p. (mixed m. p.). Heating for 2 hours in a similar acetylating experiment gave 0·12 g. of inactive diacetyldihydro-α-toxicarol, m. p. 226° alone or mixed with an authentic specimen.

When 1·7% methyl-alcoholic potassium hydroxide (4 c.c.) was added to (*A*) *l*-dihydro-α-toxicarol or (*B*) its acetate (0·5 g.) in benzene (10 c.c.) and kept, the following rotations were observed :

Time, mins.	< 0·5	2	5	10	30	60	120	240	360	1440
(*A*) $[\alpha]_D$	+273°	+255°	+238°	+227°	+172°	+112°	+45·5°	+23·5°	+16°	0°
(*B*) $[\alpha]_D$	+112°	+ 72°	+ 70°	—	—	+ 30°	—	—	+17°	0°

This confirms the structure assigned to the acetate by showing that the asymmetry of the C7—C8 centre is intact.

An attempt to isolate the products of racemisation of dihydro-*l*-α-toxicarol was unsuccessful, probably because of the similar solubilities of the α- and β-dihydro-compounds.

Dehydrogenation of *l*-dihydro-α-toxicarol in the usual way gave dehydrodihydro-α-toxicarol, identified by the m. p. (258°) and mixed m. p. with an authentic specimen and by preparation from it of the acetyl derivative, m. p. 236° alone or mixed with an authentic specimen.

Mutarotation of Sumatrol.—Under the conditions described for *l*-dihydro-α-toxicarol, etc., the following rotations were observed for sumatrol :

Time, mins.	1	2	5	10	60	120	210	360	1440
$[\alpha]_D$	+105°	+84°	+59°	+49°	0°	−33·5°	−42°	−43·5°	−42°

$[\alpha]_D$ for a similar solution without alkali was −158°.

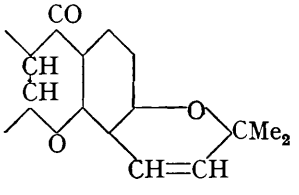
Effect of Activated Alumina on l-α-Toxicarol.—*l*-α-Toxicarol (1 g.; $[\alpha]_D$ +68° in acetone) in a little chloroform was run on to a column, 25 cm. long and 2 cm. in diameter, of B.D.H.'s "activated alumina for adsorption" (only the part retained on a 200 mesh sieve). The chromatogram was developed with chloroform until the canary-yellow zone extended down half the column; it was quite uniform. Soxhlet extraction with hot acetone–acetic acid, removal of the solvents, and dissolution of the residue in ether (5 c.c.) gave *dl*-α-toxicarol (0·06 g.), m. p. 217—219° (mixed m. p.). No other crystalline material could be isolated. A similar experiment in benzene gave 0·08 g. of *dl*-α-toxicarol as sole crystallisable product. In all experiments with activated alumina, elution of material from the column was extremely difficult. Merck's activated alumina gave similar results.

Apotoxicarol.—By Clark's method (*J. Amer. Chem. Soc.*, 1932, **54**, 2543) *dl*-and *l*-α-toxicarol gave the same (mixed m. p.) apotoxicarol, which after crystallisation from acetic acid–methyl alcohol had the recorded m. p., 246°, but contained a molecule of water of crystallisation unnoticed by Clark (Found in samples prepared from the *dl*- and the *l*-compound, respectively: C, 59·6, 58·8; H, 4·9, 4·9. $C_{18}H_{16}O_7,H_2O$ requires C, 59·3; H, 4·9%. Calc. for $C_{18}H_{16}O_7$: C, 62·5; H, 4·7%). Authenticity was confirmed by preparation of the triacetate, m. p. 206—207° (Clark gives m. p. 206°).

Action of Sulphuric Acid on the Toxicarols.—Concentrated sulphuric acid in acetic acid (cf. Wright, *loc. cit.*) transforms *l*- and *dl*-α- and β-toxicarol into substances, which crystallise with difficulty from chloroform–alcohol; these products are hard to purify owing to their sparing solubility and are hard to characterise, as they all melt above 300°. We do not know the nature of these substances, but record the following observations. All give green colours with alcoholic ferric chloride and in the Durham test. The product from *l*-α-toxicarol has $[\alpha]_D$ −208° in benzene and is racemised by alcoholic alkali. Dehydrogenation by iodine, etc., gives products, which are presumably dehydro-compounds, since they give no colour in the Durham test.

Absorption Spectra.—The determinations of absorption spectra were made on a Hilger E_3 quartz spectrograph used in conjunction with a "Spekker" photometer. The solvent used was (unless otherwise stated) ethyl alcohol specially purified until it was completely free from substances absorbing above 2200 A. Some absorption curves are shown in Fig. 2, and the exact locations of the absorption maxima, together with the intensities of absorption (as molecular extinction coefficients ε) at the maxima, are tabulated below. The determinations were made immediately after the preparation of the solutions, it having been found that several of the compounds deteriorated rapidly in alcohol (in 24 hours).

Our curve for rotenone is very similar to that reported by Seaber (*J. Soc. Chem. Ind.*, 1937, **56**, 168T). Seaber found the maximum at 2930—2940 A. (ours is at 2960 A.) and his ε_{molar} is 18,400 (ours is 16,000). These differences are possibly due to difference in the solvent, Seaber having used *iso*propyl alcohol.



The absorption of sumatrol is very similar to that of rotenone. Deguelin, as shown in the annexed partial formula, bears the same relation to toxicarol as rotenone does to sumatrol, *i.e.*, it has the same structure in ring *E* but no hydroxyl group in ring *D*. The absorption of deguelin closely resembles that of toxicarol. We consider this as confirming the structure (XXII) assigned to sumatrol (cf. p. 522).

In both pairs, rotenone–sumatrol and deguelin–toxicarol, introduction of a phenolic hydroxyl group has intensified the absorption and displaced the principal maximum 20—35 A. towards the visible region; although these displacements are not a great deal larger than the experimental error, they are probably real, since they occur in both cases.

	Wave-lengths of maxima in A.	Mol. extinct. coeff. ε.		Wave-lengths of maxima in A.	Mol. extinct. coeff., ε.
Rotenone	2960	16,000	Diacetyltoxicarol	3775	25,800
	2370	14,400		3590	27,600
Sumatrol	2980	23,000		2960	15,500
	(2350)	16,600		2580	25,400
Deguelin	2690	29,500	Dehydrorotenone	3110	18,600
	(3150)	9,300		2800	21,800
Toxicarol	2725	34,700		2365	28,000
	2970	11,300	M/20,000-Toxicarol in M/20-aqueous alcoholic NaOH	2800	27,000
Acetylrotenone	3730	27,500			
	3540	29,500			
	2500	17,300			

Comparison of diacetyltoxicarol with acetylrotenone shows a displacement of the main absorption maxima with the former substance 45—50 A. towards longer wave-lengths. In this case the extra hydroxyl group is masked. Both substances contain the conjugated system, aromatic ring *A*-C7 : C15-aromatic ring *D*. In diacetyltoxicarol the ethylenic linking of ring *E* is conjugated with ring *D* and so extends the system; it is possible that this accounts for the displacement. It is true that, by analogy with Kuhn's diphenylpolyenes, one might expect a much higher displacement (about 150 A.) for an additional ethylenic linking, but the structural analogy between these polyenes and substances of the rotenone series is far from close, and we believe the small positive displacement observed to be significant.

The maximum of absorption for toxicarol in aqueous alcoholic sodium hydroxide is not displaced by any means to the position of diacetyltoxicarol, as might be expected if complete enolisation took place. Complete, or nearly complete, enolisation is indicated by us for toxicarol in benzene–methyl alcohol containing much potassium hydroxide, but our chemical work gives no indication of the quantitative extent of enolisation under other conditions. The displacement of the principal maximum of toxicarol from 2725 to 2800 A. in aqueous alcoholic sodium hydroxide is definitely significant of some enolisation. No displacement occurred with rotenone under these conditions.

We are indebted to Messrs. Cooper, McDougall and Robertson, Ltd., Berkhamsted, for permission to publish these results.

We gratefully acknowledge our indebtedness to Mr. A. E. Gillam for determining and interpreting the absorption spectra which we report, to Mr. A. Stuart for determining and interpreting the crystallo-optical data, and to Professor I. M. Heilbron for permission to have the absorption spectra determined in his laboratory.

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[*Received, January 13th*, 1938.]