CHEMICAL EXAMINATION OF INDIAN LICHENS

Part VI. Constitution of Erythrin

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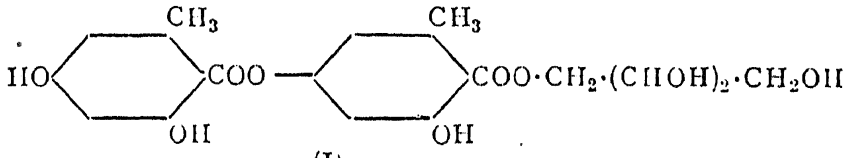
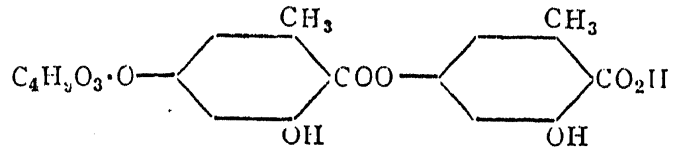
IN previous communications relating to Indian lichens the occurrence of erythrin in *Roccella montagnei* along with roccellic acid, lecanoric acid, montagnetol, orcinol and erythritol was recorded.[1] This compound has also been known to be the chromogen of many lichens belonging to the *Roccella* genus (*e.g.*, *Roccella tinctoria*) and also in certain others like *Lecanora tartarea, Dendographa leucophæa, Aspicilia calcarea, Parmelia olivetorum* and *Evernia furfuracea*.[2] Its constitution has remained so far unsettled, though its molecular formula is definitely known to be $C_{20}H_{22}O_{10}$, H_2O. From a consideration of its reactions several alternative constitutions were suggested. Hesse[3] was of the opinion that it existed in two easily interconvertible tautomeric forms: (1) the erythrityl ester form (I), (2) the erythric acid form in which the erythrityl group is attached to an ether linkage (II). Zerner[4] criticised this idea of tautomerism as untenable and the formulæ as unsatisfactory. After referring to another constitution suggested by Luynes (III) he advocated the adoption of structure (IV) as the most satisfactory. No unequivocal proof has, however, been given so far in support of any of these formulations.



(I)



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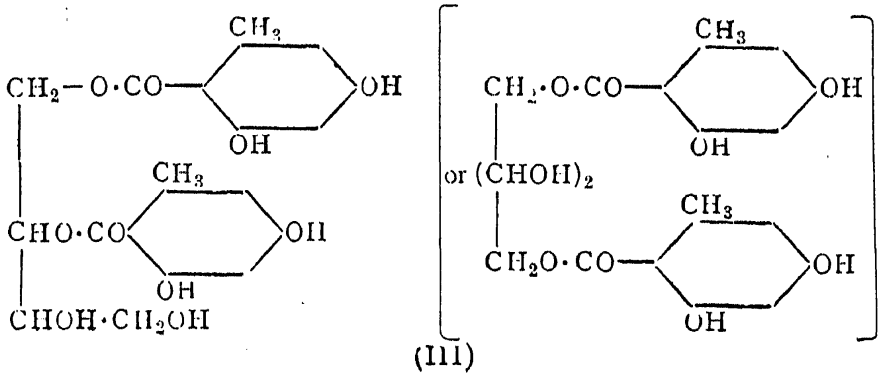
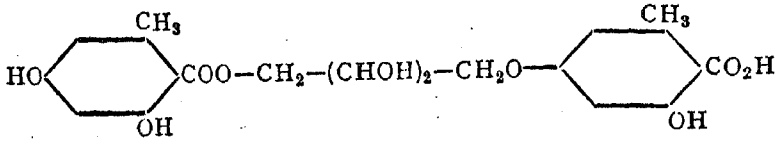
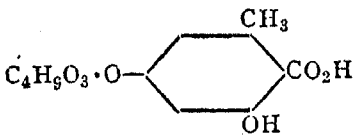


(III)



(IV)



(V)

The important properties and reactions which these formulæ were intended to satisfy were as follows :

(1) By treatment with baryta, erythrin yielded orcinol, erythritol and carbon dioxide.

(2) By gentler decomposition using boiling water or alcohol picroerythrin was formed along with orsellinic acid when water was employed, and esters of orsellinic acid when alcohols were employed.

(3) Erythrin and picroerythrin were considered to be acidic in nature.

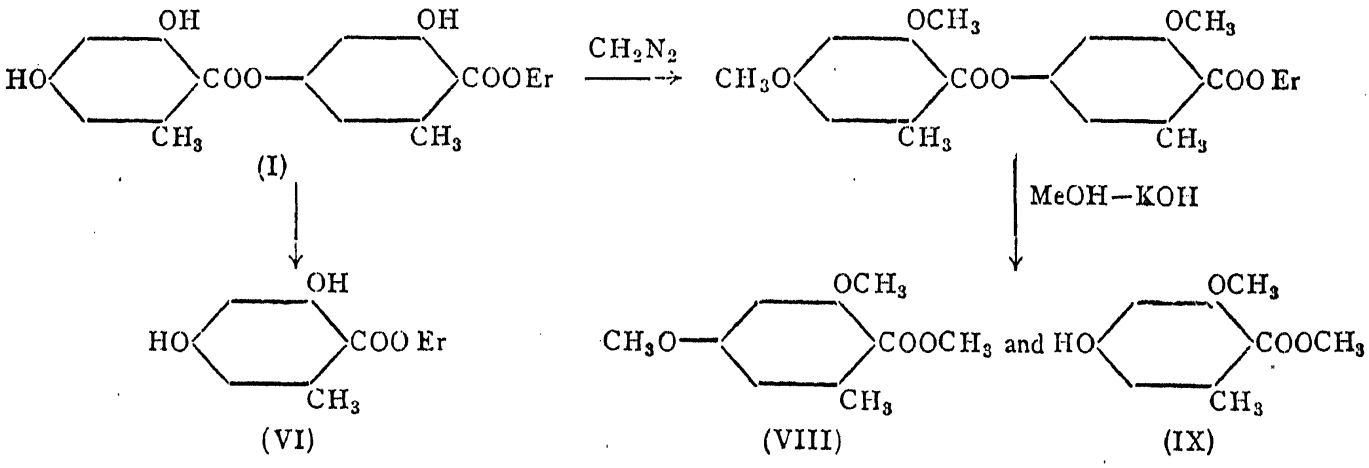
(4) Picroerythrin (also known as erythroorsellinic acid) underwent further decomposition with baryta water into erythritol, orcinol and carbon dioxide and was considered to have formula (V).

Though both formulæ (II) and (IV) explain all these properties and reactions satisfactorily, the occurrence of lecanoric acid along with erythrin was considered in Part I[1] as support for Hesse's formula (II).

A careful examination of a pure sample of erythrin shows that it does not have a free carboxylic group. Although it dissolves fairly readily in lime water and sodium carbonate it is found to be insoluble in sodium bicarbonate. On the other hand, lecanoric acid dissolves readily in aqueous bicarbonate with brisk evolution of carbon dioxide. The feeble acid properties of erythrin are obviously due to the presence of a number of phenolic hydroxylic groups and there is no carboxyl group in the molecule. A sample of picroerythrin obtained by the decomposition of erythrin with water is again found to be non-acidic. It is soluble in water to form a neutral solution and does not give effervescence with sodium bicarbonate. From its melting point and principal reactions it is found to be identical with inactive or racemic montagnetol. By carefully regulating the decomposition of erythrin with water a sample of optically active picroerythrin (*d*-montagnetol) could also be obtained. It is therefore clear that active montagnetol is the first product of decomposition and it subsequently racemises. This close relationship is in accordance with the occurrence of erythrin and montagnetol together in

the lichens. It may be noted here that erythrin exhibits optical activity being dextrorotatory just as montagnetol, and the molecular rotations of the two in acetone solution are very close (montagnetol 3427, erythrin 3376).

The constitution of montagnetol has been recently established as the erythrityl ester of orsellinic acid (VI) from a detailed study of all its properties and reactions[5] and now it is shown that montagnetol and picroerythrin are identical. Hence formula (V) has to be discarded. On account of this and also on account of the fact that erythrin is not a carboxylic acid, the formulæ (II) and (IV) become untenable. The other possibilities, (I) and (III) indicate that erythrin is a neutral compound and contains within the molecule the montagnetol unit. Of these (III) does not contain a lecanoric acid unit and further, on hydrolysis, it may not be expected to yield picroerythrin readily, since both the orsellinic acid units may be expected to be liberated with almost equal facility. On the other hand, formula (I) is free from these objections and explains all the properties of erythrin and its relationship with other compounds satisfactorily. This conclusion has been verified by methylating erythrin with excess of diazomethane and hydrolysing the product. A trimethyl ether of erythrin (VII) is first formed and when it is hydrolysed with alcoholic potash it gives rise to a mixture of esters which has been separated into the methylester of dimethyl orsellinic acid (VIII) and the methyl ester of isoeverninic acid (IX). The esters were also hydrolysed to the corresponding acids. The various stages of the decompositions can be represented as below :



[Er = $C_4H_9O_3$, erythrityl group]

As the result of our detailed study of the components of *Roccella montagnei* and the establishment of the constitutions of montagnetol and erythrin a possible evolution of these substances in the lichen could be suggested. Though orsellinic acid does not occur free in nature, it seems to be obviously the primary product and on account of its instability it does not

survive. A small part of it decomposes to orcinol; but the main bulk seems to be stabilised (1) by conversion into the erythrityl ester, montagnetol, (2) by depside formation as lecanoric acid, and (3) by further esterification of lecanoric acid to produce erythrin. The alternative method for the formation of erythrin by the combination of orsellinic acid and montagnetol is also possible. It could be inferred that under the conditions prevailing in the lichen erythrityl ester formation is largely favoured since lecanoric acid occurs only occasionally and in smaller amounts and the bulk of the components are accounted for by montagnetol and erythrin. It is remarkable however that the aliphatic lichen acid, roccellic acid, is found free and its erythrityl ester has not yet been isolated.

Experimental

Properties of erythrin.—A pure sample of erythrin crystallised from aqueous acetone in the form of bundles of colourless needles melting at 156–57°. It was very sparingly soluble in water and the aqueous solution was neutral to litmus. Though lime water and sodium carbonate dissolved it, it was insoluble in sodium bicarbonate solution. $[\alpha]_D$, + 8·0. It is not markedly bitter in taste.

Preparation of picro-erythrin.—Erythrin (0·2 g.) was boiled under reflux with 20 c.c. of water. Complete solution was effected in about an hour. The heating was, however, continued for one more hour. The solution was then extracted with ether repeatedly in order to remove orsellinic acid and orcinol, and the aqueous solution concentrated on a water-bath. A viscous liquid was first obtained and it underwent change into a crystalline solid on keeping. When recrystallised from a mixture of acetone, ether and benzene, it was obtained in the form of colourless rectangular tablets melting at 156–57°. It gave a purple-violet colouration with ferric chloride and was found to be identical in every respect with racemic montagnetol. The mixed melting point was unchanged.

If the boiling was discontinued as soon as a clear solution was obtained and the solution worked up, *d*-montagnetol, melting at 136°, could be obtained, but it was always accompanied by small amounts of the racemic variety also and purification was very difficult.

Methylation of erythrin—preparation of trimethyl erythrin.—Erythrin (1 g.) was dissolved in anhydrous methyl alcohol (50 c.c.) and treated with excess of ethereal diazomethane in two lots in the course of two days. After the first day as much of the ether as possible was removed by gentle heating and the second lot of ethereal diazomethane added. The solvents were finally distilled off and the semi-solid residue crystallised from methyl

alcohol. It came out in the form of colourless, fibrous needles, which turned to a glassy mass at 110–12° and ran down into a clear liquid at 118–20°. It gave no colour with alcoholic ferric chloride solution and was insoluble in aqueous sodium hydroxide. [Found: CH_3O, 20·8%; required for 3 MeO groups in $C_{23}H_{28}O_{10}$, 20·0%.]

Decomposition of trimethyl erythrin.—Trimethyl erythrin (0·4 g.) was carefully heated with methyl alcoholic potash (4%, 20 c.c.) for two hours on a water-bath, the temperature being kept at about 55°. The solution was diluted with an equal amount of water and repeatedly extracted with ether. When the ether solution was concentrated it yielded a viscous liquid which slowly turned solid on keeping for several hours. It could then be recrystallised from alcohol or benzene, and from the latter it came out as colourless prisms melting at 44–45°. It gave no colour with ferric chloride. [Found: OCH_3, 44·2%; required for 3-OCH_3-in $C_{11}H_{14}O_4$, 43·3%.] The ether-soluble product was therefore identified as the methyl ester of dimethyl orsellinic acid. A small sample of it was hydrolysed by boiling with 5% aqueous-alcoholic (1 : 1) potassium hydroxide solution for 15 minutes. After leaving the solution overnight it was acidified and repeatedly extracted with ether. The ether extract yielded on evaporation a colourless crystalline solid which crystallised from methyl-alcohol in the form of sheaves of needles and melted at 140–41° with slow decomposition. It gave no colour with alcoholic ferric chloride and was found to be identical with an authentic sample of dimethyl orsellinic acid.

The original aqueous solution obtained from the decomposition of trimethyl erythrin and left after the removal of the ester of dimethyl orsellinic acid by ether extraction, was at this stage acidified and repeatedly extracted with ether. The combined ether extract was shaken with aqueous sodium bicarbonate (solution X) in order to remove any acid that might have been formed. When the ether solution was then evaporated, a semi-solid residue was left. Since the quantity was small, it was directly hydrolysed by heating with 10 c.c. of 5% aqueous potash for 15 minutes on a boiling water-bath and subsequently allowing it to stand at room temperature for 1 hour. It was then acidified and repeatedly extracted with ether. On evaporating the ether solution a crystalline solid was obtained. It was repeatedly washed with benzene. The solid was in the from of colourless rectangular plates and prisms melting with decomposition at 174–75° [Found: -OCH_3, 17·7%; $C_9H_{10}O_4$ requires for 1-OCH_3, 17·0%.] With alcoholic ferric chloride it gave only a reddish-yellow colour. The substance was therefore identified as isoeverninic acid.

The bicarbonate extract (solution X) was carefully examined as below: It was acidified with hydrochloric acid and repeatedly extracted with ether. From the ether extract a solid was obtained which had an indefinite melting point and which could, however, be separated by treatment with benzene. The benzene-insoluble portion melted at 174–75° and proved to be isoeverninic acid, whereas the benzene-soluble protion melted at 140° with slight decomposition and was found to be dimethyl orsellinic acid.

Summary

The constitution of erythrin has been definitely established. The new important observations now made are as follows: (1) it is not a carboxylic acid ; (2) it is optically active ; (3) picroerythrin is identical with montagnetol ; (4) trimethyl erythrin a compound in which all the phenolic hydroxyl groups are methylated is obtained by the action of diazomethane; (5) on hydrolysis with alcoholic potash the methyl ether yields the ester of dimethyl orsellinic acid and the ester of isoeverninic acid. Consequently, it is concluded that erythrin is the erythrityl ester of lecanoric acid. This is in agreement with the occurrence of lecanoric acid and montagnetol along with erythrin in the lichens.

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4. Zerner .. *Monatsh.*, 1914, **35**, 1021.
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