100. *Analogues of Rotenone and Related Compounds. Part I. Chromeno-(3' : 4' : 4 : 3)-coumarins.*

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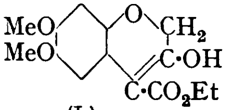
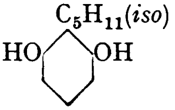
It is well established that members of the rotenone group of natural compounds possess, in addition to other physiological properties, an exceptionally high degree of toxicity to fish and certain insects. Some of the related natural furanocoumarins of the bergapten type also are capable of acting as fish poisons and it appeared that an investigation of the toxicity of analogous pyrones, dihydropyrones, pyrans, and furano-compounds would yield results of considerable interest and would, if sufficiently extensive, contribute considerably to our knowledge of structure in relation to this particular aspect of physiological activity. Moreover the rotenone molecule is complex, comprising three heterocyclic systems, and little is known regarding the features which are essential for toxicity and those, if any, which may be considered to contribute an auxotoxic effect (compare Gersdorff, *J. Amer. Chem. Soc.*, 1930, **52**, 5051; 1933, **55**, 1147; 1934, **56**, 979). To this problem a study of compounds embodying one or more of the heterocyclic systems present in the rotenone molecule (*i.e.*, sections of the rotenone molecule) would appear to offer a fertile approach. Accordingly, we have initiated a comprehensive investigation on the synthesis of analogues of rotenone and related compounds with the object of making a comparative study of their physiological properties when a sufficiently representative range of substances has been obtained. This topic is cognate to experiments on the synthesis of rotenone which are proceeding in these laboratories.

The present communication deals with the synthesis by general methods of a series of chromeno-(3' : 4' : 4 : 3)-coumarins (type III), some of which are isomeric with the chromeno-(3' : 4' : 2 : 3)-chromones previously described (Robertson, J., 1933, 489, 1163; King and Robertson, J., 1935, 993); as an example, the condensation of the enol-ester (I) with tetrahydrotubanol (II) according to the method of Pechmann gave rise to *7-hydroxy-6' : 7'-dimethoxy-8-*iso*amylchromeno-(3' : 4' : 4 : 3)-coumarin* (III), isomeric with tetrahydrodehydrorotenone (*loc. cit.*).


(I.)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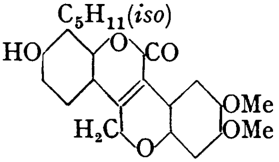
(III.)

The condensation of (I) and of ethyl 3-hydroxy-8-methoxy-Δ^3-chromene-4-carboxylate with phenols of the resorcinol type proceeded remarkably well by the sulphuric acid method. The failure to effect interaction between ethyl 3-hydroxy-7-methoxy-Δ^3-chromene-4-carboxylate and resorcinol or phloroglucinol by this means led us to employ alcoholic hydrogen chloride as the condensing agent, a procedure first used by Robinson and his co-workers (J., 1918, **113**, 879) and more recently extended by Appel (J., 1935, 1031), whose memoir appeared while the present work was in progress. Where comparison has been possible, the products obtained by the latter method are generally cleaner and the yields occasionally better.

The chromeno-(3' : 4' : 4 : 3)-coumarins are in general more intensely coloured and less soluble in organic solvents than the analogous chromenochromones of the dehydrorotenone type (*loc. cit.*). With concentrated sulphuric acid the coumarins derived from the ester (I) form dark red or purple solutions which become pale yellow on the addition of water, whereas those from ethyl 3-hydroxy-7-methoxy- and ethyl 3-hydroxy-8-methoxy-Δ^3-chromene-4-carboxylate form bright yellow solutions which, except that of *7-hydroxy-7'-methoxy-8-*iso*amylchromeno-(3' : 4' : 4 : 3)-coumarin*, are non-fluorescent in daylight; the solution of the last coumarin exhibits an intense green fluorescence.

EXPERIMENTAL.

7-Hydroxy-6′ : 7′-dimethoxychromeno-(3′ : 4′ : 4 : 3)-coumarin.—(A) Resorcinol (0·7 g.) was gradually added to a solution of ethyl 3-hydroxy-6 : 7-dimethoxy-Δ^3-chromene-4-carboxylate (Robertson and Rusby, this vol., p. 212) (0·4 g.) in 85% sulphuric acid (7 c.c.) at 0° and the resulting dark brownish-red mixture was kept for 48 hours at room temperature ; a considerable amount of crystalline material gradually separated. The mixture was poured on ice, the aqueous liquor saturated with ammonium sulphate, and next day the *coumarin* was collected, washed with a little water, and crystallised from dilute alcohol (charcoal), forming a *hydrate* (0·25 g.) in slender yellow needles, m. p. 245° with slight sintering at 240°, having a negative ferric reaction (Found in material dried in a vacuum at room temperature : C, 62·9 ; H, 4·6. $C_{18}H_{14}O_6,H_2O$ requires C, 62·8 ; H, 4·7%. Found in a specimen dried in a high vacuum at 120° : C, 65·9 ; H, 4·4. $C_{18}H_{14}O_6$ requires C, 66·3 ; H, 4·3%). The compound is readily soluble in acetone and aqueous sodium hydroxide and sparingly soluble in water, ethyl acetate, and acetic acid. Refluxed with acetic anhydride (5 c.c.) and sodium acetate (0·1 g.) for ½ hour, the coumarin (0·18 g.) yielded the *acetate* (0·13 g.), which separated from a large volume of alcohol in bright yellow, elongated plates, m. p. 220—221° (Found in material dried in a high vacuum at 120° : C, 65·6 ; H, 4·6. $C_{20}H_{16}O_7$ requires C, 65·2 ; H, 4·4%). The use of pyridine and acetic anhydride at room temperature gave the same derivative.

(B) Dry hydrogen chloride was slowly led into a solution of the ester (0·8 g.) and resorcinol (1·2 g.) in methyl alcohol (20 c.c.) maintained at room temperature for 1 hour. The mixture was then heated at 50° for 5 minutes to redissolve a small amount of solid, cooled, saturated with hydrogen chloride, kept for 4 days, and warmed at 50° for 10 minutes. Next day the solid (0·3 g.) was collected, washed, and crystallised from dilute alcohol, forming yellow needles, m. p. and mixed m. p. 245° with sintering at 240°. The acetate had m. p. 220—221°, alone or mixed with an authentic specimen.

7 : 8-Dihydroxy-6′ : 7′-dimethoxychromeno-(3′ : 4′ : 4 : 3)-coumarin, prepared from the ester (0·4 g.) and pyrogallol (0·7 g.) with 85% sulphuric acid (7 c.c.), separated from dilute alcohol as a *hydrate* in yellow needles, m. p. 274° (decomp.) after sintering at 268°, sparingly soluble in water or hot benzene and readily soluble in acetone or warm ethyl acetate (Found in a specimen dried in a vacuum : C, 60·2 ; H, 4·5. $C_{18}H_{14}O_7,H_2O$ requires C, 60·0 ; H, 4·5%. Found in material dried in a high vacuum at 120° : C, 63·2 ; H, 4·2. $C_{18}H_{14}O_7$ requires C, 63·2 ; H, 4·1%). With aqueous sodium hydroxide this coumarin gives an orange solution and with alcoholic ferric chloride an olive-green coloration. The *diacetate* crystallised from much alcohol in aggregates of elongated yellow plates, m. p. 232—233° (Found : C, 61·7 ; H, 4·3. $C_{22}H_{18}O_9$ requires C, 62·0 ; H, 4·3%).

5 : 7-Diacetoxy-6′ : 7′-dimethoxychromeno-(3′ : 4′ : 4 : 3)-coumarin.—Prepared by the Pechmann method, the hydroxy-coumarin, which was only very sparingly soluble in the usual solvents, was not obtained analytically pure. On being concentrated, a solution of the reaction product in much alcohol (charcoal) deposited partly purified material in microscopic clusters of yellow needles, m. p. above 300°, readily soluble in alkali, having a negative ferric chloride reaction, and yielding the *diacetate*, which crystallised from acetic acid or a large volume of alcohol in bright yellow rods, m. p. 190° (Found : C, 62·0 ; H, 4·3. $C_{22}H_{18}O_9$ requires C, 62·0 ; H, 4·3%).

*7-Hydroxy-6′ : 7′-dimethoxy-8-*iso*amylchromeno-(3′ : 4′ : 4 : 3)-coumarin* was obtained from tetrahydrotubanol (Robertson, *loc. cit.*) (0·5 g.) and the ester (0·5 g.) with 85% sulphuric acid (8 c.c.), and separated from alcohol in bright yellow, irregular, elongated plates, m. p. 230—231°, readily soluble in acetone or ethyl acetate and having a negative ferric reaction (Found in material dried in a high vacuum at 120° : C, 69·4 ; H, 6·1. $C_{23}H_{24}O_6$ requires C, 69·7 ; H, 6·1%). The *acetate* formed lustrous, bright yellow plates, m. p. 137°, from alcohol (Found in material dried at 120° in a high vacuum : C, 68·5 ; H, 6·0. $C_{25}H_{26}O_7$ requires C, 68·5 ; H, 5·9%).

The following 7′-methoxychromenocoumarins were prepared from ethyl 3-hydroxy-7-methoxy-Δ^3-chromene-4-carboxylate (Robertson and co-workers, this vol., p. 422) by the alcoholic hydrogen chloride method. A solution of the ester (0·3 g.) and the requisite phenol (0·4 g.) in methyl alcohol (3 c.c.) was saturated with hydrogen chloride, kept for 2 days at room temperature, and warmed on the steam-bath for ½ hour. Twenty-four hours later, the crystalline product (0·2—0·25 g.) was collected and purified. The acetyl derivatives were prepared by means of warm acetic anhydride and pyridine.

7-Hydroxy-7′-methoxychromeno-(3′ : 4′ : 4 : 3)-coumarin separated from methyl alcohol in yellow rods, m. p. 266°, sparingly soluble in acetone and having a negative ferric reaction (Found in material dried in a high vacuum at 110°: C, 68·8; H, 4·1. $C_{17}H_{12}O_5$ requires C, 68·9; H, 4·1%). The *acetate* formed yellow rods, m. p. 222°, from acetic acid (Found: C, 67·4; H, 4·1. $C_{19}H_{14}O_6$ requires C, 67·4; H, 4·2%).

7 : 8 : *Dihydroxy*-7′-*methoxychromeno*-(3′ : 4′ : 4 : 3)-*coumarin* formed tiny yellow needles, m. p. 254°, from alcohol, soluble in acetone and giving an olive-green coloration with alcoholic ferric chloride (Found: C, 65·3; H, 3·9. $C_{17}H_{12}O_6$ requires C, 65·4; H, 3·9%). This coumarin was also prepared in small yield by means of 86% sulphuric acid. The *diacetate* crystallised from acetic acid in yellow needles, m. p. 231·5° (Found: 63·5; H, 3·9. $C_{21}H_{16}O_8$ requires C, 63·6; H, 4·1%).

5 : 7-*Dihydroxy*-7′-*methoxychromeno*-(3′ : 4′ : 4 : 3)-*coumarin* separated from alcohol in yellow rods, m. p. 309°, soluble in acetone and having a negative ferric reaction (Found in material dried in a high vacuum at 110°: C, 65·1; H, 4·0. $C_{17}H_{12}O_6$ requires C, 65·4; H, 3·9%). The *diacetate* formed pale yellow rods from acetic acid, m. p. 253° (Found: C, 63·9; H, 4·2. $C_{21}H_{16}O_8$ requires C, 63·6; H, 4·1%).

7-*Hydroxy*-7′-*methoxy*-8-iso*amylchromeno*-(3′ : 4′ : 4 : 3)-*coumarin* crystallised from alcohol in tiny yellow needles, m. p. 228°, soluble in acetone or acetic acid and having a negative ferric reaction (Found: C, 72·1; H, 6·0. $C_{22}H_{22}O_5$ requires C, 72·1; H, 6·0%). The yield of this coumarin was increased by about 60% when the condensation was carried out in ethyl instead of methyl alcohol. The *acetate* formed yellow rods, m. p. 219°, from acetic acid (Found: C, 70·4; H, 5·7. $C_{24}H_{24}O_6$ requires C, 70·6; H, 5·9%).

The condensation of ethyl 3-hydroxy-8-methoxy-Δ^3-chromene-4-carboxylate (Robertson and co-workers, *loc. cit.*) with resorcinol, pyrogallol, and phloroglucinol was carried out by the sulphuric acid method and by the alcoholic hydrogen chloride method (yields approximately the same); in the latter process it was found that when ethyl alcohol was replaced by methyl alcohol the yields diminished.

7-*Hydroxy*-8′-*methoxychromeno*-(3′ : 4′ : 4 : 3)-*coumarin* formed clusters of pale yellow needles, m. p. 269°, from much alcohol, having a negative ferric chloride reaction and giving an almost colourless solution in aqueous sodium hydroxide (Found: C, 68·6; H, 4·1. $C_{17}H_{12}O_5$ requires C, 68·9; H, 4·1%). The *acetate* crystallised from acetic acid in pale yellow needles, m. p. 232° (Found: C, 67·2; H, 4·1. $C_{19}H_{14}O_6$ requires C, 67·4; H, 4·2%).

7 : 8-*Dihydroxy*-8′-*methoxychromeno*-(3′ : 4′ : 4 : 3)-*coumarin* was obtained from acetic acid in slender, pale yellow prisms, m. p. 277°; the ferric chloride reaction was olive-green and the solution in aqueous sodium hydroxide orange-red (Found in a specimen dried at 120° in a high vacuum: C, 65·3; H, 3·9. $C_{17}H_{12}O_6$ requires C, 65·4; H, 3·9%). The *diacetate* formed clusters of pale yellow needles, m. p. 246° (decomp.) (Found: C, 63·7; H, 4·2. $C_{21}H_{16}O_8$ requires C, 63·6; H, 4·1%).

5 : 7-Dihydroxy-8′-methoxychromeno-(3′ : 4′ : 4 : 3)-coumarin could not be satisfactorily purified; dilution of an alcoholic solution with water gave clusters of yellow microscopic prisms decomposing at about 290°, having a negative ferric chloride reaction, and forming an orange solution in aqueous sodium hydroxide. The well-crystallised *diacetate* separated from acetic acid in pale yellow needles, m. p. 224° (Found: C, 63·4; H, 4·1. $C_{21}H_{16}O_8$ requires C, 63·6; H, 4·1%).

7-*Hydroxy*-8′-*methoxy*-8-iso*amylchromeno*-(3′ : 4′ : 4 : 3)-*coumarin* (0·5 g.) was prepared from the requisite ester (0·5 g.) and tetrahydrotubanol (0·5 g.) in alcohol (6 c.c.) by means of hydrogen chloride in the course of 3 days, and crystallised from alcohol and then acetic acid, forming pale yellow needles, m. p. 245°, having a negative ferric reaction and giving a pale yellow solution in aqueous sodium hydroxide (Found in material dried at 110° in a high vacuum: C, 71·9; H, 5·7. $C_{22}H_{22}O_5$ requires C, 72·1; H, 6·0%). The *acetate* separated from alcohol in straw-coloured needles, m. p. 188° (Found: C, 70·8; H, 6·1. $C_{24}H_{24}O_6$ requires C, 70·6; H, 5·9%).

Phenoxyacetic-2-*acetic Acid*.—The initial material, ethyl 2-aldehydophenoxyacetate (Robertson, *loc. cit.*), for the preparation of phenoxyacetic-2-pyruvic acid has now been found, after repeated purification from light petroleum (b. p. 80—100°), to melt at 47—48° as stated by Dumont and Kostanecki (*Ber.*, 1909, **42**, 911).

(A) The azlactone of this aldehyde (Robertson, *loc. cit.*) (25 g.) was hydrolysed with boiling 10% aqueous sodium hydroxide (250 c.c.) in the course of 6 hours. After the addition of water (50 c.c.), the benzoic acid was removed in the usual manner with sulphur dioxide, concentrated hydrochloric acid (50 c.c.) added, and the mixture heated on the water-bath for 2 hours, basified with powdered sodium hydroxide (20 g.), treated at room temperature with 30% hydrogen peroxide (7 c.c., added gradually), kept for 1 hour, heated at 50—55° for 15 minutes, cooled, and acidified with excess of sulphur dioxide. The phenoxyacetic-2-acetic acid thus precipitated was most conveniently purified with the aid of amyl ether, from which it separated in slender needles, m. p. 154—155°, unchanged by repeated purification (compare La Forge, *J. Amer. Chem. Soc.*, 1933, **55**, 3040, and Kostanecki and co-workers, *Ber.*, 1909, **42**, 827, who respectively give m. p. 154° and 158—159°). Esterification of this acid with hot alcohol and sulphuric acid gave an almost theoretical yield of the *ethyl* ester, which formed slender prisms, m. p. 48—49°, from light petroleum (b. p. 80—100°) (Found : C, 63·2; H, 6·9. $C_{14}H_{18}O_5$ requires C, 63·2; H, 6·8%).

(B) To a well-stirred mixture of the bisulphite compound of ethyl 2-aldehydophenoxyacetate (from 10 g. of aldehyde) in a saturated aqueous solution of sodium hydrogen sulphite (200 c.c.), sodium cyanide (5 g.) in water (10 c.c.) was added at 0° in the course of 1 hour, and 3 hours later the cyanohydrin was isolated with ether, being obtained as a colourless solid (7·2 g.), m. p. 64—65°. This compound (3 g.) was treated in chloroform (3 c.c.) with thionyl chloride (3 c.c., added slowly) and 24 hours later the solvent was evaporated, leaving the chloro-nitrile as a brown oil, which could not be economically purified by distillation in a vacuum. Zinc dust (0·8 g.) was gradually added to a warm solution of the chloro-nitrile (2·5 g.) in alcohol (2 c.c.), acetic acid (1·2 c.c.), and water (1·5 c.c.) and the mixture was heated on the water-bath for ½ hour, filtered, concentrated, cooled, treated with dilute hydrochloric acid, and extracted with ether. Evaporation of the dried extracts left ethyl phenoxyacetate-2-acetonitrile as a pale yellow oil (1·8 g.), b. p. 201—203°/25 mm. On being refluxed with alcohol (50 c.c.) and sulphuric acid (25 c.c.) for 10 hours, this nitrile (10 g.) was converted into the ethyl phenoxyacetate-2-acetate (11·5 g.), m. p. and mixed m. p. 48—49° after crystallisation from light petroleum.

7-*Hydroxychromeno*-(3' : 4' : 4 : 3)-*coumarin*.—A solution of the foregoing ester (5 g.) in toluene (25 c.c.) was gradually added to powdered sodium (0·6 g.) in the same solvent (25 c.c.), and the mixture heated on the water-bath for 1½ hours, cooled, and treated with excess of 2% aqueous sodium hydroxide. The liquid was filtered to remove a small amount of solid, the toluene layer separated, and the alkaline liquor extracted with ether to remove remaining traces of toluene, and acidified with dilute hydrochloric acid. On isolation with ether, the resulting ethyl chroman-3-one-4-carboxylate was obtained as a pale yellow oil (1 g.), which gave a prussian-blue coloration with alcoholic ferric chloride. Acidification of an aqueous solution of the solid separating from the toluene–water mixture gave a further quantity (0·5 g.) of the ester.

From a solution of the ester (1 g.) and resorcinol (1 g.) in alcohol (10 c.c.) saturated with hydrogen chloride, the *coumarin* (0·2 g.) gradually separated in the course of 3 days; a further quantity (0·1 g.) was obtained by concentration of the alcoholic liquors. The substance crystallised from alcohol in pale yellow needles, m. p. 276—278° (Found : C, 72·3; H, 3·9. $C_{16}H_{10}O_4$ requires C, 72·2; H, 3·8%). The *acetate* formed slender, pale yellow prisms, m. p. 214—215°, from ethyl acetate (Found : C, 70·3; H, 4·1. $C_{18}H_{12}O_5$ requires C, 70·1; H, 3·9%).

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