SYNTHESIS OF NATURAL PRODUCTS BY THE OXIDATION OF PHENOLS

T H E S I S

presented to the University of Glasgow

for the degree of Doctor of Philosophy

by

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1961

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ACKNOWLEDGMENTS

I wish to thank Dr. A. I. Scott for his help and advice during the course of this work. His unfailing interest and guidance have been a constant source of encouragement to me.

I am indebted to the Department of Scientific and Industrial Research for a maintenance award.

Thanks are also due to Mr. J.M.L. Cameron and his staff and to Dr. R.I. Reed for microanalyses and mass spectrometric determinations.

CONTENTS

SUMMARY

Introduction - The oxidative coupling of phenols, as a biogenetic route to many natural products is discussed and an account of the recent evidence in support of the theory is given.

Part I - The synthesis of the depside dihydropicrolichenic acid from olivetol aldehyde is described. Several reagents were tried with a view to causing the oxidative coupling to picrolichenic acid. The vital coupling reaction was finally achieved by using manganese dioxide suspended in benzene.

A partial resolution of picrolichenic acid was obtained via the quinine methohydroxide salt.

Part II - The total syntheses of the mould metabolites, geodin and erdin, were attempted. Two routes to the intermediate benzophenones, dihydrogeodin and dihydroerdin, were unsuccessful. The first route required the condensation of a dichloro-p-orsellinic acid with a suitable derivative of methyl 3-hydroxy-5-methoxybenzoate by a Friedel and Craft's reaction. The second route reversed the rôles of the reactants by using the anhydrides of 5-benzyloxy- and 5-hydroxy-3-methoxyphthalic acid and attempting to condense them with 2,6-dichlororcorcinol.

Partial syntheses of geodin and erdin were achieved by oxidative coupling of the dihydro compounds, obtained from natural geodin and erdin, with alkaline potassium ferricyanide.

Part III - The synthesis of colchicine by oxidative coupling of a phenolic precursor, 1-(3',4'-dimethoxy-5'-hydroxyphenyl)-3-(β-tropolonyl)-propane, was attempted. The precursor was obtained by condensation of a suitably substituted phenylacetaldehyde with the anhydride of 2-carboxy-4-hydroxy-3-oxocycloheptatrienyl-acetic acid with subsequent pyrolysis, reduction and hydrolysis. All attempts to induce ring closure to the tricyclic system of desacetylamidocolchiceine by a variety of oxidants were unsuccessful.

INTRODUCTION

INTRODUCTION

Advances in the synthesis of natural products have been stimulated in recent years by the chemist's desire to accomplish the construction of the molecule by a route which parallels the proposed biosynthesis of the compound. The success of such a synthesis often provides indirect evidence of the correctness of a postulated biogenetic sequence, and furthermore is frequently adaptable to the preparation of the proper intermediates labelled with isotopic atoms which may be used in the appropriate feeding experiments. In his monograph "The Structural Relationships of Natural Products"[1] Sir Robert Robinson foreshadowed many of the recent developments in "biogenetic-type" synthesis, as well as contributing the classic example of tropinone synthesis[2]. Recent work by van Tamelen and his collaborators in the realm of alkaloid synthesis[3,4,5] and by Freudenberg[6] in the lignin series is representative of the signal success which may be achieved by the organic chemist in his simulation of natural processes. We now consider the advances which have been made in the search for laboratory analogy with one significant biosynthetic step - the formation of C-C or C-O bonds by the oxidation of phenolic compounds.

Several classes of phenolic compounds have been recognised which require the coupling of phenol radicals as

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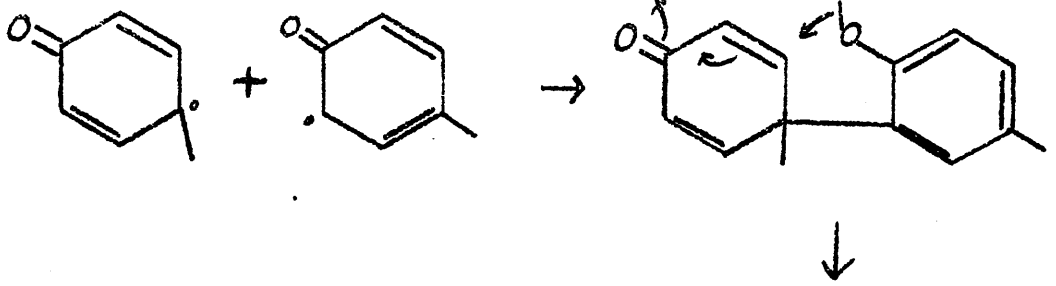

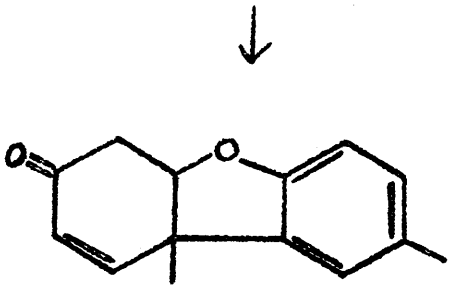

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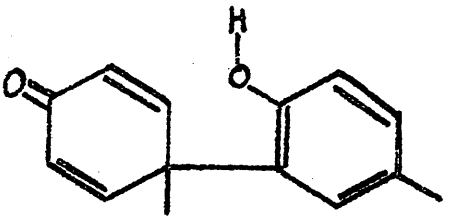

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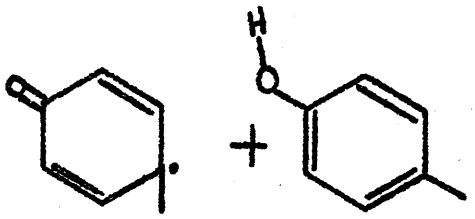
(ii)



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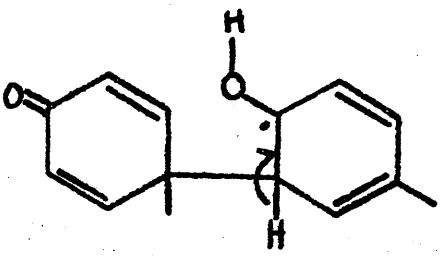
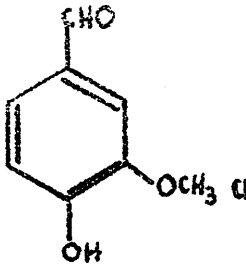


an essential biogenetic step in their formation.

Pummerer and his collaborators[7] were the first to draw attention to the role of radicals as intermediates in phenol oxidation *in vitro*. The concept of the intervention of such processes in biosynthesis was extended and discussed by Barton and Cohen[8] and by Erdtman and Wachtmeister[9]. The reviews of these authors[8,9] indicate the many classes of complex natural phenols which, from inspection of their structural formulae, are consistent with the mechanism of phenol oxidation. Among alkaloids, lichen acids, mould metabolites, lignans, tannins and many other natural products we find illustrations of this biogenetic theme. In the sequel we consider the recent work on the realisation of such oxidative processes in the laboratory.

The oxidation of phenols by one-electron transfer oxidising agents[10] affords phenol radicals which are stabilised by resonance over the *ortho* and *para* positions of the aromatic ring. The phenol radicals may be converted to stable products by several processes. The coupling to form dimers by carbon-carbon, carbon-oxygen or oxygen-oxygen bonding is one important process. (Throughout this thesis *radical coupling* (as in(i)) is chosen to represent this oxidation rather than the *substitution* mechanism (ii). Experimental distinction between these alternatives has yet to be supplied).

Carbon-carbon coupling can be *ortho-ortho*, *ortho-para*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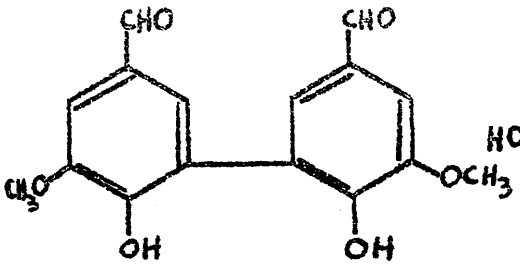
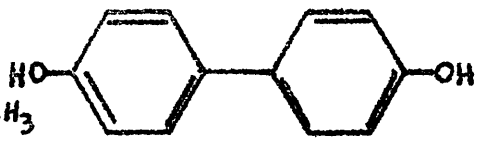
OCH3
OH

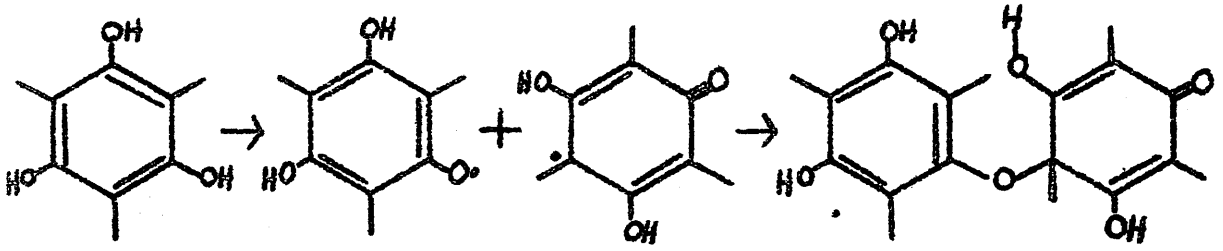
1


CHO
CHO
CH3O
OCH3
OH
OH

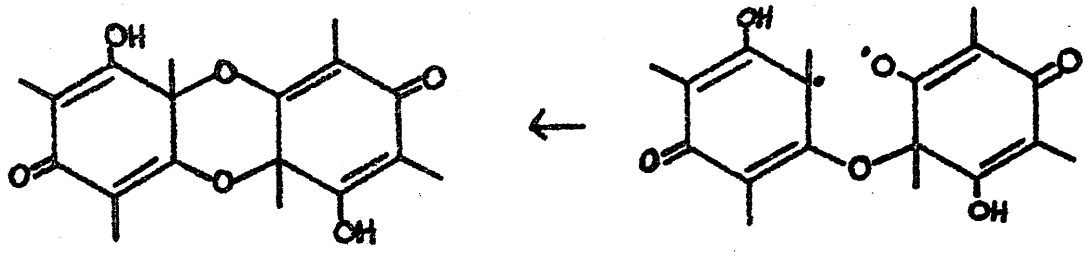
2


HO
OH

3


OH
HO
OH
HO
O
HO
O
H
O
OH

7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OH
OH
O
O
O
OH

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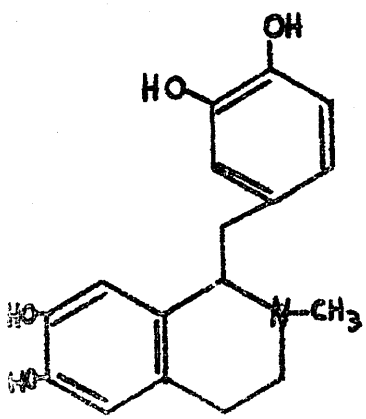
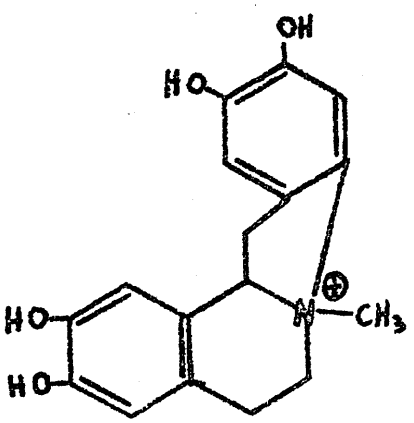
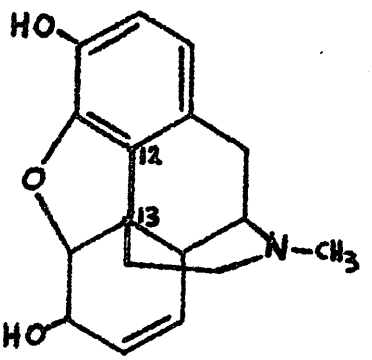

OH
HO
HO
HO
N
CH3

9


OH
HO
HO
HO
N
CH3

10


HO
O
12
13
N
CH3
HO

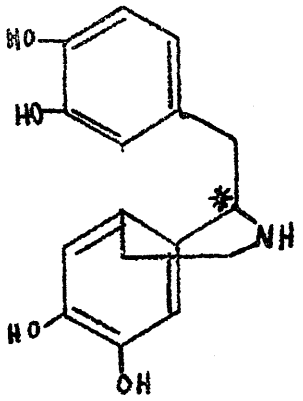
11

or para-para. An example of ortho-ortho coupling is the oxidation of vanillin (1) to give the dimer (2)[11]. The isolation of 4,4'-dihydroxydiphenyl (3)[12] from the oxidation of phenol is an instance of para-para coupling. The oxidation of p-cresol (4) gives a neutral product which from its structure (6) must arise from ortho-para coupling via the intermediate (5)[13] [see (i)].

An example of carbon-oxygen coupling is illustrated by the formation of cedrone (8)[14] from trimethylphloroglucinol (7). The mechanism of formation of cedrone requires either two successive diradical couplings or one tetraradical coupling. The first scheme (illustrated) is acceptable provided that enolised 1:3-diketones can be oxidised like phenols.

Alkaloids of the morphine type have long been thought to arise from the intramolecular oxidative coupling of the benzylisoquinoline alkaloids[1,15,16]. However when laudanosoline (9) was oxidised under mild conditions[15,17] the product of the reaction was not of the morphine type but was dehydrolaudanosoline (10). Although this variant was unknown at that time a methyl ether of dehydrolaudanosoline was in fact isolated twenty years later[18].

The biosynthesis of morphine (11) from the benzyl-isoquinoline type has been verified by the use of isotopically labelled synthetic norlaudanosoline (12)[19]. Norlaudanosoline



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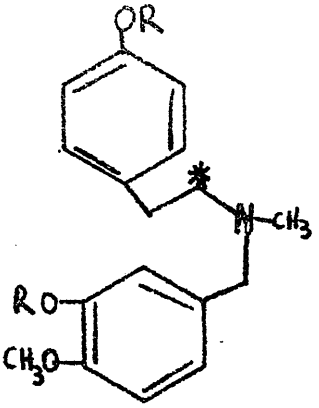

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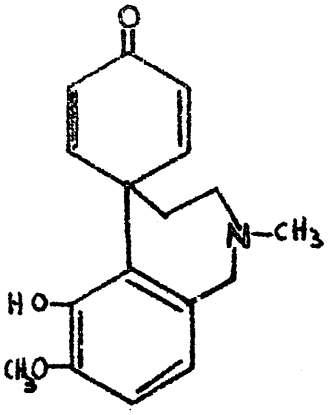

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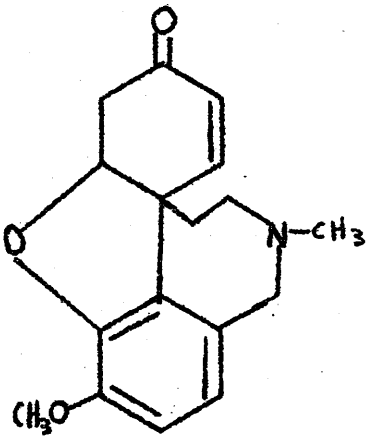

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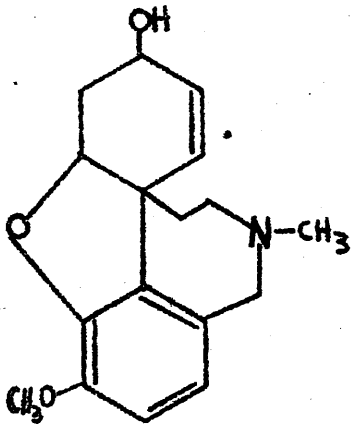

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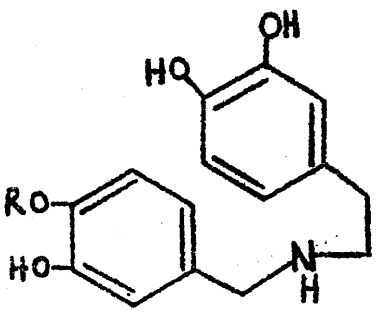

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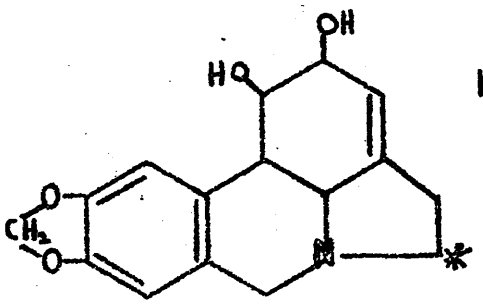

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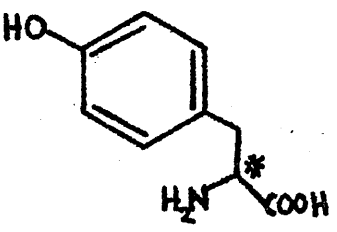

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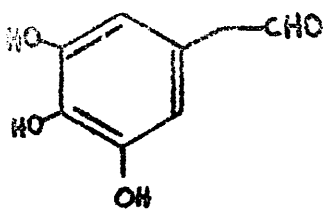
was prepared with a ^{14}C label at the position shown with an asterisk (12). The morphine isolated from Papaver somniferum plants which had been fed with labelled nor-laudanosoline was found to be strongly radioactive. This leaves little doubt that the bond between positions 12 and 13 of morphine is formed by an oxidative coupling reaction.

The Amaryllidaceae alkaloids are another group which probably result from the oxidative coupling of phenolic precursors.

Galanthamine (16) and lycorine (18) are two examples which could be derived as shown from (13, R=H) and (17) respectively. The postulated intermediate, narwedine (15), for galanthamine has in fact been isolated[20]. Belladine (13, R=CH_3) has also been found to occur naturally[21].

Barton and Kirby[22] have completed a synthesis of galanthamine which followed exactly the proposed biogenesis (13, R=H→16). The above authors also reported that the phenol (13, R=H) labelled with ^{14}C at the N-methyl group was incorporated by King Alfred daffodils and that the galanthamine isolated was radioactive.

(2-^{14}C) Tyrosine (19) has been incorporated by the appropriate plants to give radioactive galanthamine[22] and lycorine[22,23]. The lycorine obtained by Battersby, Binks and Wildman had all the activity located at the position marked with an asterisk (18).



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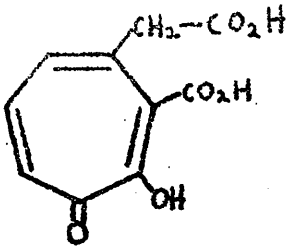

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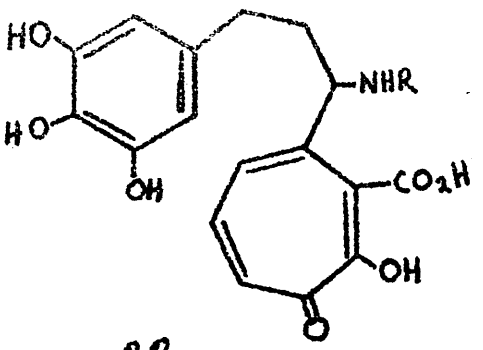

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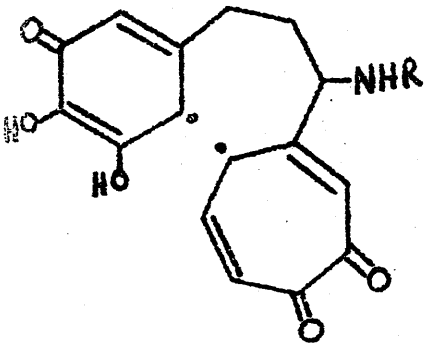

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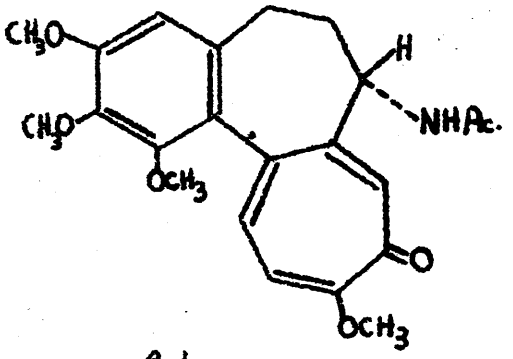

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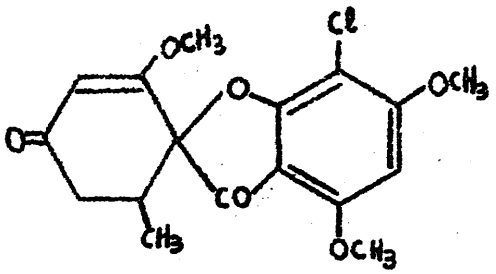

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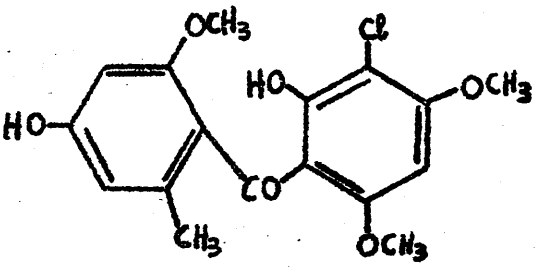

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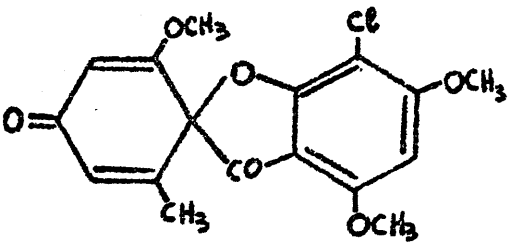

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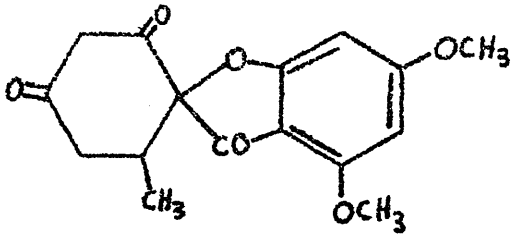
More recently it has been shown that norbelladine (13; R,CH_3=H) labelled as shown is incorporated into lycorine[23a], galanthamine and other Amaryllidaceae alkaloids[22a]. The lycorine obtained was shown to be labelled exclusively as in (18)[23a].

These results further indicate the intervention of aromatic precursors derivable from shikimate followed by oxidative coupling in the biogenesis of alkaloids.

Several schemes for the biogenesis of the alkaloid colchicine (24) have been proposed[24,25,26,27]. Scott's biogenesis[27] envisages an intermediate such as (22) arising from the precursors (20) and (21) or their biogenetic equivalents. If the aromatic nature of the tropolone system allows it to form radicals in the same manner as a phenol then under oxidative conditions the biradical (23) could form and hence give the tricyclic system of colchicine by pairing of the radicals.

The biogenesis of the physiologically active mould metabolite griseofulvin[28] (25) is considered to follow the poly-β-ketone pattern to the benzophenone (26). The spirane formation would then take place by oxidative coupling to give (27) and partial reduction would give griseofulvin. The methylation and chlorination steps take place at undetermined stages.

Labelling experiments by Birch <u>et al</u>[29] support the head-to-tail incorporation of acetic acid units in the



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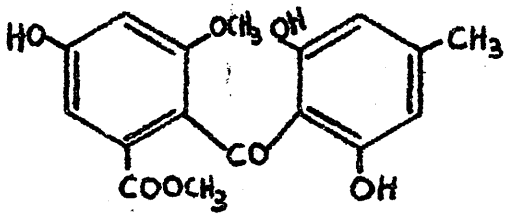

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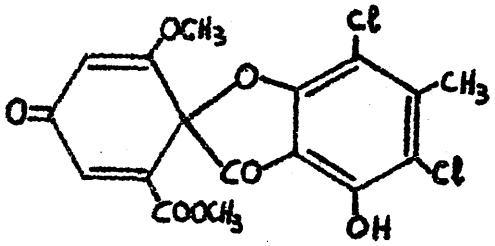

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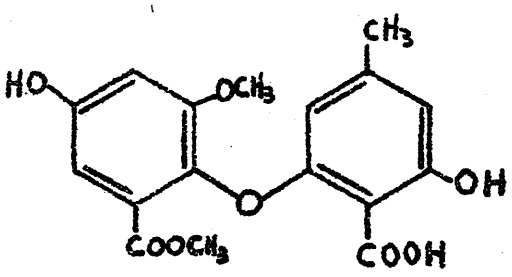

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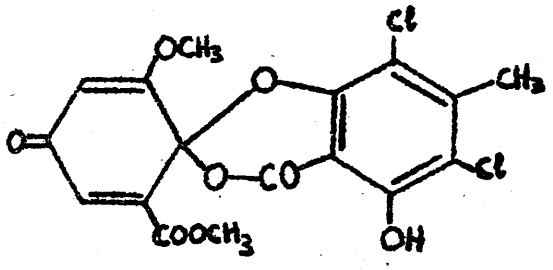

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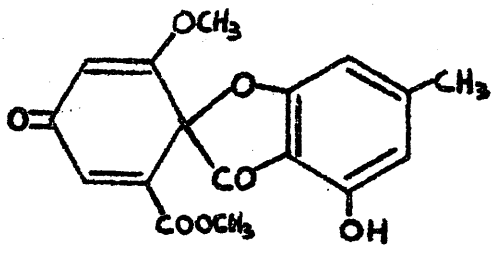

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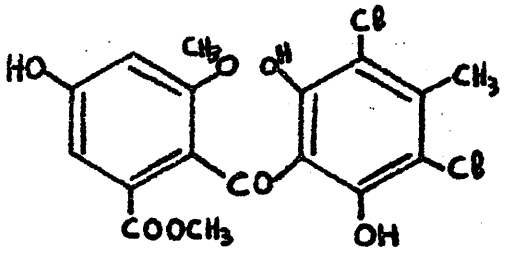

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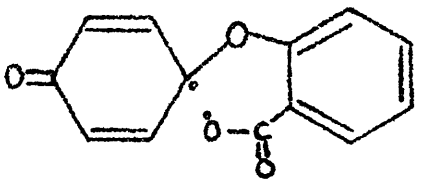
expected manner.

Synthetic work by Scott *et al*[30,31] resulted in the formation of the benzophenone (26) which was successfully oxidised to the spiro-dienone (27). The spiro-dienone was hydrogenated selectively to give (±) griseofulvin. Two other syntheses of griseofulvin have also been reported[32,33].

Further experiments indicated that the biogenesis was correct. (^{14}C)-Labelled natural griseofulvin was converted to the dehydro compound (27). The latter on being fed to *Penicillium patulum* was incorporated into the griseofulvin produced to the extent of 30%[34]. A careful examination of the *P. patulum* mother liquors resulted in the isolation of the compounds (26), (27) and (28) among others[35]. These closely related compounds occurring in the same medium provide overwhelming evidence for the biogenesis of griseofulvin.

The metabolites sulochrin[36], geodin[37], asterric acid[38] and geodoxin[39] have been isolated from various micro-organisms and their structures have been shown to be (29)[36], (30)[40-43], (31)[38] and (32)[39] respectively. Sulochrin and geodin are reminiscent of the intermediates in the scheme for the biogenesis of griseofulvin and, neglecting the presence of chlorine, they are obviously related by a phenol coupling step.

Experiments, *in vitro*, have resulted in the oxidation of sulochrin (29) to dechlorogeodin[38] (33) and of dihydrogeodin



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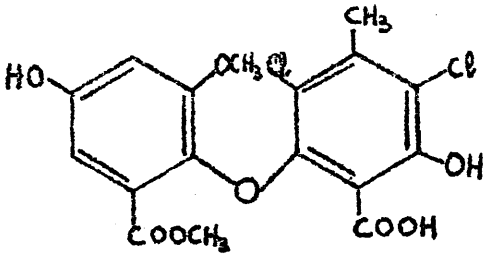

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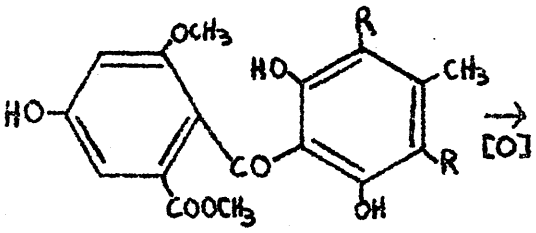
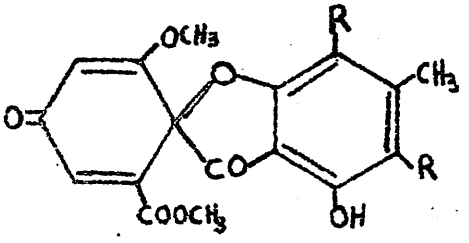
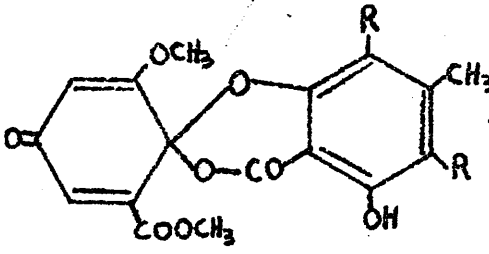


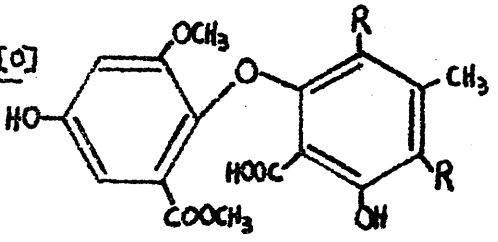
R=H, SULOCHRIN



R=Cl, GEODIN

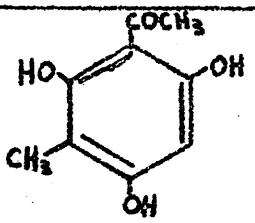


R=Cl, GEODOXIN



R=H, ASTERRIC ACID

CHART I



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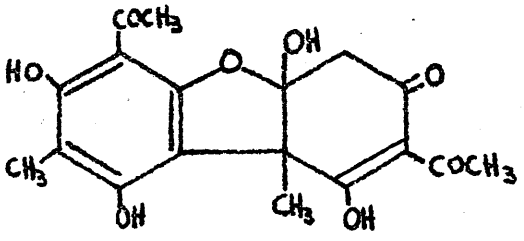

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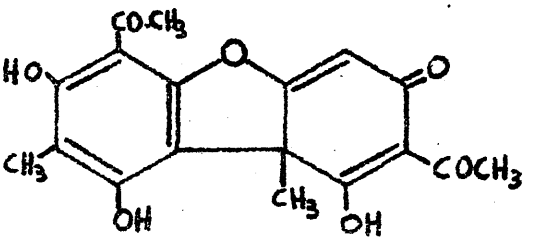


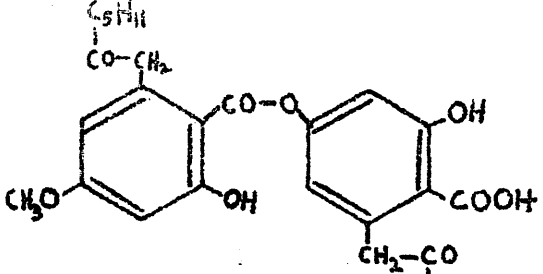
39

(34) to geodin[44] (30). The similar conversion of O-methyldihydrogeodin to O-methyl geodin has also been achieved[31].

Asterric acid (31) and geodoxin (32) can be related by an oxidative coupling through a carboxy radical of the type (35). Asterric acid was obtained in excellent yield from sulochrin by the acid catalysed hydrolysis of dechlorogeodin[38] (33) and was subsequently oxidised to the corresponding dechloro analogue of geodoxin[45]. Geodoxin has been prepared by an oxidative coupling of geodin hydrate[45] (36) which is the appropriate chlorinated analogue of asterric acid. A reasonable sequence for the biogenesis of these compounds would thus be the formation of sulochrin from acetate units, oxidation to geodin, hydrolysis to asterric acid and further radical coupling to give geodoxin as illustrated in Chart I., the introduction of chlorine taking place at an undetermined stage.

The lichen substances produce several interesting types of phenols which can arise by an oxidative coupling mechanism.

The accepted structure of the lichen product usnic acid (39) indicated that it should be a coupling product of C-methylphloracetophenone (37) arising in a manner reminiscent of the formation of Pummerer's ketone (6). Barton et al[13] succeeded in isolating (±) usnic acid by oxidation of C-methylphloracetophenone under carefully controlled conditions to (38) followed by dehydration.



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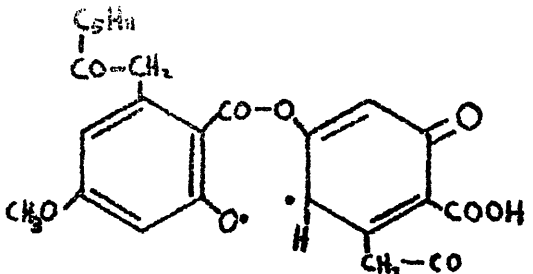

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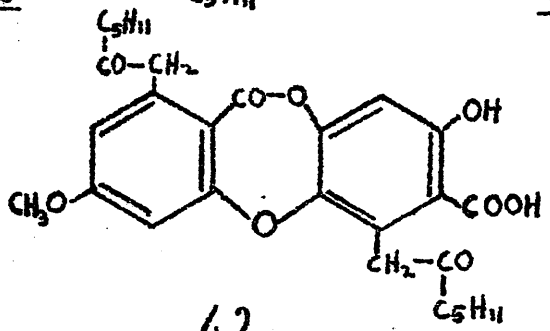

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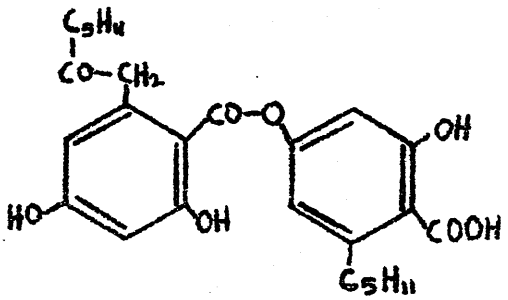

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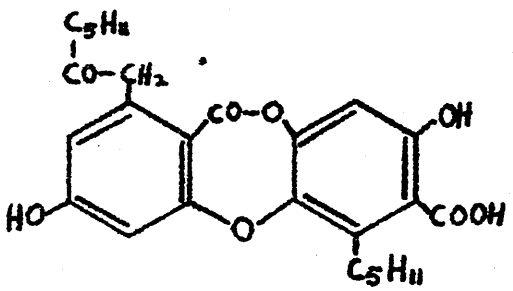

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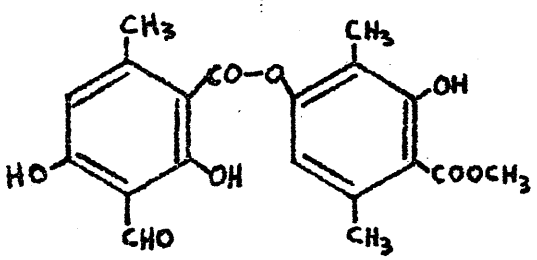

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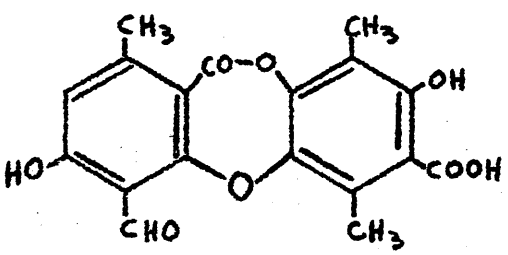

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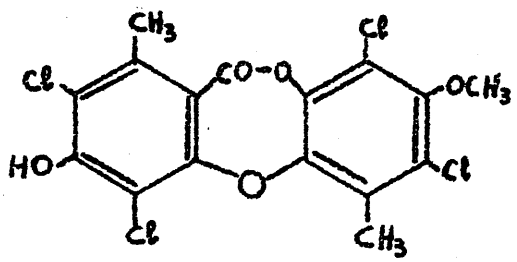

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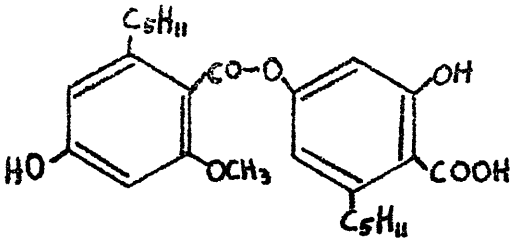
Two of the main classes of lichen substances are the depsides and the depsidones[46]. Microphyllic acid (40) is an example of a depside and α-collatolic acid (42) is the corresponding depsidone. The depsidone is almost certainly derived from the depside *via* the diradical (41) by an oxidative coupling reaction. Other examples of the occurrence of closely related depsides and depsidones are given by olivetoric acid (43) - physodic acid (44) and atranorin (45) - virensic acid[47] (46).

Laboratory analogy for the proposed biogenesis is supplied by the successful synthesis[48] of the depsidone diploicin (47) from a totally synthetic depside precursor by phenol oxidation.

If the hydroxyl group *ortho* to the carbonyl group of the depside ester link is blocked in some manner, for example by methoxyl, then the normal C-O coupling cannot occur. However the alternative C-C coupling to give a spiro-dienone product resembling the griseofulvin type can take place. This mode of coupling is illustrated by the successful synthesis[49] of picrolichenic acid[50] (50) by oxidative coupling of the synthetic depside (48) *via* the intermediate diradical (49).

A major group of plant substances contain the C_6-C_3 unit and it was suggested[8] that all compounds whose structures are based on polymerised C_6-C_3 units are formed by phenol coupling.



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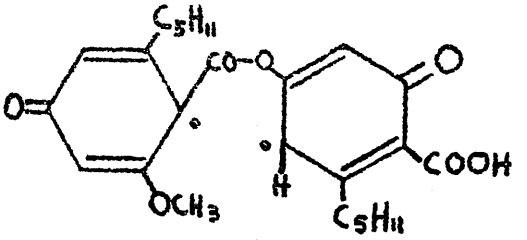

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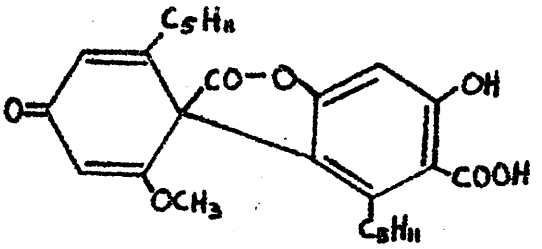

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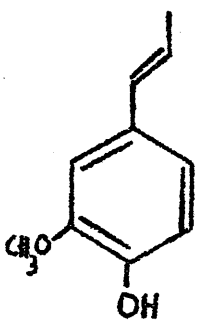

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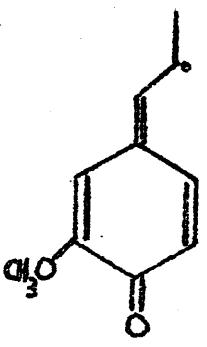

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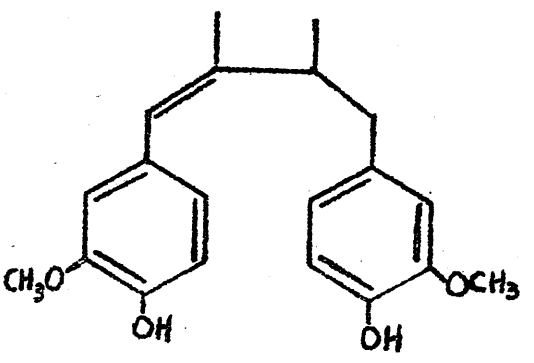

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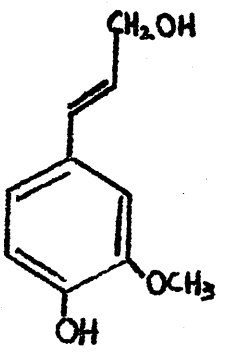

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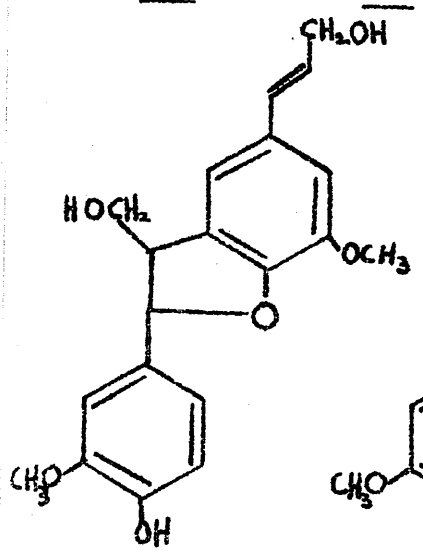

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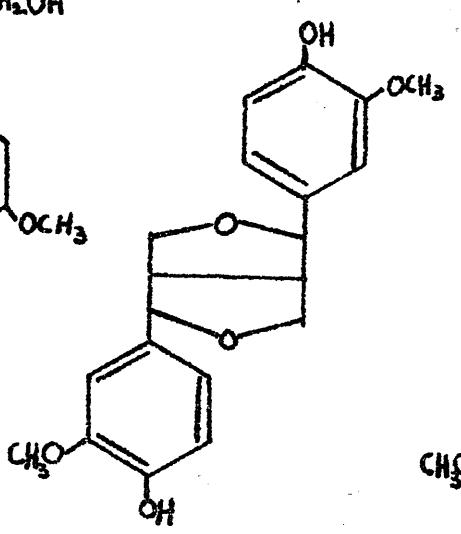

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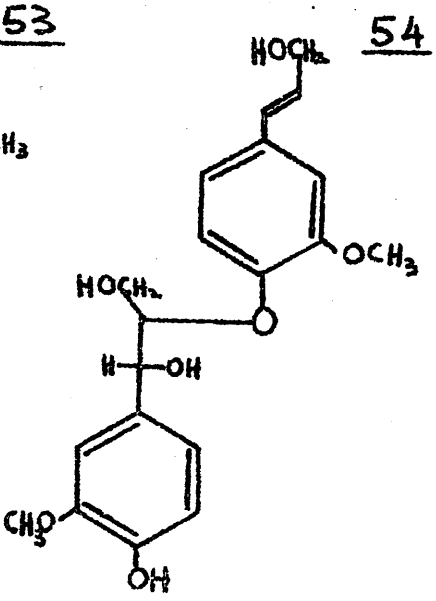

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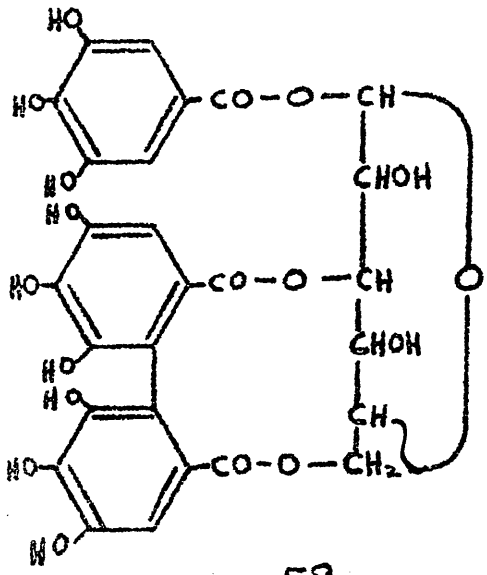
The lignins, which consist of two C_6-C_3 units coupled in the β positions all have phenolic hydroxyls in the para positions. Guaiaretic acid (53) can be formed by β-β coupling of two radicals (52) from isoeugenol (51) followed by reduction.

Freudenberg[6] considers that coniferyl alcohol (54) is the primary building unit in lignin biosynthesis. He has shown that oxidation of coniferyl alcohol with mushroom laccase or horse-raddish peroxidase produces lignin identical with natural lignin. By interrupting the process of lignification several intermediates have been isolated which from their structures must arise from oxidative coupling of coniferyl alcohol in various ways. The major intermediates identified were (55), (56) and (57).

The tannins[51] are a group of naturally occurring compounds whose structures are based on glucose esterified with gallic acid or acids derived from gallic acid by phenolic coupling. Corilagen (58)[51] is an example of a tannin.

The biflavonyls represent yet another class of phenolic compounds which are obviously derived by an oxidative coupling step, in this case a flavone is the obvious precursor. An example of these C_{30} compounds is provided by the yellow pigment ginkgetin (59)[52].

Gossypol (60), the pigment of cotton seed, is obviously a coupled product of the formyl naphthalene (61). The synthesis[53]



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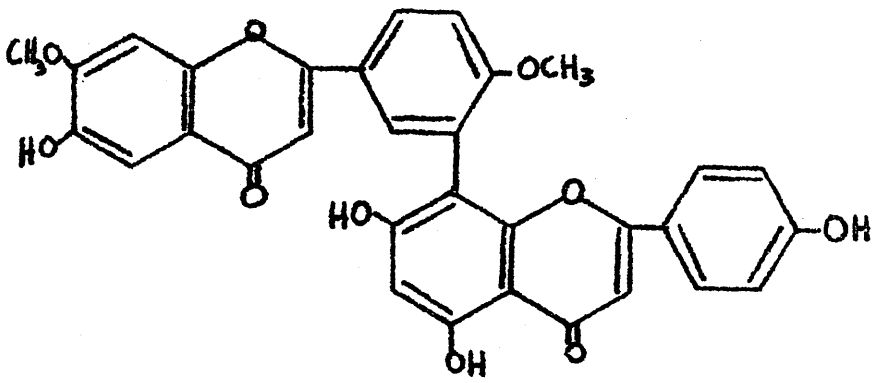

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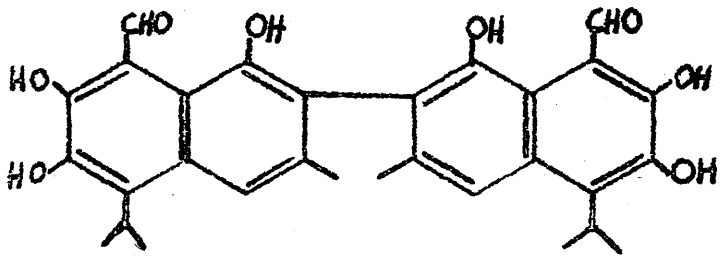

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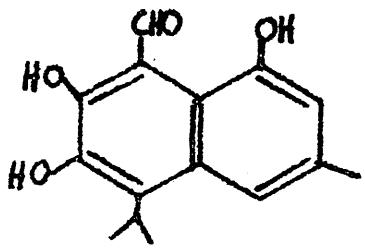

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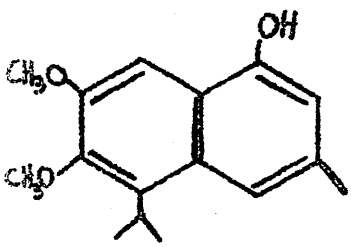

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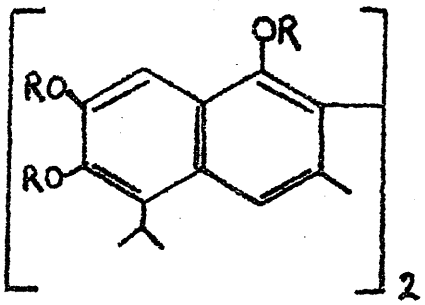

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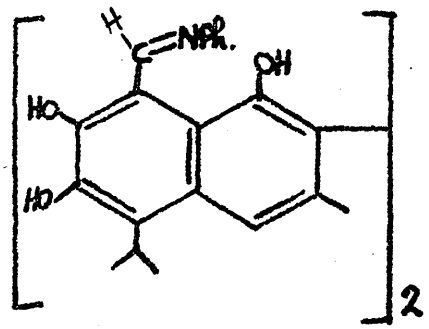

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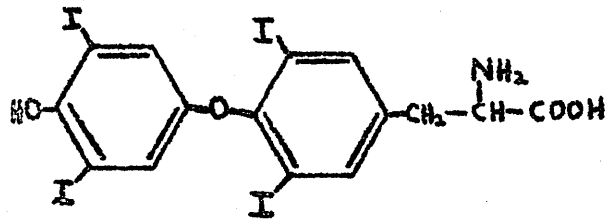

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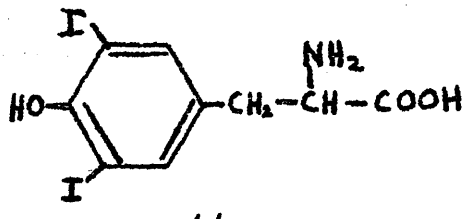
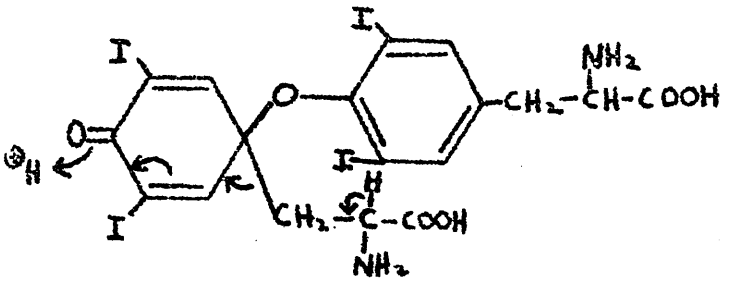


66

of gossypol has been achieved by dimerisation of the napthalene (62) followed by methylation to give apogossypol hexamethyl ether (63, R=CH_3) which was hydrolysed to apogossypol (63, R=H). Apogossypol was converted to the derivative (64) from which gossypol (60) was obtained by hydrolysis.

Thyroxine (65) can be derived from diiodotyrosine (66)[54] *via* the quinol ether (67) by C-O coupling and elimination of the amino acid side chain. This reaction has been accomplished *in vitro*[55] but in very small yield. The yield of (65) is greatly increased if the amino groups of diiodtyrosine are protected by acetylation[55]. Experiments[56] on model compounds such as (68) and (69) have produced the required analogue (70) of the intermediate quinol ether. The quinol ether can be decomposed to the thyroxine analogue (71) with the liberation of isobutene.



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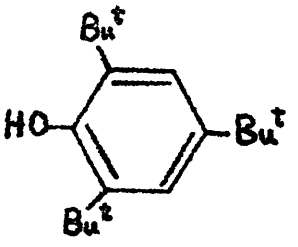

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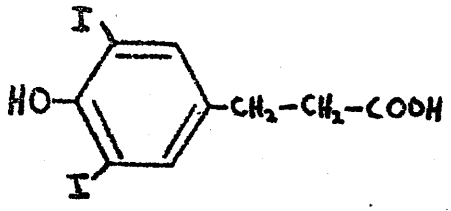

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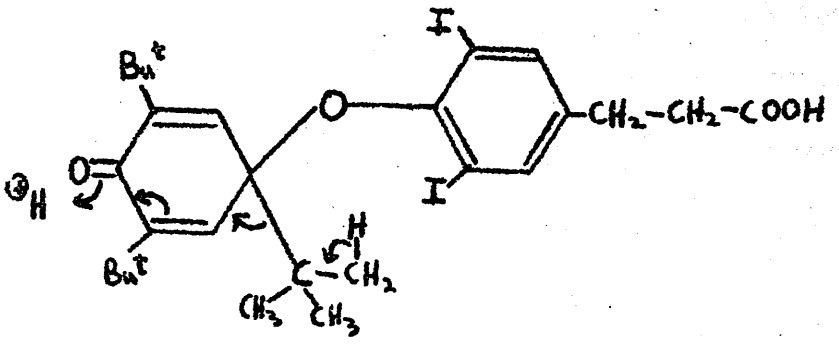

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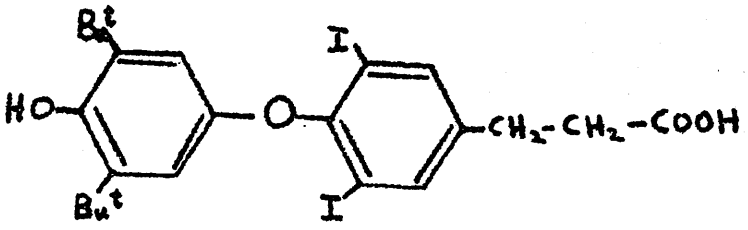

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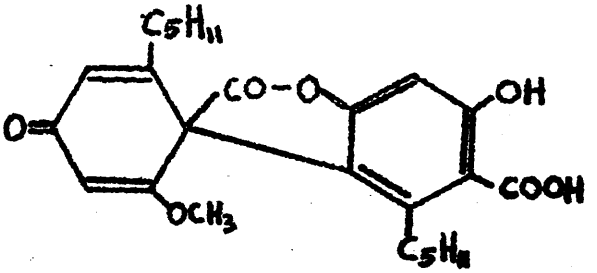
PART I

SYNTHESIS OF PICROLICHENIC ACID



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HISTORICAL

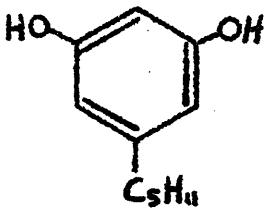
Picrolichenic acid, the bitter principle of the crustose lichen Pertusaria amara (Ach.) Nyl., was first isolated by Zopf[57] in 1900. It has a bitter taste similar to that of quinine and at one time was thought to be effective in the treatment of malaria[58].

Zopf obtained the acid in a reasonable state of purity and suggested the empirical composition $C_{17}H_{20}O_5$. The elegant investigations by Erdtman and Wachmeister[50] indicated that the empirical composition was in fact $C_{25}H_{30}O_7$ and that picrolichenic acid had the structure (50).

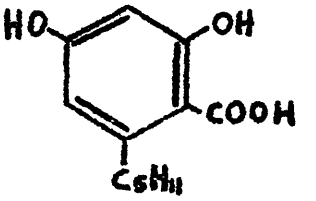
The acid, which is optically inactive, dissolved slowly with the evolution of a gas in aqueous sodium bicarbonate solution from which it can be recovered unchanged. It gave an intense violet colour with ethanolic ferric chloride solution but no colouration with Gibb's reagent[59] (2,6-dichloroquinone-monochloroimide), bis-diazotised benzene[60] or bleaching powder[46]. However after short treatment with cold aqueous alkali it gave red colourations with the benzidine reagent and with bleaching powder. This resembles the behaviour of certain easily hydrolysed depsides and depsidones[46].

On heating to the melting point the acid decomposed with the evolution of a gas containing carbon dioxide.

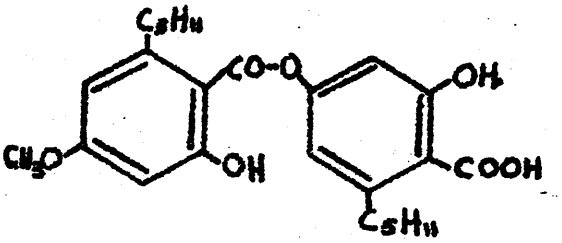
Short treatment with diazomethane furnished a monomethyl


HO
OH
C_5H_{11}

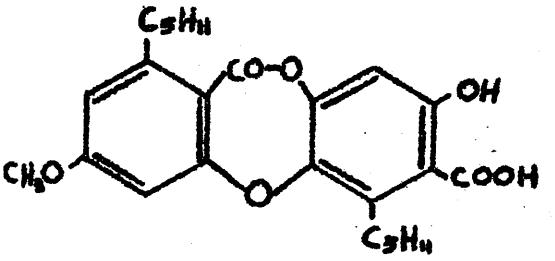
72


HO
OH
COOH
C_5H_{11}

73


C_5H_{11}
CO-O
OH
CH_3O
OH
COOH
C_5H_{11}

74


C_5H_{11}
CO-O
OH
CH_3O
O
COOH
C_5H_{11}

75

ester which was soluble in dilute alkali. Prolonged methylation with diazomethane yielded a neutral compound $C_{27}H_{34}O_7$.

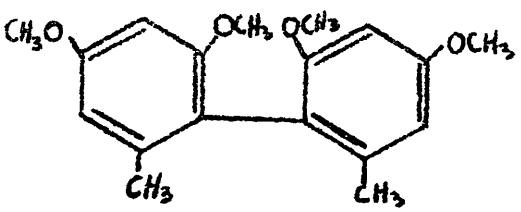
Picrolichenic acid reacted in the cold with piperidine yielding a piperidide, $C_{30}H_{41}O_7N$, which gave a violet ferric test and which, on treatment with diazomethane, furnished a dimethyl derivative containing a phenolic hydroxyl group.

On oxidation with permanganate picrolichenic acid afforded about 1.5 moles of volatile acids. The main component was n-caproic acid.

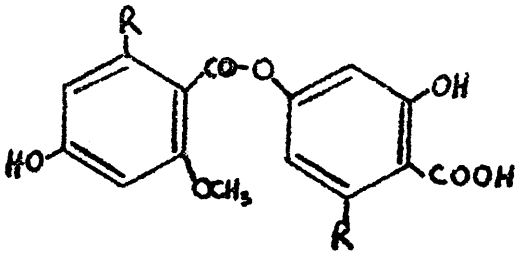
These results, along with the analyses, indicated the presence in picrolichenic acid of a free carboxyl group, a lactone group, a phenolic hydroxyl group ortho to a carboxyl function, a methoxyl group and two n-pentyl chains. Assuming the presence of two six-membered carbocyclic rings, all the carbon atoms and six of the seven oxygen atoms are accounted for.

The presence of two n-pentyl chains in picrolichenic acid points to a relationship with olivetol (72), a fairly widespread unit in lichen acids. The empirical composition, $C_{25}H_{30}O_7$, agrees with that of a dehydrogenation product of a monomethylated depside $C_{25}H_{32}O_7$ derived from two molecules of olivetol carboxylic acid (73). Perlatolinic acid[61] (74) is of this type and picrolichenic acid might be a related depsidone such as (75).

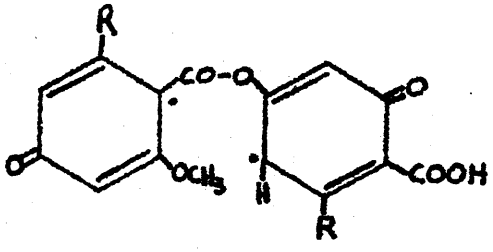
Picrolichenic acid after treatment with cold N-sodium hydroxide solution and then acidification gave a gum which rapidly evolved carbon dioxide. The product (A) gave a violet ferric test



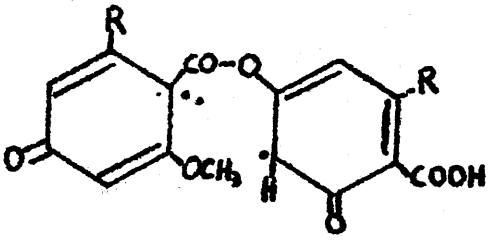
76



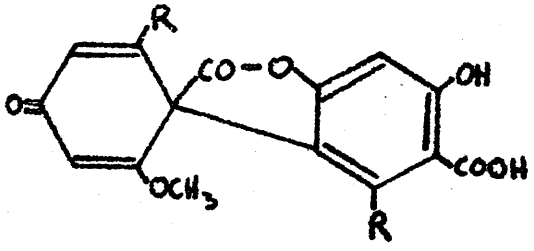
77



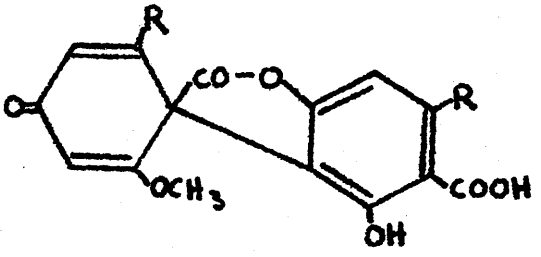
78a



78b



79



80

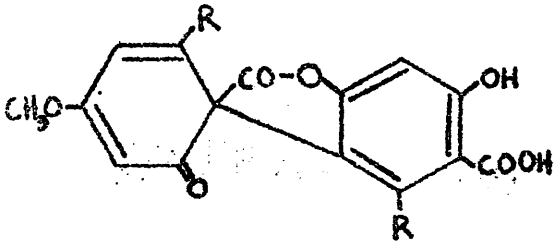
and was found to be a monocarboxylic acid containing a methoxyl group, $C_{22}H_{28}O_3(OCH_3)COOH$. The acid (A) melted with decomposition, losing one mole of carbon dioxide and forming an oily phenol (B). On simultaneous decarboxylation and demethylation with hydrobromic acid, the acid (A) furnished a phenol identical with a phenol (C), $C_{22}H_{30}O_4$, obtained directly from picrolichenic acid under similar conditions. Like phenol (B) it gave no colour with ferric chloride.

The phenols (B) and (C) afforded the same tetramethyl ether, $C_{12}H_4(C_5H_{11})_2(OCH_3)_4$. The composition of the tetramethyl ether and its bromination products clearly showed that phenol (C) and its monomethyl ether (B) were diphenyl derivatives. The ultra-violet spectrum of the tetramethyl ether was very similar to that of the tetramethyl ether of a symmetrical diorcinol (76).

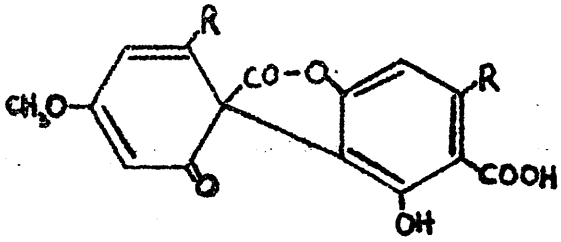
The acid (A) was obviously a carboxy derivative of phenol (B) with the carboxyl in the ortho position to a free hydroxyl group.

These results indicated that picrolichenic acid contained two six-membered rings joined by a carbon-carbon bond as well as by an ester linkage. This rules out the normal depsidone structure, illustrated by (75).

As indicated in the introduction (pp. 9,10.) the oxidative coupling of depsides such as (77), in which the phenolic hydroxyl group ortho to the ester linkage is protected in some manner, would furnish the diradical (78a) or (78b) which could be stabilised by carbon-carbon coupling with formation of the δ-lactone (79) or



81



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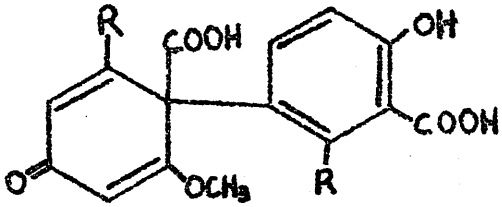
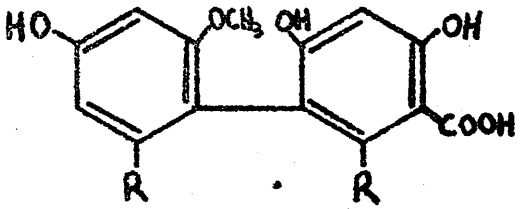


83



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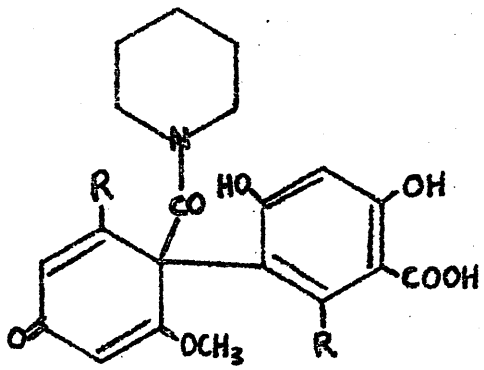

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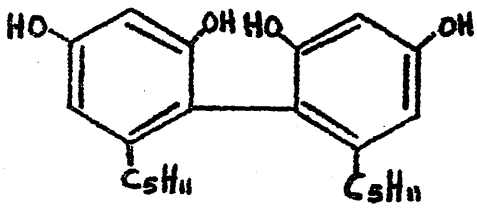

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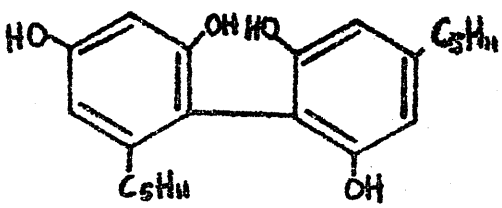
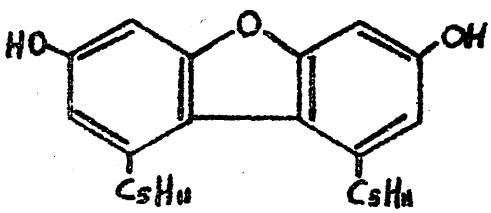


87



88

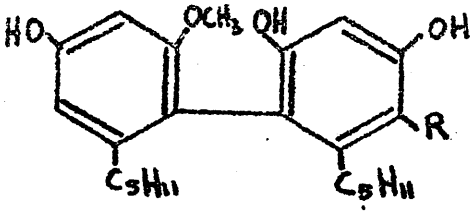
(80). There is, of course, the possibility that a depside which is not blocked may also yield products by carbon-carbon coupling (81, 82) rather than by the normal carbon-oxygen coupling.

The properties expected for γ-lactones of these types are compatible with the known chemical behaviour of picrolichenic acid. Alkaline hydrolysis of a lactone (79) would yield a dicarboxylic acid (83) which, being a vinylogue of a β-keto acid, should be easily decarboxylated and aromatised to a monocarboxylic acid (84) with a diphenyl structure. On the other hand the dienone structure would be expected to be retained if the lactone ring opens on aminolysis to form an amide. This is exactly what was observed when picrolichenic acid was treated with piperidine. On this basis (79), (80), (81) and (82) ($R=nC_5H_{11}$) would appear to be plausible structures for picrolichenic acid and a structure like (85) for the piperidide.

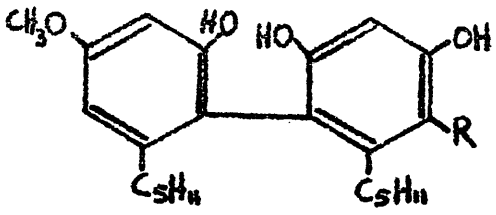
Two alternative structures (86, 87) are possible for the phenol (C).

When treated with zinc chloride for a few minutes at 240° phenol (C) gave a phenol (D), $C_{12}H_4O(C_5H_{11})_2(OH)_2$, which on methylation gave a dimethyl ether. The colour reaction with Gibb's reagent[59] and the ultraviolet spectrum of the dimethyl ether indicated that phenol (D) was the 3,7-dihydroxydibenzofuran (88).

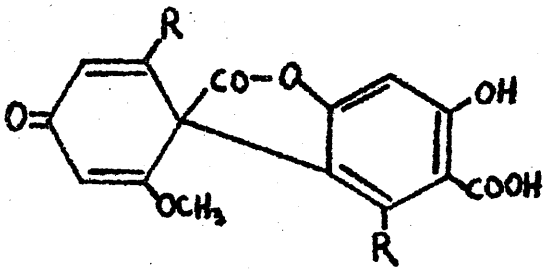
The structure (88) of phenol (D) was definitely settled by oxidation of its dimethyl ether with permanganate. The crude


HO
OCH3
OH
OH
R
C5H11
C5H11

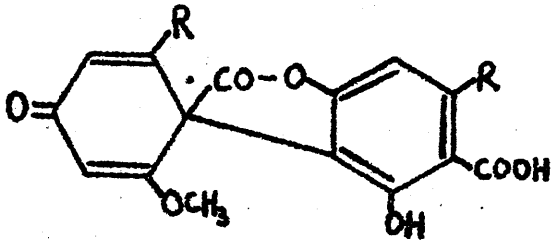
89


CH3O
HO
HO
OH
R
C5H11
C5H11

90


R
CO-O
O
OH
COOH
OCH3
R

79


R
CO-O
O
R
COOH
OCH3
OH

80

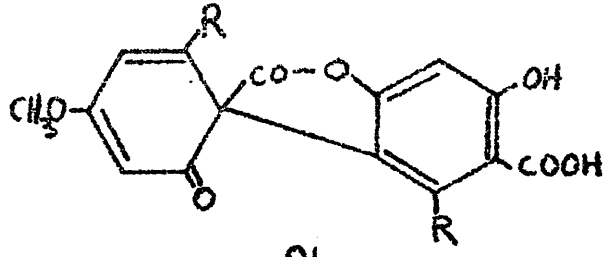
oxidation product was, paper chromatographically, identical with 3,7-dimethoxydibenzofuran-1,9-dicarboxylic acid[62] and on methylation if afforded a dimethyl ester identical with 3,7-dimethoxy-1,9-dicarbomethoxydibenzofuran[62]. Accordingly phenol (C) must have the structure (86).

Two structures (89, R=H) and (90, R=H) are possible for the phenol (B), the monomethyl ether of (C), and two alternative structures (89, R=COOH) and (90, R=COOH) remain for the monocarboxylic acid (A) in view of its violet ferric chloride and easy decarboxylation. These alternatives are also biogenetically plausible, being related to orsellinic acid.

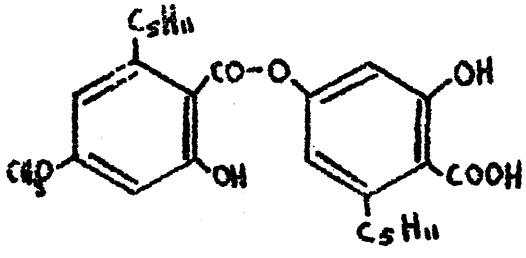
Picrolichenic acid must therefore be either (79, R=C_5H_{11}) or (81, R=C_5H_{11}) and the alternatives (80, R=C_5H_{11}) and (82, R=C_5H_{11}) must be rejected.

Additional evidence in favour of a dienone-γ-lactone structure was provided by the infrared spectra of picrolichenic acid and its derivatives.

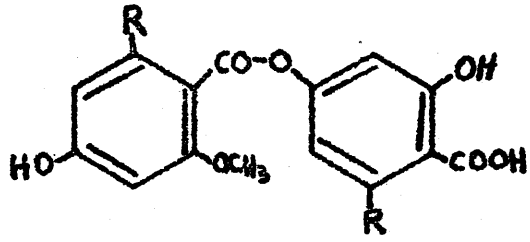
Picrolichenic acid, methyl picrolichenate and methyl O-methylpicrolichenate all showed strong absorption at 1820-1825 $cm.^{-1}$ but the piperidide showed no absorption in this region. This absorption is in accord with a β,γ-unsaturated γ-lactone in which the double bond forms part of an aromatic ring[63]. The two remaining carbonyl functions of picrolichenic acid gave rise to a single band at 1665-1670 $cm.^{-1}$ Methyl O-methylpicrolichenate,


R
CO-O
OH
CH3O
COOH
O
R

81


C5H11
CO-O
OH
CH3O
OH
COOH
C5H11

74


R
CO-O
OH
HO
OCH3
COOH
R

77

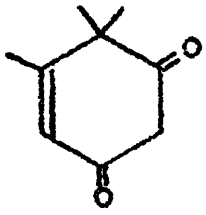
however, showed two strong bands at 1725-1730 cm.$^{-1}$ and 1660-1670 cm.$^{-1}$ which may be ascribed to the carboxymethyl group and the dienone carbonyl group, respectively. As is illustrated by examples from steroid chemistry[64] cross conjugated as well as linear conjugated dienones absorb strongly in a narrow region at 1660-1670 cm.$^{-1}$

The infrared spectrum of the piperidide of picrolichenic acid showed a broad absorption in the 1650-1725 cm.$^{-1}$ region with a maximum at 1690-1700 cm.$^{-1}$, indicating overlapping of the carbonyl bands.

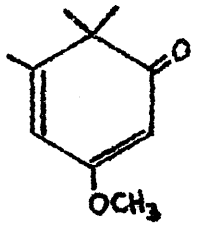
Picrolichenic acid and its piperidide showed broad absorption in the 2500-2800 cm.$^{-1}$ region. These compounds as well as methyl picrolichenate also showed a broad hydroxyl band at about 3450 cm.$^{-1}$ Methyl O-methylpicrolichenate had no hydroxyl absorption.

Two alternatives (79, R=C_5H_{11}) and (81, R=C_5H_{11}) remain for picrolichenic acid. The structure (79, R=C_5H_{11}) seems more probable from biogenetic considerations. The widespread occurrence of depsidones in lichens indicates that dehydrogenation of depsides such as (74) usually proceeds with carbon-oxygen coupling. However in the case of depsides methylated as in (77) only carbon-carbon coupling is possible.

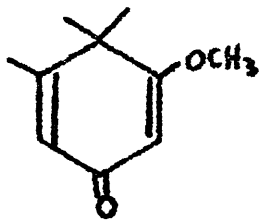
Since no direct conjugation exists between the two moieties of picrolichenic acid information about the structure of the cyclohexadienone part can be obtained by substracting the ultra-



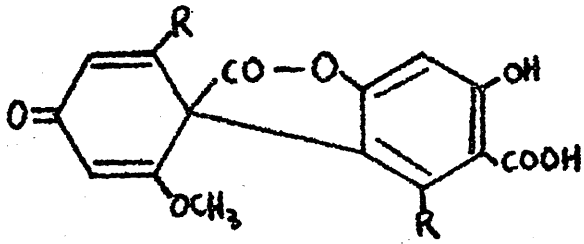
91



92



93



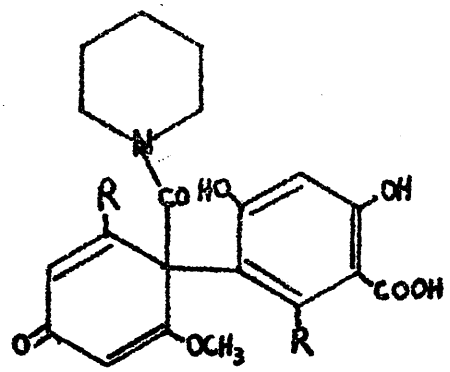
79

violet spectrum of the aromatic part from that of the acid itself. Difference curves were obtained by subtracting the ultraviolet curve of orsellinic acid from the curves for picrolichenic acid and the piperidide. The two curves which were obtained gave an approximate spectrum of the methoxy cyclohexadienone chromophore of picrolichenic acid.

Linear conjugated cyclohexadienones absorb at longer wavelengths in the ultra-violet region than the cross conjugated types. Jeger et al[65] studied a pyrolysis product of α-amyrin containing the partial structure (91) which afforded two isomeric monomethyl ethers one of which showed a very broad maximum at 330mμ.(log ε =3.18) and was alloted the linear conjugated formula (92). The isomer which showed a maximum at 250 and 278mμ.(log ε =4.2 and 3.7 resp.) was given the cross conjugated partial structure (93).

Both the difference curves obtained above showed maxima near 235mμ.(log ε =4.4, 4.1) and 280mμ.(log ε =3.8, 3.15) corresponding roughly to the two maxima of the cross conjugated compound (93) which again supports the structure (79, R=C_5H_{11}) for picrolichenic acid. The difference curve obtained from the piperidide also showed a distinct maximum at 325mμ, but this maximum is less intense (log ε =3.25) and much narrower than the broad maximum of the compound containing structure (92).

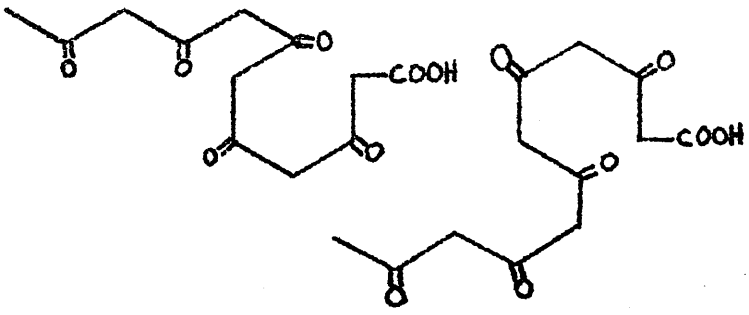
The chemical and physical evidence so far appears to be in



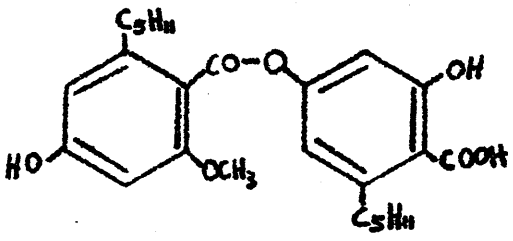
85

harmony with a structure (79, R=C_5H_{11}≡50) for picrolichenic acid.

The structure of the piperidide would then be (85, R=C_5H_{11}).



94



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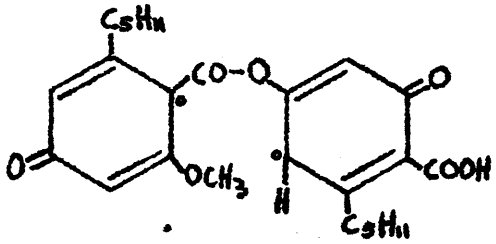

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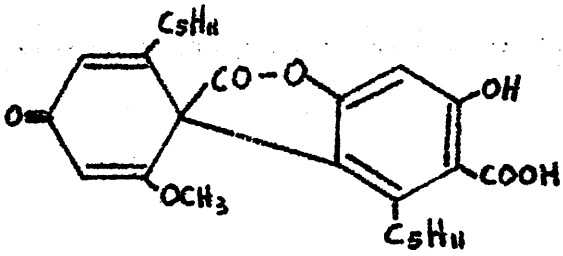

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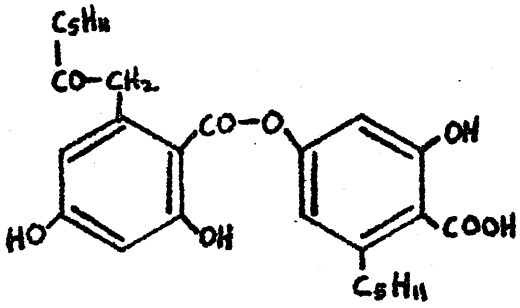

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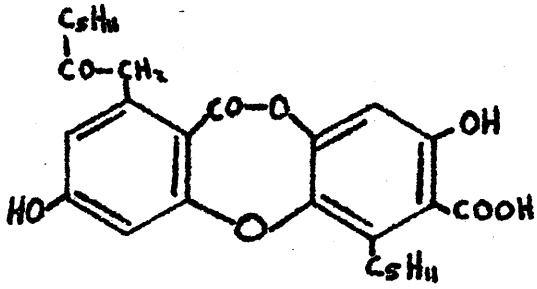


44

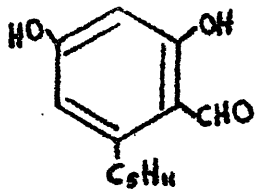
THEORETICAL

The most probable biogenetic pathway leading to the rather unusual spiro-lactone moiety of picrolichenic acid (50) involves the condensation of a poly-β-ketone[66] chain of six acetate units (as 94) followed by self-esterification, reduction and appropriate methylation to give the depside (48). The unusual methylation pattern of (48) prevents the operation of the C-O coupling[8,9] involved in the normal depside-depsidone formation which occurs with olivetoric acid and physodic acid (43→44). Instead the diradical (49) generated from (48) must lead by C-C coupling to picrolichenic acid (50) as suggested by Erdtman and Wachtmeister[50].

Methylation of hydroxyl in the ortho position to the depside link has been observed in boninic acid[67] and diffractaic acid[68] and in the tridepside, umbilicaric acid[69].

It was considered that a rational synthesis of picrolichenic acid by such a radical pairing reaction would offer considerable support for the biogenetic hypothesis, and, at the same time provide indirect confirmation of the depside-depsidone relationship. In the particular case of picrolichenic acid a synthesis by such a route would verify the proposed structure (50).

The starting material which appeared to be most useful



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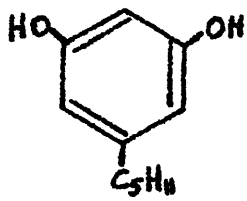

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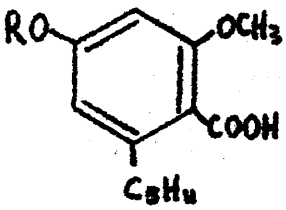

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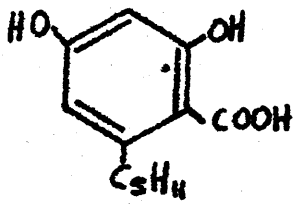

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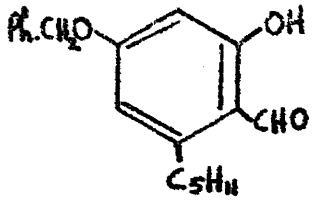
for the synthesis was olivetol aldehyde (95). This compound is ideally suited, for it can be used for construction of both components of the required depside system. Olivetol (96) was prepared[70] in good yield from 3,5-dihydroxybenzoic acid and then converted to olivetol aldehyde[71] by means of zinc cyanide[72] and dry hydrogen chloride.

Entry into the depside series by condensation of a suitably substituted acid (97) with olivetol aldehyde (95) using a modification of Asahina's general synthetic method[46,73] was now examined. Olivetol carboxylic acid (98) was prepared according to Asahina and Yosioka[74] from olivetol aldehyde by O-carbethoxylation, oxidation with neutral potassium permanganate and alkaline hydrolysis. The overall yield of the acid was low and therefore another route to a suitable derivative of (97) was sought.

Resorcinol type phenols, which may also be written in the tautomeric form of a β-diketone, are susceptible to oxidation conditions and it was therefore desirable to have the phenolic hydroxyls of (95) protected in some manner before oxidation of the formyl group. A methyl ether in the 2-position was required but monomethylation would be expected to occur predominantly on the 4-hydroxyl. A free hydroxyl group in the 4-position is required to perform the oxidative coupling reaction so that a protecting function must be found which will be stable to the conditions used in constructing the required depside and which



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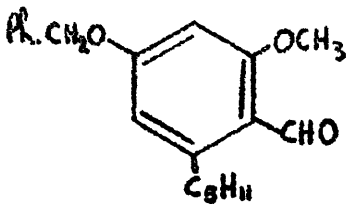

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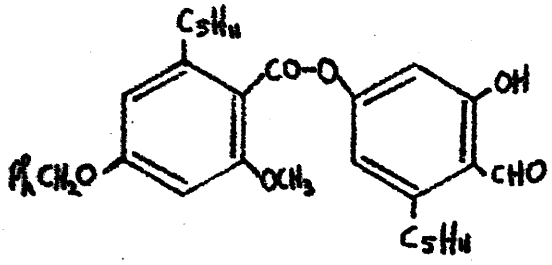

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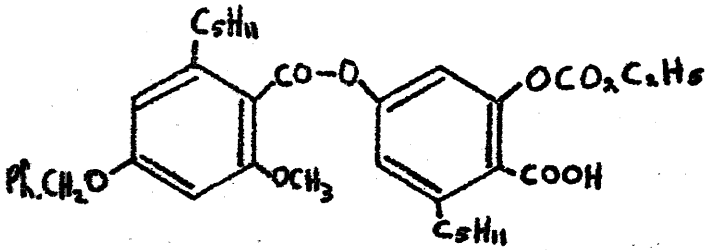

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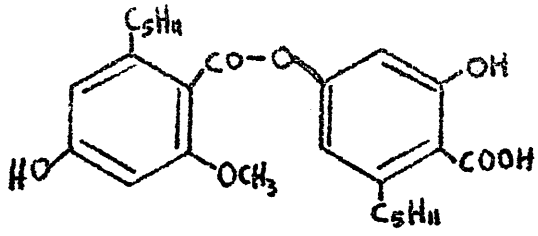
can then be readily removed without further degradation of the depside. The benzyl group was chosen as a suitable protecting function, readily removed by hydrogenolysis.

Benzylation of (95) with excess benzyl chloride and one mole of potassium hydroxide in aqueous ethanol gave an oil. Chromatography on silica gel furnished the monobenzyl derivative which was identified as the 4-benzyl compound (99) since it gave a red colour with 2% ethanolic ferric chloride solution and the infrared spectrum showed a maximum at 1640 (bonded CHO)cm.$^{-1}$ Methylation of (99) with dimethyl sulphate[75] furnished the methyl ether (100) which gave no colour with ferric chloride and absorbed at 1680 (aromatic CHO) cm.$^{-1}$ Oxidation of (100) with neutral potassium permanganate gave the required protected olivetol carboxylic acid (97, R=$PhCH_2$-) which absorbed at 1700 (-COOH) cm.$^{-1}$

The acid chloride of (97, R=$PhCH_2$-) was condensed with the 4-hydroxyl of olivetol aldehyde by means of pyridine in ether[73]. The depside (101) was isolated from the reaction after chromatography on silica gel. The product had ν max. 1750 (depside-CO.O-) and 1640 (bonded CHO) cm.$^{-1}$ and it gave a red ferric chloride colour. Carboethoxylation[76] of the phenolic hydroxyl group and oxidation with neutral potassium permanganate afforded the protected depside (102) ν max. 1770 (-O-$CO_2C_2H_5$), 1750 (depside-CO.O-) and 1700 (COOH) cm.$^{-1}$ The depside (102) was successively hydrogenated[77] to remove the benzyl group and



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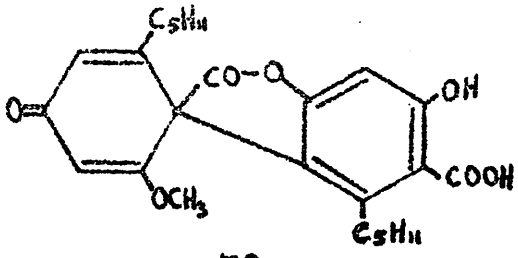

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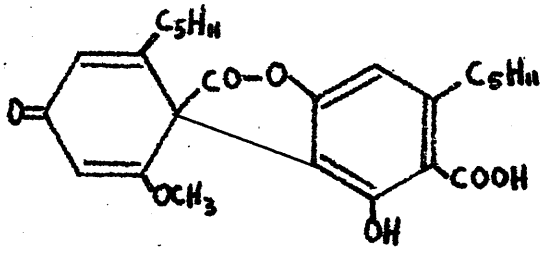

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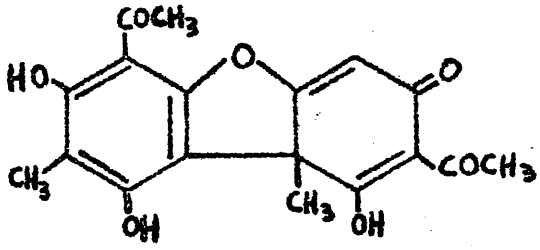

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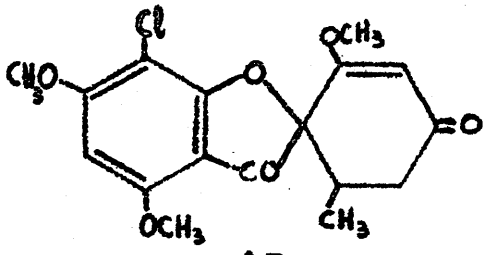

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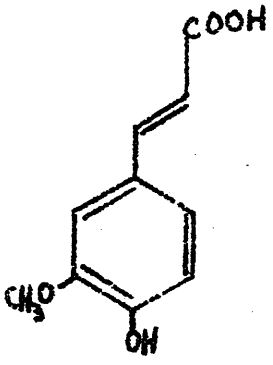
briefly treated with alkali to hydrolyse the O-carbethoxyl grouping[73]. The resultant depside, dihydropicrolichenic acid (48), showed the expected light absorption at 3300 (-OH), 1740 (depside-CO.O.) and 1655 (bonded-COOH) cm.$^{-1}$ with λ max. 250, 291 mμ (ε 13,000, 7,600).

There now remained the problem of the oxidative coupling of dihydropicrolichenic acid (48) to picrolichenic acid (50). It was conceivable that the coupling might occur with the position ortho to the hydroxyl group in the right hand ring of (48) to give the isomeric acid (103), but the necessary elaboration required to remove this ambiguity was not considered expedient.

Since picrolichenic acid exhibits a distinctive absorption maximum at 1820 cm.$^{-1}$ in the infrared due to the β,γ-unsaturated lactone chromophore[50,63], it was hoped that the appearance of a maximum at 1820 cm.$^{-1}$ and the concomitant disappearance of that at 1740 cm.$^{-1}$, due to the normal depside link in (48), on spectroscopic analysis of a reaction, would provide evidence that the properly dehydrogenated acid was indeed being formed.

Alkaline potassium ferricyanide has been used successfully in the syntheses of usnic acid (39)[13] and griseofulvin (25)[30,31]. However dihydropicrolichenic acid (48) was recovered unchanged from this treatment under conditions varying from 0° to 70° and from 40 minutes to 24 hours. No infrared absorption at



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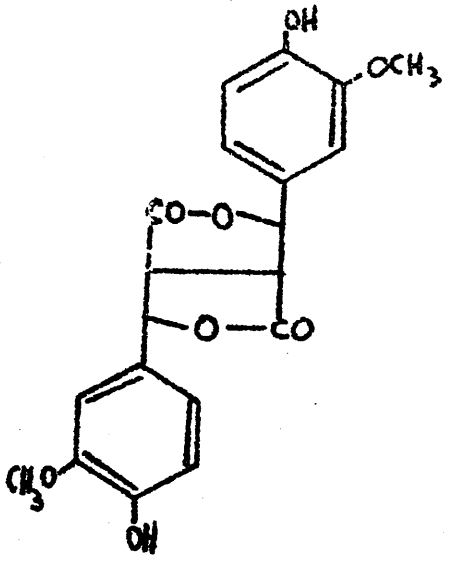

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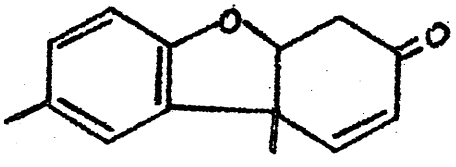

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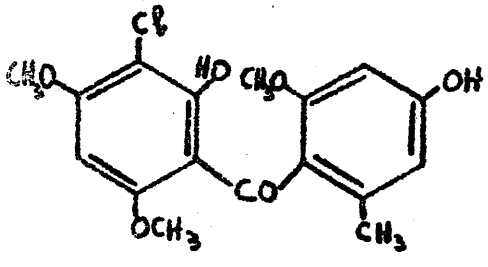

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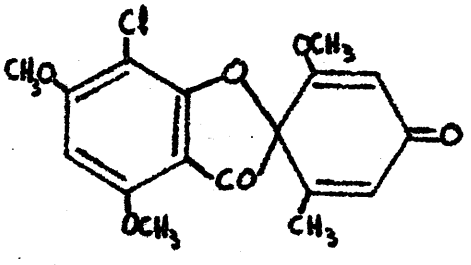

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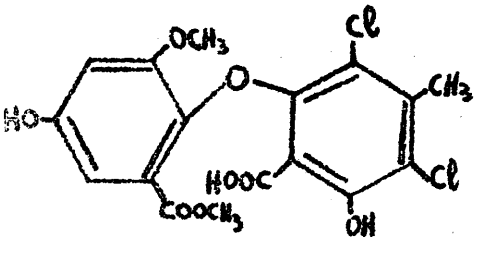

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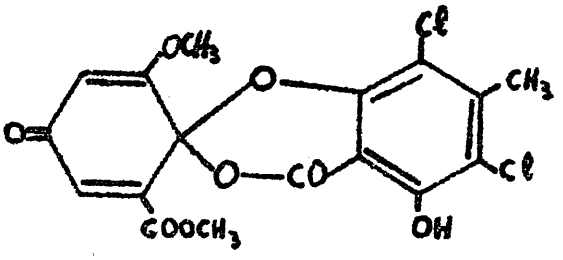

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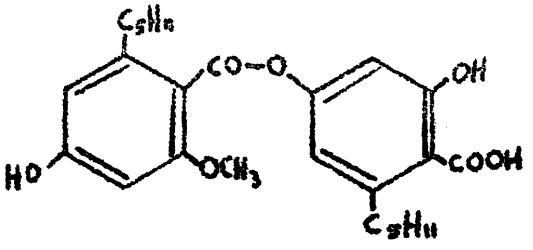
1820 cm.$^{-1}$ was observed in any of the experiments.

It was thought that the oxidation potential of the ferricyanide-ferrocyanide system might not be high enough and several other systems with higher oxidation potentials[78] were tried.

Dehydrogenation of ferulic acid (104) to give (105) has been achieved[79] by means of alcoholic ferric chloride solutions but under similar conditions (48) was recovered unchanged. No evidence of coupling was obtained from further oxidation attempts by electrolytic oxidation at a smooth platinum electrode or by using dichlorodicyanoquinone[80].

Model experiments were performed with the quinquevalent vanadium salt[81], ammonium metavanadate. Using this reagent p-Cresol gave Pummerer's ketone (6) and dihydrodehydrogriseofulvin (26) furnished dehydrogriseofulvin (27) but once again no evidence for the production of picrolichenic acid could be observed on treatment of (48) under the same conditions. Ceric sulphate treatment of dihydropicrolichenic acid left the starting material unchanged although p-cresol was once again successfully oxidised to Pummerer's ketone by this reagent. The successful coupling of geodin hydrate (36) to geodoxin (32) had been achieved by means of lead dioxide[45] in a neutral medium. Treatment of (48) with lead dioxide in refluxing benzene or toluene resulted in a resinous product and 30% recovery of the



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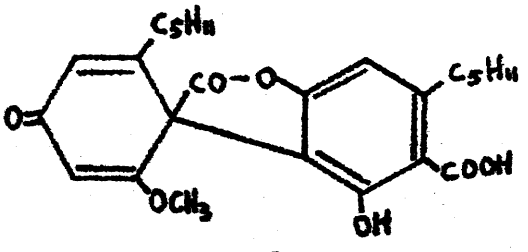

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starting material. No spectroscopic evidence of coupling was obtained.

The use of manganese compounds was now investigated. When a solution of dihydropicrolichenic acid (48) in benzene at 20°, containing fifteen equivalents of manganese dioxide in suspension, was subjected to spectroscopic control, it was found that the carbonyl band at 1740 cm.$^{-1}$ decreased in intensity and a new band appeared at 1820 cm.$^{-1}$, the position expected for the spiro-lactone chromophore. Increasing the molar ratio of the reagent or the temperature of the reaction led to rapid deterioration of the required spectrum, a broad band at 1700 - 1720 cm.$^{-1}$ replacing the 1820 and 1740 cm.$^{-1}$ bands. When the solution resulting from six successive treatments under the above conditions was rapidly chromatographed over silica gel and eluted with benzene-ether (49:1) there resulted a solid m.p.178° (with evolution of gas) which was undepressed on admixture with authentic picrolichenic acid (m.p.178°) and had infrared bands in chloroform solution at 1820 (spiro-lactone), 1670 (bonded -COOH and dienone) and 1607 (aromatic) cm.$^{-1}$ and λmax. 245, 270-277mμ. (ϵ 24,000; 7,800) in which it corresponded exactly with the data for the natural acid.

The infrared spectra (in KCl) of the natural and synthetic acids were identical in every respect.

No evidence was obtained for the presence of the isomeric acid (103).

The structure of picrolichenic acid is thus confirmed in every detail and the postulated biogenesis supported in some measure by the synthesis.

Picrolichenic acid has a quaternary centre at the spiro position but the natural material is optically inactive. A rotatory dispersion determination (589 to 350mμ.), which was kindly provided by Prof. G. Ourisson, indicates that it is in fact racemic. It is not possible to write an ionic mechanism for the racemisation and a radical mechanism resulting in the splitting of the C-C bond between the two rings is probably operative.

Attempts to effect a resolution via the quinine methohydroxide salt[82] have only been partially successful. Picrolichenic acid forms a salt m.p. 192-194°; $(\alpha)_D^{EtOH}$ - 100° which proved difficult to crystallise and no optically pure material was obtained. However regeneration of picrolichenic acid by dilute sulphuric acid from two different crops of the salt afforded impure samples of the acid with rotations of $(\alpha)_D^{EtOH}$ - 9.5° and $(\alpha)_D^{EtOH}$ + 15.2°.

EXPERIMENTAL

Olivetol (96).

This compound was prepared as described by Suter and Weston[70].

Olivetol aldehyde (95).

Dry hydrogen chloride was passed rapidly through a stirred solution of olivetol (21 g.) in anhydrous ether (250 c.c.) containing zinc cyanide (19 g.) in suspension. The reaction was maintained at room temperature. After saturation with hydrogen chloride (2.5 hr.) the passage of gas was discontinued and the reaction mixture was stood overnight at room temperature. Anhydrous ether was added to precipitate any dissolved aldimine hydrochloride. After decantation of the ether and washing twice with ether the residual paste was refluxed with water (150 c.c.) for 1 hr. The oily product, obtained by ether extraction of the cooled solution, afforded, after chromatography on silica gel (600 g.) and elution with benzene-ether (19:1, 5 l.), the required aldehyde (12.5 g.) as needles (from light petroleum), m.p. 66-68°. ν max. (Nujol) 3200 (free OH) and 1640 (bonded CHO) cm.$^{-1}$ (lit.[71] m.p. 66-67°).

Olivetol carboxylic acid (98).

This compound was prepared as described by Asahina and Yosioka[74].

4-Benzyloxy-2-hydroxy-6-pentylbenzaldehyde (99).

2,4-Dihydroxy-6-pentylbenzaldehyde (5 g.) and benzyl chloride (5 c.c.) were heated under reflux for 4 hr. in aqueous ethanol (25%; 25 c.c.) containing potassium hydroxide (1.6 g.). Removal of the ethanol under reduced pressure followed by acidification and recovery in ether gave an oil which on chromatography over silica gel (100 g.) and elution with light petroleum-benzene (9:1; 1600 c.c.) gave the mono-benzyl derivative (3.9 g; 55%), b.p. 190°/0.5 mm., $n_D^{18°}$ 1.5852; ν max (film) 1640 cm.$^{-1}$ (bonded CHO) (Found: C, 76.65; H, 7.45. $C_{19}H_{22}O_3$ requires C, 76.5; H, 7.45%). The compound gives a red colour with 2% ethanolic ferric chloride solution.

4-Benzyloxy-2-methoxy-6-pentylbenzaldehyde (100).

The monobenzyl-aldehyde (3.9 g.), dimethyl sulphate (1.5 c.c.), and potassium carbonate (12 g.) were stirred under reflux in dry acetone (100 c.c.) for 16 hr., addition of dimethyl sulphate and potassium carbonate having been made in four portions during the first 2 hr. Filtration of the cooled solution, evaporation, and isolation in ether gave the benzyl methyl ether (100) (3.6 g; 88%) as plates (from methanol), m.p. 51-52°; νmax. (Nujol) 1680 cm.$^{-1}$ (aromatic CHO) (Found: C, 76.95; H, 7.85. $C_{20}H_{24}O_3$ requires C, 76.9; H, 7.75%).

4-Benzyloxy-2-methoxy-6-pentylbenzoic Acid (97, R=$PhCH_2$-).

To a stirred solution of 4-benzyloxy-2-methoxy-6-pentylbenzaldehyde (1.8 g.) in acetone (500 c.c.) was added potassium permanganate (7 g.) in water (100 c.c.) during 2 hr. at 45°, then the mixture was stirred (at this temperature) for 6 hr. Sulphur dioxide was passed through the cooled solution, and the acetone removed. Separation into an acidic and a neutral fraction and isolation of the acid in ether gave an oil. Chromatography of this on silica gel (15 g.) and elution with benzene-ether (19:1; 250 c.c.) afforded the required acid (700 mg.; 37%) as needles (from light petroleum), m.p. 62-63°; $\nu_{max.}$ (Nujol) 1700 cm.$^{-1}$ (CO_2H) (Found: C, 73.35; H, 7.45. $C_{20}H_{24}O_4$ requires C, 73.15; H, 7.35%).

4-Formyl-3-hydroxy-5-pentylphenyl 4-Benzyloxy-2-methoxy-6-pentylbenzoate (101).

4-Benzyloxy-2-methoxy-6-pentylbenzoyl chloride (prepared from the above acid (5 g.) by the oxalyl chloride-benzene method) was dissolved in anhydrous ether (500 c.c.) containing 2,4-dihydroxy-6-pentylbenzaldehyde (3.3 g.). The solution was cooled to 0° and pyridine (20 c.c.) added. After 16 hr. at room temperature the solution was acidified and the acidic fraction isolated as an oil, which after chromatography on silica gel and elution with light petroleum-benzene (2:3; 2 l.) gave the required ester as a viscous oil (6.8 g.; 85%); this had $\nu_{max.}$ (film) 1750 (depside C=O) and 1640 (bonded CHO) cm.$^{-1}$ and gave a red ferric colour. The 2,4-dinitro-phenylhydrazone had m.p.

152-153° (from ethanol-ethyl acetate) (Found: C, 65.65; H, 6.0; N, 8.15. $C_{38}H_{42}N_4O_9$ requires C, 65.3; H, 6.05; N, 8.0%).

4-Carboxy-3-ethoxycarbonyloxy-5-pentylphenyl 4-Benzyloxy-2-methoxy-6-pentylbenzoate (102).

The foregoing aldehyde was converted into the O-ethoxycarbonyl derivative (ethyl chloroformate-pyridine at -20° for 2 hr.). Oxidation of this with potassium permanganate, the conditions described above being used, gave the required O-ethoxycarbonyl-acid (102) (45%) as prisms (from benzene-light petroleum), m.p. 107-109°; ν max. (in carbon tetrachloride) 1770 (-O.CO_2Et), 1750 (depside), and 1700 (CO_2H) cm.$^{-1}$ (Found: C, 69.1; H, 6.8. $C_{35}H_{42}O_9$ requires C, 69.3; H, 7.0%).

Dihydropicrolichenic Acid (48).

The acid (102) (1.0 g.) was hydrogenolysed in ethanol (50 c.c.) over palladium-charcoal (10%; 200 mg.). After the consumption of 1 mole of hydrogen the solvent was removed and replaced by acetone (5 c.c.). When this solution was treated with sodium hydroxide solution (N; 10 c.c.) for 10 min. and the resulting acidic fraction isolated in ether, the required acid formed prisms (460 mg.; 65%), m.p. 117-118° (from benzene); λmax. 250 and 291 mμ.(ε 13,000 and 7600); ν max. (potassium chloride disc) 3400 (OH), 1725 (depside C=O), 1650 (bonded CO_2H); ν max. (in chloroform) 3300, 1750, and 1655 cm.$^{-1}$ (Found: C, 68.05; H, 7.1. $C_{25}H_{32}O_7$ requires C, 67.55; H, 7.25%).

Oxidation experiments.

(1) Potassium ferricyanide.

A solution of potassium ferricyanide (89 mg.) in distilled water (10 c.c.) was added dropwise over 40 mins. to a stirred solution of dihydropicrolichenic acid (60 mg.) in distilled water (15 c.c.) at 0° and in an atmosphere of nitrogen. The solution was stirred for a further 30 mins. Acidification of the bicarbonate solution and isolation in ether gave a quantitative recovery of unchanged dihydropicrolichenic acid. No trace of absorption at 1820 $cm.^{-1}$ in the infrared was observed.

Similar results were obtained when the reaction was attempted under different conditions of temperature, time and quantity of oxidant. (See Table 1.).

(2) Ferric Chloride

Aqueous ferric chloride solution (5%; 3 c.c.) was added to dihydropicrolichenic acid (25 mg.) in ethanol (10 c.c.). Acidification and removal of the ethanol after 14 hr. furnished an ether soluble product which had an infrared spectrum in chloroform identical with the starting acid.

The recovered material was treated again for 14 days with the same negative result.

(3) Anodic oxidation at a platinum electrode.

Dihydropicrolichenic acid (22.4 mg.) was subjected to

oxidation at a platinum anode [(3 x 2 cm.); 110v D.C.] in a solution of sulphuric acid in acetic acid (0.1 molar) for 4 hr.

The recovery of acid was quantitative.

(4) Ammonium vanadate

(a) Pummerer's ketone (6).

Conc. sulphuric acid (10 c.c.) was added over 30 mins. to a stirred suspension of p-cresol (2 g.) and ammonium metavanadate (15 g.) in water (200 c.c.) at 0°. The solution was made alkaline and an ether extract gave a gum. Chromatography of the gum on alumina (grade V) (50 g.) and elution with light petroleum ether - benzene (1:1) furnished white prisms (230 mg.) m.p. 223-224° (from ethanol). ν max. (in carbon tetrachloride) 1690 $cm.^{-1}$ The product was identical in melting point, mixed melting point and infrared spectrum with an authentic sample of Pummerer's ketone.

(b) Dehydrogriseofulvin (27).

Dihydrodehydrogriseofulvin (26, 200 mg.) in aqueous acetic acid (30 c.c., 1:1) was stirred with ammonium metavanadate (2 g.) and conc. sulphuric acid (1 c.c.) at 0° for 15 mins. The ether extract of the reaction mixture was washed with saturated sodium bicarbonate solution and then alkali. The neutral extract furnished a solid, ν max. (in chloroform) 1715 and 1670 $cm.^{-1}$ which was identical with the infrared spectrum of (-) dehydrogriseofulvin.

(c) Attempted oxidation of dihydropicrolichenic acid (48).

Dihydropicrolichenic acid (150 mg.) in aqueous acetic acid (25 c.c; 2:3) was stirred with ammonium metavanadate (2 g.) at 20° for 24 hr. The ether extract, after several washings with water to remove acetic acid, gave a gum with an infrared spectrum in chloroform solution identical with the starting dihydro acid.

(5) Ceric sulphate.

(a) Pummerer's ketone (6).

p-Cresol (2 g.), ceric sulphate (15 g.) and conc. sulphuric acid (10 c.c.) were stirred in water (200 c.c.) at 0° for 30 mins. The neutral product from the reaction after chromatography on alumina (40 g.; grade V) and elution with light-petroleum-benzene (2:3) was identical to Pummerer's ketone.

(b) Attempted oxidation of dihydropicrolichenic acid (48).

Dihydropicrolichenic acid (100 mg.), in aqueous acetic acid (20 c.c., 2:3) was stirred with ceric sulphate (2 g.) at 20° for 24 hours. The ether extract furnished a gum with an infrared spectrum in chloroform solution identical to the starting dihydro acid.

The recovered material was recycled under the same conditions for 14 days. The infrared spectrum again indicated only dihydropicrolichenic acid and no evidence for the presence of coupled product.

(6) Lead dioxide.

(a) Pummerer's ketone (6).

p-Cresol (2 g.) was refluxed in benzene (50 c.c.) with lead dioxide (5 g.) for 3 hours. The gum obtained after filtration and removal of the solvent *in vacuo* was dissolved in ether and washed with alkali. The neutral material obtained was identified as Pummerer's ketone.

(b) Attempted oxidation of dihydropicrolichenic acid (48).

Dihydropicrolichenic acid (200 mg.) was refluxed and stirred with lead dioxide (500 mg.) in benzene (50 c.c.) for 16 hours. After filtration, washing with ether, and removal of the solvent *in vacuo* a gum (25 mg.) was obtained. The lead dioxide was extracted with sodium carbonate solution. Acidification and ether extraction gave more gum (35 mg.). Both extracts showed only the dihydro acid in their infrared spectra.

A similar result was obtained when the reaction was carried out in boiling toluene.

(7) Dichlorodicyanoquinone.

Dihydropicrolichenic acid (100 mg.) and dichlorodicyanoquinone (110 mg.) were refluxed in benzene (30 c.c.) for 16 hours. After removal of the benzene *in vacuo* the residue was digested with chloroform. The infrared spectrum of this solution showed only the presence of the dihydro acid and no absorption at 1820 $cm.^{-1}$

(8) Manganese dioxide.

Picrolichenic Acid (50).

Dihydropicrolichenic acid (30 mg.) was treated with manganese dioxide (J. Woolley Co.; 15 equivs.; 90 mg.) in benzene (30 c.c.) with stirring for 30 min. The manganese dioxide was removed by filtration and extracted with sodium carbonate solution, and the acidified solution was extracted with ether. The combined benzene and ether extracts were evaporated to leave a gum, which had in its infrared spectrum (in chloroform) a new band at 1820 cm.$^{-1}$, while the intensity of the band at 1750 cm.$^{-1}$ had decreased in proportion to the increase in intensity of the 1660 cm.$^{-1}$ band. Five more treatments of the mixture under the same conditions (with spectroscopic control) gave a product with maximum intensity of the 1820 cm.$^{-1}$ band. Chromatography on a short column of silica gel (300 mg.) and elution with benzene-ether (49:1) gave a solid fraction (5-7 mg.; average of several experiments), which on crystallisation from ether-petroleum gave prisms, m.p. 178° (with evolution of carbon dioxide) which was identical in m.p. mixed m.p., and ultraviolet and infrared absorption with authentic picrolichenic acid; λ max. 245 and 270-277 mμ. (ε 22,000 and 7800); ν max. (potassium chloride disc) 1820 (spirolactone) 1670 cm.$^{-1}$ (bonded CO_2H and dienone superimposed).

When the reaction was carried out at 50-60°, or in the presence of 40, 55, or 100 equivalents of manganese dioxide in benzene or chloroform solution, a new band in the infrared spectrum rapidly grew at 1700-1710 cm.$^{-1}$ and no crystalline product was

isolable from such a mixture. This effect was also observed when picrolichenic acid (20 mg.) was treated for 24 hr. with manganese dioxide (200 mg.) in benzene (15 c.c.) at room temperature, or for 30 min. at 50°.

The following oxidising conditions led to quantitative recovery of dihydropicrolichenic acid from the reaction mixture.

TABLE 1.

Oxidant (moles)		Solvent	Time	Temp.
$K_3Fe(CN)_6$	(2) ...	Na_2CO_3-H_2O	1 hr.	0°
"	(2) ...	"	2 hr.	20
"	(2) ...	"	2 hr.	70
"	(5) ...	"	24 hr.	20
$FeCl_3$	(5) ...	EtOH-H_2O	14 hr.	20
"	(5) ...	"	14 days	20
PbO_2	(3) ...	Benzene	24 hr.	80 *
"	(3) ...	Toluene	2 hr.	110 *
$Ce(SO_4)_2$	(3) ...	AcOH-H_2SO_4(aq.)	24 hr.	20
NH_4VO_3	(5) ...	"	24 hr.	20
Dichlorodicyanoquinone	(2) ...	Benzene	24 hr.	80

* 30% recovery of starting material; residue resinous.

Oxidation at a smooth platinum anode (3 x 2 cm.) in acetic

acid-sulphuric acid mixtures at 110v (D.C.) gave back dihydro-picrolichenic acid in quantitative yield.

Attempted resolution of picrolichenic acid (50).

Natural racemic picrolichenic acid (500 mg.) in ethanol (10 c.c.) was treated with 0.27N-aqueous quinine methohydroxide[82] (4.2 c.c., 1 equiv.). The solvents were removed in vacuo and the residual solid, m.p. 192-194°, $(\alpha)_D^{EtOH}$ - 100° was taken up in ethyl acetate (5 c.c.). The addition of ether precipitated the salt (300 mg.) $(\alpha)_D^{EtOH}$ - 108°. Further addition of ether afforded another crop of the salt (180 mg.) $(\alpha)_D^{EtOH}$ - 78.5° which on recrystallisation from ethyl acetate-ether gave prisms (120 mg.) m.p. 192-194°, $(\alpha)_D^{EtOH}$ - 83°. Attempts to recrystallise portions of the salts did not furnish any material with a constant rotation. The first crop of the salt and the recrystallised second crop were acidified with 10% W/W sulphuric acid with shaking in the presence of ether. Recovery from the ether gave meringues with rotations of $(\alpha)_D^{EtOH}$ - 26.4° and $(\alpha)_D^{EtOH}$ + 15.2° respectively. Recrystallisation of the levorotatory sample afforded picrolichenic acid (30 mg.) m.p. 175-178° (from acetonitrile) with a rotation of $(\alpha)_D^{EtOH}$ - 9.5°. Optically pure picrolichenic acid was not obtained.

PART II

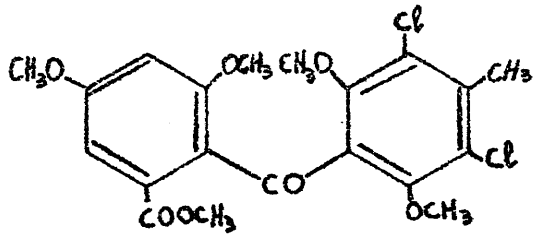
SYNTHETIC APPROACHES TO GEODIN AND ERDIN

HISTORICAL

Raistrick and Smith[37] showed that a strain of Aspergillus terreus Thom, when grown on Czapek-Dox solution containing glucose as a sole source of carbon and potassium chloride as a sole source of chlorine, produced two new metabolic products geodin ($C_{17}H_{12}O_7Cl_2$) and erdin ($C_{16}H_{10}O_7Cl_2$). These compounds were the first recorded metabolic products containing chlorine to be isolated from the lower fungi. Aspergillus flavipes produces a substance which was thought to be a new antibiotic[83] but it is in fact identical with (+) geodin[43]. Estin, an antibiotic produced by Penicillium estinogenum, was also identified[84] as (+) geodin.

Geodin is regarded as a moderately potent antibiotic substance active in the main against gram positive bacteria only, while erdin has not been observed to inhibit bacteria[85].

The first structural proposals were forwarded by Raistrick and his collaborators[42]. Both geodin and erdin, on catalylic hydrogenation[40], gave dihydro derivatives which on complete methylation with diazomethane yielded the same compound ($C_{15}H_5O_2Cl_2(OCH_3)_5$), a neutral substance. Hydrolysis of the fully methylated product afforded a monobasic acid ($C_{15}H_6O_3Cl_2(OCH_3)_4$), one methoxyl group being lost. Further work by Raistrick et al[41] indicated that dihydrogeodin was the methyl ester of dihydroerdin and that the fully methylated product was a



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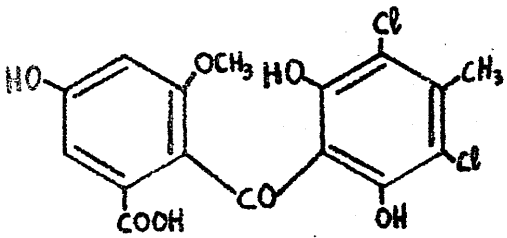

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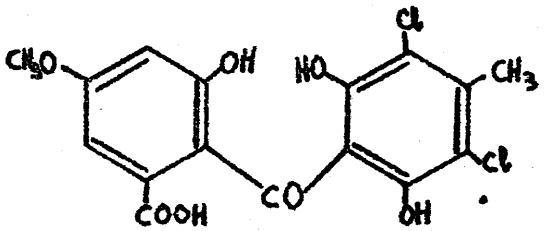

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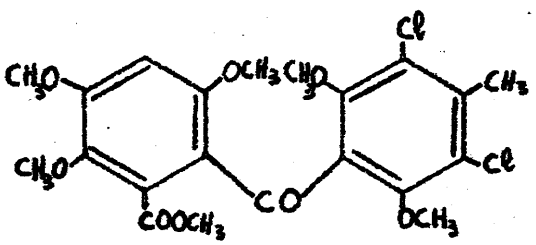


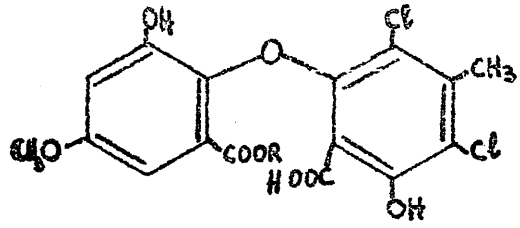
109

tetramethyl ether ester with the empirical formula ($C_{14}H_5O\ Cl_2(OCH_3)_4(COOCH_3)$).

Hydrolytic fission with 80% sulphuric acid and treatment with hydriodic acid of the dihydro compounds[41] gave rise to derivatives of 3,5-dihydroxybenzoic acid and 2,6-dichloro-p-orsellinic acid. These results along with the analytical data suggested that the dihydro derivatives were hydroxylated benzophenones. This hypothesis was confirmed by the synthesis[41] of the fully methylated benzophenone (106) by a Friedel and Craft's reaction between methyl 3,5-dimethoxybenzoate and 2,6-dichlorodimethoxy-p-orsellinic acid. The synthetic material was identical with trimethyldihydrogeodin. Dihydroerdin wasthus shown to have either the structure (107) or (108) and hence dihydrogeodin was the corresponding methyl ester. Of these the structure (107) was preferred[42] because dihydrogeodin did not readily give a blue colouration with Gibb's reagent[59]. However exceptions to the Gibb's test have been observed[86,87].

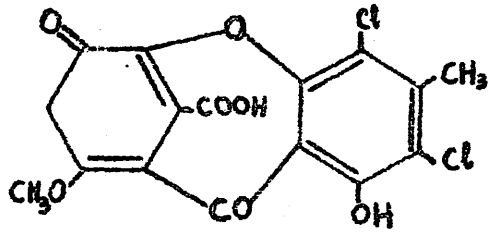
Methylation of geodin with dimethyl sulphate[40] furnished the pentamethyl ether (109).

Erdin and geodin on treatment with 80% sulphuric acid afforded the dibasic erdin hydrate and its monomethyl ester geodin hydrate[42]. Methylation of the hydrates yielded the same product $C_{20}H_{20}O_8Cl_2$. The molecular formula was accounted for by the two benzene rings, two carboxyl groups, five methoxyl groups and one methyl side chain. Assuming that the chlorine atoms



110 R=CH_3, Geodin hydrate

R=H, Erdin hydrate



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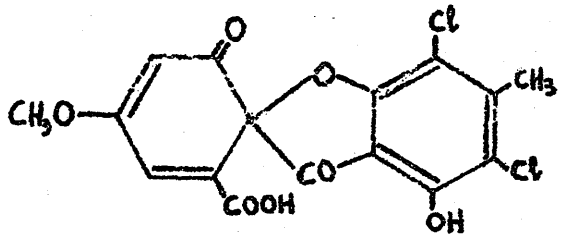


112

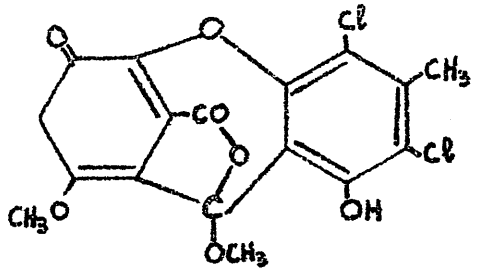
are attached to the benzene ring in the usual way the remaining oxygen could only be placed as an ether bridge between the two rings.

Since the hydrates gave a blue colour with Gibb's reagent the formula (110) was proposed for geodin and erdin hydrates.

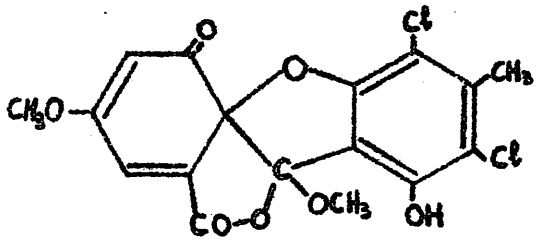
Although the carbonyl groups of the dihydro derivatives of geodin and erdin were inactive geodin readily formed an oxime. This suggested that geodin, and erdin, probably contained a second keto group. Since they both gave addition compounds with diazomethane[37] a quinonoid or potential quinonoid structure in the non-chlorinated ring was feasible. The absorption spectra of geodin and erdin (λmax. 284 mμ., ε15,000) are not incompatible with this proposal.

Hydriodic acid treatment of geodin and erdin caused the evolution of carbon dioxide and two isomeric substances norgeodin (A) and norgeodin (B) with the composition ($C_{14}H_{10}O_5 \cdot H_2O$) were isolated. The yellow norgeodin (A) was probably a xanthone.

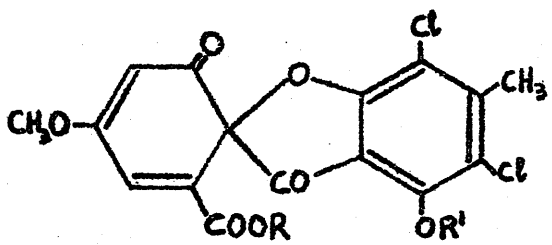
This work by Raistrick et al. showed that the two six-carbon rings were joined by an ether linkage and a carbonyl group. Structures (111) and (112) were proposed as alternatives for erdin[42]. The para quinonoid structure corresponding to (112) was ruled out because the product (110, R=H) obtained by acid treatment gave an immediate reaction with Gibb's reagent. Structure (111) was doubtful since it would be expected to revert


Cl
CH3
CO
O
Cl
CH3O
C
OH
OCH3

113


Cl
CH3O
CH3
Cl
C
CO-O
OCH3
OH

114


Cl
CH3O
CH3
CO
Cl
COOR
OR'

115

to the phenolic form with loss of ketonic properties. This structure does however account for the optical inactivity of erdin. Attempts to resolve erdin by means of brucine and d-α-phenylethylamine were unsuccessful. Structure (112) would be stable but should be resolvable. It is unsatisfactory since dihydroerdin should give a positive Gibb's reaction.

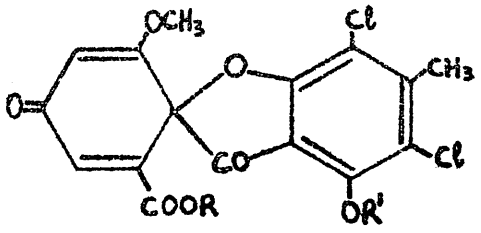
Geodin, which is dextrorotatory, appeared to be a methyl ester of erdin. The pseudo-ester structures (113) and (114) were suggested to account for the apparently isomeric but not identical products obtained by diazomethane treatment[37] of geodin and erdin.

Barton and Scott[43] reinvestigated the structures of geodin and erdin and by their elegant experiments established the correct structures. Erdin was successfully resolved by means of quinine methohydroxide, furnishing (+) erdin and thus excluding structure (111). Racemisations of (+) geodin and (+) erdin were readily achieved in dioxan containing 2% of hydrogen chloride.

Structures (111) and (113) were also incompatible with Bredt's rule.

The infrared absorption band at 1728 cm.$^{-1}$ for geodin favours a normal ester rather than the pseudo ester which would be expected to absorb at about 1780 cm.$^{-1}$

One can therefore write formulae[(115) or (116); R=CH_3, R'=H)] for geodin and[(115) or (116); R=R'=H)] for erdin.



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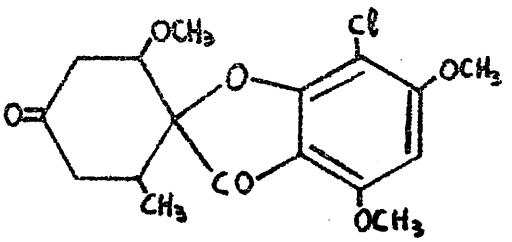

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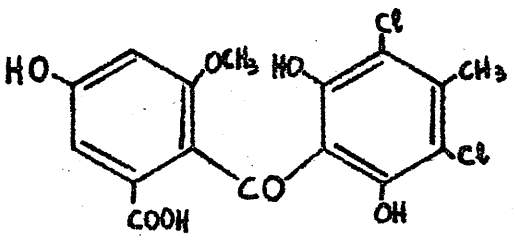

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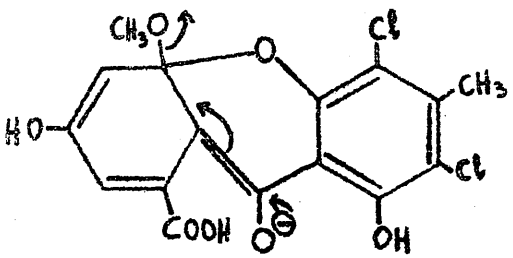

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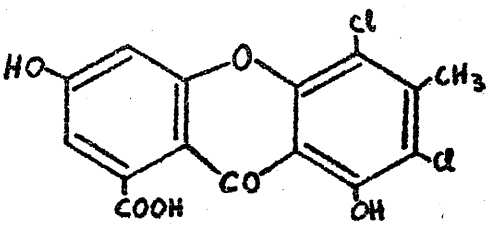


118

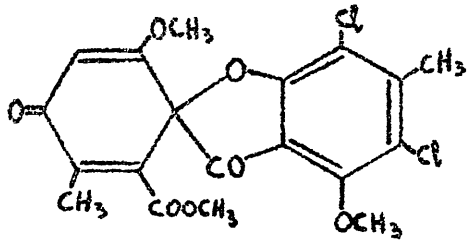
Geodin and erdin show intense ultraviolet absorption at 284mμ. A subtraction curve of the spectra of the model compound dihydrogriseofulvin[88] (117) from that of erdin gave a curve showing λ max. 241 mμ.(ε 14,000). This is fully consistent with a cross-conjugated dienone such as (116) but not with the linearly extended dienone such as (115) which would be expected[65] to absorb beyond 300 mμ.

Dihydroerdin, on treatment with N-alkali, gave 2,6-dichlororcorcinol and 5-hydroxy-3-methoxyphthalic acid. This result is in accord with a structure (116; R=R'=H) for erdin and hence (116; R=CH_3, R'=H) for geodin. These slight modifications of the formulae are in agreement with the infrared data for geodin (1728, 1665, 1630 cm.$^{-1}$) and erdin (1717, 1667, 1639 cm.$^{-1}$).

The dihydro compounds must therefore be as in (107).

The alkaline oleavage of dihydroerdin[43] afforded an acidic by-product, $C_{15}H_8O_6Cl_2$, which contained no methoxyl group. Methylation with diazomethane gave a methyl ester dimethyl ether suggesting the presence of two phenolic hydroxyl groups. The compound showed ultraviolet and infrared characteristics indicative of a xanthone. It was formulated as (118) being produced from (107) as in (119). This further confirms the orientation of the methoxyl group in erdin.

Additional chemical evidence in favour of the normal ester



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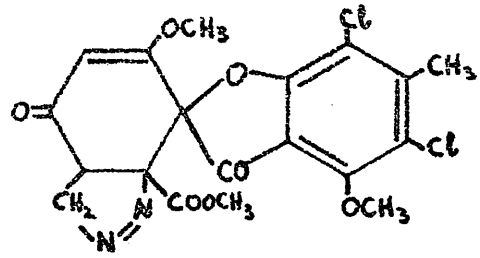

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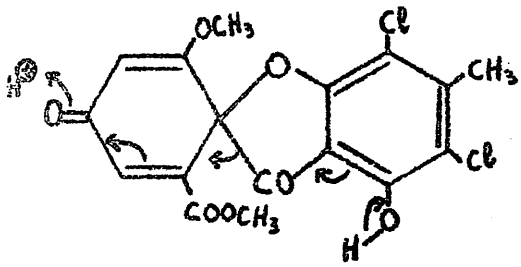

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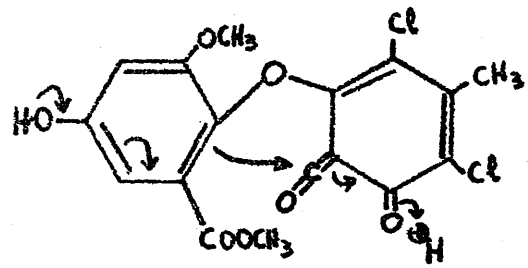

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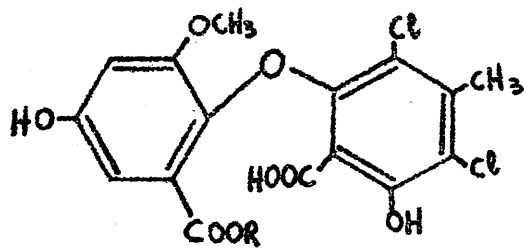
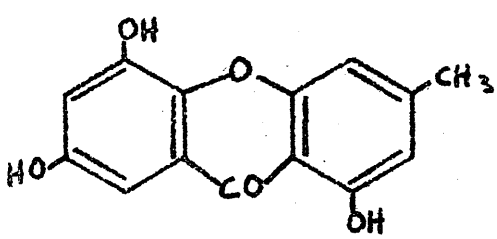


124 R = CH_3, Geodin hydrate.
R = H, Erdin hydrate.



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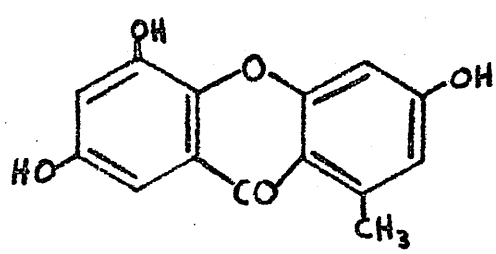


126

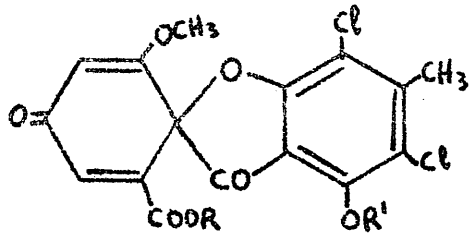
formulation was secured as follows. The pyrazolines obtained by the action of diazomethane on (+) geodin and (±) erdin had identical infrared absorption in solution and the pyrazolines from (+) geodin and (+) erdin were identical in every respect. Similarly the pyrazolines from (±) geodin and (±) erdin were the same compound. Raistrick[37] had claimed that the pyrazolines obtained from (+) geodin and (±) erdin were isomeric. This observation is due to using optically active geodin but racemic erdin.

Pyrolysis of the pyrazoline gave a compound whose analysis indicates that is is homogeodin methyl ether (120). The pyrazoline is thus best represented as (121).

The racemisation of the quaternary centre in (+) geodin and (+) erdin is thought to proceed by a mechanism such as (122) ⇌ (123)[43] which is dependant on the existence of the phenolic hydroxyl group in the chlorinated ring. Support for this mechanism is given by the observation that geodin monoacetate (116; R=CH_3, R′=OAc) and geodin methyl ether (116, R=R′=CH_3) were stable to the racemisation conditions required for (+) geodin and (+) erdin.

Geodin and erdin hydrates can now be formulated as (124). Barton and Scott also proposed that norgeodin A andB may be formulated as the xanthones (125) and (126).

A partial synthesis of (±) geodin methyl ether (116,



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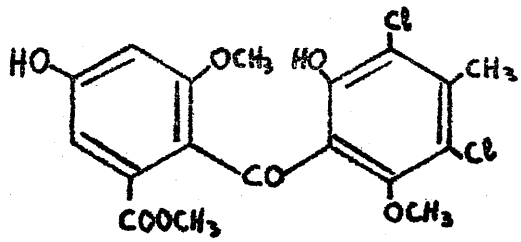
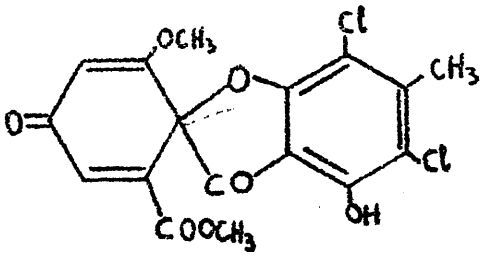


127

R=R'=CH_3) was carried out by Scott[31]. Dihydrogeodin methyl ether (127) was obtained by hydrogenation of (±) geodin methyl ether and reconverted to the spiro-dienone by treatment with alkaline potassium ferricyanide.



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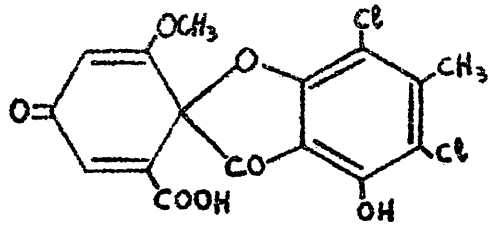

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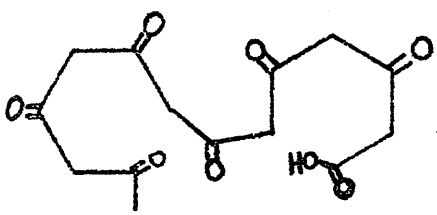

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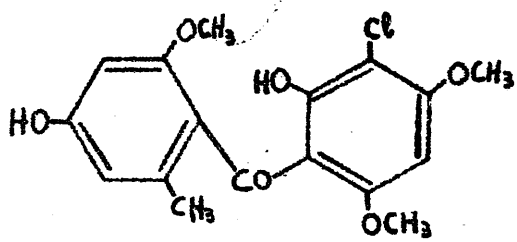

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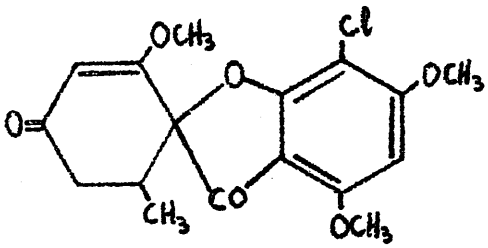

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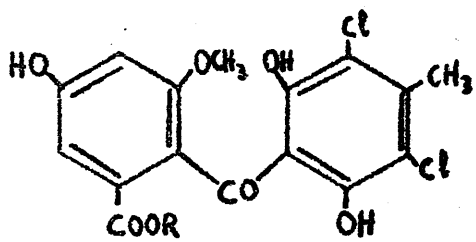

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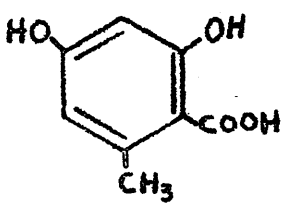

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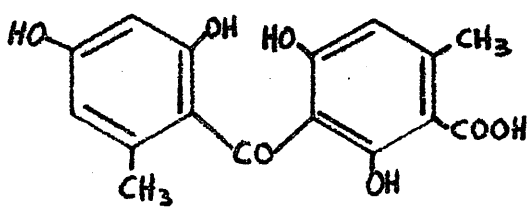


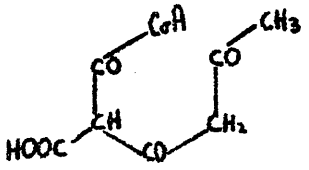
132

THEORETICAL

The structures of geodin (30) and erdin (128) although very similar to that of griseofulvin (25) cannot be formed by the same biogenetic route. Griseofulvin is derived from the condensation of seven acetic acid units in a head-to-tail fashion, possibly through a poly-β-keto acid (129), followed by cyclisation, O-methylation and chlorination in undetermined sequence leading to the benzophenone (26) which undergoes oxidative C-O coupling and partial hydrogenation to give griseofulvin (25)[29,30,34]. Geodin and erdin cannot be formed in this manner from a linear polyacetate chain because in order to obtain the C-methyl group in the aromatic ring a biogenetic "Grignard" type reaction must be postulated and this has no precedent.

It is possible to obtain the benzophenone (130) from linear polyacetate units by proposing the condensation of two molecules of orsellinic acid (131), which is derivable from acetate[89], to give (132) followed by decarboxylation and preferential oxidation of one C-methyl substituent. Alternatively the mould metabolite 3,5-dihydroxyphthalic acid[90] could be utilised along with one molecule of orsellinic acid and subsequent decarboxylation.

Geodin and erdin are derivable from a single polyacetate chain if a branched chain[91] is used. Recent work[92,93] has



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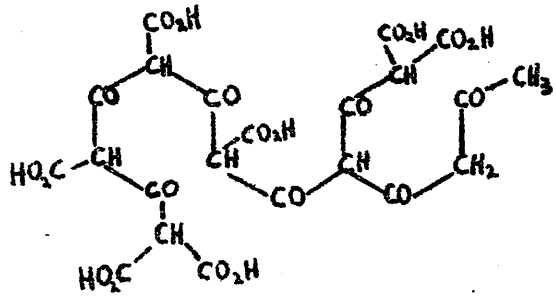

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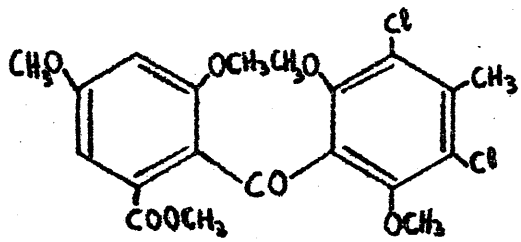

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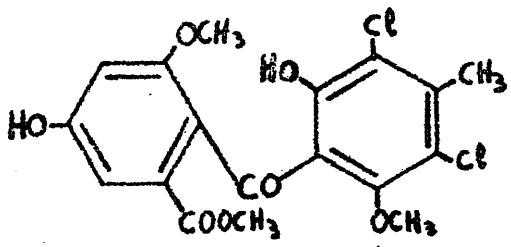

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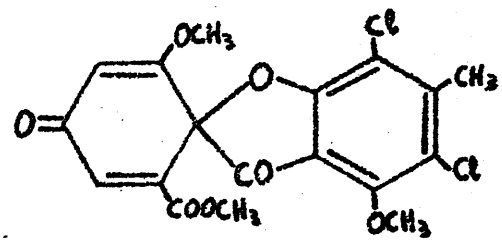


135

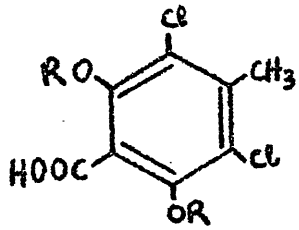
shown that malonate units play an important role on the biosynthesis of aromatic substances. Condensation of one molecule of acetyl-coenzymeA with two molecules of malonyl-coenzyme A and decarboxylation would give acetoacetylmalonyl-coenzyme A (133). If it is supposed that both of the carboxy residues combine with further malonate units a branched structure (134) could be derived which by cyclisation, decarboxylation, chlorination and oxidation in undetermined sequence, would give the required benzophenone (130).

Intramolecular oxidative C-O coupling of the benzophenone would then furnish the spiro-dienone system of geodin and erdin.

A synthesis of geodin or erdin which involved oxidative coupling of (130) would therefore provide indirect evidenceof the correctness of the postulated biogenesis.

Support for the practicability of such a synthesis has been provided by Raistrick's synthesis[41] of trimethyldihydrogeodin (106) and by the successful coupling of O-methyldihydrogeodin (127) to O-methylgeodin (135) due to Scott[31].

The first route to the benzophenone (130, R=CH_3) which was attempted envisaged the preparation of suitably protected derivatives of dichloro-p-orsellinic acid (136, R=H) and methyl 3-hydroxy-5-methoxybenzoate (137, R=H) and their subsequent condensation to the required product by means of a Friedel and Craft's reaction. Both of the parent compounds were prepared according to the literature methods.



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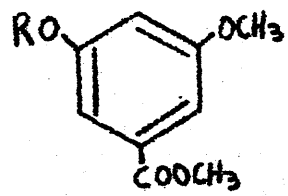
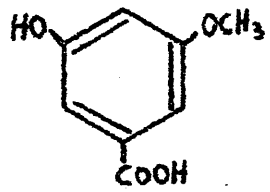


137

In Raistrick's synthesis[41] of trimethyldihydrogeodin the acid chloride of dichlorodimethoxy-p-orsellinic acid (136, R=CH_3) was condensed with methyl 3,5-dimethoxybenzoate (137, R=CH_3) by means of anhydrous aluminium chloride in the absence of any solvent. The crude reaction product was re-methylated because of the demethylation caused by the aluminium chloride. When these conditions were applied to the O-carbethoxyl derivative (137, R=-$CO_2C_2H_5$) prepared from (137, R=H) by means of ethyl chloroformate and pyridine[76] the products obtained were gums which could not be crystallised. Effervescence was observed during the reaction and it is possible that the acid chloride was being hydrolysed back to the acid (136, R=CH_3) which could then undergo acid catalysed decarboxylation. Further attempts to effect the condensation using milder conditions in solvents were also unsuccessful. The method of Shah[94] using zinc chloride and phosphorous oxychloride on the acid, thus forming the acid chloride <u>in situ</u>, was also tried without avail. The infrared spectra and ferric chloride colours of the crude products indicated that demethylation had also occurred.

The O-carbethoxyl grouping appeared to be stable to the reaction conditions. However, attempts to prepare (136, R=-$CO_2C_2H_5$) gave only mixtures of partially substituted material and the starting dihydroxy acid.

Diacetyldichloro-p-orsellinic acid (136, R=Ac) was



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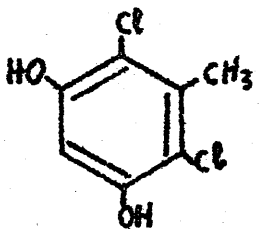

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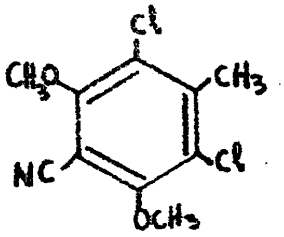
prepared from (136, R=H) by means of acetic anhydride in pyridine. Condensations with (137, R=$CO_2C_2H_5$) under various conditions were attempted but in no case was any evidence of the presence of the required benzophenone obtained. In all cases (136, R=Ac) was isolated and the presence of (137, R=$CO_2C_2H_5$) was identified spectroscopically. In one case after an alkaline work up (136, R=H) and (138) were isolated.

The Shah procedure is particularly applicable to the preparation of hydroxy benzophenones and xanthones. However attempts to condense (136, R=H) with (137, R=$CO_2C_2H_5$) and methyl 3,5-dihydroxybenzoate resulted only in quantities of yellow froth which did not yield any crystalline products. Alkaline treatment of the product from the first of these experiments yielded 2,6-dichlororcorcinol (139) as the only crystalline material.

Failure to achieve the condensation is not readily accounted for in view of Raistrick's singular success. The successful case, however, did allow for a mixture of products being obtained due to demethylation, this being overcome by subsequent methylation. In the present case the extremely vigorous conditions of Raistrick would undoubtably be detrimental to the required product. The use of the unprotected acid (136, R=H) is not suitable due to the difficulty of forming acid chlorides of the salicylic acid type[95], while use of the methyl and acetyl derivatives is unsatisfactory because of their sensitivity to acid conditions. The unsymmetrical substitution of (137) adds to the problems by offering alternative



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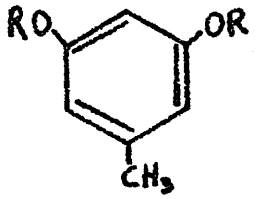

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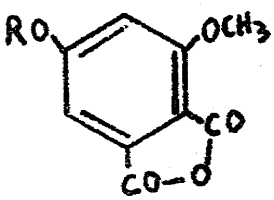

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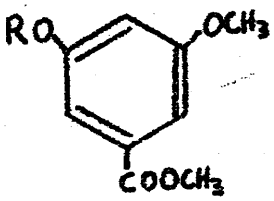

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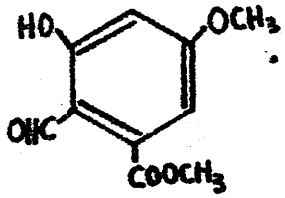

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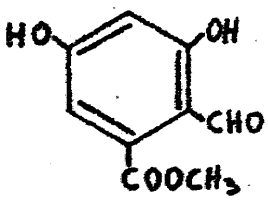

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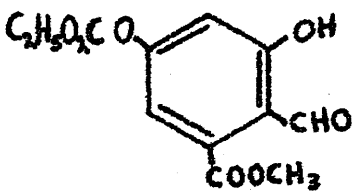


145

sites for the condensation.

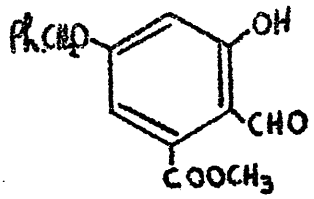
Calam and Oxford[96] were unsuccessful in condensing the nitrile (140) with orcinol (141, R=H) and its dimethyl ether (141, R=CH_3) under the milder conditions of a Hoesch reaction.

In the hope of circumventing some of these difficulties it was decided to attempt the alternative mode of condensation by using the chlorine free ring as the acylating agent in the form of an anhydride of the type (142) and condensing it with 2,6-dichloroorcinol (139). The chlorine containing ring contains only one site at which the acylation can occur.

In general the more hindered carbonyl of the anhydride is the one which is utilised in formation of a benzophenone[97,98].

A Gatterman reaction on (137, R=H) afforded a compound which was identified as (143) by the deep red ferric chloride colour obtained due to the ortho hydroxy aldehyde structure. It had infrared maxima at 1730 ($COOCH_3$) and 1665 (bonded CHO) $cm.^{-1}$ Methyl 2-formyl-3,5-dihydroxybenzoate (144) was prepared according to the procedure of Birkinshaw and Bracken[99]. Attempts to prepare 3,5-dihydroxyphthalic acid from (144) by fusion[99] with caustic potash (90%) were unsuccessful.

A route to (142) had to be devised. As in the synthesis of picrolichenic acid it was considered necessary to protect the phenolic hydroxyls of (144) before oxidation of the formyl group. O-carboethoxylation furnished the 5-ethoxycarbonyloxy compound



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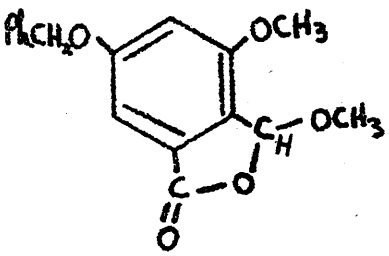

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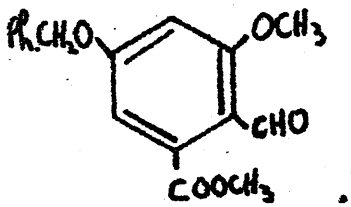

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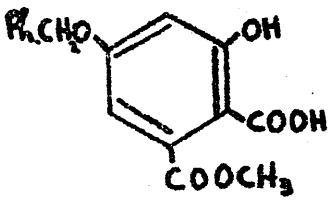

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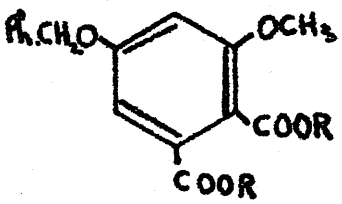

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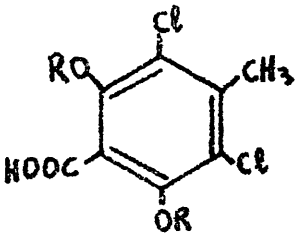
(145) ν max. 1770 ($-OCO_2C_2H_5$), 1740 ($COOH_3$) and 1660 (bonded CHO)cm.$^{-1}$ All attempts to methylate this compound resulted in hydrolysis of the O-carbethoxyl group and subsequent methylation of the liberated phenol. Benzylation of (144) with benzyl chloride in alcoholic potassium hydroxide solution afforded the 5-benzyl derivative (146) which gave a red ferric chloride colour and had ν max. 1740 ($-COOCH_3$) and 1660 (bonded CHO) cm.$^{-1}$ Methylation of (146) with dimethyl sulphate and potassium carbonate gave after chromatography on silica gel two products. The major product analysed correctly for the required formyl methyl ether but it did not afford a 2,4-dinitrophenylhydrazone. The infrared spectrum contained only one maximum at 1760 cm.$^{-1}$ instead of the expected νmax. at 1730 ($-COOCH_3$) and 1680 ($-CHO$) cm.$^{-1}$ Pseudo esters absorb[100] at 1760-1770 cm.$^{-1}$ and can be obtained under these conditions. Thus the product isolated is most probably the pseudo ester (147). The other product, obtained in small yield, had infrared absorption maxima at 1730 and 1680 cm.$^{-1}$ and is probably the normal methyl ester (148).

The hydroxy formyl compound (146) was oxidised with neutral potassium permanganate to the hydroxy acid (149), νmax. 1730 ($COOCH_3$) and 1650 (bonded COOH) cm.$^{-1}$ which gave a red ferric chloride colour. Methylation of (149) afforded the methyl ether diester (150, R=CH_3) ν max. 1720 ($-COOCH_3$) cm.$^{-1}$ which was readily hydrolysed to the diacid (150, R=H) νmax. 1690 (COOH) and 1670



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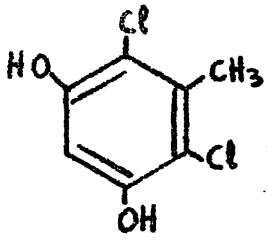

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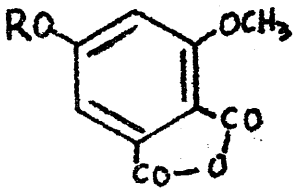

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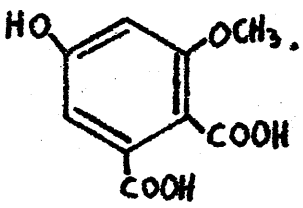

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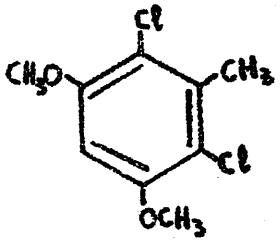

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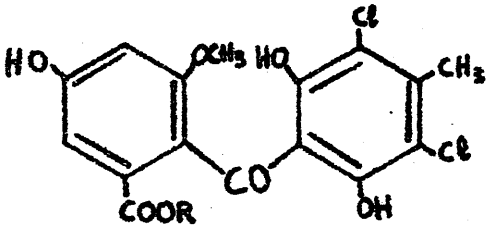


130

(COOH ortho to methoxyl) cm.$^{-1}$ The anhydride (142, R=PhCH$_2$-) was obtained by short treatment of the diacid with acetic anhydride. The anhydride had infrared maxima at 1830 and 1765 (anhydride) cm.$^{-1}$

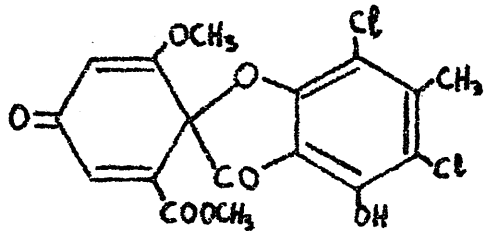
2,6-Dichlororcorcinol (139) was obtained from (136, R=H) by acid catalysed decarboxylation.

Attempts to condense the anhydride (142, R=PhCH$_2$-) with dichlororcorcinol (139) under a variety of conditions provided no evidence of benzophenone formation. In all cases dichlororcorcinol was recovered. The benzyl group appeared to be very susceptible to the Friedel and Craft conditions and no benzyl containing products wereisolated. However the hydroxy diacid (151) was obtained in every case.

5-Hydroxy-3-methoxyphthalic anhydride (142, R=H) was obtained by hydrogenation of (142, R=PhCH$_2$-) over palladium charcoal. Attempts to condense (142, R=H) with dichlororcorcinol were also unsuccessful.

Calam and Oxford[96] were similarly unsuccessful when they tried to condense the anhydride (142, R=CH$_3$) with (152). The 2,6-substitution in the dichlororcorcinol component must have a considerable influence on the position between them and a severe steric effect must arise from the highly substituted nature of the reactants.

Dihydrogeodin (130, R=CH$_3$) and dihydroerdin (130, R=H) are readily obtained from the naturally occurring (+) geodin and (±) erdin by catalytic hydrogenation[40]. Therefore it was decided



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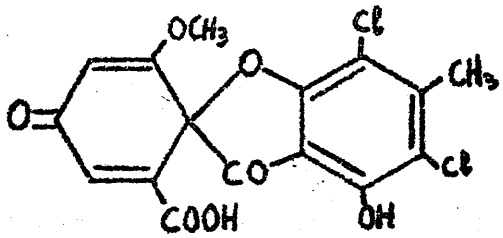

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to simulate the oxidative stage of the biogenisis by oxidation of the benzophenones obtained from the naturally occurring material.

Dihydrogeodin was oxidised with potassium ferricyanide by the "usnic acid" procedure[13]. Acidification and isolation in ether furnished a froth which crystallised to give (±) geodin (30) in 25% yield, identical in melting point, mixed melting point (225-227°), ultraviolet (λ max. 284 mμ(ε 17,200)) and infrared spectra (ν max. 3390, 1723, 1660, 1625 and 1607 cm.$^{-1}$) with (±) geodin obtained by racemisation[43] of (+) geodin.

Dihydroerdin was treated in the same manner and on crystallisation from chloroform (±) erdin (128), identical in melting point, mixed melting point (210-212°), ultraviolet (λ max. 283 mμ(ε 20,000)) and infrared spectra (ν max., 3350, 1720, 1660 and 1608 cm.$^{-1}$) with natural racemic erdin wasisolated in 16% yield.

The postulated biogenesis of geodin and erdin is thus supported by these partial syntheses.

EXPERIMENTAL

3-Hydroxy-5-methoxybenzoic acid (138).

This compound was prepared as described by Hauthner[101].

Methyl 3-hydroxy-5-methoxy benzoate (137, R=H).

The acid (138) (5 g.) was esterified with methanol (30 c.c.) and conc. sulphuric acid (2 c.c.) in the usual manner. The neutral extract furnished white needles (3.5 g.) m.p. 94-95° (from water). ν max. (Nujol) 3400 (free OH) and 1700 (CO_2CH_3) $cm.^{-1}$ (Found: C, 59.1; H, 5.1; OCH_3, 33.75. $C_9H_{10}O_4$ requires C, 59.35; H, 5.55; OCH_3, 34.05%).

Methyl 3-ethoxycarbonyloxy-5-methoxybenzoate (137, R=$CO_2C_2H_5$).

The ester (137, R=H) (2 g.) was dissolved in pyridine (10 c.c.) and cooled to -20°. Ethyl chloroformate (3.2 c.c.) was added gradually and the solution was kept at room temperature overnight (silica gel guard tube). The solution was acidified with hydrochloric acid (6N) at 0°. The ether extract of the acid solution gave an oil which on distillation gave the required O-carbethoxy compound (1.7 g.) b.p. 124°-125° (.08 mm.) $R.I^{25°}$ 1.5087. ν max. (film) 1765 (-O-$CO_2C_2H_5$) and 1725 (CO_2CH_3) $cm.^{-1}$ (Found: C, 56.95; H, 5.15. $C_{12}H_{14}O_6$ requires C, 56.7; H, 5.55%).

p-Orsellinic acid (136, R, Cl=H).

This compound was prepared as described by Robertson and

Robinson[102]. A better product was obtained by recrystallising from aqueous acetic acid.

Dichloro-p-orsellinic acid (136, R=H).

Standard solution of chlorine in acetic acid.

Chlorine was bubbled into acetic acid until the weight increase was approximately sufficient. Thesolution was standardised by titrating the iodine liberated from potassium iodide by 1 c.c. of the chlorine-acetic acid solution against 0.1N sodium thiosulphate solution using starch indicator.

p-Orsellinic acid (6 g., 1 m.) was dissolved in acetic acid (150 c.c.) and the calculated amount of chlorine-acetic acid solution (2.2 m.) was added. The solution was stood at room temperature overnight. After reducing the volume of acetic acid to 50 c.c. the cooled solution afforded the chlorinated acid as needles (6.1 g.) m.p. 215-216°. ν max. (Nujol) 3500 (free OH), 3300 (bonded OH) and 1680 (COOH) $cm.^{-1}$ It gave a blue ferric colour. (Found: C, 40.55; H, 2.8; Cl, 29.65. $C_8H_6O_4Cl_2$ requires C, 40.55; H, 2.55; Cl, 29.9%).

Methyl dichlorodimethoxy-p-orsellinate.

Dichloro p-orsellinic acid (1 g.), dimethyl sulphate (2.5 g.) and potassium carbonate were stirred and refluxed overnight in dry acetone (K_2CO_3). The cooled solution was filtered and, after removal of the acetone in vacuo, water (100 c.c.) was added.

Filtration of the precipitated needles afforded the required methylated compound as needles (1.1 g.) m.p. 85-86°. (lit.[96] 87-88° (from aqueous ethanol)).

Dichlorodimethoxy-p-orsellinic acid (136, R=CH_3).

Methyl dichlorodimethoxy-p-orsellinate (1 g.) was refluxed with methanol (10 c.c.) and 0.5N sodium hydroxide (15 c.c.) for 3 hr. The acidified solution, after removal of methanol in vacuo, gave the acid as needles (.8 g.) m.p. 120-121° (from water). (lit.[96] m.p. 121-122°).

Diacetyldichloro-p-orsellinic acid (136, R=Ac).

Dichloro-p-orsellinic acid (1 g.) and acetic anhydride (2 c.c.) were dissolved in pyridine (10 c.c.) and heated on a steam bath for 1.5 hr. The pyridine was neutralised at 0° with hydrochloric acid (6N.). An ether extract of the acid solution gave a solid which had a positive reaction with alcoholic ferric chloride solution and ν max. at 3500 (-OH), 1745 (O-acetate) and 1660 (bonded CO_2H) $cm.^{-1}$ Thesolid was retreated with acetic anhydride and pyridine as before. The solid now isolated had a negative ferric reaction and recrystallised from aqueous ethanol as needles (.8 g.) m.p. 150°. ν max. (Nujol), 1776 (O-acetate) and 1700 (CO_2H) $cm.^{-1}$ (Found: C, 45.05; H, 3.15. $C_{12}H_{10}Cl_2O_6$ requires C, 44.85; H, 3.15%).

Methyl 2-formyl-3-hydroxy-5-methoxybenzoate (143).

Dry hydrogen chloride was passed rapidly through a stirred solution of methyl-3-hydroxy-5-methoxybenzoate (2.5 g.) in anhydrous ether (25 c.c.) containing zinc cyanide (3.7 g.) in suspension. The reaction was maintained at 5°. After saturation with hydrogen chloride (2.5 hr.) the passage of gas was discontinued. After decantation of the supernatant ether and washing twice with anhydrous ether the residual syrup was refluxed with water (50 c.c.) for 30 mins. The cooled solution was filtered and the solid precipitate was recrystallised from water to give white needles (1.3 g.) m.p. 85-86°; ν max. (Nujol) 1720 (CO_2H_3) and 1660 (bonded CHO) cm.$^{-1}$ It gave a red ferric colour. (Found: C, 57.05; H, 5.0. $C_{10}H_{10}O_5$ requires C, 57.15; H, 4.8%).

Methyl 3,5-dihydroxy-2-formylbenzoate (144).

This compound was prepared as described by Birkinshaw and Bracken[99].

Methyl 5-ethoxycarbonyloxy-2-formyl-3-hydroxybenzoate (145).

The formyl ester (144) (2 g.) was dissolved in pyridine (10 c.c.) and cooled to -20°. Ethyl chloroformate (610 mg.) was added gradually and the solution was kept at room temperature overnight. The ether extract of the acidified solution furnished a semi-solid which was dissolved in a benzene-ether (6:1) solution. Chromatography on silica gel (60 g.) and elution with benzene -ether (4:1) (200 c.c.) afforded the required O-carbethoxy compound

as needles (610 mg.) m.p. 68-69° (from aqueous ethanol), ν max. (Nujol) 1770 (-O-$CO_2C_2H_5$), 1740 (CO_2CH_3) and 1660 (bonded CHO) cm.$^{-1}$ It gave a red ferric colour. (Found: C, 54.5; H, 4.6. $C_{12}H_{12}O_7$ requires C, 53.75; H, 4.5%).

Methyl 5-benzyloxy-2-formyl-3-hydroxybenzoate (146).

The formyl ester (144) (11 g.) and benzyl chloride (10 c.c.) were heated under reflux for 4 hr. in aqueous ethanol (25%; 100 c.c.) containing potassium hydroxide (3.4 g.). Filtration of the cooled solution gave a solid which had a red ferric colour. m.p. 117-118° (from ethanol). Removal of the ethanol from the filtrate in vacuo followed by acidification and recovery in ether gave an oil. Digestion of the oil with benzene gave a residue which wasthe starting dihydroxy compound (144). Chromatography of the benzene soluble material on silica gel (200 g.) and elution with benzene afforded more of the required monobenzyl compound, m.p. 117-118°; ν max. (Nujol) 1740 (CO_2CH_3) and 1660 (bonded CHO) cm.$^{-1}$ Total yield was 6.5 g. (Found: C, 67.15; H, 5.25. $C_{16}H_{14}O_5$ requires C, 67.1; H, 4.95%).

5-Benzyloxy-2-formyl-3-methoxybenzoic acid pseudo methyl ester. (147).

The benzyl compound (146) (1 g.) was stirred and refluxed overnight with dimethyl sulphate (.4 c.c.) and potassium carbonate (3 g.) in acetone (30 c.c.). The solution became bright yellow. After filtration and removal of the acetone in vacuo a gum with a negative ferric test was obtained. Chromatography on silica

gel (30 g.) and elution with benzene-ether (19:1) (100 c.c.) afforded a solid which recrystallised from light petroleum as needles (250 mg.), m.p. 110-111°. ν max. (Nujol) 1760 (pseudo ester) cm.$^{-1}$ (Found: C, 69.95; H, 5.7. $C_{17}H_{16}O_5$ requires C, 70.0; H, 5.35%). The compound is probably the pseudo ester methyl ether of (146).

Further elution with benzene-ether (19:1) gave gums with infrared maxima at 1760, 1730 and 1670 cm.$^{-1}$ Successive fractions indicated a decrease in intensity of the 1760 cm.$^{-1}$ band until after 75 c.c. a fraction was obtained which afforded a small quantity of crystalline material m.p. 160-167° (from benzene-ether); ν max. (Nujol) 1730 (CO_2CH_3) and 1670 (CHO) cm.$^{-1}$ which is probably the normal ester methyl ether of (146).

Methyl 5-benzyloxy-2-carboxy-3-hydroxybenzoate (149).

To a stirred solution of methyl 5-benzyloxy-2-formyl-3-hydroxybenzoate (4 g.) in acetone (1½) was added potassium permanganate (12 g.) in water (200 c.c.) during 4 hr. at 45°, then the mixture was stirred (at this temperature) for 1 hr. Sulphur dioxide was passed through the cooled solution and the acetone removed. The ether soluble material was extracted with saturated sodium bicarbonate solution. The ether fraction gave the starting formyl compound (1.2 g.) and the acidic fraction afforded the required acid (580 mg.) as prisms m.p. 168° (from ethyl acetate-light petroleum). ν max. (Nujol) 1730 (CO_2CH_3)

and 1650 (bonded CO_2H). It gave a red ferric colour. (Found: C, 63.55; H, 4.85. $C_{16}H_{14}O_6$ requires C, 63.55; H, 4.65%).

Dimethyl 5-benzyloxy-3-methoxyphthalate (150, R=CH_3).

The hydroxy acid (149) (1 g.) was stirred and refluxed overnight with dimethyl sulphate (.68 c.c.) and potassium carbonate (8 g.) in acetone (100 c.c.). The fully methylated product was isolated in the usual manner as prisms (915 mg.) m.p. 94-95° (from ethyl acetate - light petroleum); ν max. (Nujol) 1720 (CO_2CH_3) cm.$^{-1}$ (Found: C, 65.75; H, 5.55. $C_{18}H_{18}O_6$ requires C, 65.45; H, 5.5%).

5-Benzyloxy-3-methoxyphthalic acid (150, R=H).

The diester (150, R=CH_3) (915 mg.) was hydrolysed by methanol-4N sodium hydroxide solution (20 c.c.; 1:1) for 6 hr. After removal of the methanol *in vacuo* and acidification the required diacid was isolated in ether. Recrystallisation from ethyl acetate gave needles (750 mg.) m.p. 143-145°. ν max. (Nujol) 1690 (free CO_2H) and 1670 (CO_2H ortho to methoxyl) cm.$^{-1}$ (Found: C, 63.3; H, 4.9. $C_{16}H_{14}O_6$ requires C, 63.55; H, 4.65%).

5-Benzyloxy-3-methoxyphthalic anhydride (142, R=$PhCH_2$-).

The diacid (150, R=H) (1 g.) was refluxed in acetic anhydride (10 c.c.) for 30 mins. Ether (25 c.c.) was added to the cooled solution and the precipitated solid filtered to give the required anhydride as prisms (720 mg.) m.p. 185-186°; ν max.

(Nujol) 1830 and 1765 (anhydride) cm.$^{-1}$ (Found. C, 67.3; H, 4.15. $C_{16}H_{12}O_5$ requires C, 67.6; H, 4.25%).

2,6-Dichlororcorcinol (139).

Dichloro-p-orsellinic acid (2 g.) in 24 N-sulphuric acid (100 c.c.) was heated at 155° for 15 mins. The cooled solution was diluted with water (100 c.c.) and extracted with ether. After washing with bicarbonate and removal of the ether *in vacuo* a grey solid (1.3 g.) was obtained which, after sublimation (85°; .01 mm.) furnished the required decarboxylated compound (1 g.) as feathery needles. m.p. 165-166°. (lit.[41] m.p. 164°).

5-Hydroxy-3-methoxyphthalic acid (151).

5-Benzyloxy-3-methoxyphthalic acid (125 mg.) was hydrogenated over palladium charcoal (10%) in ethyl acetate (10 c.c.) until one mole of hydrogen had been absorbed. Filtration and evaporation of the ethyl acetate afforded the required hydroxy diacid (50 mg.) as prisms. m.p. 242-245° (sublimes at 185°); νmax. 3300 (free OH), and 1690 (CO_2H) cm.$^{-1}$

5-Acetoxy-3-methoxyphthalic anhydride (142, R=Ac).

The hydroxy diacid (151) (23 mg.) in acetic anhydride (.5 c.c.) was refluxed for 20 mins. Ether (3 c.c.) was added to the cooled solution and the precipitated solid filtered to give the required product (18 mg.) as prisms. m.p. 160-161°; νmax. (Nujol) 1840 and 1765 (anhydride) and 1785 (O-acetate) cm.$^{-1}$ (Found: C, 56.6; H, 3.85. $C_{11}H_8O_6$ requires C, 55.95; H, 3.4%).

5-Hydroxy-3-methoxyphthalic anhydride (142, R=H).

5-Benzyloxy-3-methoxyphthalic anhydride (60 mg.) was hydrogenated over palladium charcoal (10%) in ethyl acetate until one mole of hydrogen had been absorbed. Filtration and evaporation of the ethyl acetate *in vacuo* furnished the required hydroxy anhydride (35 mg.) as prisms. m.p. 245-247°. ν max. (Nujol). 3400 (free OH), 1840 and 1760 (anhydride) $cm.^{-1}$ (Found: C, 55.2; H, 3.7. $C_9H_6O_5$ requires C, 55.7; H, 3.1%).

Attempted condensation of the acid (136, R=H, CH_3, Ac) with (137, R=CH_3, $CO_2C_2H_5$).

General method: Equimolar amounts of the two reactants were heated with freshly fused zinc chloride (4 moles) and phosphorous oxychloride (8 moles) at 75° for 2 hr. The reaction mixtures were added to conc. hydrochloric acid and crushed ice. The ether extract of the acid solution was washed with saturated sodium bicarbonate solution and the ether was removed *in vacuo*. The residues were chromatographed on silica gel and the products identified by melting points and infrared spectra. The results of these experiments are shown in table 2.

A similar reaction was attempted between methyl 3,5-dihydroxybenzoate and dichloro-*p*-orsellinic acid. The products were not identified.

Table 2.

Reactants	Compounds Identified
(136, R=CH_3); (137, R=$CO_2C_2H_5$)	(137, R=$CO_2C_2H_5$)
(136, R=Ac); (137, R=$CO_2C_2H_5$)	(136, R=Ac); (137, R=$CO_2C_2H_5$).
(136, R=H); (137, R=$CO_2C_2H_5$)	(139); (137, R=$CO_2C_2H_5$)
(136, R=CH_3); (137, R=CH_3).	(137, R=CH_3)

Attempted condensation of (136, R=H) with (137, R=CH_3).

(a) The acid (136, R=H) (3 g.) and the ester (137, R=CH_3) (3 g.) were refluxed with boron trifluoride-ether complex (2.5 c.c) in ether (15 c.c.) for 24 hr. Addition of dilute hydrochloric acid and isolation of the products in ether afforded the starting materials only.

(b) The acid (136, R=H) (3 g.) and ester (137, R=CH_3) were stirred vigorously in tetrachloroethane (30 c.c.) while boron trifluoride was passed through the solution for 10 mins. at 50°. The reaction was stirred for a further 30 mins. and then it was poured into water (100 c.c.) containing sodium acetate (14 g.). After stirring for 45 mins. the solution was extracted with ether and the ether was washed with bicarbonate and alkali. The bicarbonate extract afforded 3,5-dimethoxy-benzoic acid (380 mg.)

m.p. 182°; ν max. 1680 cm.$^{-1}$ and dichloro-p-orsellinic acid (50 mg.) No evidence for the presence of a benzophenone wasobtained.

Attempted condensation of the acid chloride of (136, R=CH_3, Ac) with (137, R=$CO_2C_2H_5$).

General Method: The acid in dry benzene was treated with oxalyl chloride at room temperature overnight, protected by a silica gel drying tube. The benzene and excess oxalyl chloride were removed in vacuo. The other reactant and the appropriate catalyst were added to the acid chloride in a solvent. After the treatment specified in table 3 the reaction mixtures were added to ice and conc. hydrochloric acid and extracted with ether. The residues obtained were chromatographed on silica gel and the products identified by melting point and infrared spectra.

Table 3.

Reactants	Conditions	Compounds Identified
(137, R=$CO_2C_2H_5$); (136, R=CH_3).	Aluminium chloride, carbon disulphide 0°-20°, 24 hr.	(137, R=$CO_2C_2H_5$)
	Aluminium chloride, no solvent. 10 mins. at 100° then 2 days at 20°.	(137, R=$CO_2C_2H_5$).
(137, R=$CO_2C_2H_5$); (136, R=Ac).	Aluminium chloride, nitrobenzene, 15 mins. at 100° then 2 days at 20°.	(137, R=$CO_2C_2H_5$); (136, R=Ac).
	Aluminium chloride, nitrobenzene 3 days at R.T. (Product treated with N-NaOH, 2 hrs.)	(136, R=H); (138).

Attempted condensation of the anhydride (142, R=$PhCH_2$-,H) with 2,6-dichlororcorcinol (139).

General method: 2,6-Dichlororcorcinol (139) (1 mole) was added to the anhydride (1 mole) and catalyst (3 moles) in the appropriate solvent. After the treatment specified in table 4, the reaction mixtures were added to ice and conc. hydrochloric acid. The ethyl acetate extract of the acid solution was washed with 4N-caustic soda. Acidification of the alkali extract and isolation in ethyl acetate afforded the products which were generally separated by solvents or chromatography on silica gel. The products were identified by melting point and infrared spectra.

Table 4

Reactants	Conditions	Compounds Identified
(142, R=$PhCH_2$); (139)	Aluminium chloride, benzene, 16 hr. at 20° then 8 hr. at 60°.	(139); (151)
	Aluminium chloride, methylene chloride, 2 hr. at 20°.	(142, R=H); (139)
	Aluminium chloride, nitrobenzene, 6 days at 20°.	(139); (151)
	Aluminium chloride, nitrobenzene, 4.5 hr. at 70°.	(139); (151)
(142, R=H); (139)	Aluminium chloride, nitrobenzene, 7 days at 20°.	(139); (151)
	Stannic chloride, nitrobenzene, 7 days at 20°.	(139); (151)

Dihydrogeodin (130, R=CH_3).

This compound was prepared as described by Raistrick and Smith[37].

(±) Geodin (30).

Dihydrogeodin (250 mg.) and sodium carbonate (700 mg.) were dissolved in distilled water (15 c.c.) and cooled to 5°. Nitrogen was passed through the stirred solution as potassium ferricyanide (440 mg.) in distilled water (10 c.c.) was added dropwise during 10 mins. The stirring was continued for a further 10 mins. The solution was acidified with conc. hydrochloric acid and isolation in ether gave a froth which on trituration with ether afforded a solid residue m.p. 220-225° (from chloroform-ether); ν max. (Nujol) 3400, 1715, 1650 and 1620 $cm.^{-1}$ Chromatography of some of the froth (90 mg.) on silica gel (3 g.) and elution with benzene-ether (19:1) (80 c.c.) furnished more of the required product (20 mg.) m.p. 225-227° (from chloroform-ether) which was identical in m.p., mixed m.p. and ultraviolet and infrared absorption with authentic racemic geodin; λ max. (in ethanol) 284 mμ. (ϵ 17,000); ν max. (KCl disc.), 3390 (OH), 1723 (CO_2CH_3), 1660 (dienone C=O) and 1625 (coumaranone C=O) $cm.^{-1}$

Dihydroerdin (130, R=H).

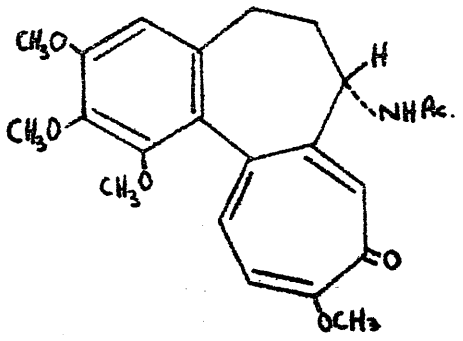
This compound was prepared as described by Raistrick and Smith[37].

(±) Erdin (128).

Dihydroerdin (50 mg.) and sodium carbonate (200 mg.) were dissolved in distilled water (10 c.c.) and cooled to 5°. Nitrogen was bubbled through the stirred solution as potassium ferricyanide (400 mg.) in distilled water (15 c.c.) was added dropwise during 5 mins. The stirring wascontinued for a further 15 mins. The solution was acidified with conc. hydrochloric acid and isolation in ether gave a froth with a crude infrared spectrum in chloroform at 1705, 1650 and 1620 $cm.^{-1}$ Trituration of the froth afforded a solid which on recrystallisation from chloroform furnished the required product (8 mg.) m.p. 209-212° which was identical in m.p. mixed m.p. and ultraviolet and infrared absorption with natural racemic erdin; λ max. (in ethanol) 282 (ε 20,000), 353 (ε, 360)$_{\text{inflex.}}$; ν max. (KCl disc.), 3350 (OH), 1720 (CO_2H), 1660 (dienone C=O), 1608 (coumaranone C=O) $cm.^{-1}$

PART III

SYNTHETIC APPROACHES TO COLCHICINE



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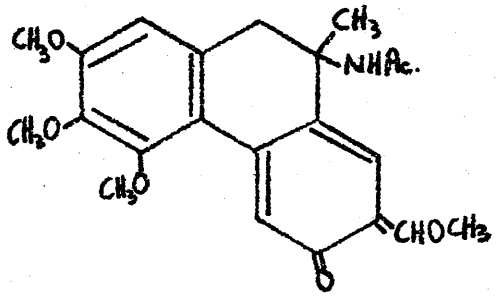

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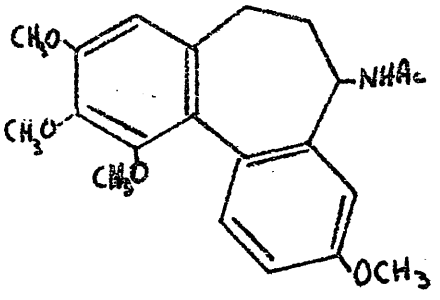
HISTORICAL.

The main source of the alkaloid colchicine (24), whence it derives its name, is Colchicum autmnale. Although it is extremely toxic by virtue of its paralysing action on the central nervous system it has been a focus of biological investigation for many years on account of its tumour inhibiting effect. This is brought about by the alkaloids property of arresting cell division in the early metaphase. (For reviewson colchicine see references 103, 104 and 105).

Crystalline colchicine was first obtained by Zeisel[106] and by Houdes[107] but this was a solvated form containing chloroform of crystallisation. Pure crystalline colchicine, free from solvent, was first described by Clewer, Green and Tutin[108]. Colchicine has a molecular formula $C_{22}H_{25}O_6N$ and Windaus[109] proposed the dihydrophenanthrene structure (153). The evidence for the presence of the phenanthrene ring system in colchicine was based on the isolation of 9-methylphenanthrene by treatment of colchinol methyl ether with zinc dust. The salicylaldehyde enol ether structure in ring C was proposed to explain the ready hydrolysis of one methoxyl group in colchicine to give methanol and colchiceine, which, unlike colchicine gave an intense ferric chloride reaction. Remethylation of colchiceine with diazomethane[110] gave colchicine and an isomer isocolchicine which Windaus



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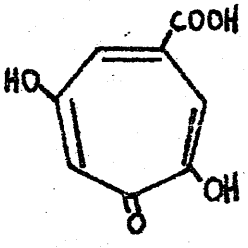

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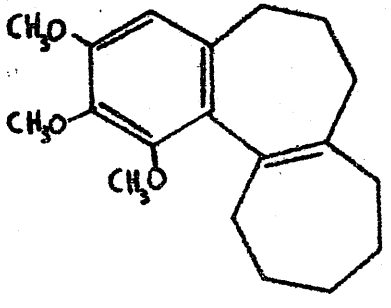

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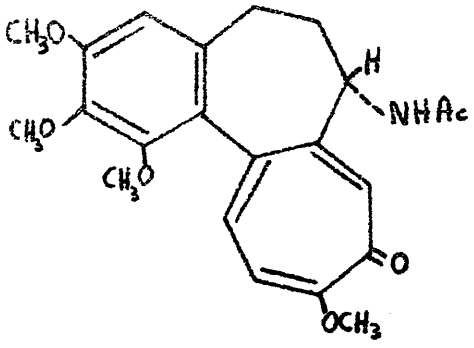
regarded as cis and trans isomers about the hydroxymethylene double bond.

The ring A substitution pattern was established by the identification of 3,4,5-trimethoxyphthalic acid as an oxidation product of colchicine[111]. A seven membered ring structure was proposed[112] for ring B and this was shown to be correct by the synthesis of N-acetylcolchinol methyl ether (154) by two independant routes. The first route produced the naturally derived optically active form[113] and the second the racemic modification[114]. The position of the nitrogen-containing substituent was thus located in this ring.

Dewar's inspired suggestion of tropolone as a new aromatic system in order to explain the puzzling properties of stipitatic acid[115] (155) was later extended to cover ring C of colchicine[116]. The tropolone sturcture immediately gave a satisfactory solution to the problem of the facile rearrangement of colchicine to benzene derivatives. The ferric colour given by colchiceine was readily explained and also the existence of the isomeric compound, isocolchicine.

The tropolone structure, of course, implied that ring C was also a seven membered ring.

The isolation of (156) from the reduction of a colchicine derivative by Rapoport et al.[117] and its synthesis by Löwenthal[118] furnished final proof of the carbon skeleton of colchicine.



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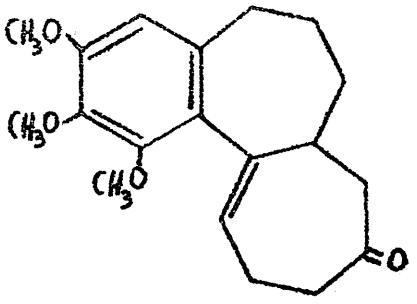

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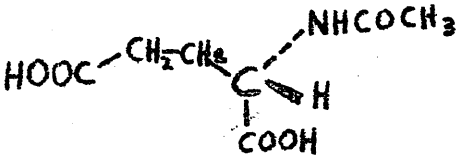

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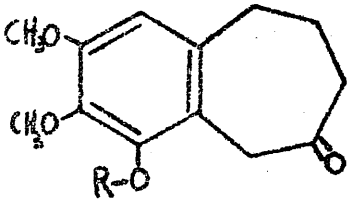

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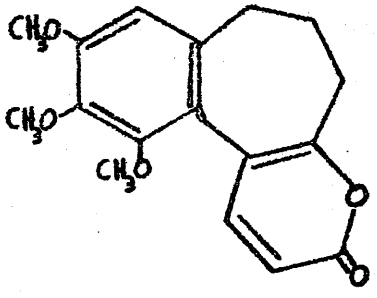

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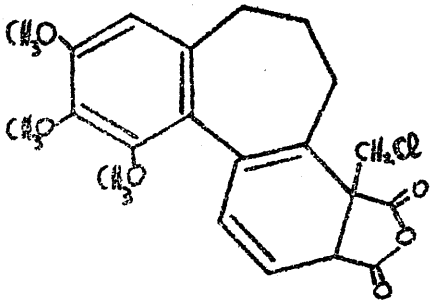

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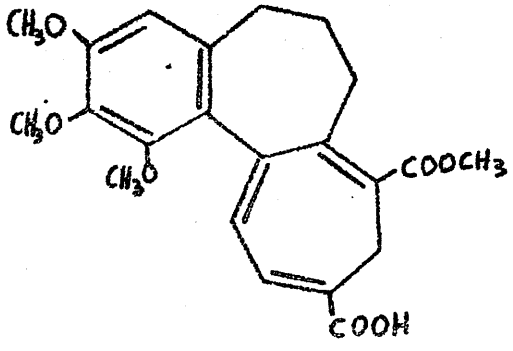

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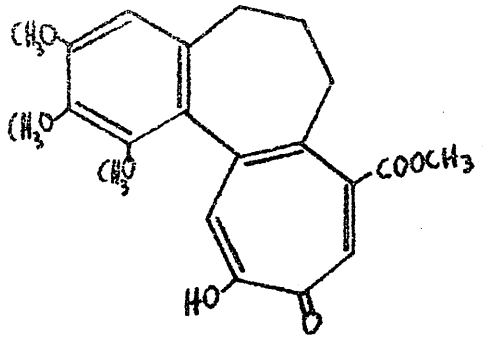
A puzzling feature only recently resolved concerns the relative positions of the methoxyl and carbonyl groups in ring C of colchicine. Several proposals based on mechanistic[119, 120] and spectroscopic[121] evidence were forwarded which all suggested that the formulation (24) was the true representation of colchicine and that isocolchicine was the corresponding compound with the methoxyl and carbonyl positions interchanged. Confirmation of this was supplied by the X-Ray measurements of Pepinsky et al.[122] Muller and Velluz[123], however, proposed that the positions of the methoxyl and carbonyl groups were the reverse of that shown in (24). Recent reinterpretation of their results by Forbes[124] is in full support of structure (24). Final chemical proof was provided by the unambiguous synthesis[125] of (157) a degradation product[126] of colchicine which retained the tropolone carbonyl.

The absolute configuration of colchicine was established by Corrodi and Hardegger[127]. The acetylamido substituent was shown to be α to ring B by degradation of colchicine to N-acetyl-L-glutamic acid (158), and thus the absolute configuration is represented by expression (24).

Many approaches[128] to the synthesis of colchicine culminated in the successful syntheses carried out independantly by three groups of workers.

Eschenmoser et al.[129] started with the β-benzosuberone (159, R=H) and converted it by addition of propiolic ester, an



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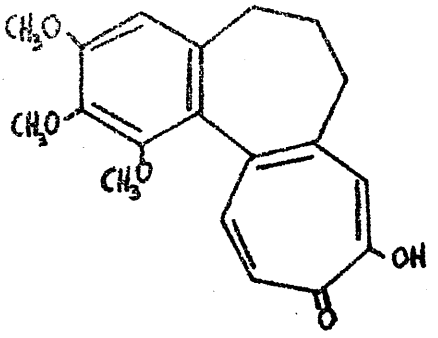

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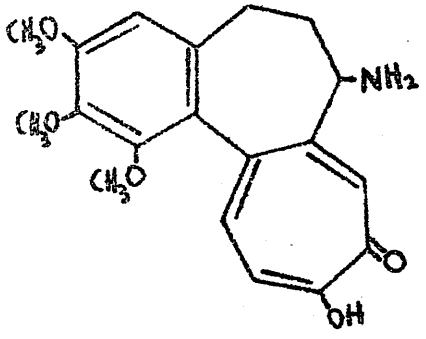

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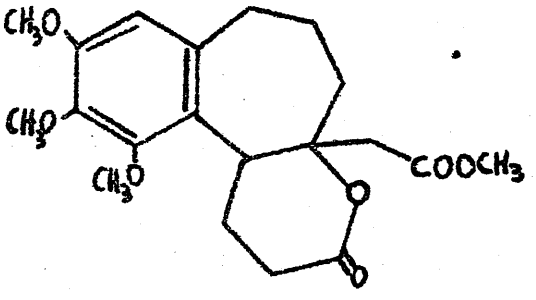

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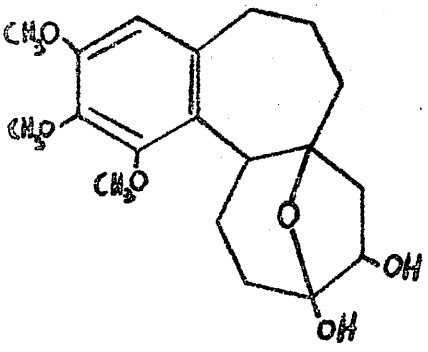

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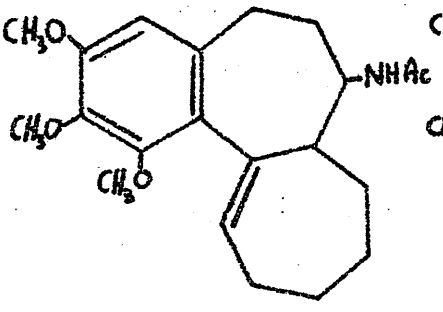

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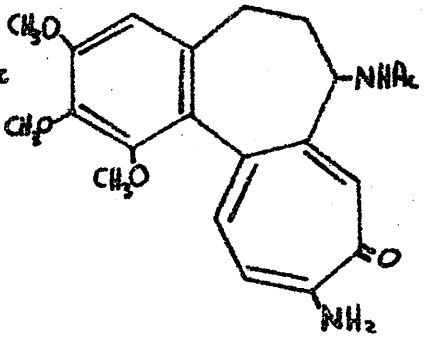


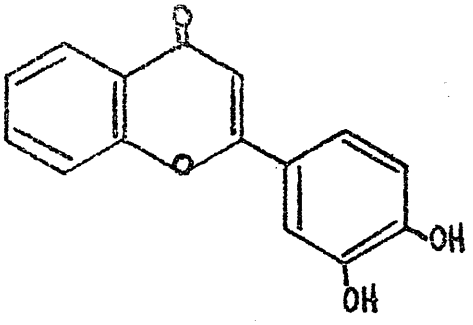
169

internal Michael reaction and lactonisation to the unsaturated lactone (160) which was condensed with chloromethylmaleic anhydride to give (161). Ring expansion of the diester derived from (161) afforded the 6,7,7 ring system (162) which by treatment with osmium tetroxide and sodium bicarbonate gave the tropolone (163). Desacetylamidocolchiceine (164) was obtained in several steps from (163) and was subsequently converted *via* a bromo derivative to (±) desacetycolchiceine (165). (-) Colchicine (24) had previously been prepared from (165) by Corrodi and Hardegger[130], thus completing the synthesis by the Swiss group.

Van Tamelen *et al.*[131] in the United States began with the β-benzosuberone (159, R=CH_3). Addition of acrylonitrile and a Reformatsky reaction with bromoacetic ester followed by lactonisation afforded the lactone (166) which underwent a novel acyloin condensation to the tricyclic compound (167). Cupric acetate treatment of (167) and subsequent dehydration and aromatisation furnished desacetylamidocolchiceine (164) which was converted to (±) desacetylcolchiceine (165).

Nakamura[132] in Japan prepared colchiceineamide (169), which had already been converted to colchicine (24)[121], from (168). Later a successful multistage synthesis[133] of (168) was achieved thus completing a third total synthesis of colchicine.

Several proposals for the biogenesis of colchicine have been made[24-27].



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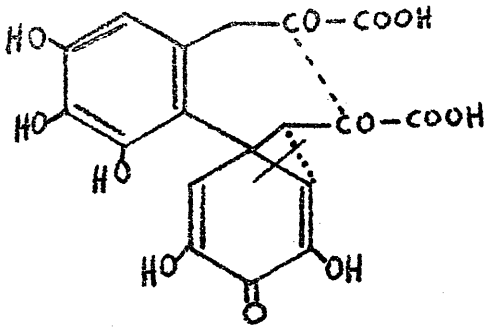

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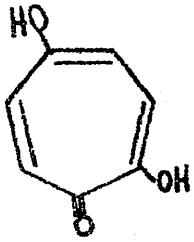

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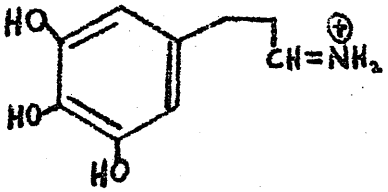

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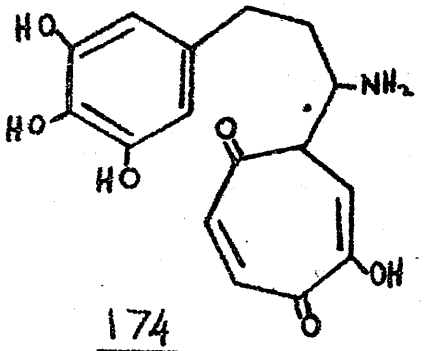

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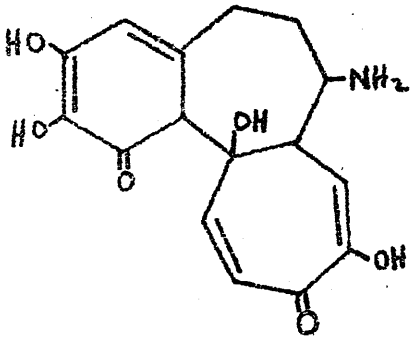

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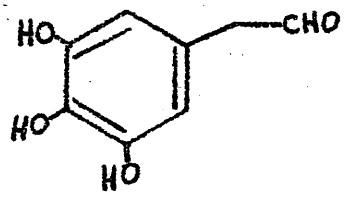

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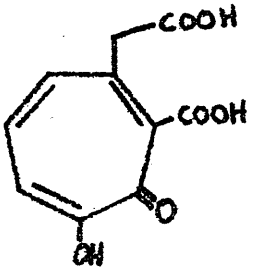

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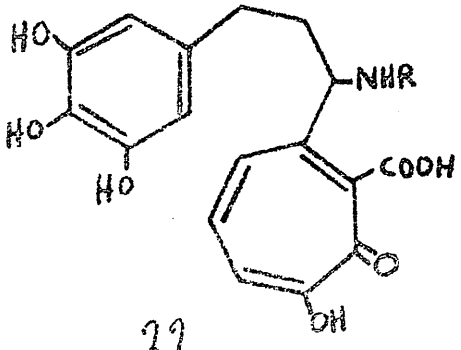

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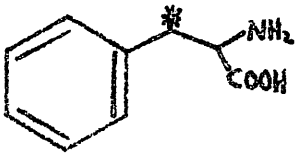

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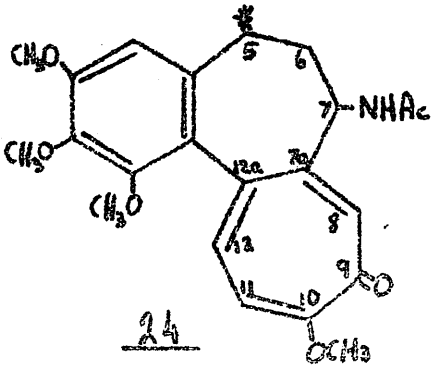


24

The Anet-Robinson scheme[24] suggested that colchicine may be biogenetically related to the flavones (170), the tropolone ring being formed by the ring enlargement of the catechol ring by the insertion of a one carbon fragment. Belleau[25] suggested that colchicine is formed by the oxidative coupling of two molecules of 3,4,5-trihydroxyphenylpyruvic acid to give a compound such as (171). The quinonoid ring of this compound then undergoes fission and cyclisation of the resultant fragments to form the carbon skeleton of colchicine. Wenkert[26] proposed that colchicine is formed by the condensation of an intact tropolone ring (172), derived from shikimate, with the protonated Schiff's base (173), derived from prephenate. The transformation to colchicine occurs through intermediates such as (174) and (175). Scott's biogenesis[27] envisages the intramolecular oxidative coupling of an intermediate such as(22) derived from the precursors(20) and (21) or their biogenetic equivalents.

Phenylalanine is a plausible precursor for the 3,4-dihydroxyphenylpropane moiety of (170) and for 3,4,5-trihydroxy-phenylpyruvic acid. The Schiff's base (173) and the aldehyde (20) might also arise from the same amino acid.

Leete and Nemeth[134] fed (3-^{14}C)-phenylalanine (176) to Colchicum byzantinum and obtained colchicine (24) labelled exclusively in the C_5 position. This result disagrees with the Anet-Robinson and Belleau schemes for the biogenesis of colchicine.

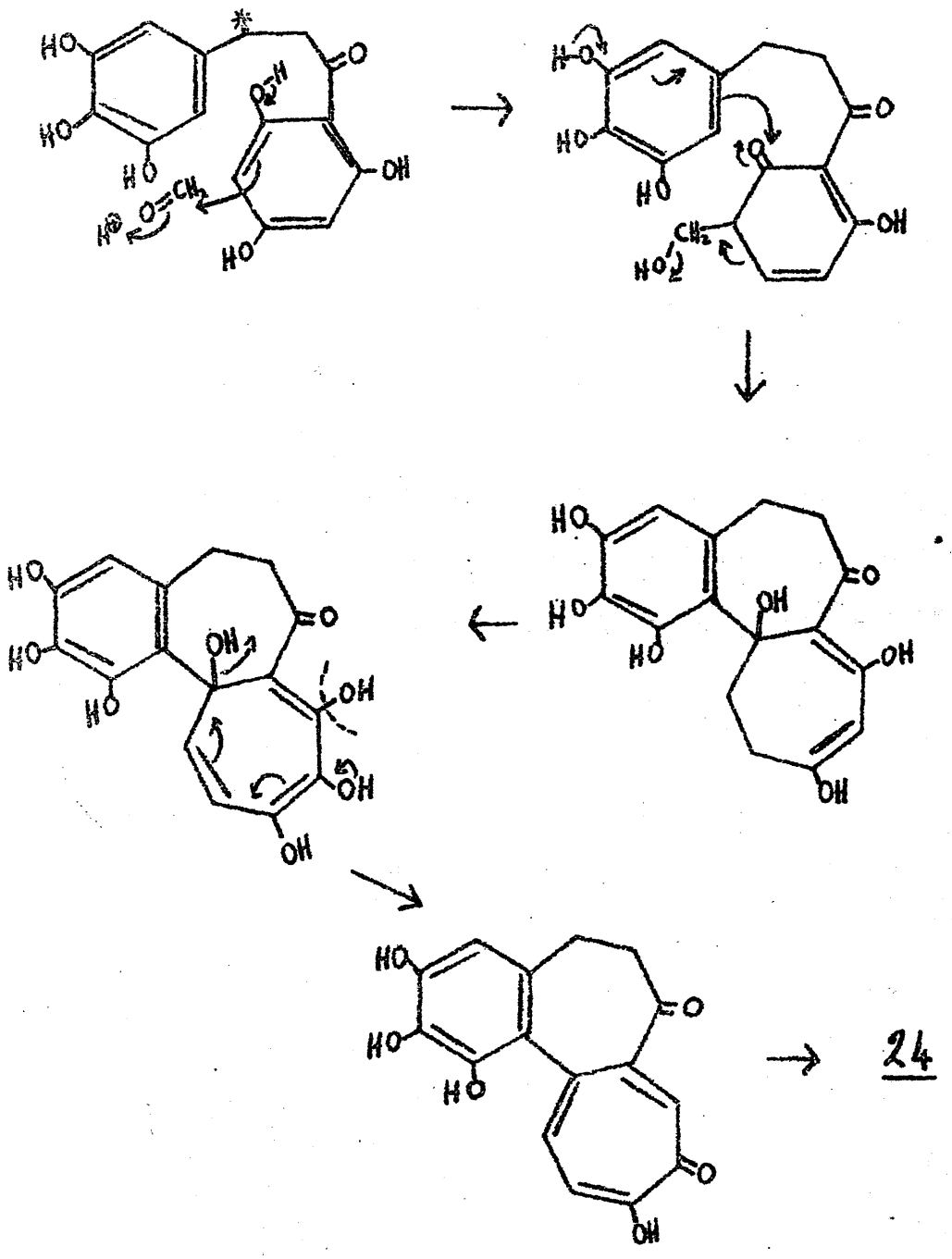
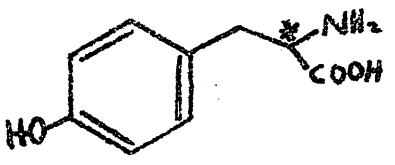


CHART 2.



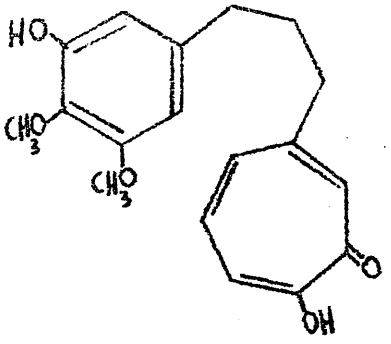
The former would be expected to cause the incorporation of activity at the C_7 carbon atom and the latter at the C_5 position and also in the tropolone ring at the C_{7a} carbon atom. The schemes of Wenkert and Scott are still compatible with this information.

A modification of the Anet-Robinson scheme was proposed[134] in which ring A is derived from the flavones while ring C is obtained by enlargement of an acetate derived aromatic ring through the intervention of formaldehyde or its biogenetic equivalent. The scheme is outlined in chart 2. (3^{14}-C)-Phenylalanine is thuscorrectly incorporated.

Battersby and Reynolds[135], however, have fed (2^{14}-C)-acetic acid to Colchicum autumnale and they found that the acetate was incorporated into the N-acetyl group of colchicine in 96% yield and that the tropolone ring was completely inactive. The tropolone ring is therefore not derived from acetate units as was proposed[134]. The same authors fed C. autumnale with (2^{14}-C) tyrosine (177) and obtained active colchicine which had 50% of the original activity in the N-acetyl group and none at the C_6 carbon atom.

It would thus appear[135] that the side-chains of the aromatic amino-acids are degraded to the C_6-C_1 state before incorporation. The C_2 fragment obtained being partially incorporated as the N-acetyl group.

The Wenkert and Scott hypotheses are still valid at this stage.



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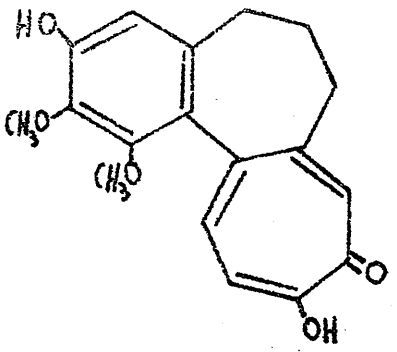

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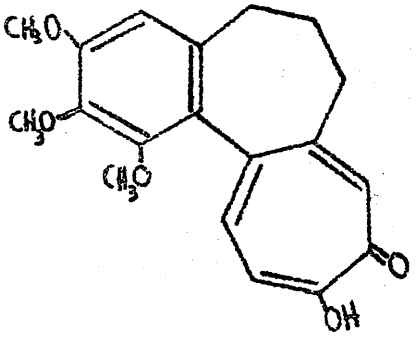

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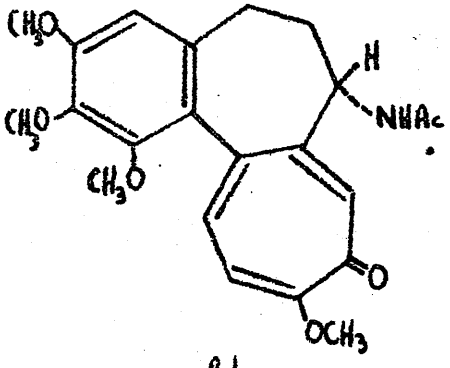

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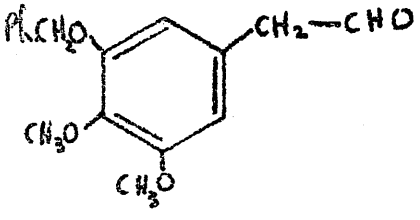

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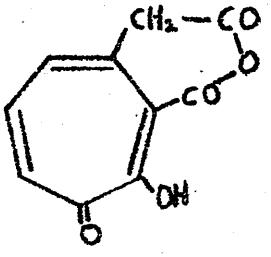

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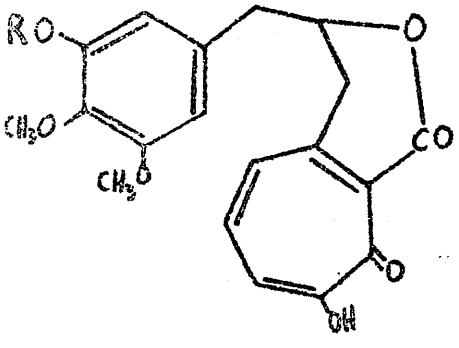

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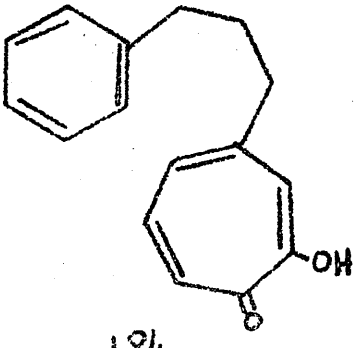


184

THEORETICAL

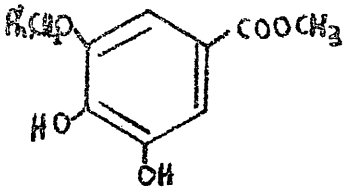
Several syntheses[129,131,133] of colchicine have now been accomplished. They were concerned, however, only with the elaboration of the gross structure of the alkaloid and none were attempted from a biogenetic point of view.

The biogenesis of colchicine proposed by Scott[27] requires the oxidative coupling of a precursor of the type (22), *via* the diradical (23), to give (178) and hence colchicine (24) (see pp. 7, 75).

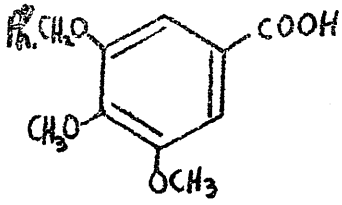
The aim of the following work wasthe synthesis of a simplified precursor (179) and its subsequent oxidation to the tricyclic product (180) which on methylation followed by hydrolysis would give desacetylamidocolchiceine (164). This would constitute a formal total synthesis of (-) colchicine since (164) has been converted by other workers[129,130,131] to (-) colchicine (24), and at the same time would supply a laboratory analogy for the biogenetic hypothesis.

The route to (179) envisaged the condensation of 3-benzyloxy-4,5-dimethoxyphenylacetaldehyde (181) with the anhydride (182) to give the lactone (183, R=$PhCH_2$) which should be readily transformed to the required oxygenated 1-(β-tropolonyl)-3-phenylpropane (179).

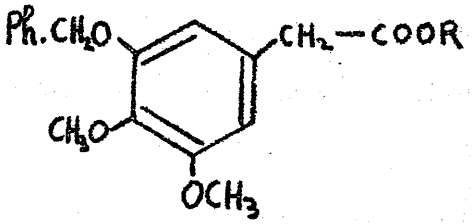
Nozoe[136] and Haworth[137,138] have done much preliminary work to establish the feasibility of such a route. Thus Nozoe prepared


Ph.CH2O
COOCH3
HO
OH

185


Ph.CH2O
COOH
CH3O
OCH3

186

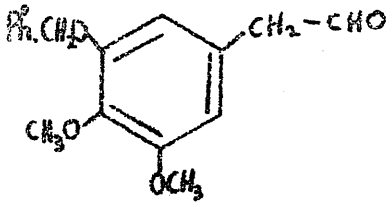

Ph.CH2O
CH2—COOR
CH3O
OCH3

187

the phenyl analogue (184)[139,140] corresponding to (179) by the above route, however he reported[136] that the attempted condensation of 3,4,5-trimethoxyphenylacetaldehyde with the anhydride (182) was unsuccessful.

However, in our hands, 3,4,5-trimethoxyphenylacetaldehyde was successfully condensed with (182) in the absence of any catalyst by using a large excess of the aldehyde and heating it with the anhydride at 100° until effervescence ceased. The lactone (183, R=CH_3) was obtained by crystallisation of the reaction mixture from benzene. It absorbed at ν max. 1730 (lactone) cm.$^{-1}$ and λ max. 255, 332, 377, 387 mμ. (log ε 4.20, 3.72, 3.83, 3.83) and it gave a green ferric colour. The isolation of the lactone was expected by analogy with Nozoe's findings[139,140] on the condensation of phenylacetaldehyde with the anhydride (182).

For the projected synthesis of the properly substituted ring A, methyl gallate was converted, via the diphenyldioxymethylene derivative, to the monobenzyl compound (185)[141] which was methylated and hydrolysed to 3-benzyloxy-4,5-dimethoxybenzoic acid (186)[142]. The acid chloride from (186) was treated with triethylamine and ethereal diazomethane to give the diazoketone. The latter was then rearranged to the homologous ester (187, R=CH_3) by stirring with silver oxide in methanol containing a few drops of triethylamine[143]. The ester was isolated by chromatography on alumina and elution with benzene. 3-Benzyloxy-4,5-dimethoxy-



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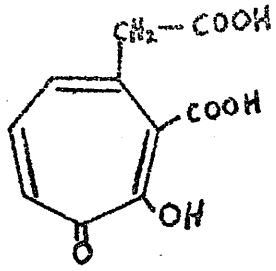

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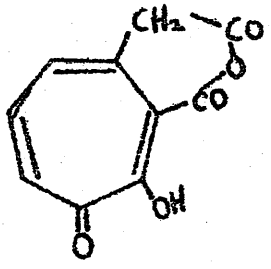

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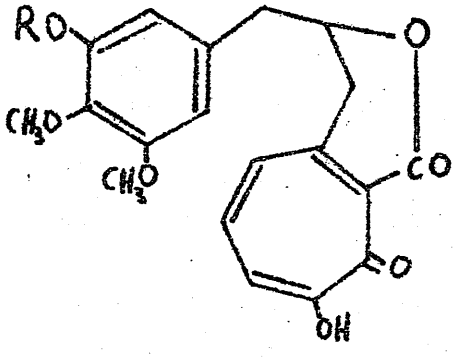


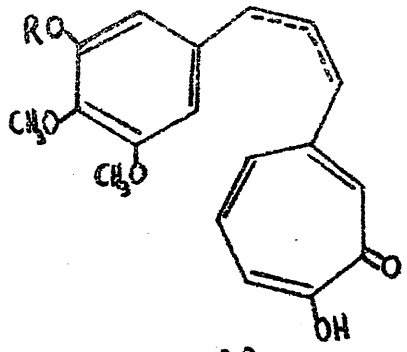
183

phenylacetic acid (187, R=H), ν max. 1740 (CO_2H) cm.$^{-1}$, was obtained by hydrolysis of the ester. Rosenmund reduction[144] of the acid chloride obtained from (187, R=H) afforded the required aldehyde (181), ν max. 1720 (CHO) cm.$^{-1}$

Purpurogallin was oxidised with hydrogen peroxide in alkaline solution to the tropolone diacid (21)[137] which was converted to the anhydride (182)[138] by heating with conc. sulphuric acid at 100° as described by Haworth et al.

As for the model trimethoxy series, the aldehyde (181) was found to condense with the anhydride (182) on heating the two components together at 100° in the absence of a catalyst. The product obtained crystallised from benzene-light petroleum to give a compound shown to be the lactone (183, R=$PhCH_2$) for the following reasons. The compound had the expected analysis, gave a green ferric colour, and showed light absorption maxima at 1730 (lactone) cm.$^{-1}$ in the infrared and at 257, 329, 377 mμ.(log ε 4.20, 3.71, 3.86) with an inflexion at 320 mμ.(log ε 3.66) in the ultraviolet. In the presence of a trace of alkali the ultraviolet absorption became λmax. 270, 348, 428 mμ.(log ε 4.10, 3.93, 4.19) with a shoulder at 278 mμ(log ε 4.06).

When shaken in chloroform solution with 2N-sodium hydroxide solution the lactone gave a sodium salt which was soluble in the chloroform layer and not in the alkali. Dissolved in benzene and reprecipitated with light petroleum the salt was obtained as an



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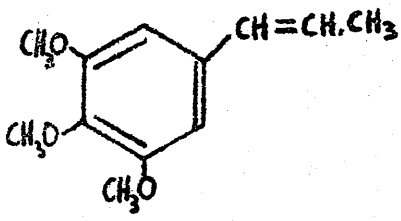

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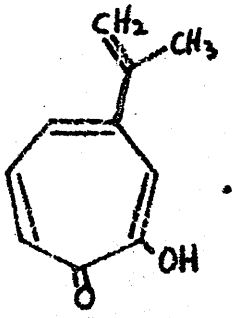
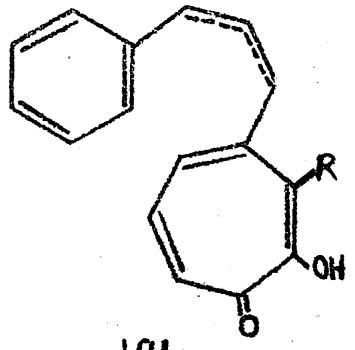


190

intensely yellow solid m.p. 135-137°, ν max. 1700 cm.$^{-1}$, λ max. 268, 354 and 429 mμ (log ε 3.90, 3.74, 4.02), shoulder at 277 mμ (log ε 3.86). The solid gave a persistent yellow flame test, leaving a white residue, and it gave back the lactone (183, R=$PhCH_2$) when suspended in ethanol and treated with hydrochloric acid.

The lactone (183, R=CH_3) was decarboxylated by heating at 190° with a trace of copper bronze until the effervescence ceased. The crude product, after filtration through a silica gel column and elution with benzene-ether (6:1), afforded a yellow solid which had a red ferric colour and a molecular weight of 328 as determined by the mass spectrum. This value of molecular weight corresponds to the unsaturated decarboxylated compound (188, R=CH_3). The infrared spectrum shows no absorption in the carbonyl region except for the tropolone carbonyl at 1610 cm.$^{-1}$ and a strong band is observed at 970 cm.$^{-1}$ which was not present in the spectrum of the lactone. The band at 970 cm.$^{-1}$ is probably due to a <u>trans</u> double bond[145]. The unsaturated compound showed ultraviolet absorption at 273, 374 mμ (log ε 4.35, 3.76).

The position of the double bond was not determined and the product may well be a mixture of double bond isomers. Comparison with the ultraviolet absorption of isoelemicin (189), λ max. 264 mμ (log ε 4.4), and β-dolabrin (190), λ max. 254, 330, 370 mμ (log ε 4.28, 3.79, 3.76)[146], suggests that (188) is more likely to be the isomer with the double bond in conjugation with the trioxygenated



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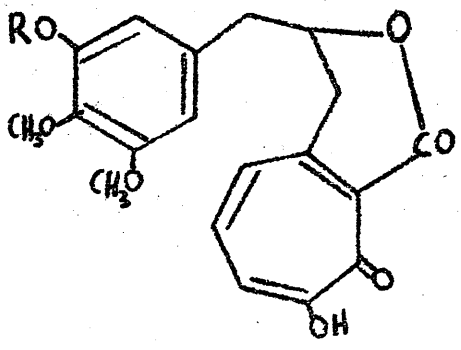


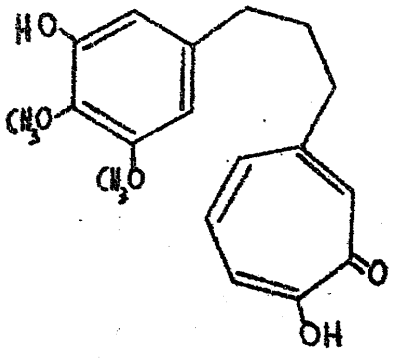
183

benzene ring.

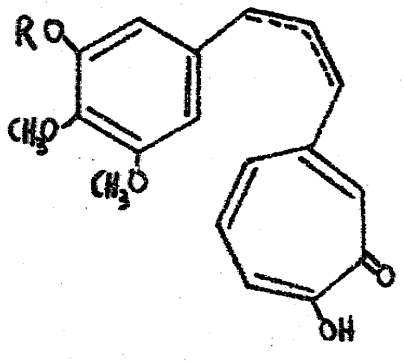
Similar treatment of the lactone (183, R=$PhCH_2$) at first did not provide any satisfactory material due to the viscous nature of the products.

Nozoe et al.[140] reported that the lactone obtained by condensation of phenylacetaldehyde and (182) afforded the unsaturated acid (191, R=CO_2H) on treatment with sodium methoxide. The acid was then readily decarboxylated to give (191, R=H) by heating at the decomposition temperature. Similar small scale treatment of the lactone (183, R=$PhCH_2$) and distillation of the product at 265° (.06 mm.) gave only a small yield (10%) of gum which had the correct infrared properties. Larger scale experiments were unsuccessful as were attempts using potassium tert-butoxide.

The lactone (183, R=$PhCH_2$-) was successfully decarboxylated to the unsaturated product (188, R=$PhCH_2$-) by rapid pyrolysis of small amounts ($\leqslant$ 100 mg.) in sublimation tubes and immediate distillation at 280° (.05 mm.). The product, obtained in 70% yield, showed infrared absorption at 960 cm.$^{-1}$ and no maxima in the carbonyl region above 1610 cm.$^{-1}$ were present. The ultraviolet spectrum was also similar to the trimethoxy compound and had λmax. 273, 365 mμ. (logε 4.35, 3.82) which became λmax. 278, 340, 394 mμ. (logε 4.46, 4.02, 3.95) in the presence of a trace of alkali. The unsaturated product gave a green ferric colour and when treated with benzylamine in ethereal solution it afforded a crystalline



179

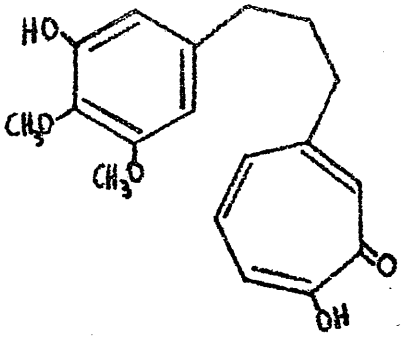


188

benzylamine salt.

Hydrogenation of (188, R=$PhCH_2$) in ethyl acetate or ethanol over palladium charcoal was expected to cause rapid reduction of the propenyl double bond and hydrogenolysis of the benzyl group to give the required hydroxy compound (179). The tropolone ring was expected to be stable to these mild hydrogenation conditions. In the event hydrogenation was found to be extremely unpredictable and highly dependant on the batch of catalyst being used. In general there was a fairly rapid uptake of about 0.7 moles of hydrogen and then a slow, steady uptake of hydrogen continued. The infrared spectrum of the hydrogenated material after successive moles of hydrogen had been absorbed showed only a weak maxima at 960 $cm.^{-1}$ and the development of strong absorption at 3400 and 1700 $cm.^{-1}$ The tropolone hydroxyl band at 3200 $cm.^{-1}$ disappeared and the band at 700 $cm.^{-1}$ which is assigned to the benzyl group remained throughout. The material obtained after absorption of four moles of hydrogen had a negative ferric test and gave a yellow precipitate with 2,4-dinitrophenylhydrazine. It had only benzenoid absorption in the ultraviolet. It thus appears that once the slow reduction of the tropolone ring has resulted in the formation of some non-tropolonoid material the further reduction of this competes with the reduction of the double bond and the hydrogenolysis of the benzyl group.

The material obtained by hydrogenation of (188, R=$PhCH_2$) in ethylacetate over palladium-charcoal (5%) until one mole of



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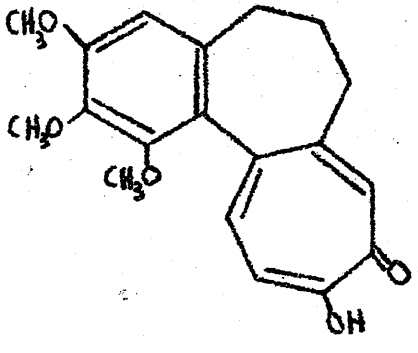
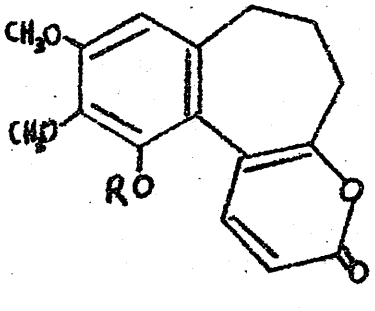


164



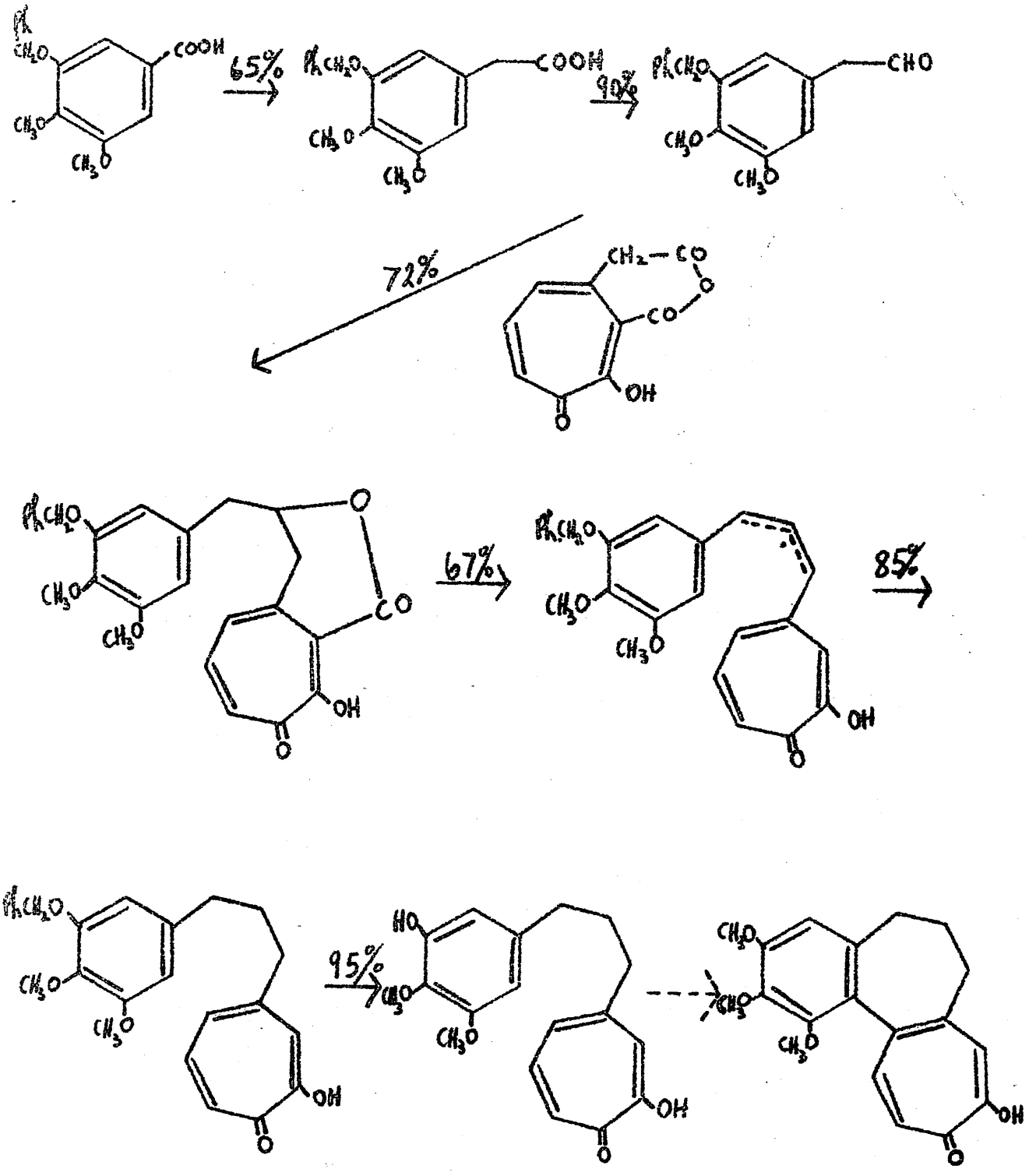
193

the distillate no longer became turbid on dilution with water[148]. The hydroxy compound (179) was an extremely viscous gum which had νmax. 3500-3200 (OH) cm.$^{-1}$; λ max. 322, 349 mμ.(log ε 3.63, 3.57) and with a trace of alkali present this became λ max. 335, 392 mμ. (log ε 3.94, 3.94). It also gave a red ferric colour. For purification purposes, attempts were made to prepared crystalline salts of (179) with benzylamine and quinine methohydroxide without success.

Desacetylamidocolchicoeine (164) shows ultraviolet maxima at 231 (shoulder), 244, 354 mμ.(log ε 4.46, 4.54, 4.25)[129]. The product (180), derivable from (179), although it possesses a free phenolic hydroxyl in the benzenoid ring would be expected to absorb at much the same wavelengths. Support for this is given by the spectra of the lactone (193, R=H) and itsmethyl ether (193, R=CH_3) which absorb at 223, 263, 330 mμ. and 223, 261, 325 mμ. respectively with almost identical ε values.

The band at 354mμ in the ultraviolet spectrum of (164) is approximately five times as intense as the inflexion at 349 mμ. in the spectrum of (179) so that the presence of coupled material in the product obtained from an oxidation of (179) should be readily detected by spectroscopic analysis.

Using the latter criterion for reaction control, the phenol (179) was heated under reflux with manganese dioxide in chloroform and at intervals over two days aliquots were taken, washed with


65%
90%
72%
67%
85%
95%

dilute sulphuric acid, and the material isolated. The ultraviolet spectrum was unchanged throughout. If the aliquots were worked up merely by filtration from suspended manganese dioxide a peak appeared in the ultraviolet at 335 mμ. of greater intensity than the tropolone bands. However, such samples were shown to contain manganese. It appears that a manganese-tropolone complex is responsible for the 335 mμ. peak. The tropolone anion normally has strong bands at 335 mμ. and 392 mμ. The longer wavelength band of the anion was not observed in these manganese containing compounds. A similar result was obtained when lead dioxide was used instead of manganese dioxide.

An attempt to perform the coupling using potassium ferricyanide as the oxidant gave a gum in poor yield with an ultraviolet spectrum resembling starting material. Treatment of (179) with palladium-charcoal (10%) at 360° for 30 minutes in an attempt to cause dehydrogenation to the tricyclic series gave a product, the spectrum of which was similar to starting material except that the 322 mμ. peak was slightly more intense which is probably due to a palladium-tropolone complex.

EXPERIMENTAL

Trimethylgalloyl chloride

Thionyl chloride was purified by distillation first from quinoline then from linseed oil[149].

Trimethyl gallic acid (20 g.) was heated under reflux with thionyl chloride (20 c.c.) and dry benzene (36 c.c.) for 30 mins. A further quantity of thionyl chloride (8 c.c.) was then added and the reflux was continued for a further 3.5 hr. The solvent and reagent were removed *in vacuo* and the residual solid was triturated with light petroleum to remove the last traces of thionyl chloride. The *acid chloride* (20 g.) obtained had ν max. (Nujol) 1750 (C=O) $cm.^{-1}$ m.p. 76-78° (lit.[150] m.p. 76-78° (from light petroleum-benzene)).

3,4,5-Trimethoxyphenylacetic acid.

Triethylamine (3.5 c.c.) (dried over KOH) was added to a stirred solution of diazomethane in ether (100 c.c.) at -10°. (The diazomethane solution was prepared according to Org. Synth., Coll. Vol II, p.166). Trimethylgalloyl chloride (4.7 g.) in dry ether (50 c.c.) was added dropwise to the cooled solution over 15 min. with stirring. The mixture was stirred overnight at room temperature then filtered. The solid residue was stirred with water, and the insoluble material added to the ethereal solution

which was then evaporated at room temperature under reduced pressure. The diazo-ketone (4.1 g.) was obtained as a light yellow solid, m.p. 95-101° (decomp.) ν max (film) 2120 (N≡N) and 1700 (C=O) cm.$^{-1}$

The crude diazoketone (4.1 g.) in methanol (75 c.c.) was treated at 60° with a few drops of triethylamine and a slurry of silver oxide (1.2 g.) in methanol (10 c.c.). The solution was kept at 60° for 45 mins., then refluxed for 1 hr. After treatment at reflux with charcoal, the solution was filtered and evaporated, and the residue was dissolved in ether and washed with saturated sodium bicarbonate solution. The neutral product was filtered in benzene through Grade III alumina (30 g.) and eluted with benzene (150 c.c.) to give a pale yellow oil (3.65 g.). The oil was heated under reflux with a 20% solution of potassium hydroxide in 50% methanol for 3 hr. The acidic material, which was isolated in the usual manner, was recrystallised from light petrol-benzene to give the required acid (2.42 g.) m.p. 116-118° (lit.[151] m.p. 120°).

3,4,5-Trimethoxyphenylacetaldehyde

3,4,5-Trimethoxyphenylacetic acid (7.92 g.) dissolved in benzene (80 c.c.) on warming. The solution was treated with oxalyl chloride (25 c.c.) and was set aside at room temperature for 18 hr. in a flask fitted with a silica gel drying tube. Evaporation of the solvent and excess oxalyl chloride in vacuo gave the acid chloride (8.75 g.) as an oil, ν max. (film) 1800 cm.$^{-1}$

The acid chloride (8.75 g.) in redistilled xylene (75 c.c.) (sodium dried) containing Quinoline-S solution (0.16 c.c.) was hydrogenated over palladium-barium sulphate (5%) (16. g.) at 125° with magnetic stirring. After 5 hr. the evolution of hydrogen chloride ceased. The filtered solution was diluted with ether, washed with dilute sodium bicarbonate solution and then with brine, dried and evaporated. After decolourisation in ethereal solution with charcoal, the required aldehyde (6.2 g.) was obtained as a yellow oil, ν max. (film) 1730 (CHO) cm.$^{-1}$ The semicarbazone, recrystallised twice from methanol had m.p. 186.5-188°. (lit.[152] m.p. 191° from water). (Found: C, 53.9; H, 6.2; N, 15.4. $C_{12}H_{17}N_3O_4$ requires C, 53.9; H, 6.4; N, 15.7%).

Purpurogallin.

This compound was prepared as described by Evans and Dehn[153].

2-Carboxy-4-hydroxy-3- oxocycloheptatrienylacetic acid (21).

Purpurogallin (15 g.) was dissolved in aqueous potassium hydroxide solution (820 c.c.) (23%; W/V). The solution was heated to 90-95°, and then to the stirred solution was added dropwise hydrogen peroxide (37.5 c.c; 100 vol.) at such a rate that the temperatureof the solution kept above 90°. The solution was cooled in ice and water to room temperature, then sodium metabisulphite (7.5 g.) was added, followed by dilute sulphuric acid (405 c.c.) (40%; W/V). The precipitated potassium sulphate

was filtered off and the clear, red, acidic solution was extracted continuously with ether for 48 hr. Removal of the ether gave a gummy solid which was washed by trituration with ether until the washings were almost colourless. The product (4 g.) was the required acid and was sufficiently pure for the next stage. m.p. (from acetic acid) 182-184° (decomp.) (lit.[137] m.p. 183-184°).

The Anhydride (182).

The preceding diacid (21, 1.87 g.) was finely ground and warmed with conc. sulphuric acid (10 c.c.) for 5 min. on the steambath. The clear solution was poured into water (400 c.c.) and the red precipitate was immediately collected by filtration at the water pump, acid-resistant filter paper being advisable. The solid was washed successively with water (3 x 50 c.c.), methylated spirits (3 x 20 c.c.), then ether (3 x 20 c.c.) and dried *in vacuo* at 100°. The red anhydride (920 mg.) obtained had a m.p. 201-205°. (lit.[138] m.p. 205-208°).

Diphenyldichloromethane.

This compound was prepared as described by Norris, Thomas, and Brown[154].

Methyl 3-benzyloxy-4,5-dihydroxybenzoate.

This compound was prepared as described by Jurd[141].

3-Benzyloxy-4,5-dimethoxybenzoic acid (186).

Methyl 3-benzyloxy-4,5-dihydroxybenzoate (43 g.), dimethyl

sulphate (34 c.c.) and potassium carbonate (180 g.) in acetone (700 c.c.) were refluxed and stirred overnight. The mixture was filtered and after evaporation to 50 c.c., water (250 c.c.) was added. Ether extraction of the aqueous material afforded a pale yellow oil which solidified below 100°. The oil was heated under reflux with methanol (250 c.c.) and 4N sodium hydroxide solution (250 c.c.) for 3 hr. After removal of the methanol, the solution was acidified and the precipitated solid was recrystallised from ethanol to give the required acid (40 g.), m.p. 176-177°. (lit.[142] m.p. 172°).

3-Benzyloxy-4,5-dimethoxyphenylacetic acid. (187, R=H).

3-Benzyloxy-4,5-dimethoxybenzoic acid (186, 20.1 g.) was stirred with dry benzene (150 c.c.), oxalyl chloride (25 c.c.) and a trace of dimethyl formamide for 5 hr. Removal of the benzene and excess oxalyl chloride in vacuo gave the acid chloride as a solid melting below 100°, ν max. (film) 1755 cm.$^{-1}$ The acid chloride in dry ether (500 c.c.) was added dropwise over 30 mins. to ethereal diazomethane (400 c.c.) containing triethylamine (KOH dried) (12 c.c.) at -10°, with stirring. The mixture was stirred overnight at room temperature. The precipitated triethylamine hydrochloride was filtered off and the solution was evaporated in vacuo at room temperature to give a bright yellow oil with ν max. (film) 2080 (N≡N) and 1675 (C=O) cm.$^{-1}$ which is in accord with a diazoketone.

The diazoketone in dry methanol (200 c.c.) was heated at 60° with a slurry of silver oxide (13.6 g.) in methanol (70 c.c.) and a few drops of triethylamine. After 15 mins. the mixture was heated under reflux for 1.5 hr. After treatment at reflux with charcoal, the solution was filtered and evaporated, and the residue was dissolved in ether and washed with sodium bicarbonate solution. The neutral product was chromatographed on Grade III alumina and eluted with benzene (1 l) to give the required ester as a brown oil (15.3 g.), ν max. (film) 1750 (CO_2CH_3) cm.$^{-1}$

The ester was heated under reflux with methanol (150 c.c.) and aqueous potassium hydroxide solution (150 c.c; 40%) for 4 hr. The acid product (13.5 g.) was an oil which solidified to give the required homologous acid, m.p. 108.5-109° (from light petroleum-benzene), ν max. (Nujol) 1740 (CO_2H) cm.$^{-1}$ (Found: C, 67.6; H, 6.0. $C_{17}H_{18}O_5$ requires C, 67.5; H, 6.0%).

3-Benzyloxy-4,5-dimethoxyphenylacetaldehyde (181).

3-Benzyloxy-4,5-dimethoxyphenylacetic acid (187, R=H, 13.5 g.) in dry benzene (150 c.c.) wastreated with oxalyl chloride (25 c.c.) and a trace of dimethyl formamide and left overnight at room temperature. After removal of the solvent and excessreagent in vacuo the acid chloride was obtained as an oil, ν max. (film) 1810 cm.$^{-1}$

The acid chloride in freshly distilled xylene (sodium dried) (75 c.c.) was hydrogenated over 5% palladium-barium sulphate (2.8 g.) in the presence of Quinoline-S solution (0.28 c.c.) with magnetic

stirring at 120°-130°. The evolution of hydrogen chloride ceased after 4 hr. The cooled solution was filtered, diluted with ether and washed with aqueous sodium hydroxide solution (2%). The neutral extract furnished the required aldehyde as an oil (11.52 g.), ν max. (1730 (CHO) cm.$^{-1}$ The semicarbazone was formed and recrystallised from methanol, m.p. 156-157°. (Found: C, 62.7; H, 6.1; N, 12.3. $C_{18}H_{21}N_3O_4$ requires C, 63.0; H, 6.2; N, 12.2%).

The lactone (183, R=CH_3).

The anhydride (182, 1 g.) was heated on a steam bath with 3,4,5-trimethoxyphenylacetaldehyde (4.8 g.) until effervescence ceased (ca. 30 min.). The dark homogeneous oil was dissolved in benzene, and filtered, and the solution was evaporated to 25 c.c. A fawn solid (1.15 g.) was precipitated which was recrystallised from benzene to give the required lactone (790 mg.) as pale yellow needles, m.p. 170-171°. The lactone gave a deep green ferric colour and showed light absorption at ν max. (chloroform) 1720 (lactone), (Nujol) 1730 (lactone) cm.$^{-1}$; λ max. 255, 332, 377, 387 mμ.(log ε 4.20, 3.72, 3.83, 3.83). (Found: C, 64.35; H, 5.65; $-OCH_3$, 24.6. $C_{20}H_{20}O_7$ requires C, 64.5; H, 5.4; OCH_3, 25.0%).

Pyrolysis of the Lactone (183, R=CH_3).

The preceding lactone (183, R=CH_3) (50 mg.) was heated at 190-200° with copper bronze (10 mg.) until the effervescence ceased (ca. 10 min.), then for a further 5 min. The dark melt

was filtered in benzene solution through silica gel (1 g.). Elution with benzene-ether (6:1, 25 c.c.) followed by charcoal treatment gave a pale yellow gum (24 mg.), which crystallised on scratching and had a red ferric colour. The solid on recrystallisation from aqueous methanol afforded the required unsaturated product (188, R=CH_3) as a granular yellow solid, m.p. 108-112°, λmax. 273, 374 mμ.(log ε 4.35, 3.76), M(mass spectrometric) 328, ($C_{19}H_{20}O_5$ requires M, 328), ν max. (Nujol) 968 $cm.^{-1}$ (trans CH=CH).

The Lactone (183, R=$PhCH_2$).

The anhydride (182) (1.90 g.) was heated on a steam bath with 3-benzyloxy-4,5-dimethoxyphenylacetaldehyde (181, 11g.) for 30 mins. until the effervescence ceased. The melt was dissolved in warm benzene (70 c.c.) and filtered while still hot. The solution was concentrated to 45 c.c. and light petrol (15 c.c.) was added. After 16 hr., the yellow solid (2.71 g.), m.p. 139-143°, was filtered off. The mother liquors were evaporated and dissolved in a little chloroform, then diluted greatly with ether, and shaken with 4N-sodium hydroxide solution. The bulky brown precipitate was filtered off, dissolved in chloroform and shaken with 6N-sulphuric acid. The chloroform layer afforded a gum (1.16 g.) which on crystallisation from acetone-light petrol gave more of the yellow solid (260 mg.) m.p. 142-144°. The total

yellow solid (2.97 g.) had ultraviolet and infrared spectra in accord with the required lactone, m.p. 145-146° (from acetone); νmax. (Nujol) 1730 (lactone) cm.$^{-1}$; λmax. 257, 329, 377 mμ. (log ε 4.20, 3.71, 3.86) inflexion at 320 mμ. (log ε 3.66); in the presence of a trace of alkali the ultraviolet had λmax. 270, 348, 428 mμ. (log ε 4.10, 3.93, 4.19), shoulder at 278 mμ. (log ε 4.06). It had a green ferric colour. (Found: C, 69.5; H, 5.65. $C_{26}H_{24}O_7$ requires C, 69.65; H, 5.4%).

The first time the above condensation was carried out, the lactone crystallised from benzene-light petroleum as a solvate, m.p. 75-79°. (Found: C, 72.90, 73.70; H, 6.55, 6.35%). In its reactions this behaved like the unsolvated form, and its mass spectrum had peaks at 448 (lactone molecules), 404 (decarboxylated product) and 78 (benzene). The solvate was not encountered in the other experiments.

Pyrolysis of the Lactone (183, R=$PhCH_2$).

The preceding lactone (183, R=$PhCH_2$; 100 mg.) was mixed with copper bronze (20 mg.) and placed in a sublimation tube (8 mm. diameter) which was then evacuated to less than 0.05 mm. The tube was then placed vertically in a sublimation block (pre-heated to 270-280°) until vigorous frothing abated (2-3 min.). The tube was then set horizontally and the material distilled along a short length (4-5 cm.) of the sublimation tube. After ten

minutes the tube was cooled and the yellow distillate was washed out with ether (leaving some ether insoluble residue). It is important to have no hold up in the tube, such as a glass wool plug, and to avoid prolonged heating of the reaction mixture. The product was obtained in yields of 55-65 mg. in this way. The infrared and ultraviolet spectra were in accord with the required unsaturated product (188, R=$PhCH_2$), ν max. (carbon disulphide) 955 (trans CH=CH) $cm.^{-1}$, (film) 960 $cm.^{-1}$; λ max. 273, 365 mμ.(log ε 4.35, 3.82); in the presence of a trace of alkali the ultraviolet had λmax. 278, 340, 394 mμ.(log ε 4.46, 4.02, 3.95). It gave a red ferric colour.

The product (188, R=$PhCH_2$) when treated in ethereal solution with benzylamine gave the benzylamine salt as a gum which crystallised when scratched. Recrystallisation from ethylacetate gave a yellow granular solid, m.p. 110-112°, λmax. 272, 368, 390 (shoulder) mμ.(log ε 4.41, 3.87, 3.70). (Found: C, 75.2; H, 6.5; N, 2.5. $C_{32}H_{33}O_5N$ requires C, 75.1; H, 6.5; N, 2.75%).

The free tropolone was readily obtained by shaking an ethylacetate solution of the salt with 6N. sulphuric acid.

Action of base on the Lactone (183, R=$PhCH_2$).

The lactone (183, R=$PhCH_2$; 50 mg.) was heated under reflux with a solution of sodium (1 g.) in dry methanol (10 c.c.) for 2 hr. The lactone passed into solution slowly. The alkali

soluble fraction (45 mg.), which was isolated in the usual way, was a yellow gum which solidified on trituration with ether, m.p. above 100° over a range of temperature (vigorous effervescence above 150°). The infrared spectrum indicated the presence of carboxyl. It had a positive ferric test. The product (27 mg.) on pyrolysis at 265° (.06 mm.) for 10 min. gave the unsaturated compound (188, R=$PhCH_2$, 5 mg.).

A similar result was obtained with potassium tert-butoxide in tert-butanol at reflux for 20 min.

Hydrogenation of the pyrolysis Product (188, R=$PhCH_2$).

The unsaturated material (188, R=$PhCH_2$) (400 mg.), when shaken under hydrogen in ethyl acetate (40 c.c.) with 5% palladium charcoal (100 mg.) absorbed 1 mole of hydrogen in ca. 7 hr. The product was chromatographed on silica gel (20 g.) and eluted with ether (200 c.c.) to give the product (192) (336 mg.) as a yellow viscous oil which gave a red ferric colour. Distilled twice at 254° (0.07 mm.) it had λ max. 323, 348 mμ. (log ε 3.48, 3.42); in the presence of a trace of alkali the ultraviolet had λmax. 335, 392 mμ (log ε 3.86, 3.86) with an inflexion at 266 mμ. (log ε 4.10). The infrared spectrum in carbon disulphide showed the presence of benzyl ν max. 700 $cm.^{-1}$ and absence of the ethylenic band of the precursor at 960 $cm.^{-1}$ (Found: C, 73.3; H, 6.4. $C_{25}H_{26}O_5$ requires C, 73.85; H, 6.45%).

If hydrogenation was continued, the compound slowly absorbed several moles of hydrogen to give products which still contained benzyl, but which had a carbonyl band at 1700 cm.$^{-1}$, gave a precipitate with 2,4-dinitrophenylhydrazine and had no colouration with ferric chloride. A similar result wasobtained when ethanol or acetic acid was used as solvent.

Hydrogenation of the benzylamine salt of (188, R=$PhCH_2$) in ethanol with 5% palladium-charcoal resulted in a more rapid uptake of 1 mole of hydrogen (ca. 3 hr.). The reduced product (192) was obtained by acid treatment of the benzylamine salt. A crystalline benzylamine salt of the reduced compound could not be prepared.

Debenzylation of the reduced Compound (192).

A solution of the benzyl ether (192, 118 mg.) in acetic acid (20 c.c.) was added dropwise to a solution of dilute hydrochloric acid (25%, 30 c.c.) and acetic acid (10 c.c.) at 135°. As the ether was added the benzyl alcohol formed was distilled out with water and acetic acid. The distillation was continued until the distillate no longer became turbid on dilution with water. The cooled solution was extracted with chloroform and the chloroform layer was washed well with water to remove acetic acid. Evaporation of the solvent gave a dark brown gum. (90 mg.) which had ν max. (film) 3500-3200 (-OH) cm.$^{-1}$ and no absorption at 700 cm.$^{-1}$,

λ max. 322, 349 mμ. (log ε 3.63, 3.57); in the presence of a trace of alkali the ultraviolet had λ max. 335, 392 mμ. (log ε 3.94, 3.94). It gave a red ferric colour. The material is presumably the required hydroxy compound (179).

Attempts to prepare a crystalline derivative for purification purposes were not successful (benzylamine and quinine methohydroxide were tried).

Attempted Oxidative Coupling of the Hydroxy-Compound. (179).

(1) Manganese dioxide.

The phenol (179, 5 mg.) was heated under reflux with manganese dioxide (20 mg.) in chloroform (15 c.c.). At intervals over 2 days, aliquots were taken, washed with 6N sulphuric acid and then with brine, dried, evaporated, and the ultraviolet done on the residue dissolved in ethanol. The spectrum was unchanged throughout, aliquots being taken at 2.5, 5, 16 and 37 hr.

Similar results were obtained when the reaction was carried out at room temperature and when several runs were made with fresh quantities of manganese dioxide being used for each run. Increasing the scale resulted in diminished yields due to the difficulty in extracting the material from the manganese dioxide.

If the experiment was worked up merely by filtration of the manganese dioxide and evaporation of the solvent, a peak appeared at 335 mμ in the ultraviolet of greater intensity than the tropolone

bands. However, such samples were shown to contain manganese.

Test for manganese - The material (0.5 mg.) was treated with a drop of conc. nitric acid and heated for 3 min. on the steam bath. After the addition of 2 drops of water, the mixture was cooled and a few milligrammes of sodium bismuthate were added with shaking. A distinct permanganate colour developed rapidly.

(2) Potassium ferricyanide.

The phenol (179, 42 mg.) dissolved in a solution of sodium carbonate (312 mg.) in de-aerated, distilled water (25 c.c.) on warming. The solution was cooled to room temperature, and stirred under nitrogen during the dropwise addition of a solution of potassium ferricyanide (15.6 c.c., 1%) in de-aerated, distilled water. After a further 1.5 hr. with stirring under nitrogen, the solution was acidified with 6N sulphuric acid and extracted with chloroform. The chloroform solution gave a gum (26 mg.), the ultraviolet of which resembled starting material. The gum was, however, much less soluble than the starting material.

Treated in methylene chloride-methanol with excess of ethereal diazomethane, it gave a neutral fraction (19.7 mg.) which was heated on the steambath with 4N-sodium hydroxide solution (5 c.c.) and a few drops of methanol. The gummy, alkali-soluble material so obtained appeared to contain little tropolone, as shown by the ultraviolet spectra in ethanol, or in ethanol-alkali.

(3) Palladium-charcoal.

Heated at 360° for 30 min. with 10% palladium-charcoal, the phenol (179) gave a product the ultraviolet of which was very similar to the starting material.

(4) Lead dioxide.

The phenol (179, 10 mg.) was heated under reflux with lead dioxide (100 mg.) in chloroform (30 c.c.) for 3 hr. The lead dioxide was filtered off and the chloroform solution washed with 6N-sulphuric acid. Evaporation of the chloroform afforded a gum (3 mg.) which had an ultraviolet absorption maximum at 335 mμ in chloroform. It is probably a lead complex of the tropolone moeity.

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