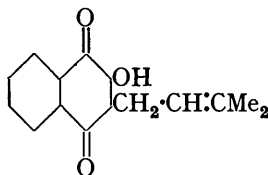


321. *Dunnione. Part I.*

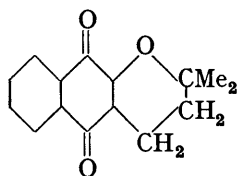
By J. R. PRICE and SIR ROBERT ROBINSON.

The orange pigment of *Streptocarpus Dunnii* Mast. (cf. Robinson and Robinson, *Biochem. J.*, 1934, **28**, 1718) occurs as a deposit on the leaves. This substance, termed *dunnione*, has been characterised as a  $\beta$ -naphthaquinone closely resembling  $\beta$ -lapachone (IV). The conversion of *dunnione* into various isomerides and oxidation products has been studied and the results of the investigation are best interpreted by means of the formula (VIII). The constitutional problem has not yet, however, been completely solved.

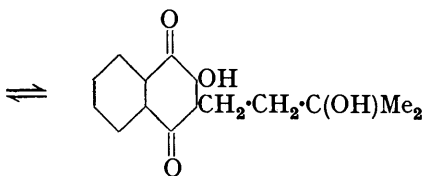
In a preliminary notice (*Nature*, 1938, **142**, 147) of this work, the name *dunnione* was suggested for the colouring matter which occurs as an orange-red deposit on the leaves, stems and flowers of *Streptocarpus Dunnii* Mast. The substance is readily purified, has the formula  $C_{15}H_{14}O_3$ , is optically active, and yields phthalic acid on oxidation. No hydroxyl or carboxyl groups are present, but there is at least one carbonyl group as shown by the formation of a *semicarbazone* and 2 : 4-*dinitrophenylhydrazone*. Reducing agents cause a rapid disappearance of the colour, which is regenerated by aeration, and reductive acetylation gives a *diacetyldihydro*-derivative. This behaviour indicates quinonoid character and the colour and solubility in sodium bisulphite solution suggest an *o*-quinone. This is confirmed by the ready condensation of *dunnione* with *o*-phenylenediamine with elimination of  $2H_2O$  and probable formation of a *phenazine* derivative. The third oxygen atom of *dunnione* is not present as methoxyl; nor is it ketonic, since diacetyldihydro*dunnione* was recovered unchanged (except for a small amount of *dunnione* semicarbazone) after prolonged treatment with semicarbazide. The function of this third oxygen atom is shown by the behaviour of *dunnione* with acids and alkalis in comparison with that of lapachol (I) and  $\alpha$ - and  $\beta$ -lapachones (II and IV) under the same conditions. Lapachol dissolves in alkali, giving a red solution of the sodium salt of an *isopentenyl*-2-hydroxy-1 : 4-naphthaquinone. This salt is evidently a chelate metal compound, because it can be extracted from its aqueous solution by such solvents as *iso*amyl alcohol and ethyl acetate. Careful neutralisation regenerates lapachol and, with an excess of acid, partial conversion into  $\beta$ -lapachone ensues. When  $\beta$ -lapachone is treated with alkali, it dissolves slowly at the ordinary temperature and more rapidly on warming. The chroman ring opens, isomerisation to a 1 : 4-quinone follows, and a red solution of the sodium salt of hydroxyhydrolapachol (III) is produced. This salt resembles that of lapachol in its behaviour. When its solution is just acidified, hydroxyhydrolapachol is regenerated, and this redissolves in alkalis *immediately*, in contrast to  $\beta$ -lapachone, which is formed by the action of an excess of acid on the sodium salt of (III). *Dunnione* dissolves slowly in cold alkali to a



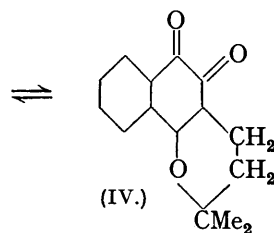
(I.)



(II.)



(III.)



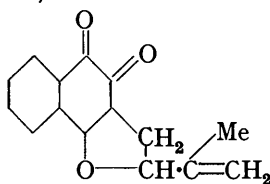
(IV.)

red solution similar in colour to that of lapachol or hydroxyhydrolapachol, and the behaviour in respect of extraction by organic solvents is the same as that of lapachol. Careful neutralisation gives a yellow solution, which returns *immediately* to red with alkali. The action of an excess of acid on the red solution regenerates *dunnione*.

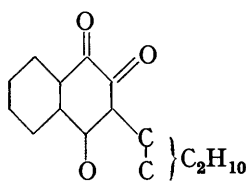
When heated for a short time with concentrated hydrochloric acid,  $\beta$ -lapachone is converted into the corresponding 1 : 4-quinone,  $\alpha$ -lapachone (Hooker, J., 1892, **61**, 611).

Dunnione behaves similarly, yielding a yellow isomeride,  $\alpha$ -dunnione, m. p. 121—122°.  $\alpha$ -Dunnione dissolves in aqueous-alcoholic alkali to a red solution, from which dunnione can be recovered by acidification.

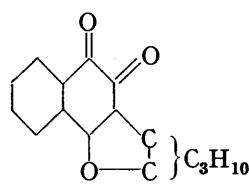
The ultra-violet absorption spectrum of dunnione has been examined by Cooke, Macbeth, and Winzor (this vol., p. 878), who confirm the 1 : 2-quinone structure, as the absorption closely resembles that of  $\beta$ -lapachone, bromo- and hydroxy- $\beta$ -lapachones and dehydroiso- $\beta$ -lapachone (V). The third oxygen atom of dunnione is probably, therefore, in a chroman or coumaran ring and the relation of dunnione to  $\alpha$ -dunnione is the same as that of  $\beta$ -lapachone to  $\alpha$ -lapachone. The formula of dunnione can now be written as (VI) or (VII).



(V.)



(VI.)



(VII.)

In one respect dunnione differs profoundly from lapachol, lomatiol, and their derivatives, whose properties have been closely studied by Hooker and his co-workers (see *J. Amer. Chem. Soc.*, 1936, **58**, 1163 and earlier papers). A solution of dunnione or  $\alpha$ -dunnione in alkali changes, on boiling for a few minutes, from red to yellow and acidification affords a new yellow substance, m. p. 161—162°, isomeric with dunnione, which we propose to designate *alloodunnione*. Also, when dunnione is kept in 5% aqueous sodium hydroxide at room temperature for two days, it is partly converted into *alloodunnione*, which is obtained from the yellow solution (initially red) on acidification. The remainder of the dunnione, curiously enough, is changed into  $\alpha$ -dunnione. This fact is less surprising when it is noted that the oxide ring of  $\alpha$ -dunnione is difficult to open compared with, *e.g.*,  $\alpha$ -lapachone. Aqueous alcoholic alkali is necessary to obtain a solution of the red sodium salt, and also  $\alpha$ -dunnione remains unchanged after standing in concentrated sulphuric acid at room temperature for two hours.

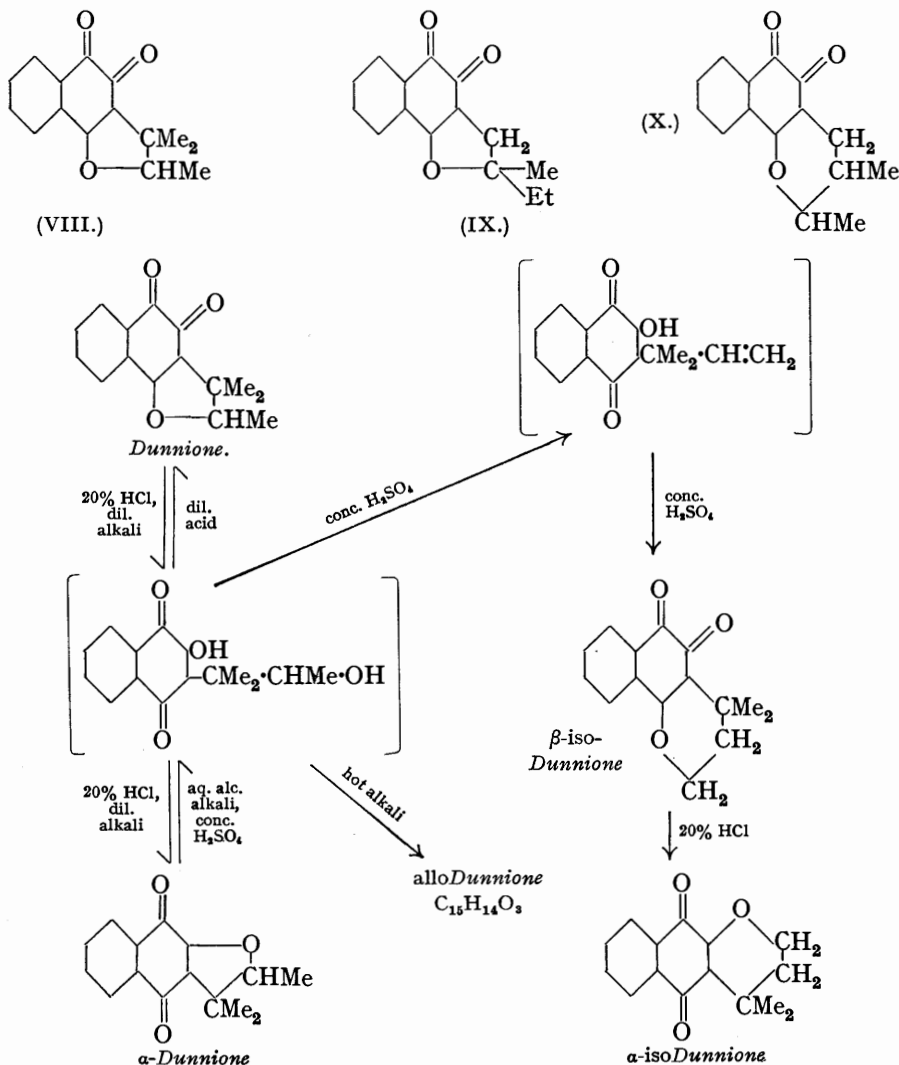
The formation of *alloodunnione* involves a rearrangement not paralleled in the lapachol and lomatiol series, and it is evidently the intermediate produced by alkali fission of the oxide ring of dunnione or  $\alpha$ -dunnione which undergoes the rearrangement. The substance cannot be a derivative of a 2-hydroxy-1 : 4-naphthaquinone, because (1) the alkaline solution is yellow, (2) the ultra-violet absorption, examined by Cooke, Macbeth, and Winzor (*loc. cit.*) under the name "*isodunnione*," is unlike that of any of the naphthaquinones studied by those workers, and (3) it gives an acid,  $C_{12}H_{12}O_4$ , on oxidation with alkaline peroxide under conditions whereby lapachol and hydroxyhydrolapachol yield only phthalic acid and acetone.

Dunnione was oxidised with alkaline peroxide before it was realised that conversion into *alloodunnione* would take place under the conditions of the experiment. Except for a small amount of phthalic acid, the products of the oxidation were the same as in the case of *alloodunnione*, namely, acetaldehyde and the acid  $C_{12}H_{12}O_4$ . More vigorous oxidation of *alloodunnione* (hydrogen peroxide in acetic acid) gave phthalic acid.

Hooker (*loc. cit.*) found that  $\alpha$ -lapachone is recovered unchanged from a freshly prepared solution in concentrated sulphuric acid, but after standing for a few minutes it is completely converted into  $\beta$ -lapachone.  $\alpha$ -Dunnione, however, was recovered unchanged after its solution in sulphuric acid had been kept for two hours at room temperature. When the solution was heated at 100°, the colour changed from intense red to brownish-orange and after pouring into water another new substance, m. p. 129—131°, was isolated. This is isomeric with dunnione and its colour and solubility in aqueous sodium bisulphite indicate that it is a 1 : 2-quinone. It evidently differs structurally from dunnione, as it is markedly sensitive to light (*cf.* dehydroiso- $\beta$ -lapachone; Hooker, *J. Amer. Chem. Soc.*, 1936, **58**, 1181) whereas dunnione is not. In addition the red alkaline solution does not change in

colour on boiling, as does that of dunnione. For these reasons, the substance is termed  $\beta$ -isodunnione. Heating with hydrochloric acid converts it into a yellow isomeride, m. p. 118—119°, termed  $\alpha$ -isodunnione. As only small quantities of these two compounds have been available, no derivatives have yet been prepared. Dunnione itself is unchanged by heating with concentrated sulphuric acid at 100° for an hour.

On account of the ease of isomerisation in acid solution, the amount of acetic acid produced by chromic acid oxidation may not be an accurate index of the number of side-chain methyl groups in the dunnione molecule. Subject to this limitation, the following results are of some assistance. The figures for dunnione and *alldunnione* are substantially the same, namely, *ca.* 1.3 mols. of acetic acid, suggesting that there is one side-methyl group and a *gem*-dimethyl group. The  $C_{12}$  acid from *alldunnione* gave *ca.* 0.5 mol. of acetic acid. Since acetaldehyde is eliminated during the formation of the acid, this figure is in good agreement with expectation for the remaining *gem*-dimethyl group;  $\beta$ -lapachone gave a similar result, 0.59 mol. of acetic acid.

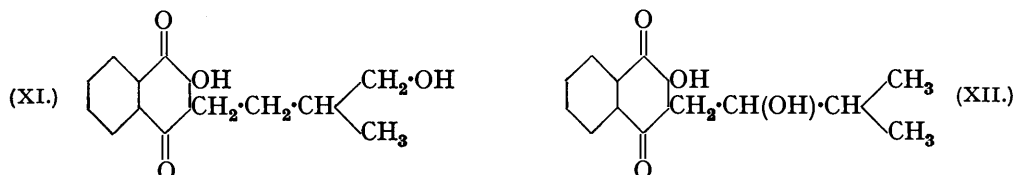


These data are insufficient to determine the structure of dunnione with finality. But if we assume that the  $C_5$  side chain has the usual isoprene configuration, the possible formulæ are limited to eleven, two of which, corresponding to  $\beta$ -lapachone and *iso*- $\beta$ -lapachone, can

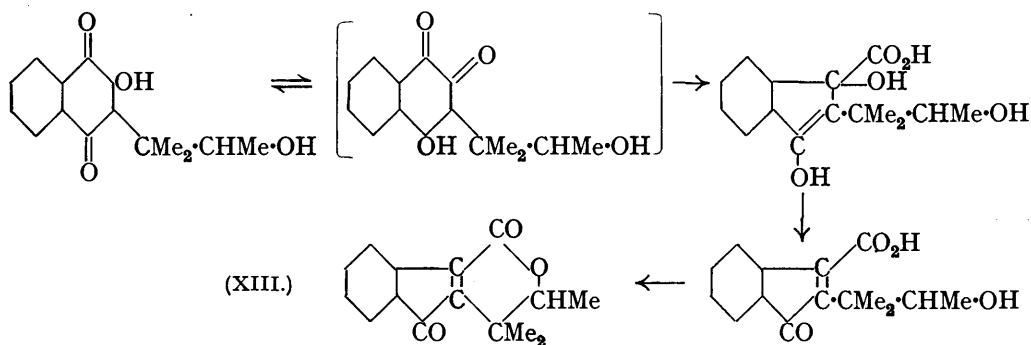
be eliminated. Since the amount of acetic acid produced by chromic acid oxidation is practically the same for both dunnione and *alloodunnione*, we may infer that the change dunnione  $\rightarrow$  *alloodunnione* does not affect those carbon atoms which are oxidisable to acetic acid. The formation of acetaldehyde by oxidation of *alloodunnione* is then of some significance for the structure of dunnione. It enables us to eliminate six of the remaining nine possible formulæ, leaving (VIII), (IX), and (X).

Of these three, (VIII) is preferable, as it agrees best with the results of oxidation. Structural rearrangements leading to the formation of  $\alpha$ -dunnione and the two *isodunniones* may then be tentatively formulated as on p. 1524.

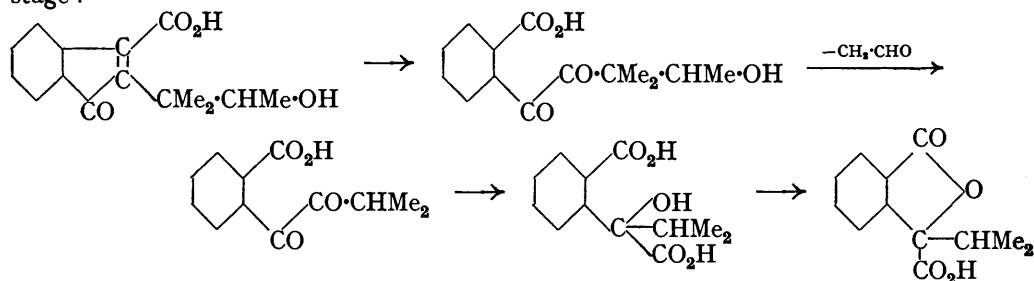
The formation of the  $C_{12}H_{12}O_4$  acid, together with acetaldehyde, by oxidation of *alloodunnione* supports formula (VIII), as this is the only one of the three structures (VIII, IX, and X) which will account for the presence of twelve hydrogen atoms in the oxidation product. The suggested enlargement of the ring from a coumaran to a chroman by the action of concentrated sulphuric acid on  $\alpha$ -dunnione is analogous to the effect of sulphuric acid on the three known hydroxy-*isoamyl* derivatives of 2-hydroxy-1:4-naphthaquinone (Hooker, *J. Amer. Chem. Soc.*, 1936, **58**, 1181). These three substances, hydroxyhydro-lapachol (III), hydrolomatiol (XI), and hydroisomatiol (XII), all give  $\beta$ -lapachone as principal product or as one of the principal products. According to Hooker, this indicates a general tendency towards the formation of a six-membered ring.



The behaviour on acidification of an alkaline solution of *alloodunnione* indicates the presence in the molecule of a ring system which is opened by alkalis and closed by acids, and a tentatively suggested structure and mechanism of formation for *alloodunnione* (XIII) are set out below :



On this basis the  $C_{12}$ -acid would be produced as follows, the actual removal of acetaldehyde being due to hydrolysis of an intermediate and not to oxidation at the last stage :



The suggested formation of acetaldehyde is clearly a reversed aldol condensation which would certainly be facilitated by the quaternary carbon attached to the  $\cdot\text{CH}(\text{OH})\cdot$  group. Synthetic experiments are in progress with the object of checking these views.

The 2:4-dinitrophenylhydrazones of  $\alpha$ - and  $\beta$ -lapachone were prepared in order to ascertain whether mono- or di-derivatives were formed with Brady's reagent under the usual conditions.

#### EXPERIMENTAL.

*Isolation of Dunnione.*—Leaves of *Streptocarpus Dunnii* were extracted 5–10 times with water at room temperature, each extraction taking 24–48 hours. The leaves of the plant are large (2 feet long by 18 inches wide, approximately) and 10–20 l. of extract were obtained from each leaf. The orange-coloured solution was shaken with benzene, which completely extracted the colouring matter, and the benzene was dried and evaporated. The residue was dissolved in a small quantity of alcohol and poured into half-saturated aqueous sodium bisulphite, which was shaken and filtered. *Dunnione* was recovered by addition of an excess of aqueous sodium carbonate, and was again isolated by means of benzene; after twice crystallising from light petroleum (b. p. 60–80°), it formed large, orange-red needles, m. p. 98–99°, not raised by further crystallisation. The yield from one leaf varied from 0.5 to 2 g. [Found: C, 74.6; H, 5.7; MeO, 0.0; C-CH<sub>3</sub>, 8.6; *M* (Rast), 261. C<sub>15</sub>H<sub>14</sub>O<sub>3</sub> requires C, 74.4; H, 5.8; 1 C-CH<sub>3</sub>, 6.2%; *M*, 242].  $[\alpha]_D^{25} + 310^\circ$  (*c*, 4.163 in chloroform). The substance is readily soluble in the simple alcohols, acetic acid, acetone, ether, benzene and chloroform, but is sparingly soluble in water and light petroleum and can be crystallised from either of these solvents. It dissolves in concentrated sulphuric acid to an orange solution, from which it can be recovered unchanged (m. p. and undepressed mixed m. p.), after heating for an hour at 100°, by addition of water.

*Dunnione* dissolves in aqueous sodium bisulphite to a colourless solution, from which it is regenerated by addition of alkali. Reducing agents bring about a rapid decolorisation of its solution, but the colour returns on shaking in air.

The *semicarbazone* crystallises from aqueous methyl alcohol in yellow needles, m. p. 232–233° (Found: C, 64.4; H, 5.7; N, 14.4. C<sub>15</sub>H<sub>17</sub>O<sub>3</sub>N<sub>3</sub> requires C, 64.2; H, 5.7; N, 14.0%). The 2:4-dinitrophenylhydrazone separates from acetic acid solutions in orange-red needles, m. p. 266–268° (Found: N, 13.4. C<sub>21</sub>H<sub>18</sub>O<sub>6</sub>N<sub>4</sub> requires N, 13.3%).

*Diacetyldihydrodunnione.*—*Dunnione* was recovered unchanged (m. p. and mixed m. p.) after being heated with acetic anhydride in the presence of pyridine at 100°. Boiling with acetic anhydride and anhydrous sodium acetate gave a dark bluish-green amorphous solid which could not be crystallised.

Acetylation was then carried out with hot acetic anhydride and a little pyridine in the presence of zinc dust. The product was crystallised several times from methyl alcohol and obtained as colourless, equidimensional prisms, m. p. 143–144°.  $[\alpha]_D^{25} + 16.7^\circ$  (*c*, 3.236 in chloroform) [Found: C, 68.9; H, 6.0; Ac, 27.1. C<sub>15</sub>H<sub>14</sub>O(OAc)<sub>2</sub> requires C, 69.5; H, 6.1; Ac, 26.2%].

In order to determine whether diacetyldihydrodunnione contains a carbonyl group, 0.5 g. was heated on the steam-bath for 4 hours with semicarbazide hydrochloride (0.5 g.), sodium acetate (0.75 g.), water (1 c.c.), and ethyl alcohol (15 c.c.). After keeping overnight, the mixture was diluted with water, and the precipitate collected and crystallised from methyl alcohol. The product was a mixture of two kinds of crystals, which were separated by hand. Concentration of the mother-liquor afforded a further crop, which was also hand-picked and combined with the main fraction. (a) Colourless prisms, twice crystallised from methyl alcohol, m. p. and mixed m. p. with diacetyldihydrodunnione, 141–143°; (b) yellow needles, in much smaller quantity than (a), twice crystallised from methyl alcohol, m. p. and mixed m. p. with dunnione semicarbazone, 232–233°.

*Dunnione and o-Phenylenediamine.*—A mixture of dunnione (0.5 g.), *o*-phenylenediamine (0.3 g.), and glacial acetic acid (5 c.c.) was heated to boiling and then kept for 10 minutes with occasional gentle heating. The yellow precipitate thrown down on dilution with water was collected, washed with water, and crystallised thrice from methyl alcohol, being obtained as yellow prisms, m. p. 140–141° (Found: C, 79.6; H, 5.7; N, 8.7. C<sub>21</sub>H<sub>18</sub>ON<sub>2</sub> requires C, 80.3; H, 5.7; N, 8.9%). As in the case of  $\beta$ -lapachone, the occurrence of a second form consisting of yellow needles, m. p. ca. 125°, was observed. The *azine* dissolves in alcohol or acetone to a yellow solution exhibiting a strong green fluorescence; its solutions in concentrated mineral acids have an intense red colour.

*β-Lapachone and o-Phenylenediamine.*—The condensation product, prepared as in the case of dunnione, crystallised from methyl alcohol and finally from aqueous acetic acid, as flat yellow plates, m. p. 133.5—134°. The lower-melting form described by Hooker (*J. Amer. Chem. Soc.*, 1936, 58, 1190) was also isolated.

*Behaviour of Dunnione with Alkalis, and Comparison with Lapachol Derivatives.*—Dunnione is almost insoluble in cold aqueous sodium carbonate or dilute sodium hydroxide, but dissolves slowly on standing with more concentrated alkali (e.g., 5% NaOH) to a red solution. This was best prepared by shaking a concentrated solution of dunnione in light petroleum with aqueous sodium hydroxide. The solution had the following properties:

(i) When it was shaken with *isoamyl* alcohol, the red colour was wholly transferred to the alcoholic layer; with ethyl acetate the colour was distributed between the two solvents, and benzene extracted none of the colouring matter.

(ii) Treatment of the *isoamyl*-alcoholic solution from (i) with a few drops of 1% hydrochloric acid gave a yellow solution and the red colour was immediately regenerated on addition of sodium carbonate or hydroxide.

(iii) On addition of an excess of acid to the red alkaline solution, the colour changed through yellow to orange, *i.e.*, the normal colour of an aqueous solution of dunnione. In an experiment on a larger scale, dunnione was dissolved in the minimum amount of methyl alcohol, and a large excess of aqueous hydroxide (5%) added. After 2—3 minutes the mixture was shaken with benzene to remove unchanged dunnione and the red aqueous layer was acidified with hydrochloric acid. On extraction with benzene, concentration, and crystallisation from light petroleum, dunnione was recovered, m. p. and mixed m. p. 98—99°.

(iv) On heating, it became straw-coloured, and acidification afforded a yellow crystalline precipitate. Complete extraction of this yellow substance was effected with benzene. When the benzene solution was shaken with alkali, only a part of the yellow substance was transferred to the alkali, and re-acidification again resulted in partial conversion into the alkali-insoluble product. The conversion was completed by repeating two or three times the alkali extraction and re-acidification.

A solution of lapachol in sodium hydroxide had the same red colour as that of dunnione referred to above. Its properties in respect of tests (i)—(iii) were identical with those of the dunnione solution, but after boiling for long periods the colour remained unchanged, and *β*-lapachone was recovered by addition of an excess of acid. Both *α*- and *β*-lapachone on warming with alkali gave a red solution whose properties were similar to that of lapachol.

*alloDunnione.*—This substance was prepared by heating dunnione with aqueous sodium hydroxide (10%) until the red colour disappeared. It was found best to work with a series of small batches of 0.2—0.5 g., as otherwise some decomposition occurred. The hot solution was acidified with concentrated hydrochloric acid, cooled, and extracted with benzene, and the benzene washed with alkali, which was re-acidified and extracted as described above. The combined benzene solutions were dried and concentrated, and the residue crystallised from light petroleum. The substance separated in yellow needles, m. p. 161—162° [Found: C, 74.3; H, 5.9; C-CH<sub>3</sub>, 7.7; *M* (Rast), 266. C<sub>15</sub>H<sub>14</sub>O<sub>3</sub> requires C, 74.4; H, 5.8; 1 C-CH<sub>3</sub>, 6.2%; *M*, 242].

*alloDunnione* is generally less readily soluble than dunnione, but nevertheless dissolves freely in most organic solvents; it is sparingly soluble in light petroleum and almost insoluble in water. It dissolves in cold concentrated sulphuric acid to an orange-red solution, from which it can be recovered unchanged (m. p. and mixed m. p.). It is insoluble in cold aqueous sodium bisulphite solution. Decolorisation takes place on treatment of an alcoholic solution with zinc and hydrochloric acid. It dissolves in 5—10% sodium hydroxide on heating, giving a yellow solution, and regeneration by acidification of this solution follows the same course as in the preparation from dunnione. It was recovered unchanged (m. p. and mixed m. p.) when condensation with *o*-phenylenediamine was attempted. It was also recovered unchanged after attempted acetylation with acetic anhydride and pyridine.

*alloDunnione 2:4-dinitrophenylhydrazone* separated from acetic acid in yellow-orange needles, m. p. 315° (decomp.), after darkening at about 290° (Found: N, 13.7. C<sub>21</sub>H<sub>18</sub>O<sub>6</sub>N<sub>4</sub> requires N, 13.3%).

*α-Dunnione.*—When dunnione was kept with aqueous sodium hydroxide (5%) at room temperature for 2 days, a part dissolved to a red solution, the colour of which changed slowly to yellow; the remainder was converted into a yellow solid, deposited at the bottom of the vessel. The mixture was shaken with benzene, and the benzene layer washed with a little 1% hydrochloric acid, dried, and concentrated. The residue crystallised from light petroleum in

flat, yellow plates, m. p. 121—122°, mixed m. p. with *alloodunnione*, ca. 110° (Found: C, 74.5; H, 6.2.  $C_{16}H_{14}O_3$  requires C, 74.4; H, 5.8%). The alkaline liquid after extraction with benzene was acidified and yielded, on being worked up in the usual manner, *alloodunnione*, m. p. 161—162°.

$\alpha$ -*Dunnione* was also prepared as follows. *Dunnione* was heated under reflux for 2 hours with 20% hydrochloric acid, and the solution filtered, cooled, and shaken with benzene. The benzene was washed with sodium carbonate solution and concentrated, and the residue twice crystallised from light petroleum. The product had m. p. 120—122° alone or mixed with a specimen prepared as above. When *dunnione* was heated with hydrobromic acid (*d* 1.49) at 70° for an hour, it was recovered unchanged (m. p. and mixed m. p.).

$\alpha$ -*Dunnione* dissolved in concentrated sulphuric acid to a brilliant red solution, from which it was recovered unchanged after keeping at room temperature for 2 hours (m. p. and mixed m. p.). When boiled with 5% aqueous sodium hydroxide,  $\alpha$ -*dunnione* slowly dissolved and *alloodunnione*, m. p. and mixed m. p. 160—162°, was obtained from the solution after acidification. Evidently the change to *alloodunnione* takes place more rapidly than solution of the  $\alpha$ -*dunnione*, as the alkaline solution was yellow throughout. However, when  $\alpha$ -*dunnione* was dissolved in a little alcohol and 5% aqueous sodium hydroxide added, a deep red solution was obtained which behaved in the same way as an alkaline solution of *dunnione*. This red solution was shaken with benzene to remove any unchanged  $\alpha$ -*dunnione*, acidified with hydrochloric acid, and extracted with benzene. After washing with sodium carbonate, the benzene was concentrated, and the residue crystallised from light petroleum; m. p. 95—97°, mixed m. p. with *dunnione* 96—98°.

$\alpha$ -*Dunnione* 2:4-dinitrophenylhydrazone crystallised from acetic acid in reddish-orange plates, m. p. 278—280° (Found: N, 13.2.  $C_{21}H_{18}O_6N_4$  requires N, 13.3%).

$\beta$ -*isoDunnione*.— $\alpha$ -*Dunnione*, dissolved in concentrated sulphuric acid, was heated at 100° for  $\frac{1}{2}$  hour, the colour changing from intense red to brownish-orange. The solution was cooled, poured into water, and extracted with benzene, and the extract washed with aqueous sodium carbonate. The residue after concentration of the benzene was twice crystallised from light petroleum and obtained as orange-red needles, m. p. 129—131° (Found: C, 74.4; H, 5.7.  $C_{16}H_{14}O_3$  requires C, 74.4; H, 5.8%). The substance dissolves in aqueous sodium bisulphite to a colourless solution, from which it is regenerated by alkalis. It is markedly sensitive to sunlight, turning brown after a few minutes' exposure. On heating with aqueous sodium hydroxide it dissolves to a red solution, which, in contrast to that obtained from *dunnione*, remains red after boiling for 5 minutes.

$\alpha$ -*isoDunnione*.— $\beta$ -*isoDunnione* was heated under reflux for  $\frac{1}{2}$  hour with 20% hydrochloric acid, and the solution diluted with 2 vols. of water and allowed to cool. The yellow crystals which separated were recrystallised from aqueous alcohol (needles) and then had m. p. 118—119° (mixed m. p. with  $\alpha$ -*dunnione*, ca. 90°) (Found: C, 74.3; H, 5.8.  $C_{15}H_{14}O_3$  requires C, 74.4; H, 5.8%).

$\alpha$ -*Lapachone* 2:4-dinitrophenylhydrazone crystallised from acetic acid as flat, brownish-orange plates, m. p. 277—278° (Found: N, 13.6.  $C_{21}H_{18}O_6N_4$  requires N, 13.3%).  $\beta$ -*Lapachone* 2:4-dinitrophenylhydrazone separated from acetic acid in orange needles, m. p. 283—285°, after sintering at about 250° (Found: N, 13.5.  $C_{21}H_{18}O_6N_4$  requires N, 13.3%).  $\beta$ -*Lapachone* (Found: C-CH<sub>3</sub>, 3.67. Calc. for 1 C-CH<sub>3</sub>, 6.2%).

*Oxidation Experiments*.—(a) *Dunnione* (1 g.) was heated at 70—80° with water (1 l.) containing a little sulphuric acid, and aqueous potassium permanganate was added until no further oxidation took place. The cooled solution was extracted thrice with ether, the combined extracts dried and distilled, and the residue washed with light petroleum to remove a little oily material. There remained 0.4 g. of a white solid which after repeated crystallisation from water had m. p. 198—200°, alone or mixed with phthalic acid (Found: C, 57.9; H, 4.0. Calc. for  $C_8H_6O_4$ : C, 57.8; H, 3.6%).

(b) *alloodunnione* was treated in warm acetic acid solution with 3% hydrogen peroxide until the colour became pale yellow. The solvent was removed under reduced pressure, the residue taken up in benzene, and the benzene extracted with sodium hydroxide solution. The alkaline extract was acidified and shaken with ether, which was separated, dried, and concentrated. The residual white solid (positive fluorescein reaction) was boiled with aniline, and the product isolated in the usual manner; m. p. 206—207°, mixed with phthalanil, m. p. 207—208°.

(c) A mixture of *dunnione* (1 g.), aqueous sodium hydroxide (8 c.c. of 10%), and water (50 c.c.) was boiled in a distilling flask, and hydrogen peroxide (3%) added drop by drop until the solution became straw-coloured. The distillate and vapour were passed into a solution of

2 : 4-dinitrophenylhydrazine in 5% aqueous sulphuric acid, and the resulting precipitate twice crystallised from alcohol. It had m. p. 162—164°; mixed m. p. with acetaldehyde-2 : 4-dinitrophenylhydrazone, 163—165° (Found : C, 43.6; H, 3.6. Calc. for  $C_8H_8O_4N_4$  : C, 42.9; H, 3.6%). The residue in the distilling flask was filtered and acidified, giving an immediate crystalline precipitate (A), which was collected. The filtrate was extracted with ether, the ethereal solution dried and concentrated, and the residue washed with the minimum amount of cold ether. This left a white solid (B), which was twice crystallised from water; it then had m. p. 195—197° and mixed m. p. with phthalic acid, 196—198°. (A), which was produced in greater amount than (B), was thrice crystallised from alcohol and obtained in small, colourless prisms, m. p. 205—206°. It did not give a fluorescein reaction.

(d) *allo*Dunnione was oxidised with alkaline peroxide as in (c). The end-point was more difficult to recognise and some unchanged *alldunnione* was recovered. The distillate contained acetaldehyde, identified by the m. p. and mixed m. p., 165—167°, of its 2 : 4-dinitrophenylhydrazone. The acid fraction was thrice crystallised from aqueous acetone and once from alcohol; it was then obtained in colourless prisms, m. p. 205—206°, not depressed by admixture with the product (A) from the oxidation of dunnione. The two specimens were combined and again crystallised from aqueous acetone; the m. p. was unaltered [Found : C, 65.05; H, 5.5;  $C-CH_3$ , 3.52;  $M$  (Rast), 240.  $C_{12}H_{12}O_4$  requires C, 65.45; H, 5.5; 1  $C-CH_3$ , 6.8%;  $M$ , 220].

(e) For comparison, lapachol and  $\beta$ -lapachone were oxidised with alkaline peroxide. In each case the products were phthalic acid (m. p. and mixed m. p. 196—198°; anil, m. p. and mixed m. p. 206—208°), and acetone (2 : 4-dinitrophenylhydrazone, m. p. and mixed m. p. 126—127°).

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