

## SYNTHETICAL EXPERIMENTS IN THE FLAVONE AND ISOFLAVONE GROUPS.

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(Read at Symposium, September 26-27, 1938.)

The wide occurrence of chromone derivatives in the roots, stems, bark, fruits, flowers and leaves of plants and their intimate relationship to the anthocyan group constituting the red, blue and purple pigments of fruits and flowers have stimulated wide interest in the chemical constitution of the naturally occurring representatives and methods for their synthesis. Their origin in plants is not known with certainty; they may originate at the point of vegetation, but are more frequently formed in the older tissue and their occurrence and definite localization often clearly indicate the relationship of plants in particular families, genera and species (Klein and Werner, *Z. physiol. Chem.*, 1925, **143**, 9). Their role in the plant economy is also somewhat uncertain; it has been suggested (Rouge, *Bull. Soc. Bot. Geneva*, 1921, **13**, 18) that they take part in the process of assimilation by absorbing oxygen and transporting it away from the cell. According to Ruzsuyak and Szent-Gyorgyi (*Nature*, 1936, **138**, 27) this great group of vegetable dyes is allied to the vitamins, fulfilling an important function in plant and animal life. While flavones have been isolated from many plants of medicinal value, such as *Digitalis thapsi*, *Calycopteris floribunda*, *Cuscuta reflexa* and *Saponaria officinalis*, they do not appear to possess any pharmacological properties of practical interest (cf. Mahal, *Proc. Ind. Acad. Sc.*, 1937, **5**, 186).

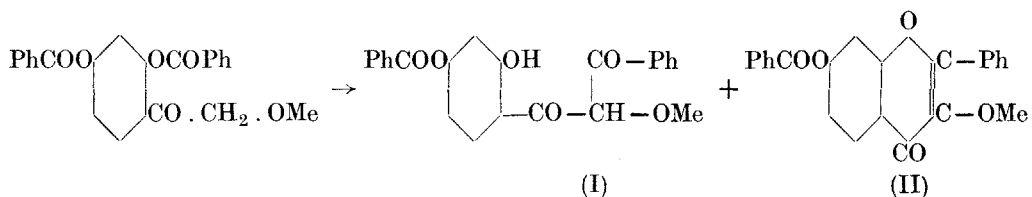
The older methods, due largely to Kostanecki, for the synthesis of flavones and flavonols had certain limitations (Venkataraman, *J. Ind. Chem. Soc., Sir P. C. Ray Commem. Vol.*, 1933, 28), which led Robinson to seek a reaction of wider utility (Allan and Robinson, *J. Chem. Soc.*, 1924, **125**, 2193 *et sequa*). The possibilities of an early observation of Tahara (*Ber.*, 1892, **25**, 1302) regarding the prolonged action of acetic anhydride on resacetophenone soon became apparent and a general synthesis of flavones and flavonols was developed by Robinson, in which *o*-hydroxyaryl alkyl ketones were heated with the appropriate acid anhydride and the sodium salt of the acid. Thus the interaction of benzoic anhydride and phloracetophenone and alkaline hydrolysis of the product yielded chrysin, the colouring matter of poplar buds (Robinson and Venkataraman, *J. Chem. Soc.*, 1926, **129**, 2344). By the application of the Robinson method in its simplest form or involving in addition methylation or complete demethylation with hydriodic acid, the following naturally occurring flavones and flavonols have been synthesised: flavone (the parent

member of the series), chrysin, acacetin, pratol, luteolin, galangin, k ampferol, fisetin, quercetin, morin, robinetin, herbacetin, tangeritin, myricetin, gossypetin and quercetaletin.

The Robinson reaction proceeded smoothly in the case of  $\omega$ -substituted ketones derived from *m*-dihydric phenols, e.g.  $\omega$ -methoxyresacetophenone and  $\omega$ -methoxyphloracetophenone, but although several naturally occurring and other flavones (unsubstituted in the 3-position) have been successfully prepared by the Robinson method, a careful study of the process revealed complexities. Under the right conditions the chromone condensation is applicable not only to resacetophenone and its derivatives, but also to *o*-hydroxyacetophenone, 2:5-dihydroxyacetophenone, 1-acetyl-2-naphthol and 2-acetyl-1-naphthol. It was found, however, in the case of 2-acetyl-1-naphthol that the major product of the reaction was a 3-acyl chromone and it has since been shown that such 3-acylation is a source of confusion and experimental difficulty in carrying out the Robinson synthesis (*J. Chem. Soc.*, 1931, 1165; 1933, 1074; Anderson, *Canadian J. Research*, 1932, 7, 285). The mechanism of the reaction and alternative procedures precluding 3-acylation were therefore investigated. Failure to effect ring closure to the  $\gamma$ -pyrone in *o*-acyl derivatives of *o*-hydroxyketones (e.g. in 2-acetyl-1-naphthyl benzoate and in 2-phenylacetyl-1-naphthyl benzoate in which the  $\omega$ -phenyl might be expected to facilitate chromone formation) indicated that the commonly assumed mechanism of the reaction (cf. Wittig, Baugert and Richter, *Ann.*, 1925, 446, 155) needed experimental verification. At least one example, however, of direct dehydration of an *o*-benzoyloxyphenyl alkyl ketone to the chromone is available in the conversion of  $\omega$ -2:4:6-tetrabenzoyloxyacetophenone to galangin tribenzoate by means of boiling alcoholic potassium acetate (Chavan and Robinson, *J. Chem. Soc.*, 1933, 368).

The action of an acid anhydride and the sodium salt of an acid on a phenolic ketone may lead to the acyl derivative of the ketone, a chromone, a 3-acylated chromone, or a coumarin. The reaction could be limited to the formation of the *o*-acyl derivative of the ketone by brief heating, preferably in the absence of the sodium salt and the presence of pyridine. The course of the reaction with regard to the other three alternatives is dependent both on the nature of the ketone and of the acid anhydride. An  $\omega$ -substituent is an aid to chromone formation, which also takes place more readily in the naphthalene than in the benzene series. With an *o*-hydroxyaryl methyl ketone, the deciding factor is essentially the acid anhydride. The anhydride of an acid such as phenylacetic acid containing a reactive methylene group yields the coumarin always. Coumarin formation being precluded in the case of aromatic acid anhydrides, flavones and 3-acyl flavones are formed. The interaction of aliphatic acid anhydrides with *o*-hydroxyaryl methyl ketones is of a complicated character and both coumarins and chromones (including 3-acyl chromones) are producible.

So far as the action of aromatic acid anhydrides on *o*-hydroxyaryl methyl ketones is concerned, the mechanism of chromone synthesis has been demonstrated by Baker (*J. Chem. Soc.*, 1933, 1381) as involving the intermediate formation of a dibenzoylmethane. While Baker achieved the transformation of *o*-acyloxyacetophenones to the dibenzoylmethanes by means of potassium carbonate in toluene at 100°, the tautomerisation of 2-acetyl-1-naphthyl benzoate in ether solution to  $\omega$ -benzoyl-2-acetyl-1-naphthol by means of sodamide at room temperature was observed simultaneously in Lahore (Mahal and Venkataraman, *Current Science*, 1933, 4, 214; *J. Chem. Soc.*, 1934, 1767). As the diketone could be cyclised to the flavone also at room temperature, the process was of interest from the point of view of the phytochemical synthesis of flavones and flavone glykosides. Other observations relating to the reaction mechanism were the isolation of a 2-hydroxy-naphthaflavanone, a type postulated by Baker as an intermediate stage in the conversion of his diketones into flavones, from the products of the interaction of 2-benzylacetyl-1-naphthol with ethyl formate and sodium (Cheema and Venkataraman, *J. Chem. Soc.*, 1932, 918); and the thermal transformation (by distillation in high vacuum) of  $\omega$ -methoxyresacetophenone dibenzoate to a mixture of the diketone (I) and the flavone (II) (Mahal, Ph.D. thesis, Panjab University, 1936).

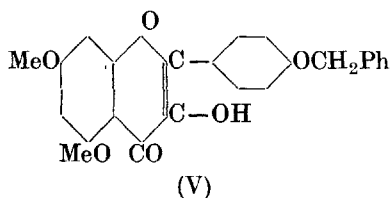
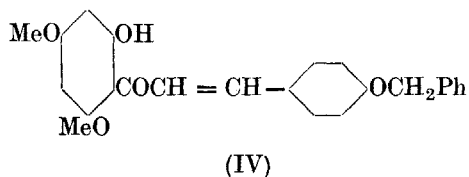
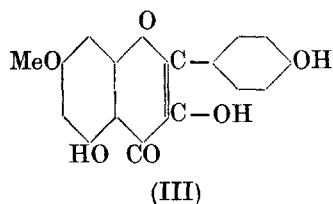


Modifying the original Robinson method by using benzyloxy or benzyloxy-derivatives in the ketone or acid anhydride parts of the reaction mixture and taking advantage of the more facile removal of the benzoyl and benzyl groups in comparison with methyl ethers, partially methylated polyhydroxyflavones—kämpferide, syringetin, diosmetin, tricetin—have been prepared (Heap and Robinson, *J. Chem. Soc.*, 1926, 2336; 1929, 68; Lovecy, Robinson and Sugasawa, *J. Chem. Soc.* 1930, 817; Gulati and Venkataraman, *J. Chem. Soc.*, 1933, 942).

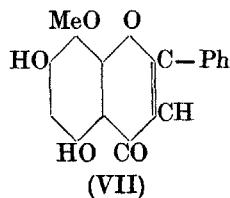
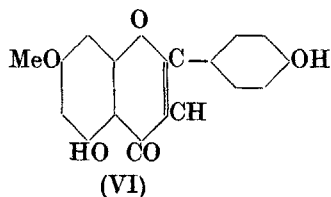
Selenium dioxide is a specific oxidising agent for the conversion of methylene to carbonyl (Müller, *Ber.*, 1933, 66, 1668; Evans, Ridgeon and Simonsen, *J. Chem. Soc.*, 1934, 137), but when 2:3-dihydro- $\alpha$ -naphthaflavone in xylene was heated with selenium dioxide, the product was the naphthaflavone and not the naphthaflavonol. *o*-Hydroxychalkones may thus be directly oxidised to flavones (Mahal, Rai and Venkataraman, *J. Chem. Soc.*, 1935, 866). 7-Methoxy-4'-hydroxyflavone was synthesised in this manner from 2-hydroxy-4-methoxyphenyl 4'-benzyloxystyryl ketone, followed by debenylation, and found to be different from a hydroxymethoxyflavone isolated by Adrian and

Trillat (*Comp. rend.*, 1899, **129**, 889) from a species of *Digitalis*. In view of the smooth dehydrogenation of chalcones and flavanones to flavones, the utility of selenium dioxide for the dehydrogenation of hydroaromatic to aromatic compounds has been examined; by heating tetrahydronaphthalene with selenium oxide at 160° for 24 hours a 30% yield of naphthalene was obtainable (Mahal, *loc cit.*).

Algar and Flynn (*Proc. Royal Irish Acad.*, 1934, Series B, 42, separate issue) have accomplished the synthesis of flavonols from chalcones by oxidation with alkaline hydrogen peroxide. Attempting to utilise this reaction for the synthesis of kempferol 7-methyl ether (III), a substance of interest with regard to the structure of rhamnocitrin (Oesch and Perkin, *J. Chem. Soc.*, 1914, **105**, 2332), 2-hydroxy-4:6-dimethoxyphenyl 4-benzyloxystyryl ketone (IV) was treated with alkaline hydrogen peroxide under a variety of conditions, but the flavonol (V), which could then be debenzylated and partially demethylated to (III), was not obtained, the pale yellow needles, m.p. 198°, exhibiting the properties of a flavanone.

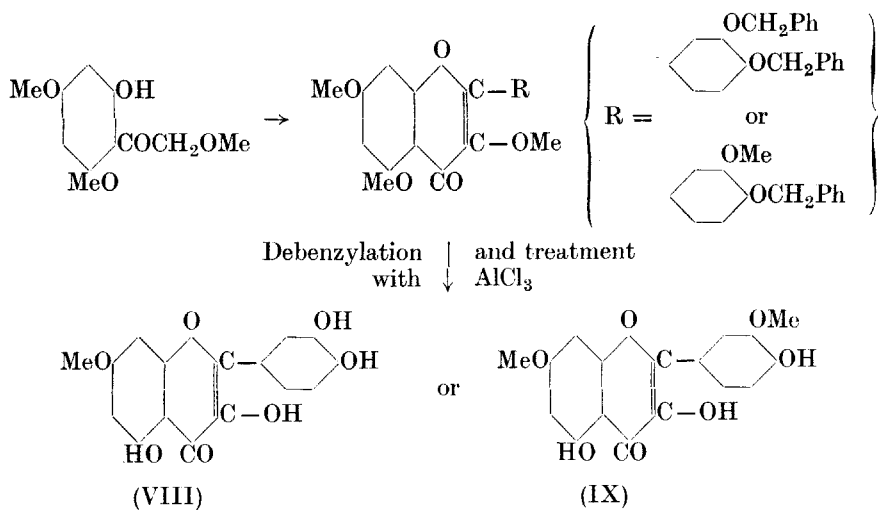


The 5-hydroxyl in a flavone is analogous to the *o*-hydroxyl of a ketone, chelation being possible in both cases; methylation can only be effected under special conditions and the methyl ether is more readily demethylated than methoxyls in other positions. Such partial demethylation of polymethoxyflavones to the 5-hydroxy derivatives can be carried out with aluminium chloride, a process which has proved useful for the synthesis of partially methylated polyhydroxyflavones. Genkwanin (VI) and wogonin (VII) have thus



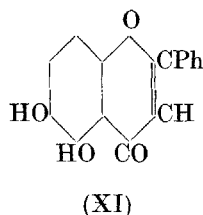
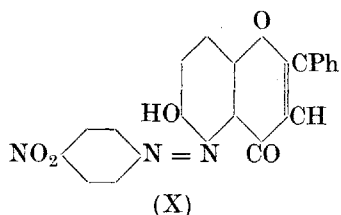
been synthesised (Mahal and Venkataraman, *J. Chem. Soc.*, 1936, 569; Mehta, Shah and Wheeler, *in print*), and the reaction has thrown light on the constitution of calycopterin, although further work has shown that the suggested structure (Mahal and Venkataraman, *Current Science*, 1935, 311) must be confirmed by additional evidence, which is now being sought. While the synthesis of 5:7:2':4'-tetrahydroxyflavone (Robinson and Venkataraman, *J. Chem. Soc.*, 1929, 66; cf. Cullinane, Algar and Ryan, *Proc. Royal Soc. Dublin*, 1928, 19, 77) leaves little doubt regarding its non-identity with lotoflavin (Dunstan and Henry, *Phil. Trans.*, 1901, 194, 515), the properties of lotoflavin trimethyl ether, which crystallised in dimorphic forms with different melting points, were more characteristic than those of lotoflavin. It would, therefore, be desirable to synthesise 5-hydroxy-7:2':4'-trimethoxyflavone, for comparison with lotoflavin trimethyl ether, by the controlled demethylation with aluminium chloride of the tetramethyl ether, which may be conveniently prepared by the selenium dioxide oxidation of 2-hydroxy-4:6-dimethoxyphenyl 2:4-dimethoxystyryl ketone.

The 3-methoxyl group in a flavone is also sensitive to hydrolysis by aluminium chloride, and this may be utilised for the preparation of methoxyflavonols, the following being feasible schemes for the synthesis of rhamnetin (VIII) and rhamnazin (IX):



The reactivity of plant dyestuffs derived from resorcinol and phloroglucinol towards diazo salts has been studied by Perkin (*J. Chem. Soc.*, 1895, 67, 933), who used it for characterising such phenolic colouring matters. While Perkin found that chrysin, apigenin, euxanthone and catechin all gave *dis-azo* dyes, the coupling of 6-hydroxyflavone with diazotised *p*-nitraniline yields a *mono-azo* dye, which in consideration of the fixation of the double

bonds in the fused benzene ring of the chromone nucleus may be assumed to have the structure (X) (Mahal and Venkataraman, *Current Science*, 1938, 6, 450). A route to the synthesis of

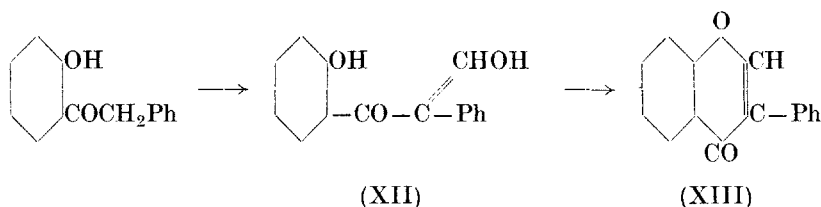


primetin (XI) then becomes possible. The coupling of chrysin or its 7-benzyl ether with a diazo salt of low coupling energy might lead to the 8-*mono*-azo dye, a reaction of potential value for the transformation of 5:7-dihydroxyflavones into the 5:7:8-trihydroxy derivatives (e.g. chrysin→norwogonin; 7-benzyl chrysin→wogonin; quercetin→gossypetin). Likewise the coupling of 5- or 8-hydroxyflavones with a suitable diazo salt may be utilised for the synthesis of the 5:8-dihydroxy derivatives. Such an orientation of hydroxyls has been suggested for the colouring matter of *Gingko biloba* (Furukawa, *Sci. Papers Inst. Phys. Chem. Res., Tokyo*, 1933, 21, 278).

Nierenstein's conversion of quercetin to gossypetin has been unconfirmed (Baker, Nodzu and Robinson, *J. Chem. Soc.*, 1930, 922) and norwogonin (Hattori, *Acta Phytochim.*, 1932, 6, 183) bore no resemblance to Nierenstein's 8-hydroxychrysin (*Ber.*, 1912, 45, 490). Attempts in this laboratory to oxidise chrysin by means of chromic anhydride and with other oxidising agents (nitric acid, potassium persulphate, selenium dioxide) did not lead to any homogeneous material other than the starting substance. Oxidation of 5-hydroxy-6-benzyl-7-benzoyloxyflavone also gave negative results. The ready oxidisability of pyrogallol trimethyl ether to 2:6-dimethoxyquinone suggested that oxidation of 7:8-dihydroxyflavone might yield norwogonin, but numerous attempts to oxidise this flavone, as well as its dimethyl and dibenzyl ethers, have been fruitless; further experiments on these lines are, however, indicated.

While flavones have been extensively studied since 1873, our knowledge of the *isoflavone* group is much more recent. The first natural product to be recognised as an *isoflavone* was prunetin (Finnemore, *Pharm. J.*, 1910, 31, 1761); Baker and Robinson (*J. Chem. Soc.*, 1926, 127, 2713) demonstrated the identity of demethyl prunetin or 5:7:4'-trihydroxy*isoflavone* with genistein, the colouring matter of dyer's broom. Since then several other *isoflavones* have been isolated from plants. The first synthesis of an *isoflavone* was carried out by Baker and Robinson (*J. Chem. Soc.*, 1925, 127, 1981) by the oxidation of 7-methoxy-2-styryl-3-phenylchromone to the 2-carboxylic acid, which was decarboxylated to 7-methoxy*isoflavone*. Genistein and daidzein

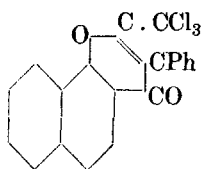
were later synthesised similarly, but in the case of irigenol (5:6:7:3':4':5'-hexahydroxyisoflavone) the oxidation of the 2-styryl derivative of the hexamethyl ether proved impracticable (Baker and Robinson, *J. Chem. Soc.*, 1929, 135, 152). Baker, Pollard and Robinson (*J. Chem. Soc.*, 1929, 135, 1468) developed a second synthesis of isoflavones involving a Hoesch reaction on the cyanhydrin of an  $\omega$ -phenoxyacetophenone, which was applied by Späth and Lederer (*Ber.*, 1930, 63, 743) to  $\psi$ -baptigenin, but was inadequate for other syntheses in the field (Baker, Morgan and Robinson, *J. Chem. Soc.*, 1933, 143, 374). The third synthesis, due to Späth and Lederer, consisted in heating derivatives of 2-hydroxyphenyl benzyl ketone with ethyl formate and sodium in a sealed tube, followed by treatment of the reaction mixture with boiling alcohol and fuming hydrochloric acid in order to effect cyclisation of the oxy-methylene compound (XII), assumed to be an intermediate product, to the isoflavone (XIII).



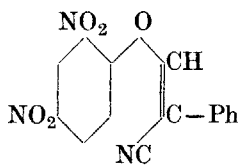
By sublimation in high vacuum, poor yields of the isoflavones ( $\psi$ -baptigenin, daidzein and formononetin) were obtained (Späth and Lederer, *loc. cit.*, Wessely, Kornfeld and Lechner, *Ber.*, 1933, 66, 685). The action of sodium on an ice-cooled solution of *o*-hydroxyphenyl benzyl ketones in ethyl formate proceeds, however, in one stage to the isoflavone (Joshi and Venkataraman, *J. Chem. Soc.*, 1934, 513); a series of isoflavones—*isoflavone*, the parent member, 7-hydroxyisoflavone,  $\alpha$ - and  $\beta$ -naphthaisoflavone, formononetin, daidzein and  $\psi$ -baptigenin—have thus been synthesised with simplicity and in excellent yields; and the method should also be applicable to irigenol along obvious lines.

Some negative experiments in the isoflavone group may be recorded. The action of trichloroacetic anhydride and potassium trichloroacetate on 2-phenylacetyl-1-naphthol gave a substance, which had the qualitative properties of 2-trichlormethyl-3-phenyl-1:4- $\alpha$ -naphthapyrone (XIV), but it could not be crystallised and hydrolysis to the 2-carboxylic acid and decarboxylation to the naphthaisoflavone were not accomplished.

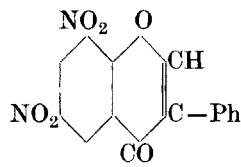
2:4-Dinitrobrombenzene condensed with  $\alpha$ -formylphenylacetonitrile in pyridine solution to give 2:4-dinitrophenoxymethylene benzyl cyanide (XV). Saturation of an ethereal solution of (XV) with hydrogen chloride gave a substance, m.p. 188°, which was not the dinitroisoflavone (XVI); nor was it the pyrone-imide (XVII),



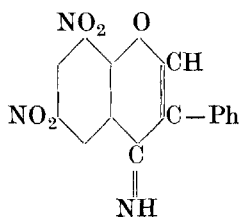
(XIV)



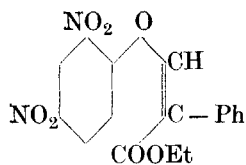
(XV)



(XVI)

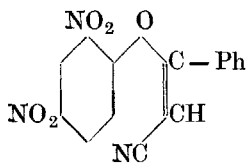


(XVII)

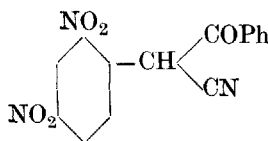


(XVIII)

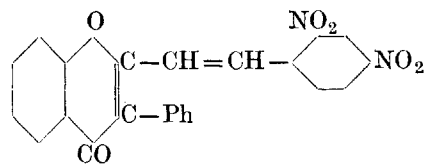
being unaffected by boiling concentrated hydrochloric acid and syrupy phosphoric acid. Similarly to (XV), the dinitrophenoxymethylene phenylacetic ester (XVIII) was prepared; attempts to effect ring closure to (XVI) were unsuccessful.



(XIX)



(XX)



(XXI)

Condensing 2:4-dinitrobenzene with benzoylacetonitrile in presence of sodium ethylate, two substances, the *O*-phenyl and the *C*-phenyl derivatives (XIX) and (XX), were isolated. Treatment of (XIX) with hydrogen chloride in ether yielded the unconverted substance or uncrystallisable material.

In the Baker-Robinson method for the synthesis of isoflavones by the oxidation of their 2-styryl derivatives, the possibility of the use of the dinitrostyryl compound (XXI) leading to more facile oxidation could not be examined, since 2:4-dinitrobenzaldehyde did not condense with 2-methyl-3-phenyl-1:4- $\alpha$ -naphthopyrone, which reacts readily with benzaldehyde, anisaldehyde and veratraldehyde (Cheema, Gulati and Venkataraman, *J. Chem. Soc.*, 1932, 926).

$\beta$ -Naphthol condenses with ethyl acetoacetate to give a chromone (Dey and Lakshminarayanan, *J. Indian Chem. Soc.*, 1932, 9, 153), but the  $\alpha$ -pyrone was obtained in the case of  $\alpha$ -formylphenylacetic ester.