[Contribution from the Department of Chemistry, University of California, Los Angeles]

SOME OBSERVATIONS ON THE STRUCTURE OF LIMONIN

T. A. GEISSMAN and VSEVOLOD TULAGIN

Received April 22, 1946

Limonin, first isolated from *Citrus* spp. (1) and since recognized in several other plants (2, 3, 4), is a member of a fairly large but relatively little-studied group of substances known generally as "bitter principles." These substances appear to form a well-defined class of naturally-occurring, nitrogen-free materials, possessing certain characteristics in common although being rather divergent in molecular size and perhaps in fundamental structure. The most common characteristic of these substances, besides their bitter taste, is that they are lactones. Where information is available it appears that the bitter principles possess complex alicyclic structures, or heterocyclic structures involving oxygen atoms in ether or acetal linkages. The presence of mobile ether linkages would be in accord with the ease with which isomerization (and in some cases degradation) of the molecules of many of these substances can be brought about by the action of acids. Indeed, this fact may bear some relationship to the observation that isomeric or closely similar bitter principles are frequently found associated in the same plant. For example, limonin has been reported to occur along with isolimonin (5), obacunoic acid (6), dictamnolide (6), obacunone (7), and dictamnolic acid (7). The bitter substances, with some exceptions, give acetone upon fusion with alkali and possess neither methoxyl, methylenedioxyl, nor acetyl groups.

Interest in the structure of liminon is heightened by the fact that it probably is concerned in the development of a bitter taste in expressed Navel orange juice, a problem of considerable economic importance to the citrus industry. It is to be hoped that precise information on the structures of limonin and of isolimonin, which is isolated along with limonin from citrus fruits, will be of value in solving the problem of producing stable, palatable Navel orange juice.

Up to the time the present work was begun the available information on the structure of limonin was scanty. Since the isolation of the bitter principle in 1841 (1) it has been investigated in a number of laboratories (2, 5, 8, 9, 10, 11, 12, 13, 14). The results of these investigations can be summarized briefly as follows:

Limonin is a dilactone having the composition $C_{26}H_{30}O_8$. It dissolves in concentrated sulfuric acid with the formation of a red-brown solution from which limonin cannot be recovered on dilution. It shows no indication of possessing a (reactive) carboxyl group or hydroxyl groups, and does not react with bromine in chloroform in the cold. Limonin does not react with Tollens' reagent nor does it give a positive Legal test. When oxidized with manganese dioxide in the presence of sulfuric acid it yields benzenepentacarboxylic acid. Upon fusion with alkali under very drastic conditions, followed by selenium dehydrogenation of the products of this treatment, 1,2,5-trimethylnaphthalene is formed. This hydrocarbon was also obtained by the zinc-dust distillation of columbin (15). Among the products of alkali fusion not subsequently treated with selenium was isophthalic acid. Limonin has been shown to be different from columbin (9), a bitter principle which it closely resembles, and identical with obaculactone, evodin, and dictamnolactone (13).

The results of the catalytic hydrogenation of limonin gave early promise of useful results, but a more recent reexamination of this method of attack has cast some doubt upon the validity of some of the previously reported results. Koller and Czerny (5) reported the isolation of "hexahydrolimonic acid" and "tetrahydrolimonin," the latter substance probably being the same as one later obtained by Feist (11). Schechter and Haller (16) were unable to duplicate these findings.[1] In view of these conflicting reports the hydrogenation of limonin was not adopted as the first point of departure in the present work.

Our first efforts were directed at the stepwise degradation of limonin by oxidative means. Limonin is quite resistant to many oxidizing agents, but once oxidation starts it tends to proceed rapidly to the formation of fragmentary oxidation products. It was expected that if the lactone linkages could be opened to hydroxy esters, the hydroxyl groups might serve as points of attack. When limonin is warmed with dilute alkali it dissolves, and upon acidification the lactone groups are immediately reconstituted, limonin being the only recoverable substance. This is in contrast to the behavior of columbin, which can be transformed into an hydroxy acid by similar treatment (17). The remarkable stability of the lactone linkages in limonin is further shown by the fact that it has been found impossible to convert the compound into an ester by any of a number of procedures, including the use of dimethyl sulfate or methyl *p*-toluenesulfonate and alkali under a variety of conditions and the use of diazomethane with ether extracts from rapidly-extracted, freshly-acidified solutions of limonin.

Limonin is oxidized to a non-separable mixture of the fragmentary products of extreme degradation by hot, dilute nitric acid, chromic anhydride in acetic acid, hot alkaline potassium permanganate, and acidic hydrogen peroxide. It is stable to cold, alkaline hydrogen peroxide and is not materially affected by being refluxed overnight in nitrobenzene solution. The use of eight to ten equivalents of potassium manganate, however, has led to a partial oxidation product of limonin, limonilic acid. This substance contains a lactone group and is a carboxylic acid. Aqueous solutions of limonilic acid are not bitter. Analyses of limonilic acid and its methyl ester, along with neutral and saponification equivalents and a molecular weight determination, agree with the formula $C_{25}H_{28}O_9$ for the acid. This corresponds to a loss of a methyl or methylene group. Since limonin has $(\alpha)_D^{25}$ $-123°$ and limonilic acid has $(\alpha)_D^{25}$ $+109°$ it is probable that the oxidative attack has taken place at an asymmetric center.

Limonilic acid is inert towards hydroxylamine and 2,4-dinitrophenylhydrazine, forms no acetate when treated with acetic anhydride and sodium acetate, and yields no iodoform with iodine and alkali. Upon fusion with alkali, limonilic acid yields about 0.6 mole of acetone. Since limonin when treated in the same

---

[1] The formation of "hexahydrolimoninic acid" by the treatment of limonin with boiling, dilute hydrochloric acid, reported by Higby (12), must be regarded as doubtful since from Higby's description it is evident that he was dealing with an amorphous substance, and since it is difficult to see how this treatment could bring about so extensive a reduction of the molecule.

way yields about 0.88 mole of acetone it is probable that the grouping which gives rise to acetone in the case of limonin is still present in limonilic acid, and that both substances possess one acetone-producing side chain. Limonilic acid is no more prone to yield crystalline derivatives than is limonin, and shows the same sensitivity towards mineral acids and to further oxidation. Because of these facts and the difficulty of obtaining large amounts of limonin for study, attention was turned to an examination of other methods of treatment of limonin in the hope that more tractable degradation products might be found.

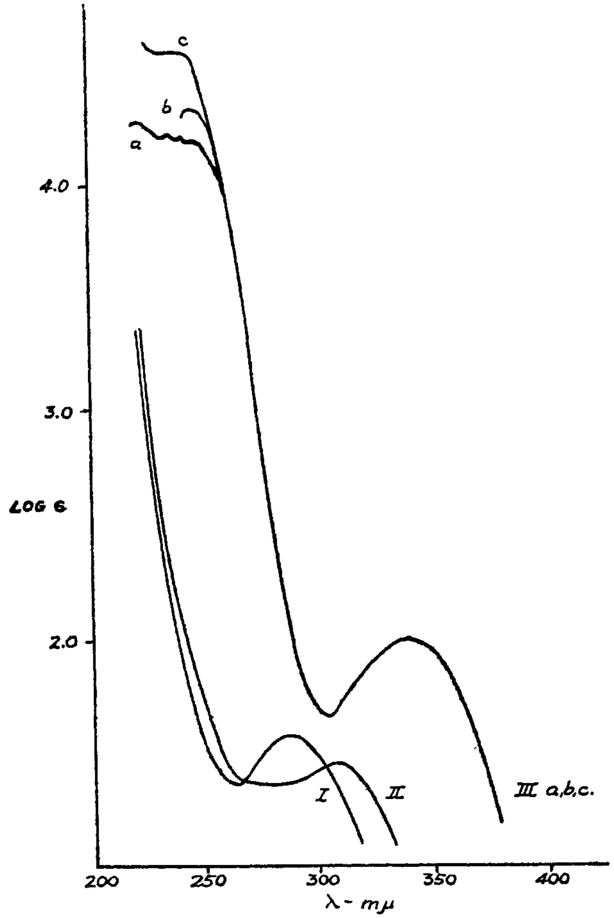


FIG. 1. Absorption Spectra of Limonin, Limonilic Acid, and Citrolin.
I. Limonin (in alcohol)
II. Limonilic Acid (in alcohol)
III. Citrolin (a) in alcohol)
(b) in chloroform
(c) in ether

Because the use of hydriodic acid has been found to yield valuable information in studies on the nature of picrotoxin (18, 19, 20), its action on limonin has been studied. The treatment of limonin with hydriodic acid in boiling acetic acid has yielded two reduction products, one (citrolin) having the composition $C_{26}H_{28-30}O_6$ and the other (desoxolimonin) having the probable composition $C_{26}H_{30}O_7$. Desoxolimonin has been obtained in very small amount only and has not been studied in detail.

Citrolin appears to undergo ready decomposition even in dilute alkaline solution. Treatment with hot 0.25 *N* potassium hydroxide for one hour causes the formation of dark-colored substances, and upon acidification of the solution neither citrolin nor any other definite products can be isolated. Titration of the alkaline solution is difficult because of an indefinite end-point and saponification values of approximately 280 are obtained. The most striking characteristic of citrolin is the ease with which acetone is split off upon treatment with dilute alkali. Acetone was readily detectable in the solution resulting from the treatment with 0.25 *N* alkali just described. No acetone is formed when limonin is treated in a similar fashion. This observation might indicate that the reduction with hydriodic acid has resulted in an attack upon the linkage responsible for the production of acetone.

Attempts to isolate definite products from the mixture resulting from the treatment of citrolin with alcoholic alkali were unsuccessful. An indication was obtained, however, that among the substances produced may be a phenolic or enolic substance.

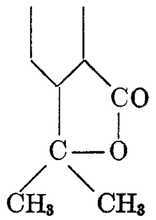
In an attempt to obtain information about the carbon skeleton of limonin, a sample of the bitter principle was heated with hydriodic acid in a sealed tube at 200°. This treatment yielded a colorless, viscous oil. This was treated with selenium in an attempt to bring about dehydrogenation to identifiable aromatic substances but the resulting tarry material could not be induced to yield a picrate.

The absorption spectra of limonin, limonilic acid, and citrolin are shown in Fig. 1.

A supplemental observation, while leading to no definite conclusions, is worthy of mention. Citrolin is oxidized with cold, dilute nitric acid to yield a very small amount of a crystalline oxidation product. Due to the poor yields obtained in this reaction the compound was not examined further.

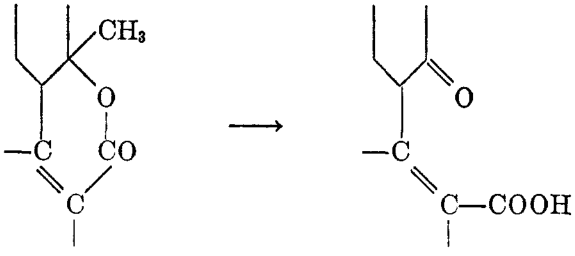
DISCUSSION

The evidence so far available indicates that limonin does not possess a bilaterally symmetrical structure. The formation of but one mole of acetone upon alkali fusion and the formation by oxidation of a lactone-carboxylic acid which also gives a mole of acetone support this conclusion. The great stability of the lactone groups in limonin, coupled with the formation of but one mole of acetone by alkali fusion, leads one to suggest that one of the lactone groupings is



I

and that the other is not identically constituted. This second lactone group is the one which disappears in the formation of limonilic acid, and since in this transformation a carbon atom is lost, it is suggested that the original lactone linkage possesses the partial structure II shown in the following equations, which show the possible course of the reactions involved in the oxidation process:

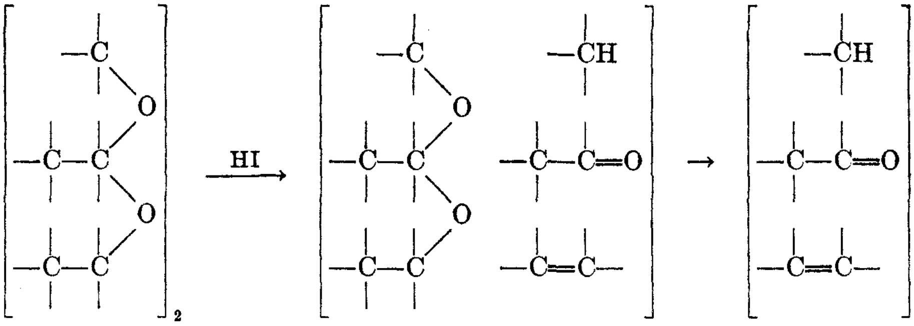


II III

The center of optical asymmetry lost in this step is evident from this equation. The inertness of the carboxyl group which it is supposed is formed in this reaction may be due to the kind and number of substituents present on the adjacent carbon atoms. That the lactone grouping II cannot be replaced by IV is indicated by the fact that limonilic acid does not have an absorption spectrum which would be expected of the resulting acid V, which is an α,β-unsaturated ketone (21).

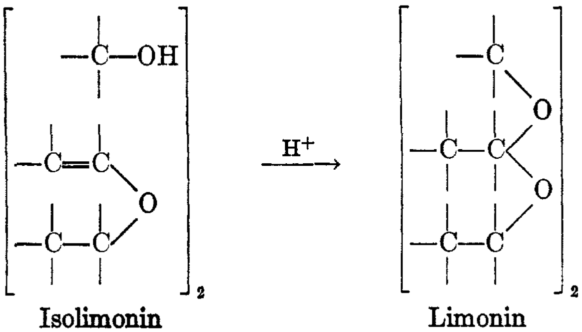
IV V

The formation of citrolin and desoxolimonin can be explained by the following partial structures, of which VI represents another part of the limonin molecule.



Limonin VI Desoxolimonin VII Citrolin VIII

The reported presence of two hydroxyl groups in isolimonin (5), coupled with the fact that isolimonin is readily isomerized to limonin by acids, is in accord with the suggestion that limonin contains the grouping shown in VI. These observations would be in accord with the suggestion that isolimonin may contain the groupings represented in IX:



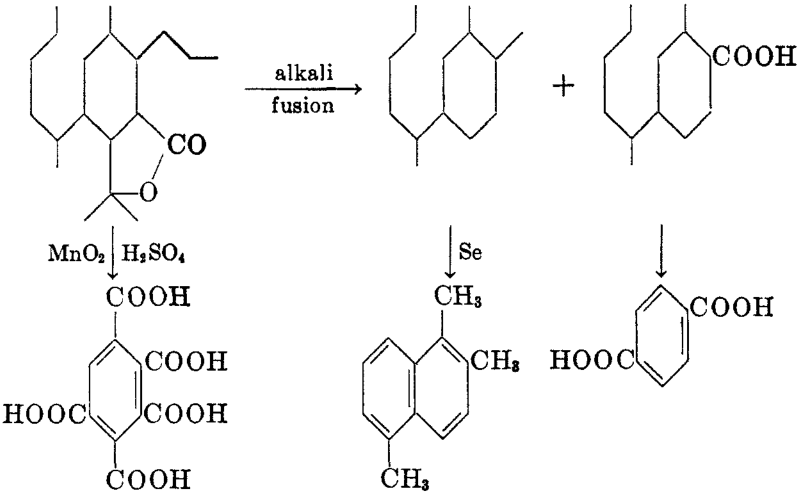
Isolimonin
IX

Limonin

If the above structures represent portions of the limonin molecule it can be seen that the completely reduced skeleton of limonin would have the composition $C_{26}H_{46}$. This differs by eight hydrogen atoms from the paraffinic $C_{26}H_{54}$, indicating that limonin may possess four rings on four double bonds, or some combination of these.

The absorption spectra of limonin and of limonilic acid, showing a sharp increase at about 240 mμ which continues indefinitely into the short ultraviolet, are characteristic of these of α,β-unsaturated acids and esters (22). The fact that limonin, although showing unsaturation to catalytic hydrogenation, does not react with perphthalic acid, is in accord with the α,β-unsaturated lactone structure. The peaks in the spectrum of citrolin at about 245 mμ are reminiscent of those in the spectra of styrene and indene and may indicate that aromatization of a ring has occurred during the treatment with hydriodic acid. It should be mentioned that picrotoxin derivatives, which possess certain structural features similar to those suggested for limonin, can undergo aromatization under similar treatment (23).

The source of 1,2,5-trimethylnaphthalene, benzenepentacarboxylic acid, and isophthalic acid, all obtained by earlier workers by the use of degradation methods mentioned above, could be a partial structure such as the following, which includes the lactone ring shown in I:



The above suggestions are tentative only, and rest upon an experimental foundation which is still a narrow one. Further work, such as C-methyl determination, possibly ozonization, a reexamination of the hydrogenation of limonin and further degradation studies on citrolin will doubtless furnish further useful data to clarify the still obscure picture.

EXPERIMENTAL

The isolation of limonin. In a 5-gallon crock was placed about 10 lbs. of dried orange pulp (stock-feed material), which was covered with water. Steam was passed in for several hours. The pulp swelled to fill the crock, and occasionally more water was added to keep the mixture fluid. The hot mash was pressed out in a fruit press and reextracted in the same way. The cooled water extract was stirred thoroughly with two gallons of benzene, using a "Lightning" mixer, and the resulting emulsion broken with the aid of sulfuric acid and methanol. After a second extraction with benzene, the benzene solutions were evaporated as far as possible on a steam-cone, at atmospheric pressure and then at 22 mm. The residue was treated with 100 ml. of glacial acetic acid which was then removed under reduced pressure. This treatment removed essential oils whose presence made crystallization of the product difficult. The residue was dissolved in glacial acetic acid and allowed to crystallize. The thick, black suspension was filtered and the dark crystalline material washed with acetic acid until it was pale brown in color. It can be purified further by recrystallization from acetic acid or from acetone-methanol.

Limonin crystallizes from acetic acid with a molecule of acetic acid (24), forming crisp, white needles. It melts with decomposition at about 302–305° (corr.), depending somewhat upon the heating rate. The melting point of the solvated material is the same, the acetic acid evidently being lost during the heating.

Limonin gives a deep red-brown color in concentrated sulfuric acid. Tests for the methylenedioxyl group were negative. The change in titer of a chloroform solution of limonin and perphthalic acid was the same as that of a blank containing no limonin. Tests for other functional groups with the usual carbonyl reagents and with acylating reagents were negative, unchanged limonin being recovered, occasionally accompanied by very small amounts of oily materials.

Attempts to methylate limonin. I. A solution of 1.10 g. of limonin in 20 ml. of 6 *N* sodium hydroxide was filtered into a separatory funnel containing 100 ml. of ice-cold ether. The funnel was cooled in ice, 30 ml. of ice-cold 6 *N* sulfuric acid was added, the whole vigorously shaken, and the ether layer separated and added immediately to an ether solution of diazomethane (excess). After allowing the solution to stand overnight the excess diazomethane was decomposed with acetic acid and the solvents evaporated. The residue was shaken with cold, dilute sodium hydroxide and benzene. Nothing was recovered from the benzene; from the aqueous solution was recovered 0.80 g. of limonin upon acidification.

II. A solution of 1.0 g. of limonin in the theoretical amount of alkali was covered with 100 ml. of a diazomethane solution in ether, the whole being contained in a 3-necked flask fitted with a stirrer and dropping-funnel. While the mixture was stirred, dilute hydrochloric acid was introduced at the bottom of the aqueous layer. At the conclusion of the experiment there was present in the aqueous layer an ether-insoluble precipitate. This proved to be limonin. Nothing was obtained when the ether layer was evaporated.

III. A cooled solution of 0.51 g. of limonin in an equivalent amount of alcoholic alkali was treated with 2 ml. of acetic acid and the solution poured immediately into an excess of diazomethane in ether. From the solution limonin (0.43 g.) was obtained along with about 50 mg. of a non-crystallizable, dark-colored oil.

IV, etc. Attempts to methylate limonin by treatment with dimethyl sulfate in alcoholic alkali, keeping dimethyl sufate in excess in some experiments and alkali in excess in others failed to yield any recoverable material but limonin. Similar experiments with methyl *p*-toluenesulfonate gave similar results. When the reagents were added in such a way as to keep the solution as nearly neutral as possible no better results were obtained.

The oxidation of limonin. I. A solution of 0.50 g. of limonin in 100 ml. of 1 *N* alkali was treated with 1 g. of sodium peroxide. After an hour the solution was heated to boiling and acidified. The precipitate which formed was limonin.

II. A solution of 0.50 g. of limonin in 100 ml. of 2 *N* alkali was treated with 15 ml. of 30% hydrogen peroxide. After 24 hours the solution was heated to boiling and acidified. Limonin was recovered.

III. Limonin was largely unaffected when 0.5 g. was allowed to stand overnight with 6 ml. of 30% hydrogen peroxide containing, first, one small drop of concentrated sulfuric acid and, later, 3 ml. of concentrated sulfuric acid. The solution after removal of unreacted limonin, yielded a small amount of water-soluble, dark-colored material which could not be purified further.

IV. Treatment of limonin with chromic anhydride in glacial acetic acid solution results in a rapid oxidation. The products of these experiments were water-soluble amorphous materials which resisted attempts at crystallization or purification.

V. A solution of 0.50 g. of limonin in 20 ml. of glacial acetic acid containing 2 ml. of concentrated nitric acid was refluxed for thirty minutes. Nitrogen oxides were evolved copiously. The solution was buffered with sodium acetate, evaporated to 5 ml., diluted with 25 ml. of water and extracted with ether. Evaporation of the ether left a considerable residue which was soluble in most of the common solvents with the exception of benzene and ligroin. In the absence of solvents it formed a glass. It dissolved in sodium bicarbonate solution with the evolution of carbon dioxide. The material could not be crystallized nor purified.

VI. *Limonilic acid.* To a solution of 3.16 g. of limonin in 100 ml. of 6 *N* sodium hydroxide was added 100 ml. of a solution 0.03 molar in potassium manganate and 6 *N* in sodium hydroxide (9 equivalents of potassium manganate). The solution was maintained at about 50° for an hour, during which the green color disappeared and a precipitate of manganese dioxide formed. The mixture was filtered and the filtrate acidified with hydrochloric acid, heated to boiling and allowed to cool, and kept at 5° overnight. The granular precipitate was collected and dried; it weighed 1.88 g. after recrystallization from methanol-water. Limonilic acid melts at about 292–293° (dec.) (uncorr.) when introduced into a bath preheated to that temperature. A mixture of limonin and limonilic acid melted at 255–260°.

Anal. Calc'd for $C_{25}H_{28}O_9$: C, 63.55; H, 5.97.
Found: C, 63.65; H, 5.84; $(\alpha)_D^{25}$ +109° (in acetone).

When limonilic acid is titrated rapidly to the first end-point with thymol blue an equivalent weight of about 450 is found. The end-point fades, however; and if excess alkali is added and the excess back-titrated with standard acid a neutral equivalent of 237 is obtained. Calc'd for $C_{25}H_{28}O_9$: eq. wt. 236 (one lactone, one carboxyl).

Limonilic acid is soluble in sodium bicarbonate solution. It gives a red-brown color in concentrated sulfuric acid similar to that shown by limonin. It is not readily affected by bromine in carbon tetrachloride nor by potassium permanganate in acetone in the cold. On treatment with hot, dilute mineral acids it decomposes to brown, amorphous products. It did not yield carbonyl derivatives with hydroxylamine nor with 2,4-dinitrophenylhydrazine and was recovered unchanged after treatment of its solution in 1 *N* alkali with iodine. Treatment of 0.30 g. of limonilic acid with a mixture of acetic anhydride, sodium acetate, and acetic acid (20 minutes reflux) resulted in the eventual recovery of 0.28 g. of limonilic acid.

Methyl limonilate. A solution of 0.55 g. of limonilic acid in 30 ml. of ethanol was treated in small portions with 100 ml. of a solution of excess diazomethane in ether. After 30 minutes the excess diazomethane was decomposed with acetic acid, the ether solution washed with sodium bicarbonate solution, dried, and evaporated at 22 mm. The residual material was taken up in 30 ml. of acetone and treated with 11 ml. of 0.1 *N* sodium hydroxide. This solution was poured into water and ether, the ether layer discarded and the aqueous layer acidified and then treated with excess sodium bicarbonate. Extraction of the resulting suspension with benzene and evaporation of the benzene solution yielded a solid which was crystallized from a mixture of 1 ml. of acetone and 4 ml. of methanol. After recrystallization from methanol the ester melted at 217–218° (uncorr.) with no decomposition.

Anal. Calc'd for $C_{26}H_{30}O_9$: C, 64.19; H, 6.22; mol. wt., 486.5.
Found: C, 64.23; H, 6.39; mol. wt. (Rast), 488.

Hydrolysis of limonin with acid. Treatment of limonin with boiling 50% hydrobromic acid in acetic acid resulted in the formation of a dark green, opaque solution from which no definite substance could be isolated. With hot, dilute hydrochloric acid in acetic acid some limonin was recovered.

Pyrolysis of limonin. A sample of 35 mg. (0.074 millimole) of limonin was heated in an evacuated, sealed ampule at 250–280° for 3 hours. The gas which formed was analyzed in a Blacet-Leighton micro gas analysis apparatus and was found to contain 0.021 millimole of carbon dioxide. Some limonin was recovered unchanged (about 50%) when another sample of 0.51 g. was heated for 3 hours at 290–300°. These results indicate that a mole of carbon dioxide is formed per mole of limonin; but because of the difficulty in recovering crystalline limonin from the tarry pyrolysis products this is subject to some uncertainty.

Alkali fusion of limonin and limonilic acid. About 50 mg. of limonin or limonilic acid was treated with 2 ml. of 75% potassium hydroxide solution and heated gradually to 350–360°. The distillate was condensed in a small bulb cooled in an ice-salt mixture.

To the distillate were added 1 ml. of alcohol, 1 ml. of a 10% solution of salicylaldehyde in alcohol, and 2 ml. of 10% ethanolic potassium hydroxide. This solution was heated at 100° for exactly one hour, cooled in ice, and the color measured at 550 mμ on a Beckman Photoelectric Spectrophotometer. The amount of acetone was calculated from a curve prepared by treating known amounts of acetone in the same way. The recovery of acetone by distillation from solutions containing known amounts was 90–95%.

Limonin:

45.7 mg. gave 0.085 millimole of acetone = 0.87 mole/mole limonin
51.3 mg. gave 0.097 millimole of acetone = 0.88 mole/mole limonin

Limonilic acid:

74.4 mg. gave 0.086 millimole of acetone = 0.54 mole/mole limonilic acid
55.5 mg. gave 0.070 millimole of acetone = 0.60 mole/mole limonilic acid

Hydriodic acid reduction of limonin: Citrolin. To a solution of 50 ml. of constant-boiling hydriodic acid in 80 ml. of glacial acetic acid was added 3.0 g. of limonin. The mixture was warmed to 60° until solution was complete. After the addition of 0.5 g. of red phosphorus the mixture was refluxed for 2 hours, then filtered through asbestos, diluted to 600 ml. with boiling water containing enough sodium bisulfite to reduce the excess iodine, clarified by filtration through "Supercel," and the filtrate allowed to cool. The material which separated was combined with a further amount obtained by extracting the dilute acetic acid filtrates with benzene and the whole dissolved in 40 ml. of acetone. An equal volume of methanol was added and the solution concentrated by distillation until crystallization began. After cooling, the solid was collected and recrystallized again in the same way. The yield was usually 500–600 mg. of long silky white needles which melted at 305° (uncorr.). It has $(\alpha)_D^{25}$ −120° (in acetone).

Anal. Calc'd for $C_{26}H_{28}O_6$: C, 71.54; H, 6.47; mol. wt., 436.5.
Calc'd for $C_{26}H_{30}O_6$: C, 71.37; H, 6.68; mol. wt. 438.5.
Found: C, 71.68; H, 6.81; mol. wt. (Rast), 426, 443.

Desoxolimonin. By evaporating the mother liquors from the crystallization of citrolin and extracting the residue with an amount of methanol just insufficient for complete solution, 50–60 mg. of another material was obtained. This material had the same m.p. as citrolin (305°, uncorr.), but a mixture of the two showed a large depression (m.p. 260–270°, uncorr.).

Anal. Calc'd for $C_{26}H_{30}O_7$: C, 68.70; H, 6.65.
Calc'd for $C_{26}H_{28}O_7$: C, 69.01; H, 6.24.
Found: C, 68.83; H, 6.57.

When 0.94 g. of citrolin was shaken for 20 hours with a solution of 20 ml. of concentrated nitric acid in 80 ml. of water, and the resulting clear solution neutralized with sodium bicarbonate and extracted continuously with ether, there was obtained a small amount of material which crystallized readily from acetic acid. Only about 5 mg. was obtained. It was not investigated further.

Saponification of citrolin. A solution of 0.50 g. of citrolin in 20 ml. of 10% ethanolic potassium hydroxide was refluxed for an hour. The solution became a deep greenish-brown. A few ml. of the solution was distilled off and was found to give a strong positive test for acetone. No definite products could be isolated, but a small amount of crude material, obtained by saturating the solution with carbon dioxide and extracting successively with ethyl acetate, benzene, and ether, rapidly decolorized bromine and gave a colored precipitate with ferric chloride.

Complete reduction of limonin. Two grams of limonin was heated in a sealed tube with 3 ml. of constant-boiling hydriodic acid at 200° for 24 hours. The contents of the tube were shaken with ether and an aqueous solution of sodium bisulfite. The colorless ether solution was evaporated, yielding about 2 g. of a colorless, viscous oil. This oil did not decolorize bromine in carbon tetrachloride and appeared to be insoluble in cold, concentrated sulfuric acid, but dissolved in cold, fuming sulfuric acid with a dark red-brown color. The oily material was heated at 330–350° for 48 hours with 3 g. of selenium. The product of this reaction was a yellow-brown tar which could not be purified and which could not be induced to yield a picrate.

SUMMARY

Three crystalline degradation products of limonin have been isolated, one, *limonilic acid*, by oxidation with potassium manganate, and two, *citrolin* and *desoxolimonin*, by hydriodic acid reduction.

On the basis of these observations and those reported by previous workers, proposals are made concerning certain structural features of the limonin molecule.

Los Angeles, Calif.

REFERENCES

(1) Bernay, *Ann.*, **40**, 317 (1841).
(2) Fujita and Wada, *J. Pharm. Soc. Japan*, **51**, 506 (1931).
(3) Keimatsu, *J. Pharm. Soc. Japan*, No. 248, 979 (1902).
(4) Thoms, *Ber. deut. pharm. Ges.*, **33**, 68 (1923).
(5) Koller and Czerny, *Monatsh.*, **67**, 248 (1936); **70**, 26 (1937).
(6) Kaku and Ri, *J. Pharm. Soc. Japan*, **55**, 222 (1935).
(7) Kaku and Ri, *J. Pharm. Soc. Japan*, **55**, 219 (1935).
(8) Schmidt, *Ann.*, **51**, 338 (1844).
(9) Paterno and Oglioloro, *Ber.*, **12**, 685 (1879).
(10) Peters and Frerichs, *Arch. pharm.*, **240**, 661 (1902).
(11) Feist and Schulte, *Ber.*, **69**, 1322 (1936).
(12) Higby, *J. Am. Chem. Soc.*, **60**, 3013 (1938).
(13) Schechter and Haller, *J. Am. Chem. Soc.*, **62**, 1307 (1940).
(14) Chen and Chen, *J. Am. Pharm. Assoc.*, **22**, 716 (1933).
(15) Feist, Kunst, and Brachvogel, *Ann.*, **519**, 124 (1935).
(16) Haller, private communication.

(17) Frey, *Ann.*, **351**, 372 (1909).
(18) Mercer, Robertson, and Cahn, *J. Chem. Soc.*, 997 (1935).
(19) Mercer and Robertson, *J. Chem. Soc.*, 288 (1936).
(20) O'Donnell, Robertson, and Harland, *J. Chem. Soc.*, 1261 (1939).
(21) Woodward, *J. Am. Chem. Soc.*, **63**, 1123 (1941).
(22) Dimroth, *Angew. Chem.*, **32**, 552 (1939).
(23) Angelico, *Gazz. chim. ital.*, **41**, ii, 337 (1911).
(24) Mayeda, *J. Pharm. Soc. Japan*, **55**, 90 (1935).