STUDIORUM PROGRESSUS

Chemical Contributions to the Mechanism of the Biological Oxidation of Tryptophan[1]

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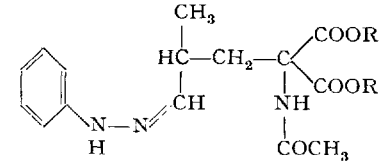
The *in vitro* oxidation of indole derivatives, such as indole[2] itself, methylketol[2], tetrahydrocarbazole[3], 2,3-cyclopentenoindole[4], 2,3-cycloheptenoindole[4], and other indoles[5] by the action of catalytically excited oxygen[6] or by autoxidation[7], by perbenzoic[8] or peracetic[9] acids and other oxidants[5] has been shown to lead through intermediate 3-hydroperoxy- or 3-hydroxyindolenine derivatives[10], the colorful chemistry of which has been demonstrated in several instances[11].

This course of oxidation was expounded in the conversion of the alkaloid cinchonamine into quinamine by the action of peracetic acid[12], a transformation that helped to settle the structural and phytochemical relationship of the two plant products.

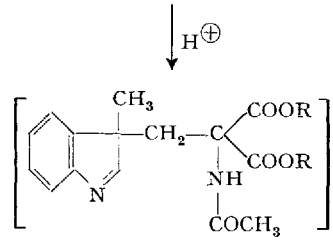
In the light of these facts it is not surprising that α-hydroxytryptophan (oxindolylalanine), III (first isolated from the hydrolysate of the toxic peptide phalloidine[13], then obtained from tryptophan by the action of peracetic acid[9] and by total synthesis[14]), originally postulated to be the first intermediate in the oxidative breakdown of tryptophan (KOTAKE, BUTENANDT), was shown not to be a normal intermediate by the important biochemical studies of KNOX and MEHLER[15], AMANO *et al.*[16], SAKAN and HAYAISHI[17], MASON and BERG[18].

The synthesis of the ACTUAL intermediate, viz. β-hydroxy-ψ-tryptophan (IVa, pag. 37) has been under investigation in these Institutes for some time. Although this aim has not been quite reached yet, some findings of interest were made.

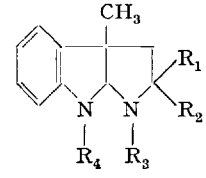
A. – β-Methyl- or β-hydroxyindolenines with an alanine side chain at pH < 6 undergo an internal acid-catalyzed condensation to yield derivatives of eseroline with alkyl or hydroxyl substituents in the angular position (C_{10}). Even acetylation of the primary amino group does not prevent this internal cyclization. Thus, it is impossible to synthesize β-methyl-ψ-tryptophan by the acid-catalyzed Fischer synthesis from the phenylhydrazone



XI



XII



XIIIa: $R_1 = R_2 = COOEt$, $R_3 = CH_3CO$, $R_4 = H$
XIIIb: $R_1 = R_2 = COOEt$, $R_3 = R_4 = CH_3CO$
XIIIc: $R_1 = R_2 = R_3 = R_4 = H$

XI of the compound resulting from the base-catalyzed addition of diethyl acetamidomalonate to α-methylacrolein[1]. Instead, one obtains derivatives of *bisnor*eseroline (XIIIc), such as XIIIa and XIIIb, compounds that open new synthetic routes into the eserine system and furnish new derivatives of medicinal interest[2].

B.—These internal condensation products, such as present in the alkaloid quinamine, are derivatives of hydroxyindolines, lack the reactive $-N=C<$ element of indolenines (comparable to a keto group) and are, therefore, resistant to further oxidative attack. These findings are in accord with the fact that a pH > 6 is required for the breakdown of tryptophan (as such or in proteins)[3] by the peroxidase-oxidase system of

[1] The preparation of this preliminary report, reviewing material that appeared or will appear in the Journal of the American Chemical Society, was kindly suggested by Prof. L. RUZICKA.—*On the Mechanism of Oxidation* V, *vide* Papers III and IV in this series: J. Amer. Chem. Soc. (in press).—From the National Heart Institute, National Institutes of Health, Washington 14, D. C., and the Converse Memorial Laboratory of Harvard University, Cambridge 38, Massachusetts, U.S.A.

[2] B. WITKOP and J. B. PATRICK, J. Amer. Chem. Soc. *73*, 713 (1951).

[3] B. WITKOP, ib. *72*, 614 (1950).

[4] J. B. PATRICK, M. ROSENBLUM, and B. WITKOP, Exper. *6*, 461 (1950); J. Amer. Chem. Soc. *73*, 2641 (1951).

[5] B. WITKOP (unpublished).

[6] B. WITKOP and J. B. PATRICK, J. Amer. Chem. Soc. *73*, 2196 (1951).

[7] R. J. S. BEER, L. MCGRATH, and A. ROBERTSON, J. Chem. Soc. *2118*, 3283 (1950).

[8] B. WITKOP and H. FIEDLER, Ann. Chem. *558*, 91 (1947).

[9] B. WITKOP, ib. *558*, 98 (1947).

[10] B. WITKOP, J. Amer. Chem. Soc. *72*, 1428 (1950).

[11] B. WITKOP and J. B. PATRICK, Exper. *6*, 183 (1950); J. Amer. Chem. Soc. *72*, 633 (1950); *73*, 1558, 2188 (1951).—S. G. P. PLANT and R. ROBINSON, Nature *165*, 36, 928 (1950).—S. G. P. PLANT and M. TOMLINSON, J. Chem. Soc. *1950*, 2127.

[12] B. WITKOP, ib. *72*, 2311 (1950).

[13] H. WIELAND and B. WITKOP, Ann. Chem. *543*, 171 (1940).

[14] M. KOTAKE, T. SAKAN, and T. MIWA, J. Amer. Chem. Soc. *72*, 5085 (1950).—J. W. CORNFORTH, R. H. CORNFORTH, C. E. DAIGLIESH, and A. NEUBERGER, Biochem. J. *48*, 591 (1951).

[15] W. E. KNOX and A. H. MEHLER, J. Biol. Chem. *187*, 419, 431 (1950).

[16] T. AMANO, M. TORII, and H. IRITANI, Med. J. Osaka Univ. *2*, 45 (1950).

[17] T. SAKAN and O. HAYAISHI, J. Biol. Chem. *186*, 177 (1950).

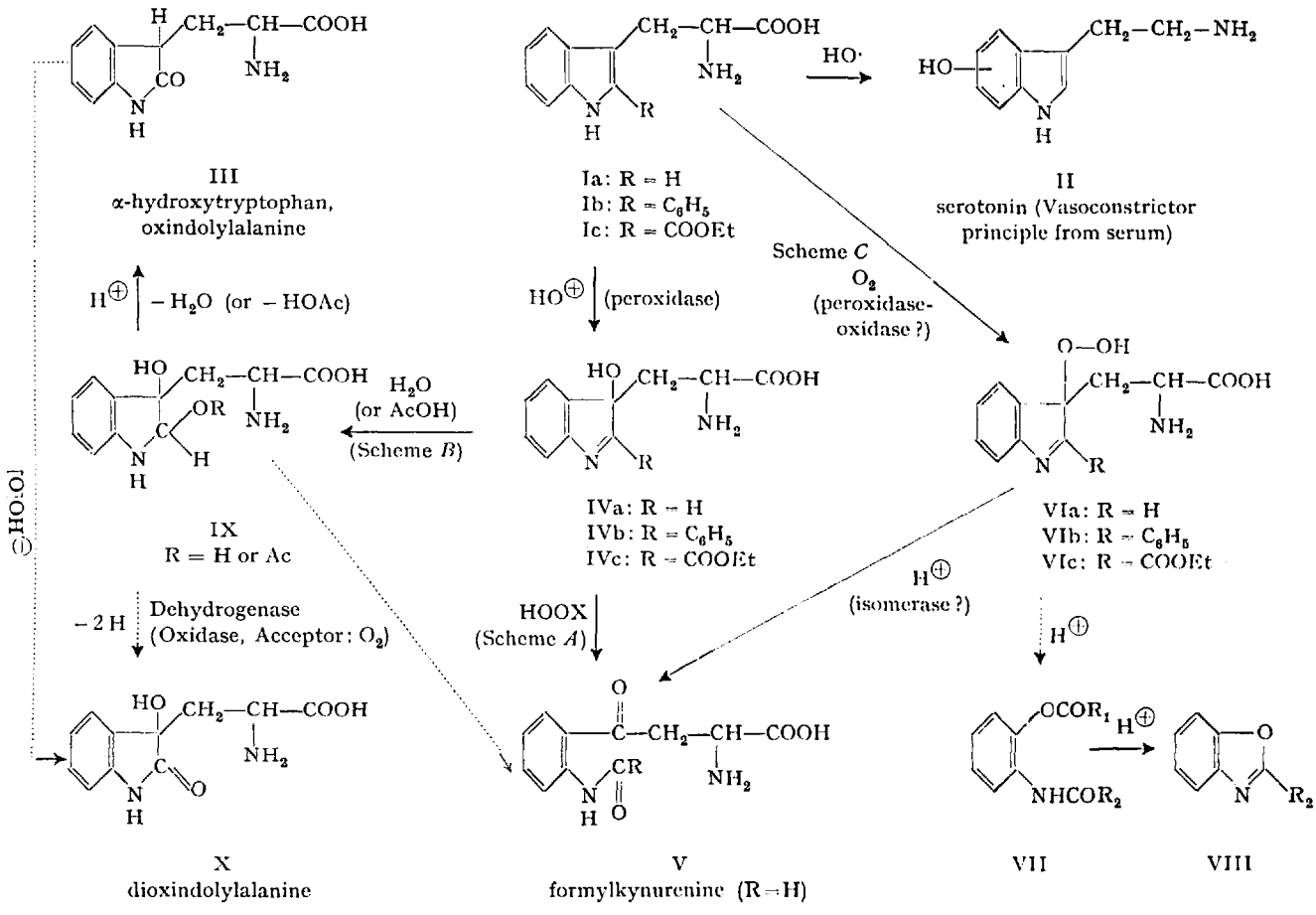
[18] M. MASON and C. P. BERG, ib. *188*, 783 (1951).

[1] Cf., D. T. WARNER and O. A. MOE, J. Amer. Chem. Soc. *71*, 2586 (1949).

[2] B. WITKOP, R. K. HILL, and H. KISSMAN, unpublished.

[3] The absorption of oxygen by solutions of serum proteins observed under the influence of light and hematoporphyrins is largely due to the presence of *tryptophan* and *tyrosine:* D. T. HARRIS, Biochem. J. *20*, 280 (1926) and ref. [4]. Likewise, it has been known for 20 years that peroxidase (from milk) oxidizes just these two amino acids: K. A. C. ELLIOT, ib. *26*, 10 (1931). The formation of the diphenyl ether linkage in *thyroxine* from diiodotyrosine and possibly the iodination of tyrosine has lately been attributed also to the action of peroxidase. Thus, the role of peroxidase in animal cells, uncertain for such a long time, is gaining more and more biological significance.

[4] H. GAFFRON, Biochem. Z. *179*, 157 (1926).

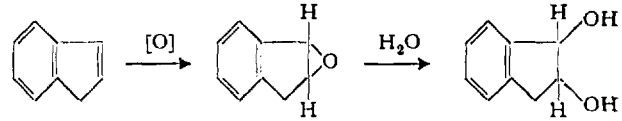


Tentative Correlation of the Breakdown Products Resulting from Tryptophan by Chemical or Biological Oxidation

KNOX and MEHLER[1] or by heavy metal-catalyzed (aut)-oxidation[2] or photooxidation with[3] or without photosensitizers[4].

C.—Formulae Ia–X give a reasonable scheme of the breakdown of tryptophan and tries to combine the enzymatic findings of KNOX and MEHLER[1] and of NEUBERGER and his school[5] with our chemical studies on the mechanism of oxidation in this series.

There are two principal pathways from β-hydroxy-ψ-tryptophan (IVa) to formylkynurenine (V). Scheme *B* starts out with the addition of water to give the glycolic intermediate IX[6]. However, this glycol IX is not likely to be the primary intermediate arising directly from tryptophan (Ia). Such a direct formation, it is true, would have an analogy in the autoxidation of indene (XIV) yielding (among other products) 2,3-oxidoindan (XV) and 2,3-dihydroxyindan (XVI)[1]. Yet indole (and its derivatives) is quite different in its chemical habitus from indene, and, if one wants to push the comparison, more like indone. In fact, it has proved impossible to prepare 2,3-oxidoindolines and where such structures have been postulated[2] revision[3] was necessary.



There still remained in the literature a number of indole epoxides such as XVIIa[4], XVIIb[5], and XVIIc[6],

[1] W. E. KNOX and A. H. MEHLER, J. Biol. Chem. *187*, 419, 431 1950).

[2] B. WITKOP (unpublished).—K. A. KUIKEN, C. M. LYMAN, and F. HALE, J. Biol. Chem. *171*, 551 (1947).

[3] H. GAFFRON, Biochem. Z. *179*, 157 (1926).

[4] F. LIEBEN, ib. *184*, 453 (1927).

[5] C. E. DALGLIESH, W. E. KNOX, and A. NEUBERGER, Nature *168*, 20 (1951).

[6] W. E. KNOX and A. H. MEHLER, J. Biol. Chem. *187*, 419, 431 (1950).—C. E. DALGLIESH, W. E. KNOX, and A. NEUBERGER, Nature *168*, 20 (1951).

[1] H. HOCK and F. DEPKE, Ber. *84*, 122 (1951).

[2] R. GOUTAREL, M.-M. JANOT, V. PRELOG, and W. I. TAYLOR, Helv. chim. acta *33*, 150 (1950).

[3] B. WITKOP, J. Amer. Chem. Soc. *72*, 2311 (1950).

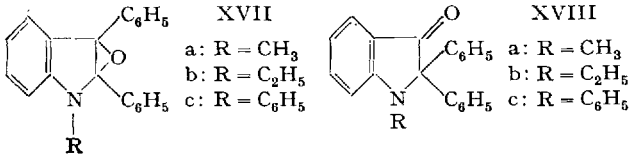
[4] M. KOHN and A. OSTERSETZER, Monatshefte *34*, 787 (1913).—F. J. MYERS and H. G. LINDWALL, J. Amer. Chem. Soc. *60*, 2153 (1938).

[5] F. J. MYERS and H. G. LINDWALL, J. Amer. Chem. Soc. *60*, 2153 (1938).

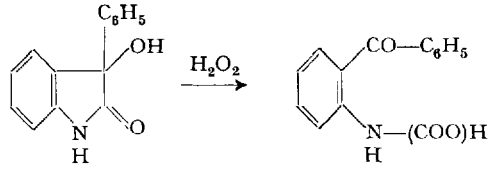
[6] R. STOLLÉ, J. prakt. Chem. *135*, 345 (1932).—W. C. SUMPTER, J. Amer. Chem. Soc. *64*, 1736 (1942).

which, however, turned out actually to be all indoxyl derivatives, XVIIIa, XVIIIb, and XVIIIc[1].

The glycolic intermediate IX[2] (corresponding to a hydrated ketone) would be a suitable substrate for a dehydrogenase (oxidase[3]) with oxygen serving as an acceptor, producing hydrogen peroxide. Dioxindolylalanine (X) would be the product of such a dehydrogenation. Whether there is another enzyme ("carboligase") catalyzing an inverse acyloin condensation, or whether there is possible a ring-chain transformation from X to formylkynurenine (V) may be left open at the moment. There is no chemical precedent for the existence of such a ring-chain tautomerism of dioxindole and its derivatives.



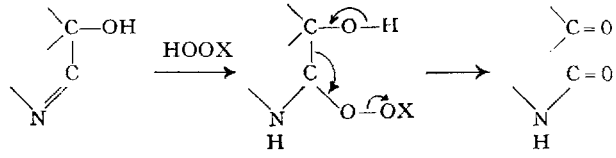
Dioxindole behaves more like a true acyloin giving isatin on mild oxidation[4] and requiring hydrogen peroxide for oxidative ring fission[5]:



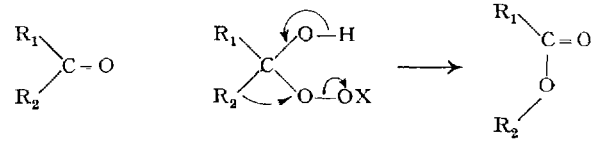
3-Phenyldioxindole 2-Aminobenzophenone

The preservation of the formyl group in formylkynurenine V militates against oxidative ring opening, the absence of chemical precedents against ring-chain tautomerism.

The stumbling block in this course of Scheme *B*, namely the breakage of $> C_2-C_3 >$, is avoided by assuming a direct glycolic cleavage by the action of the oxidase, comparable to other enzymatic ring openings, e.g. the formation of *cis,cis*-muconic acid from catechol by the action of pyrocatechase[6]. We have shown previously[1] that such a breakage occurs smoothly by the action of peracids on hydroxyindolenines in analogy to the formation of lactones from ketones. The two related reactions can be correlated in the following way (X = H, CH_3CO, C_6H_5CO, etc.):

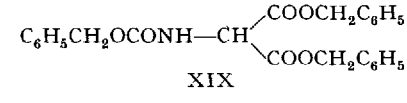


Hydroxyindolenine (ketone) intermediate with cationoid oxygen equivalent of formylkynurenine (lactone)



To summarize. Scheme *B* involving direct glycolic cleavage of IX (rather than the formation of a dioxindole intermediate X) attractively accommodates the enzymatic requirements. Scheme *A* fulfills all chemical prerequisites, but does not take due account of the enzymatic conditions, viz. the presence of a dehydrogenase (oxidase[2]), the necessity of oxygen as a hydrogen acceptor, and the formation of hydrogen peroxide in the dehydrogenation.

D.—Since it proved difficult to halt oxidation of tryptophan (Ia) under alkaline conditions at an intermediate stage, substituents such as phenyl or carbethoxy were introduced into the 2-position (Ib and Ic) in order to block internal condensation and to allow of the use of acidic conditions in the oxidation reactions. The synthetic problem, involving four different functional groups in the case of Ic, was greatly helped by the use of



XIX

dibenzyl carbobenzyloxyaminomalonate (XIX)[3], a compound which should turn out to be very useful for many synthetic reactions, since all three benzyl groups can be removed in one single step by catalytic hydrogenation. It is then hoped that the intermediate IVc will, after saponification, lose carbon dioxide with the same ease as α-picolinic acid in a base-catalyzed decarboxylation reaction[4].

E.—The nature of the blocking group *R* (Ib, Ic) greatly affects the ease and rate of autoxidation of tryptophan (Ia) reminiscent of the influence of the ring size on the rate and course of autoxidation of homologous tricyclic indoles[5]. Accordingly, the possible rôle that a cyclic chelate complex of a suitable heavy metal (copper, cobalt, iron, etc.)[6] with Ia or IVa might have on the rate and course of this oxidation is being studied.

[1] B. WITKOP and A. EK, ib. (in press).

[2] The preparation of glycols from indoles offers certain difficulties. Osmium tetroxide forms very stable complexes with tryptophan: B. HELFERICH and F. VORSATZ, Z. physiol. Chem. *239*, 241 (1936). The reaction of N-acylindoles with hydrogen peroxide in tertiary butyl alcohol, now under investigation, seems to be more promising. The formation of atroxindol from skatol, or of oxindolylalanine (III) with peracetic acid proceeds through a glycolic intermediate (equivalent to IX) followed by acid-catalyzed loss of water (cf. Table I).

[3] W. E. KNOX and A. H. MEHLER, J. Biol. Chem. *187*, 419, 431 (1950).

[4] B. KLEIN, J. Amer. Chem. Soc. *63*, 1474 (1941).

[5] S. INAGAKI, J. Pharm. Soc. Jap. *59*, 1 (1939); Chem. Abstr. *33*. 3790 (1939).—G. JACINI, Gazz. Chim. Ital. *72*, 510 (1942); *73*, 85 (1943),

[6] O. HAYAISHI and K. HASHIMOTO, Med. J. Osaka University *2*, 33 (1950); cf. R. Y. STANIER and O. HAYAISHI, The Bacterial Oxidation of Tryptophan, Science, *114*, 326 (1951); The Bacterial Oxidation of Tryptophan. III. Enzymatic Activities of Cell-Free Extracts from Bacteria Employing the Aromatic Pathway, J. Bacteriol, in press.—The example of catechol is not quite adequate: as an enediol its oxidation state is more comparable to dioxindole (cf. X). On the other hand, saturated glycols [e.g. 1,2-*trans*-dihydroxy-1,2-dihydroanthracene-1-glucuronic acid and the analogous glycols from naphthalene and phenanthrene, E. BOYLAND and G. WOLF, Biochem. J. *47*, 64 (1950)] do not undergo ring fission. This makes the formation of formylkynurenine (V) a unique biochemical reaction.

[1] B. WITKOP and J. B. PATRICK, J. Amer. Chem. Soc. *73*, 2196 (1951).

[2] W. E. KNOX and A. H. MEHLER, J. Biol. Chem. *187*, 419, 431 (1950).

[3] B. WITKOP and H. KISSMAN (unpublished).

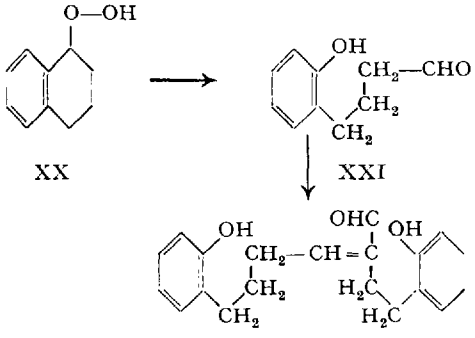
[4] Cf. B. R. BROWN and LL. HAMMICK, J. Chem. Soc., 659 (1949).

[5] J. B. PATRICK, M. ROSENBLUM, and B. WITKOP, Experr. *6*, 461 (1950); J. Amer. Chem. Soc. *73*, 2641 (1951).

[6] Copper and Cobalt (again in *alkaline* medium) in concentrations down to $5 \cdot 10^{-6}$ M catalyze still another reaction, viz. the tryptophanase reaction leading from tryptophan to indole and ammonia; E. A. DAWES and F. C. HAPPOLD, Nature *164*, 704 (1949).—F. C. HAPPOLD, Advances in Enzymology, *10*, 51 (1950).

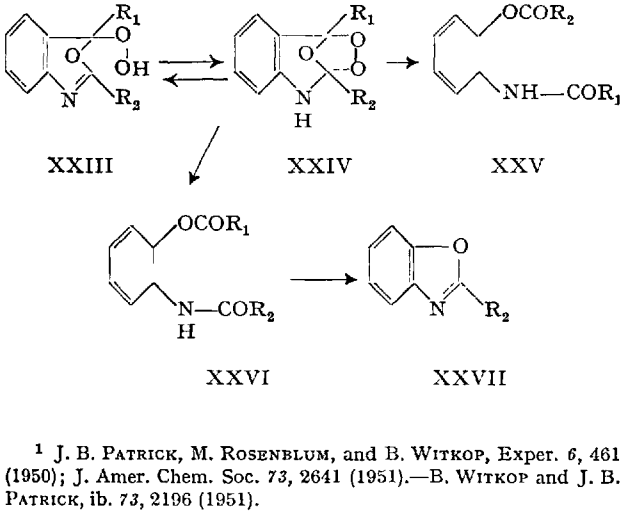
F.—The formation of hydroperoxides of type VI, so easily observed *in vitro*, might also occur *in vivo* (Table I, Scheme *C*). The ease of rearrangement, varying with the type of substituents in positions 2 and 3, into derivatives equivalent to formylkynurenine (V) has been demonstrated with model compounds[1]. A hydroperoxide-isomerase effecting the transformation from β-hydroperoxy-ψ-tryptophan (VIa) to formylkynurenine (V) would seem an attractive biological speculation apart from the possible mutagenic[2], cardiotonic[3], and carcinogenic[4] aspects of the intermediary hydroperoxide.

G.—The rearrangement of hydroperoxide intermediates equivalent to VI and in particular to VIb could also take a second course analogous to the acid-catalyzed rearrangement of tetralin hydroperoxide (XX) to the phenolic compounds XXI and XXII[5]. In fact, such a



XX XXI XXII

rearrangement was recently realized in the acid-catalyzed isomerization of the *ring-chain* tautomeric ozonide, XXIII ⇄ XXIV, to the *o*-diacylaminophenols (XXV, XXVI)[6].



XXIII XXIV XXV XXVI XXVII

[1] J. B. PATRICK, M. ROSENBLUM, and B. WITKOP, Exper. *6*, 461 (1950); J. Amer. Chem. Soc. *73*, 2641 (1951).—B. WITKOP and J. B. PATRICK, ib. *73*, 2196 (1951).

[2] A. LOVELESS, Nature *167*, 338 (1951).

[3] O. KRAYER, R. P. LINSTEAD, and D. TODD, J. Pharmacol. and Exp. Therap. *77*, 113 (1943).—O. KRAYER, Proc. Soc. Exp. Biol. Med. *53*, 51 (1943).—J. MITA, Arch. of Exp. Path. Pharmacol. *104*, 276 (1924).—H. RICHTER, Arch. für exper. Path. Pharmakol. *194*, 362 (1940).—R. MENDEZ, J. Pharmacol. and Exper. Therap. *81*, 151 (1944).

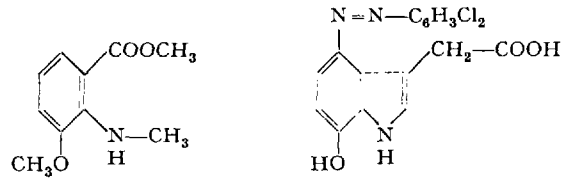
[4] H. F. PARK, J. Amer. Chem. Soc. *69*, 2248 (1947).

[5] M. S. KHARASCH, Lecture before the Swiss Chemical Society, 15 February 1950, at Zürich, cf. Angewandte Chemie *62*, 292 (1950).

[6] B. WITKOP and J. B. PATRICK, J. Amer. Chem. Soc. (in press).

If derivatives of *o*-aminophenol[1] or benzoxazole[2] were to be found as normal or abnormal animal or bacterial metabolites, their origin would have to be considered by this scheme.

H.—The introduction of the phenolic hydroxyl group, present in the "cn+-compound" (3-hydroxykynurenine), 3-hydroxyanthranilic acid[3], xanthurenic acid and in methylated form in the plant alkaloid damascenine (XXVIII), has so far been assumed to occur at the kynurenine stage. Yet the coupling product of potassium dichlorophenyldiazonium sulfonate with a



XXVIII Damascenine XXIX

pathogenic indole breakdown product isolated from the urine of a patient with liver cancer[4], if correctly expressed by XXIX, would indicate that phenolic hydroxyindoles may be formed prior to the breakdown of the pyrrole ring. A more conclusive piece of evidence for this hitherto-neglected aspect of tryptophan metabolism is furnished by *serotonin* (II), the vasoconstrictor principle isolated from beef serum[5]. The synthesis of serotonin[6], now actively pursued in these Institutes, will show whether this oxidation product is an intermediate in the chain of reactions leading to xanthurenic acid or, in an alternate pathway, leading

[1] The antibiotic properties of *o*-aminophenol may be mentioned in this connection: M. BARBER and G. A. D. HASLEWOOD, Brit. Med. J., *1944*, 754.

[2] *o*-Aminophenol as a metabolite passes readily into benzoxazoles (cf. R. T. WILLIAMS, *Detoxication Mechanisms* [John Wiley & Sons, Inc., New York 1947], pp. 144, 149) similar to the conversion of XXVI to the benzoxazole XXVII.

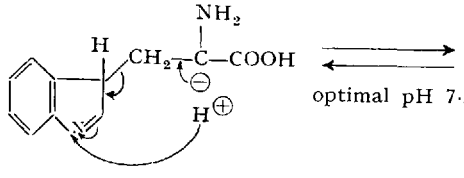
[3] The biological transformation of 3-hydroxyanthranilic acid into nicotinic acid need not be discussed here. It should be pointed out, however, that the formulation of this process according to C. MENTZER, D. MOLHO, and Y. BERGUER, Bull. Soc. Chim. France 782 (*1950*) is unlikely, since it involves the reduction of 6-hydroxynicotinic acid to nicotinic acid; a much more reasonable sequence of reactions is suggested by M. GUGGENHEIM, *Die biogenen Amine* (S. Karger, Basel-New York 1950), p. 263.

[4] L. HERMANNS and P. SACHS, Z. physiol. Chem. *114*, 79, 88 (1921); these coupling reactions with diazonium compounds have recently led R. J. WILLIAMS to the discovery of abnormal products in the urine of schizophrenic patients (Chem. Eng. News. *29*, 314 [1951]) and helped in recognizing the significance of individual metabolic patterns in human biochemistry (*cf.* American Chemical Society, 120th Meeting, Sept. 1951, Abstracts 6C).

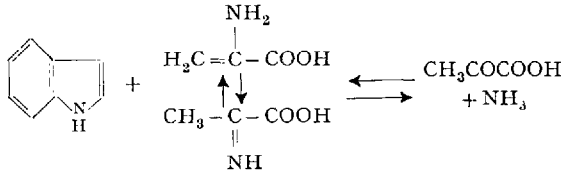
[5] M. M. RAPPORT, A. A. GREEN, and I. H. PAGE, Science *108*, 329 (1948).—M. M. RAPPORT, J. Biol. Chem. *180*, 961 (1949).

[6] Note added in proof.—According to a private communication from Dr. IRVINE H. PAGE (Cleveland Clinic Foundation, Cleveland 6, Ohio), 5-hydroxytryptamine, synthesized in the Abbott Laboratories (North Chicago, Illinois) by Dr. K. E. HAMLIN (J. Am. Chem. Soc., in press), exhibits physical and pharmacological properties identical with those of serotonin. The final confirmation of identity is awaiting the direct comparison of the synthetic with the natural product.—The recent reports from the Cleveland group on the existence of another hypertension factor, produced in the brain region and possibly related to serotonin, are of interest in connection with our work on the synthesis of further isomers and derivatives of serotonin (B. WITKOP and A. EK, unpublished).

to 5-hydroxy-, 5,6-dihydroxyindoles and from there to compounds of melanin character[1]. It would be interesting indeed, if there were to be found a biological parallelism to the fact that *ionic oxidants* (involving OH^+) oxidize the pyrrole part of indole compounds, while certain radical oxidants (involving ·OH) favor attack of the benzene ring. The position equivalent to a benzyl carbon, namely the methylene group of the alanine chain of tryptophan, has also been considered as a likely site for metal-catalyzed oxidation, a process for which radical mechanisms have been discussed[2]. A simpler and more obvious mechanism explaining the rupture of the tryptophan molecule into indole, pyruvic acid and ammonia is given in the following:



Tryptophan (*pseudo*-form)



indole α-aminoacrylic acid pyruvic acid + ammonia

The initial formation of an anion could equally well be formulated in a manner involving radical intermediates. This formulation would also account for the production of harman from tryptophan and ferric chloride[3]. Either the addition of tryptophan to aminoacrylic acid, or the condensation of the former with pyruvic acid, would lead to intermediates giving harman on oxidation and decarboxylation.

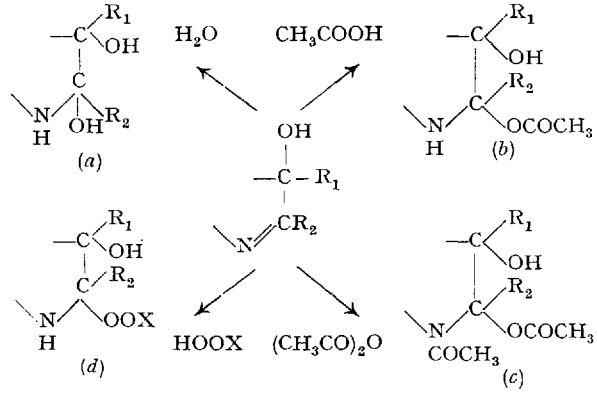
Acknowledgment: It is a pleasure to thank Drs. A. H. MEHLER and O. HAYAISHI for stimulating discussions.

Zusammenfassung

Die Oxydation von Indol-Derivaten einschließlich Tryptophan durch molekularen Sauerstoff, Autoxydation, Persäuren und Enzyme führt primär zu β-Hydroperoxy- und β-Hydroxy-ψ-indolen. Solche Hydroxyindolenine zeigen folgende charakteristische Reaktionen:

1. In saurem Medium gehen sie *inter*molekulare oder, falls geeignete Substituenten (zum Beispiel die Alanin-Seitenkette in Tryptophan) vorhanden sind, *intra*molekulare Kondensation ein zu Verbindungen vom Eserolin-Typus.

2. Sie addieren unter anderm Wasser (*a*), Essigsäure (*b*), Essigsäureanhydrid (*c*), Persäuren (*d*, X=Ac, Bz



usw.), Wasserstoffsuperoxyd (*d*, X=H) zu Produkten, die formal hydratisierten Ketonen (*a*), deren Mono- (*b*) und Ortho-Azetaten (*c*) sowie Ketonhydroperoxyden (*d*) entsprechen.

3. Die oxydative Ringspaltung, die über das Zwischenprodukt (*d*) erfolgt, ist ein Vorgang, welcher der Lactonbildung aus Ketonen mittels Persäuren und Wasserstoffperoxyd analog ist. Vom chemischen Standpunkt erklärt dieser Mechanismus den biologischen Abbau des Tryptophans zu Formylkynurenin zufriedenstellend.

4. Die Anlagerungsverbindungen von Wasser oder Essigsäure an β-Hydroxy-ψ-indole vom Typ (*a*) und (*b*) sind Glykole, die in saurer Lösung, falls Wasserabspaltung erfolgen kann (*a* R_2 = H), in Oxindole übergehen oder in alkalischem oder neutralem Medium möglicherweise oxydative Ringspaltung erleiden; ein Seitenweg zu Derivaten des Dioxindols ist möglich, würde aber nicht weiter zu Formylkynurenin führen. Die Bildung von Formylkynurenin aus Tryptophan nach Schema *B* würde den enzymatischen Versuchen von KNOX und MEHLER gerecht.

5. Als dritter Mechanismus (Schema *C* in der Übersicht auf Tafel I) für den biologischen Abbau des Tryptophans läßt sich ein β-Hydroperoxy-ψ-tryptophan nicht unbedingt ausschließen. Eine solche Verbindung könnte sich direkt zu Formylkynurenin umlagern.

Im Zusammenhang mit der Erörterung dieser Oxydationsmechanismen werden einige neue Beziehungen und Aspekte im Stoffwechsel des Tryptophans diskutiert.

[1] Cf. H. BURTON, Chem. and Ind. 313 (1948).

[2] F. C. HAPPOLD, Advances in Enzymology *10*, 62 (1950).

[3] F. G. HOPKINS and S. COLE, J. Physiol. *29*, 451 (1903).—R. ROBINSON, J. Chem. Soc., *115*, 968 (1919).