[CONTRIBUTION FROM THE LABORATORIES OF THE ROCKEFELLER INSTITUTE FOR MEDICAL RESEARCH]

The Synthesis of Nitro- and Aminoindoles Analogous to Serotonin

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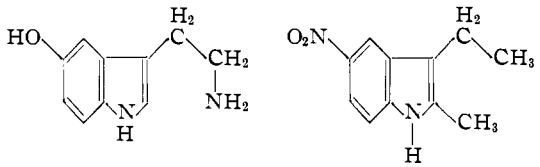
RECEIVED OCTOBER 24, 1952

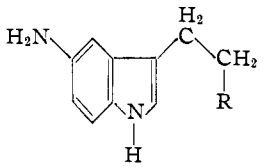
A number of new 3-alkyl- and 2,3-dialkyl-nitroindoles have been prepared by Fischer rearrangement. By the use of concentrated hydrochloric acid in a biphasic system at room temperature, aldehyde *p*-nitrophenylhydrazones have been converted to 3-alkyl-5-nitroindoles for the first time. From 3-β-chloroethyl-5-nitroindole, 5-nitrotryptamine and related bases were obtained. Although amino indoles have been relatively unknown, a number were readily prepared from the nitro compounds by reduction with hydrosulfite. Other reducing agents led to mixtures. 1,2,3-Trimethyl-5-nitroindole was formed when the sodium salt of 2,3-dimethyl-5-nitroindole was treated with methyl iodide in toluene.

The 5-aminoindoles were chosen for investigation as structural analogs of serotonin, the vasoconstrictor of serum, recently shown[1-3] to be 5-hydroxytryptamine (I). Compounds similar to (II), acting as metabolic antagonists,[4] were expected to prevent serotonin from exerting its physiological properties such as raising blood pressure. The possibility was considered that the appearance of excess serotonin might be the cause of some form of hypertension and that antagonists of serotonin might offer a therapeutic control. Expanding evidence of the widespread occurrence of serotonin[5] (or enteramine) and its possible physiological functions[6] increases the likelihood that substances able to regu-



I

III



II

late these functions will be pharmacologically useful. The first 5-aminoindoles which were prepared and tested as reported in a preliminary communication,[7] had indeed the desired ability to prevent constriction of segments of arteries caused by serotonin. Subsequently,[8] it was found that oral administration of 2-methyl-3-ethyl-5-nitroindole (III) to dogs was effective in preventing the rise in blood pressure usually induced by intravenous serotonin. The study of the relation of the structure of aminoindoles to antiserotonin activity has been extended to other compounds. The purpose of this paper is to describe the synthesis of a group of these compounds. Among the new bases are 5-aminotryptamine and its derivatives.

The relationship of the chemical structure of these nitro and amino indoles to their biological activity will be described in a subsequent paper. However, the statement may be made here that the 3-alkyl- and 2,3-dialkylaminoindoles were antimetabolites of serotonin when tested on segments of carotid arteries. Aminotryptamine was no antagonist but rather had serotonin activity. The nitroindoles and acylaminoindoles were inactive on the segments of artery, but some of these showed an effect in living animals presumably by conversion to the aminoindoles.

The desired aminoindoles were apparently to be obtained most readily from the nitro compounds provided by the Fischer indole rearrangement. Nitrophenylhydrazones of the alkyl ketones are readily converted to nitroindoles in fair yield in refluxing hydrochloric acid.[9-11] However, attempts to carry out a similar conversion of aldehyde *p*-nitrophenylhydrazones by this and other reagents have failed.[9,12,13] A solution to this problem was sought. Since Bauer and Strauss had isolated a propylidine di-2,2′-indole derivative on heating propionaldehyde *p*-nitrophenylhydrazone with hydrochloric acid, it appeared that the Fischer rearrangement took place but that free aldehyde formed by simultaneous hydrolysis of the starting material led to side reactions involving the unsubstituted 2-position. The rearrangement of *n*-butyraldehyde *p*-nitrophenylhydrazone by various reagents was investigated. The characteristic ultraviolet absorption spectrum of 5-nitroindoles (Table III) was used to determine the extent of 5-nitroindole formation. With zinc chloride the main product was of a different nature. However, it was discovered that concentrated hydrochloric acid *at room temperature* was adequate to bring about the formation of 3-ethyl-5-nitroindole. As eventually carried out, the hydrochloric acid solution of the nitrophenylhydrazone was stirred in the presence of a large benzene layer for several hours to permit extraction of the indole as it formed and to minimize side reactions. The number of products was still rather large and, although the desired indole could be obtained by crystallization from the mixture, chromatography on alumina was more effective. The yield of 3-ethyl-5-nitroindole was 20–25%. One of the by-products appeared to be 1,1-di-(3′-ethyl-

(1) M. Rapport, *J. Biol. Chem.*, **180**, 961 (1949).

(2) K. E. Hamlin and F. E. Fischer, THIS JOURNAL, **73**, 5007 (1951).

(3) M. E. Speeter, R. V. Heinzelman and D. I. Weisblat, *ibid.*, **73**, 5515 (1951).

(4) D. W. Woolley, "A Study of Antimetabolites," John Wiley and Sons, Inc., New York, N. Y., 1952.

(5) V. Erspamer and B. Asero, *Nature*, **169**, 800 (1952); B. Asero, V. Colo, V. Erspamer and A. Vercellone, *Ann.*, **576**, 69 (1952).

(6) V. Erspamer and A. Ottolenghi, *Experientia*, **8**, 232 (1952).

(7) D. W. Woolley and E. Shaw, THIS JOURNAL, **74**, 2948 (1952).

(8) D. W. Woolley and E. Shaw, *ibid.*, **74**, 4220 (1952).

(9) H. Bauer and E. Strauss, *Ber.*, **65**, 308 (1932).

(10) K. Schofield and R. S. Theobald, *J. Chem. Soc.*, 796 (1949).

(11) K. Schofield and R. S. Theobald, *ibid.*, 1505 (1950).

(12) G. K. Hughes, F. Lions and E. Ritchie, *J. Proc. Roy. Soc., N. S. Wales*, **72**, 209 (1939); *C. A.*, **33**, 6837 (1939).

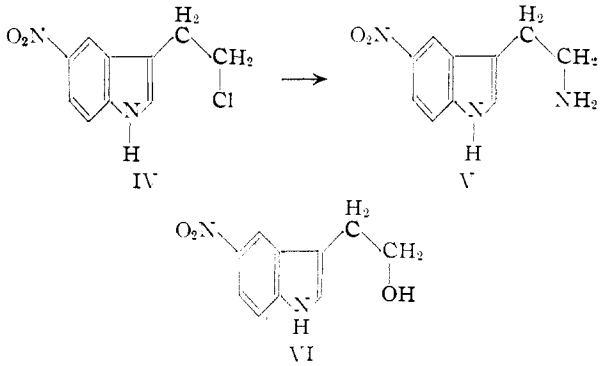
(13) H. N. Rydon and S. Siddappa, *J. Chem. Soc.*, 2462 (1951).

TABLE I
NITRO INDOLES

Indole	M.p., °C.	Yield, %	Analyses, % Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
3-Ethyl-5-nitro-	94–95[a]	25[i]	63.14	5.30	14.73	63.46	5.35	15.21
2-Methyl-3-*n*-butyl-5-nitro-	125–126[a]	21	67.21	6.94	12.06	66.93	6.85	12.36
1,2,3-Trimethyl-5-nitro-	138–139[a]	51	64.69	5.92	13.72	64.40	5.92	13.72
3-β-Chloroethyl-5-nitro-	120–121[a]	15[i]	53.47	4.04	12.47	53.81	4.09	12.41
3-β-Hydroxyethyl-5-nitro	97–98[b]	5[i]	57.98	4.79	13.57	58.29	4.89	13.59
5-Nitrotryptamine	136–139[b]	69	58.53	5.40		58.43	5.58	
3-β-Dimethylaminoethyl-5-nitro-, HCl	268–270[d]	57	53.43	5.97		53.51	6.04	
3-β-Piperidinoethyl-5-nitro-, HCl	272–273[d]	51	58.15	6.51		57.88	6.66	
3-β-N-Decahydroquinolylethyl-5-nitro-, HCl	254–256[d]	10	62.71	7.20		62.75	7.30	
3-β-(4-Imidazolylethylamino)-ethyl-5-nitro, dipicrate	207–208[c]	24	42.81	3.08		42.69	3.69	
2-Methyl-3-β-chloroethyl-5-nitro-	204–205[e]	32	55.34	4.64	11.74	55.59	4.67	11.94
2-Methyl-5-nitrotryptamine, HCl	265–266[f]	75	51.67	5.12		51.66	5.54	
2-Methyl-3-β-N-piperidinoethyl-5-nitro-, HCl	275–277[d]	88	59.36	6.85		59.55	6.80	
2,3,3-Trimethyl-5-nitroindolenine	124–125[g]	48	64.69	5.92		65.11	6.04	

[a] Solvents for recrystallization: benzene and hexane. [b] Aqueous alcohol. [c] Aqueous acetone. [d] 95% ethanol. [e] Benzene. [f] Dilute HCl. [g] Ligroin. [i] Yield given not of the m.p. described but of fraction obtained from chromatogram.

5′-nitro-2′-indolyl)-butane arising from condensation of the 3-ethyl-5-nitroindole with hydrolytically liberated butyraldehyde.

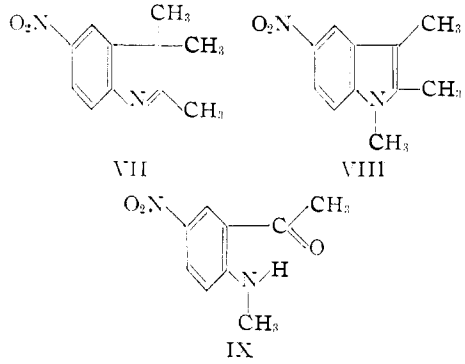


IV V VI

Under the conditions for the Fischer rearrangement described above, that is, concentrated hydrochloric acid and benzene at room temperature, γ-chlorobutyraldehyde *p*-nitrophenylhydrazone provided 3-β-chloroethyl-5-nitroindole (IV) in yields of about 15%. The corresponding alcohol VI was also formed in variable amounts due to hydrolysis of the chloro compound. From the chloroethylindole by reaction with ammonia and amines, 5-nitrotryptamine (V) and alkylated derivatives were accessible (Table I). Members of a similar series of amines bearing a 2-methyl group were prepared starting with methyl γ-chloropropyl ketone. The isolation of these amines was often facilitated by the low solubility of their hydrochlorides in water.

In the case of the unsymmetrical methyl alkyl ketones, Fischer rearrangement of the *p*-nitrophenylhydrazones has been shown in all cases examined to lead only to the 2-methyl-3-alkylindole by ring closure on the longer alkyl group.[11] In the present work only one product has been encountered in each rearrangement of a similar unsymmetrical ketone and the product has been considered to be a 2-methyl-3-alkylindole by analogy.

Alkylation of the ring nitrogen in 2,3-dialkyl-5-nitroindoles was realized by conversion of 2,3-dimethyl-5-nitroindole to 1,2,3-trimethyl-5-nitroindole (VIII) when a sodium salt of the former was refluxed with methyl iodide in toluene. Indolenines are usually formed on heating indoles with methyl iodide.[14] However, in the reaction with 2,3-dimethyl-5-nitroindole the structure of the product was established by the fact that the product was neutral, had the characteristic 5-nitroindole spectrum, and was different from the basic 2,3,3-trimethyl-5-nitroindolenine (VII) obtained from methyl isopropyl ketone *p*-nitrophenylhydrazone. Finally the methylation product, when subjected to chromic acid oxidation followed by acid hydrolysis according to the method of Schofield and Theobald,[11] lost two carbon atoms and provided a ketone the analysis of which agreed with the formula of the expected acetophenone IX.



VII VIII IX

The reduction of the 5-nitroindoles to 5-aminoindoles by a number of agents was studied. When this work was carried out the only previously reported, uncomplicated reduction of a nitroindole was the hydrosulfite reduction[9] of 2,3-dimethyl-5-nitroindole to the amine in undisclosed yield.[15–17]

(14) *Cf.* Summary by P. L. Julian, E. W. Meyer and H. C. Printy in Elderfield, "Heterocyclic Compounds," Volume 3, John Wiley and Sons, Inc., New York, N. Y., 1952.

(15) W. H. Perkin, Jr., and S. G. P. Plant, *J. Chem. Soc.*, **119**, 1825 (1921), reduced 6-nitro-1,2,3,4-tetrahydrocarbazole, in reality a 5-nitroindole, with iron and HCl to obtain 40% of the amine.

(16) Subsequently, the reduction of 2,3-dimethyl-6-nitroindole was reported by R. K. Brown, N. A. Nelson, R. B. Sandin and K. G. Tanner, THIS JOURNAL, **74**, 3934 (1952). A stannous chloride reduction followed by isolation of the base in an atmosphere of nitrogen gave a yield of 13%.

(17) S. G. P. Plant and W. D. Whitaker, *J. Chem. Soc.*, 283 (1940), reduced 2,3-dimethyl-4-nitro-7-chloroindole with tin and HCl to 2,3-dimethyl-4-aminoindole, losing the halogen. No yield was given.

TABLE II
AMINO INDOLES

Amino indoles	M.p., °C.	Yield, %	Analyses, % Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
3-Ethyl-5-amino-	116–118[a]	45	74.97	7.55	17.49	74.97	7.57	17.86
2,3-Dimethyl-5-amino-	173–174[b,d]	52	74.97	7.55		74.80	7.45	
2-Methyl-3-ethyl-5-amino-	148–149[a]	64	75.84	8.10	16.08	75.77	7.83	16.33
2-Methyl-3-*n*-butyl-5-amino-	96–98[a]	64	77.18	8.97	13.84	77.35	9.08	13.94
2,3-Dimethyl-4-amino-	156–160[a,e]	50	74.97	7.55	17.49	74.71	7.73	17.63
2,3-Dimethyl-6-amino-	117–118[a,f]	50	74.97	7.55		75.14	7.84	
2-Methyl-3-ethyl-7-amino-	110–112[a,g]	40	75.84	8.10	16.08	75.86	8.29	15.90
1,2,3-Trimethyl-5-amino-, picrate	203–205[b]	41	50.63	4.25		51.03	4.65	
5-Aminotryptamine dipicrate	204–205[c]	60	41.71	3.03	19.27	41.49	3.47	19.38
5-Amino-3-β-dimethylaminoethyl-, dipicrate	202–204[c]	38	43.57	3.50		43.71	3.68	
5-Amino-3-β-piperidinoethyl-, dipicrate	211–212[b]	67	46.23	3.88		46.51	4.04	
2-Methyl-5-aminotryptamine	112[a]	40	69.78	7.99		69.69	7.88	
2-Methyl-3-β-piperidinoethyl-5-amino-	149–151[a]	45	74.66	9.01	16.33	74.65	8.80	16.20
6-Amino-1,2,3,4-tetrahydrocarbazole	146–147[a,h]	64	77.39	7.58		77.67	7.57	

[a] Purified by distillation. [b] Recrystallized from aqueous alcohol. [c] From water. [d] Ref. 9, 178°. [e] Ref. 17, 163°. [f] Ref. 16, 119–120°. [g] Nitro compound prepared as described in ref. 11. [h] Ref. 15, 152°.

The procedure described could not be repeated and modified conditions were evolved which subsequently were used generally for nitroindoles with the results shown in Table II. In the presence of palladium-on-charcoal, 2,3-dimethyl-5-nitroindole was rapidly hydrogenated, but the amino compound thus formed melted considerably below that obtained by the hydrosulfite method, even after attempted purification. The ultraviolet absorption spectra of the amines from either method were essentially identical. Nevertheless, because of a possible persistent contamination of the catalytic reduction product with a dihydroindole, the hydrosulfite method was used exclusively thereafter.[18] With stannous chloride, a benzene-insoluble byproduct was formed in addition to the desired amine. Oxidation of the aminoindoles seemed to be favored by some metallic ions; material obtained by hydrosulfite reduction was more stable than that obtained by other methods.

A number of 5-aminoindoles (and position isomers) could be distilled or sublimed *in vacuo* with high recovery, providing perfectly white material of long stability. The eventual discoloration of bases so prepared appeared to be more a photochemical process than an oxidative one. Other bases were characterized as picrates since the hydrochlorides had no well-defined melting point. The ultraviolet absorption spectra of some aminoindoles are described in Table III.

When 2,3-dimethyl-5-aminoindole was diazotized, the resultant solution coupled with β-naphthol or N-(1-naphthyl)-ethylenediamine[19] only in strongly alkaline solutions, although the latter reagent is usually sensitive even in acidic solutions.[20] Of the other aminoindoles, the 6-amino isomer behaved similarly to the 5-, but the 4- and 7-amino-2,3-dialkylindoles gave precipitates on diazotization. Of these, only the product from the latter isomer retained some coupling ability.

TABLE III
ULTRAVIOLET ABSORPTION SPECTRA OF SOME NITRO- AND AMINOINDOLES IN 95% ETHANOL

Indole	Maxima λmμ	Maxima ε × 10⁻³	Minima λmμ	Minima ε × 10⁻³
4-Nitro-2-methyl-3-ethyl-	240–245 (plat.)	8.6	295	1.1
	340 (infl.)	2.8		
	408–412	4.35		
5-Nitro-2-methyl-3-ethyl-	276–277	21.0	240	5.65
	336–337	9.25	305	4.5
6-Nitro-2-methyl-3-ethyl-	251	9.1	265	5.9
	272	6.2	295	2.3
	340–350 (plat.)	7.1		
	395–400	9.25		
7-Nitro-2-methyl-3-ethyl-	240	10.6	250	9.25
	259–260	10.2	300	0.5
	372–378	6.28		
4-Amino-2,3-dimethyl-	230	31.0	253	3.6
	277–279	6.8	288–293 (plat.)	6.05
5-Amino-2-methyl-3-ethyl-	231–232	24.7	260	3.74
	284–286	6.84		
6-Amino-2,3-dimethyl-	235	30.9	255	4.0
	273–275	5.5	290	3.25
	307	5.1		
7-Amino-2-methyl-3-ethyl-	228	35.8	250	3.8
	275–277	8.5		
	300 (infl.)	4.5		

Experimental[21]

Fischer Indole Rearrangements.—The ketone *p*-nitrophenylhydrazones were refluxed with concd. HCl (10 ml. per g.) for three hours and the insoluble indole was filtered off, washed with concd. HCl, then with water. In the case of 2,3,3-trimethyl-5-nitroindolenine, obtained from methyl isopropyl ketone *p*-nitrophenylhydrazone by this treatment, the product remained in solution and was precipitated on neutralization of the reaction mixture. Products so obtained had a dark brown color which persisted through re-

(18) In one instance, the catalytic reduction of 2,3-dimethyl-5-nitroindole over palladium led to the isolation of what appeared to be 2,3-dimethyl-2,3-dihydro-5-nitroindole, m.p. 158–159°. Calcd. for $C_{10}H_{12}N_2O_2$: C, 62.49; H, 6.29; N, 14.58. Found: C, 62.65; H, 6.35; N, 14.73.

(19) A. C. Bratton and E. K. Marshall, Jr., *J. Biol. Chem.*, **128**, 537 (1939).

(20) The diazonium chloride of 3-aminocarbazole, structurally related to 5-aminoindoles, has been reversibly converted to an unstable diazoimine by *p*H changes; the imine coupled. *Cf.* G. T. Morgan and H. N. Read, *J. Chem. Soc.*, **121**, 2709 (1922).

(21) All melting points are uncorrected and were taken in a copper block.

crystallizations without lowering the m.p. However, in the case of 2-methyl-3-ethyl-5-nitroindole, which was required in large amounts for biological studies, it was considered desirable to remove all pigment. This was carried out by dissolving the crude indole in boiling benzene (20 cc. per g.) filtering, and treating the filtrate near the b.p. with alumina (Merck, 1.5 g. per g. of crude base). The filtrate from this was evaporated to $^1/_5$ its volume and the crystals collected. One recrystallization from alcohol raised the m.p. to 190–191°. Other examples are listed in Table I.

We are grateful to Dr. K. Schofield of the University College of the South West of England, Exeter, England, for samples of 4- and 6-nitro-2,3-dialkylindoles used for the measurement of spectra (Table III) and preparation of the corresponding amines.

Rearrangement of *n*-Butyraldehyde *p*-Nitrophenylhydrazone.—The hydrazone (10 g.) was dissolved in a suspension of *p*-nitrophenylhydrazine (5 g.) in concd. HCl (200 ml.) and a layer of benzene (200 ml.) added. The mixture was stirred for three hours when the benzene was replaced with fresh solvent for a second three hours. The combined benzene layers were washed with water, dried ($MgSO_4$), and concentrated to about 25 ml. This solution was chromatographically separated in benzene on a column (3.5 × 40 cm.) of Alumina Merck. The third yellow band to emerge consisted of the desired 3-ethyl-5-nitroindole, obtained as a crystalline residue, 2.2 g., on removal of the solvent. An orange band remaining near the top of the column was scooped out and eluted with ethanol providing, after recrystallization from ethanol, 1.1 g., m.p. 237–238°. This material also had the two peaks of absorption in the ultraviolet characteristic of 5-nitroindoles and was apparently 1,1-di-(3′-ethyl-5′-nitro-2′-indolyl)-butane.

Anal. Calcd. for $C_{24}H_{26}O_4N_4$: C, 66.34; H, 6.04; N, 12.90. Found: C, 66.46; H, 6.14; N, 12.95.

γ-Chlorobutyraldehyde *p*-Nitrophenylhydrazone and Its Rearrangement.—γ-Chlorobutyraldehyde was readily obtained from tetrahydrofurfuryl alcohol through the sequence described by Paul.[22,23] Instead of distillation of the aldehyde, the *p*-nitrophenylhydrazone was prepared directly. Thus, the ether extract from a periodic acid cleavage[23] of 245 g. of isomeric chloropentanediols was concentrated and the residue left at water-pump pressure for one hour to ensure removal of formaldehyde. The remaining material added to *p*-nitrophenylhydrazine (100 g.) in 50% acetic acid (2 l.) precipitated 103 g. of hydrazone, m.p. 101–102° (ref. 23, 110°), which was used without further purification.

The Fischer rearrangement of γ-chlorobutyraldehyde *p*-nitrophenylhydrazone (15 g.) was carried out in a 10% solution in concd. HCl stirred with two portions of benzene (750 ml.) as described above and the products resolved on alumina (7.5 × 30 cm.). The third band eluted was the desired 3-β-chloroethyl-5-nitroindole, 2.0 g., which remained as a partially crystalline residue after removal of the benzene and was used without further purification. The alumina containing the lowest band still on the column at this point was removed, and the material eluted with ethanol. This fraction consisted of the 3-β-hydroxyethyl-5-nitroindole. The first two yellow bands were not identified but yielded crystalline material. The first was 0.5 g., m.p. 134–135° (benzene and hexane): C, 54.02; H, 4.63; N, 9.03; Cl, 19.68. The second band yielded 1.63 g., m.p. 82-83° (aqueous alcohol): C, 57.03; H, 5.25; N, 9.65; Cl, 13.10.

5-Nitrotryptamine.—3-β-Chloroethyl-5-nitroindole (1 g.) in ethanol (75 ml.) was treated with concd. NH_4OH (50 ml.) in a stoppered flask at room temperature for 10 days. The alcohol was removed and the aqueous suspension of the tryptamine was acidified with 6 *N* HCl and filtered hot. Slow addition of alkali to the filtrate precipitated needles of 5-nitrotryptamine, 0.63 g., m.p. 136–139°, unchanged after recrystallization from aqueous alcohol.

3-β-Dimethylaminoethyl-5-nitroindole.—A solution of the chloro compound (IV, 1 g.) in ethanol (60 ml.) was left four days at room temperature with aqueous 25% dimethylamine (40 ml.). Isolation was carried out by removal of the alcohol under reduced pressure. The precipitated base was washed with water to remove soluble amines, then converted to the insoluble hydrochloride by addition of 6 *N* HCl.

(22) R. Paul, *Bull. soc. chim.*, [5] **8**, 911 (1941).

(23) R. Paul and S. Tchelitcheff, *ibid.*, [5] **15**, 197 (1948).

3-β-N-Piperidinoethyl-5-nitroindole.—A solution of the chloro compound (IV, 1 g.) in ethanol (20 ml.) was heated 40 hours at 50° with piperidine (5 ml.). Isolation was carried out as described in the preceding example.

3-β-(4′-Imidazolylethylamino)-ethyl-5-nitroindole.—A solution of the chloro compound (IV, 1 g.) in ethanol (50 ml.) was left three weeks at room temperature with histamine (1 g. of free base). After isolation of the free base as in the above cases, it was converted to the dipicrate with 2% aqueous picric acid. The base gave a deep red color with diazotized sulfanilic acid. Therefore the alkylation is considered to have taken place on the primary amino group of histamine rather than on the imidazole ring.

3-β-N-Decahydroquinolylethyl-5-nitroindole.—A solution of the chloro compound (IV, 1 g.) in ethanol (40 ml.) was left 17 days at room temperature with decahydroquinoline (10 ml., mixed isomers). The hydrochloride was isolated as in the above examples.

2-Methyl-5-nitrotryptamine.—2-Methyl-3-β-chloroethyl-5-nitroindole (1.5 g.) was dissolved in ethanol (75 ml.) and treated with concd. NH_4OH (30 ml.) at room temperature for seven days. The hydrochloride was isolated as in the above examples.

2-Methyl-3-β-N-piperidinoethyl-5-nitroindole.—An ethanolic solution of the chloro compound identical with that in the preceding example was left with piperidine (10 ml.) for one week at room temperature (45% yield) or refluxed for 24 hours (88% yield). The hydrochloride was isolated as in the above example.

1,2,3-Trimethyl-5-nitroindole.—To 2,3-dimethyl-5-nitroindole (3.8 g.) in toluene (300 ml.) was added an equivalent amount of ethanolic sodium ethylate (20 ml. of a molar solution) and the mixture distilled until the b.p. of toluene was reached. The system was then treated under reflux, protected against moisture, with an excess of methyl iodide until samples showed no alkalinity. The filtrate was concentrated to dryness and the residue applied in benzene to a column of alumina (3.4 × 30 cm.). The chromatogram was developed with the same solvent. The methylation product was rapidly eluted, yielding 51%, 2.1 g., of crystals, m.p. 136–139°.

Oxidation of 1,2,3-Trimethyl-5-nitroindole.—The methylation product (0.85 g.) was oxidized with chromium trioxide (0.85 g.) in glacial acetic acid as described by Schofield and Theobald[11] and the crude oxidation product hydrolyzed with aqueous alcoholic HCl[11] to yield 0.22 g. of ketone, m.p. 140–142°. Recrystallization from aqueous alcohol gave needles melting at 148–150°. A test for carbonyl function with 2,4-dinitrophenylhydrazine was positive.

Anal. Calcd. for $C_9H_{10}O_3N_2$: C, 55.67; H, 5.16; N, 14.43. Found: C, 55.68; H, 5.19; N, 14.49.

Aminoindoles by Hydrosulfite Reduction.—The following procedure is typical of that used for the bases listed in Table II. 5-Nitro-2,3-dimethylindole (5 g.) in ethanol (175 ml.) and *N* NaOH (100 ml.) was heated in a water-bath at 50° and treated with a solution of sodium hydrosulfite (25 g.) in *N*/2 NaOH (120 ml.). The almost colorless solution was filtered hot and the inorganic precipitate washed with alcohol. The combined filtrates were concentrated under reduced pressure. The product separated as the alcohol was completely removed. The aqueous suspension of the product was filtered and the base recrystallized from alcohol, yielding 2.2 g., 52%, m.p. 173–174°.

In the case of some nitroindoles a higher concentration of alcohol was required to keep the starting material in solution. When, in a reduction, the crude amine contained material not soluble in dilute acid, such contaminants were removed by filtration of an acidic solution and reprecipitation of the base with alkali. If this were not done, subsequent purifications were less effective. Generally, aminoindoles which did not crystallize in the above procedure on removal of alcohol were isolated by extraction of the oily base with ethyl acetate and eventual formation of a picrate. Crystalline bases were distilled or sublimed *in vacuo*. This was done simply by placing 100–200 mg. of the base at the lower end of a nearly horizontal tube (8–10 mm. diameter) and slowly heating with a free flame at about 1 mm. pressure. The base usually condensed about 1 inch away as a white crystalline mass.

2,3-Dimethyl-5-succinamidoindole.—An intimate mixture of 2,3-dimethyl-5-aminoindole (0.5 g.) and succinic anhydride (0.32 g.) was layered on the bottom of a 50-ml.

erlenmeyer flask which was placed for two minutes in an oil-bath at 125°. The melt was taken up in aqueous sodium carbonate. From the filtrate on gradual acidification was obtained the half amide, 0.62 g. A reprecipitation through the sodium salt gave crystals, m.p. 147–149°.

Anal. Calcd. for $C_{14}H_{16}O_3N_2$: C, 64.58; H, 6.20. Found: C, 64.26; H, 6.26.

Longer heating favored formation of the succinimide derivative, isolated as a carbonate insoluble fraction, m.p. 198–199° (from ethanol).

Anal. Calcd. for $C_{14}H_{14}O_2N_2$: C, 69.38; H, 5.82. Found: C, 69.22; H, 6.05.

NEW YORK, NEW YORK

[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, UNIVERSITY OF WISCONSIN]

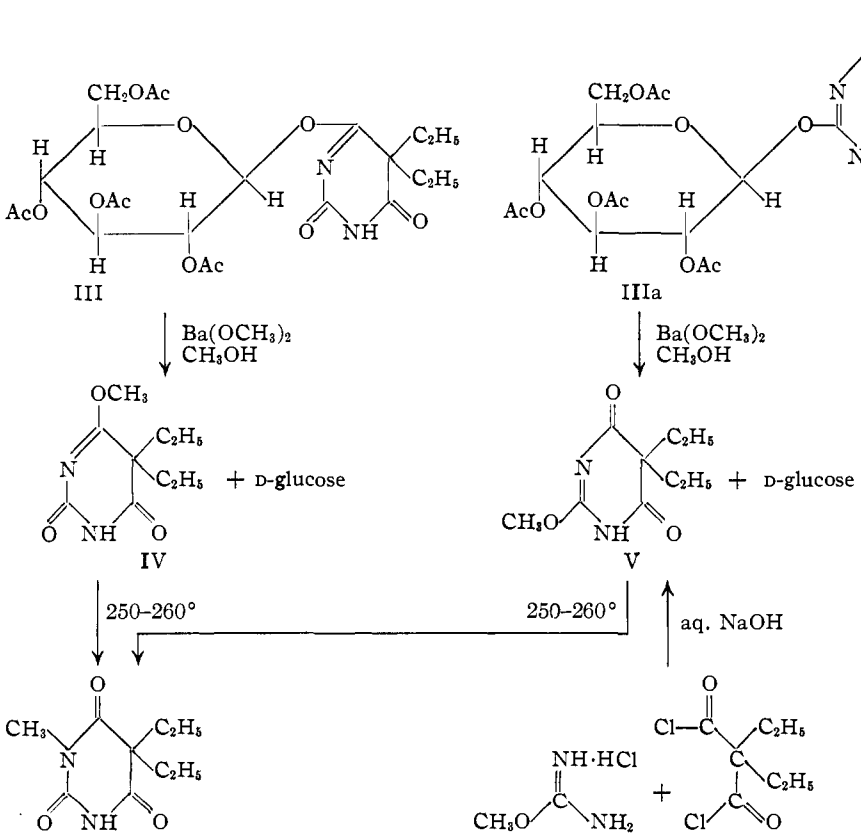
Preparation and Characterization by Alkaline Methanolysis of 5,5-Diethyl-4-(tetraacetyl-β-D-glucosyloxy)-2,6(1,5)-pyrimidinedione[1]

BY JACK A. SNYDER AND KARL PAUL LINK

RECEIVED NOVEMBER 15, 1952

The synthesis and alkaline methanolysis of 5,5-diethyl-4-(tetraacetyl-β-D-glucosyloxy)-2,6-(1,5)pyrimidinedione (III) is reported. The position of the glucosidic linkage was determined by comparison of the product of alkaline methanolysis, 5,5-diethyl-4-methoxy-2,6(1,5)-pyrimidinedione (IV), with 5,5-diethyl-2-methoxy-4,6(1,5)-pyrimidinedione (V) synthesized from diethylmalonyl chloride and O-methylisourea hydrochloride. The ultraviolet absorption spectra of IV and III are similar, but differ from that of V, indicating that rearrangement does not occur during the alkaline methanolysis of III.

Ballou and Link[2] found the ultraviolet absorption spectra of theobromine β-D-glucoside tetraacetate (I)[3] and the methoxy-3,7-dimethylpurine (II) formed by alkaline methanolysis of I to be similar (maxima at 245 and 295 mμ in methanol), but distinctly different from the spectra of theobromine and caffeine (maxima at 275 mμ). From this they concluded that the methoxyl group in II occupied the position at which the sugar molecule was attached, and because it was not known whether the glucosidic linkage in I was to position 2 or 6 of the aglucon, they suggested determination of the structure of II as a means of indicating the correct position of the linkage.



This method was applied to the characterization of 5,5-diethyl-4-(tetraacetyl-β-D-glycosyloxy)-2,6-(1,5)-pyrimidinedione (III), in which the glucosidic linkage might be to either the 2 (IIIa) or 4 (III) position. Because of the method of its preparation, III is assumed to be a β-glucoside. It is alkali-sensitive, and undergoes alkaline methanolysis in a manner similar to I.

Attempts to prepare III from the silver salt of barbital (5,5-diethylbarbituric acid) and tetraacetyl-D-glucosyl bromide in the method for the preparation of I, and from IV and barbital in the usual Koenigs–Knorr procedure were unsuccessful. The use of a catalytic amount of quinoline in the Koenigs–Knorr reaction, a modification of the Robertson procedure[4] introduced by Huebner, *et al.*,[5] gave the desired product. This modification has been used to prepare enol-glycosides of 4-hydroxycoumarins, β-keto esters, β-diketones and β-keto anil-

(1) Published with the approval of the Director of the Wisconsin Agricultural Experiment Station. Supported in part by the Research Committee of the Graduate School from funds supplied by the Wisconsin Alumni Research Foundation.

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(3) E. Fischer and B. Helferich, *Ber.*, **47**, 210 (1914).

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