cherichia coli 54, *Proteus vulgaris* 73, or *Pseudomonas aeruginosa* 77. The minimum inhibitory concentration of **2** in μg/ml for the following organisms is given in parentheses; **3** was either inactive as 50 μg/ml or was not tested, (−). An asterisk means static activity only. *Sarcina lutea* 14 (3), *Staphylococcus aureus* 15 (7−), *Corynebacterium fimi* 22 (3), *Candida albicans* 204 (4), *Saccharomyces cerevisiae* 216 (6), *Hansenula anomala* (8*−), *Aspergillus niger* 13 (9*−), *Penicillium notatum* 40 (9−), *Trichophyton mentagrophytes* 171 (2), *Mycobacterium smegmatis* 607 (2), *M. rhodochrous* 271 (2), *Micropolyspora brevicatena* 1086W/F (2*−), *Microellobosporia cinerae* 3855 (2), *Microbispora rosea* 3748 (2−), *Nocardia asteroides* 3409 (2.5), *N. madurae* 1091 (2−), *Actinoplanes sp.* W13 (2), *Streptosporangium roseum* W48 (2−).

Acknowledgment.—We wish to thank Mrs. Eva M. Fekete for valuable technical assistance and Mrs. Mary P. Lechevalier for testing the purified materials against microorganisms. We are grateful to Dr. L. E. McDaniel and his staff for large-scale production of both pigments and to Dr. Pat G. Hamilton of the Research Triangle Institute, Durham, North Carolina, for the toxicity tests. Finally, we wish to thank Professor Paul J. Scheuer for the sample of **2** from spinochrome M and Professor John C. Roberts for flaviolin trimethyl ether.

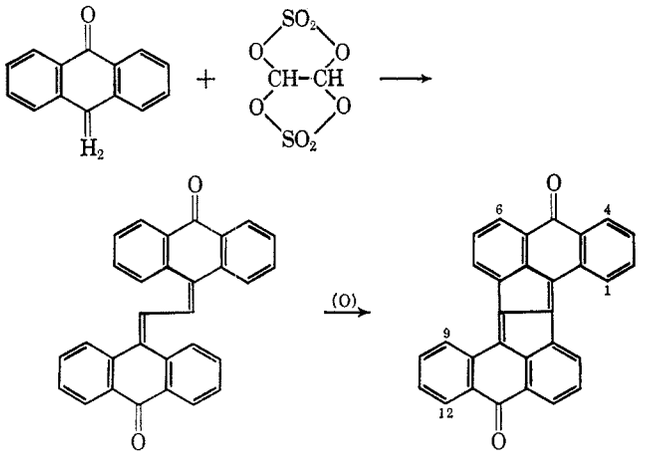
Vat Dyes from Benzimidazo[1,2-*b*]isoquinolin-5(12H)-one

MARIO F. SARTORI, AARON OKEN, AND HERMAN E. SCHROEDER

Research and Development Division Publication No. 392, Jackson Laboratory, Organic Chemicals Department, E. I. du Pont de Nemours and Company, Wilmington, Delaware

A benzimidazoisoquinolinone, prepared by Bistrzycki and Fassler[1] in 1923, is shown to be benzimidazo[1,2-*b*]-isoquinolin-5(12H)-one (II). Several routes to the corresponding dione (V) are described. The preparation and properties of heterocyclic vat dyes, analogs of acedianthrones, are reported.

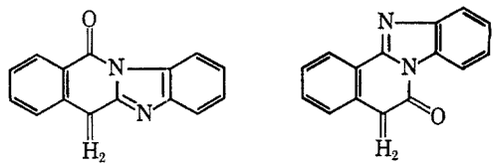
Aceanthryleno[2,1-*a*]aceanthrylene-5,13-dione (I) is the parent compound of an important class of vat dyes known as the acedianthrones. It is also noteworthy as an example of that uncommon class of compounds which contains the pentalene ring system. The method of preparation of I suggests that analogous structures, possibly useful as vat dyes, might be prepared from compounds which show a similar structural relationship



I

between a carbonyl group, a methylene group, and an aromatic ring. This paper describes the preparation of heterocyclic analogs of I from an intermediate possessing these structural features, benzimidazo[1,2-*b*]-isoquinolin-5(12H)-one (II).

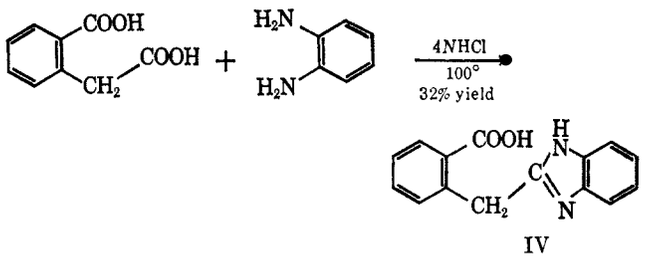
The fusion of *o*-phenylenediamine with homophthalic anhydride has been reported[1] to give a benzimidazoisoquinolinone of structure II or III.



II III

(1) A. Bistrzycki and K. Fassler, *Helv. Chim. Acta*, **6**, 527 (1923).

This ambiguity made it desirable to prepare II by an unequivocal route. First α-(2-benzimidazolyl)-*o*-toluic acid (IV) was prepared from *o*-phenylenediamine and homophthalic acid. Structure IV is assigned on the



IV

basis of the different reactivities of the two carboxyl groups of homophthalic acid. It is reported that benzoic acid and *o*-phenylenediamine heated at 100° in the presence of 4 *N* hydrochloric acid give traces of 2-phenylbenzimidazole,[2] whereas phenylacetic acid and *o*-phenylenediamine under the same conditions give 2-benzylbenzimidazole in 50–60% yield.[3]

Further evidence for structure IV is given by the similarity of the ultraviolet spectrum of IV with that of other 2-benzylbenzimidazoles (Table I). By comparison, the 2-phenylbenzimidazoles exhibit completely different spectra.

The cyclization of IV to II, was carried out in acetic anhydride at 60°. Evidence that the cyclized compound has structure II rather than III was provided by its conversion to the corresponding dione, which was identical with a sample of benzimidazo[1,2-*b*]isoquinoline-5,12-dione (V) prepared by the unambiguous route shown in Scheme I.

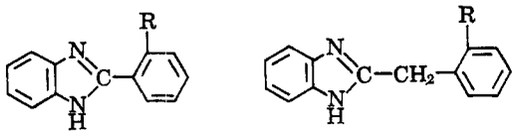
Comparison of the physical properties of II with those of the acetic acid soluble fraction of Bistrzycki's product[1] suggests that Bistrzycki's product also has structure II. The presence of a small amount of III in the reaction mixture of Bistrzycki is not excluded.

(2) M. A. Phillips, *J. Chem. Soc.*, 2395 (1928).

(3) G. K. Hughes and F. Lions, *J. Proc. Roy. Soc. N. S. Wales*, **71**, 221 (1938).

TABLE I

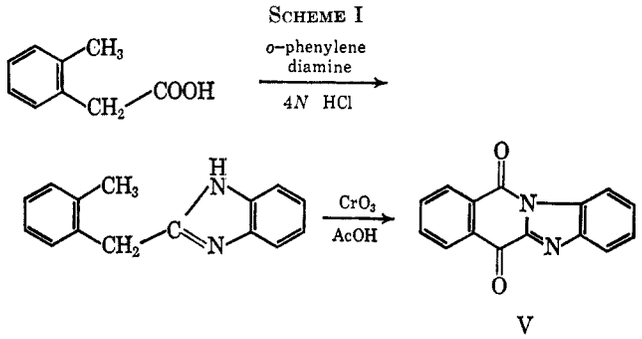
ULTRAVIOLET SPECTRA OF 2-PHENYLBENZIMIDAZOLES AND 2-BENZYLBENZIMIDAZOLES



R	λ_{max}, mμ	ε × 10^{-3}	λ_{max}, mμ[a]	ε × 10^{-3}[a]
H	303[a]	24.0[a]	274–281	9.1–10.2
CH_3	290[b]	15.8[b]	270–277	9.3–9.8
NH_2	289[c]	20.5[c]	274–281[d]	8.5–8.9
COOH	312[a,e]	27.5[a,e]	275–281	9.1–9.3

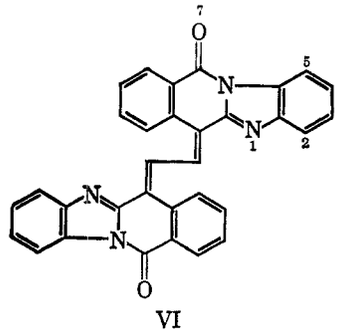
[a] The solvent used was 2-methoxyethanol. [b] The solvent used was chloroform [A. Mangini, F. Montanari, *Bull. Sci. Fac. Chim. Ind. Bologna*, **14**, 36 (1956)]. [c] The solvent was ethanol [G. Leandri, A. Mangini, F. Montanari, and R. Passerini, *Gazz. Chim. Ital.*, **85**, 769 (1955)]. [d] The 2-(*o*-aminobenzyl)-benzimidazole was obtained as white crystals (mp 240°) by catalytic reduction of 2-(*o*-nitrobenzyl)benzimidazole[3] under the conditions reported for the reduction of 2-benzyl-5-nitro-benzimidazole [B. N. Feitelson and R. Rothstein, *J. Chem. Soc.*, 2426 (1958)]. [e] Values given by *p*-(2-benzimidazolyl)benzoic acid.

SCHEME I

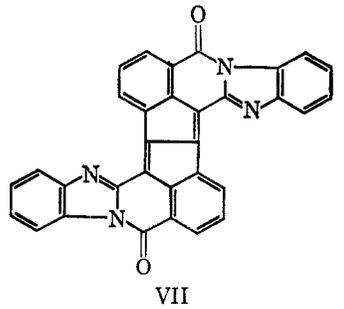


V

Benzimidazo[1,2-*b*]isoquinolin-5(12H)-one (II) reacts as an analog of anthrone with glyoxal sulfate; in dimethylformamide solution it gives a high yield of the ethanediylidene derivative (VI). Fusion of VI with sodium *m*-nitrobenzenesulfonate as the oxidizing agent in an aluminum chloride–sodium chloride melt gives a reddish violet compound (VII). This dye, when reduced with alkaline dithionite (hydrosulfite), gives a green solution of the corresponding hydroquinone anion, which shows a strong affinity for cotton and is readily reoxidized by air to the violet quinonoid form.

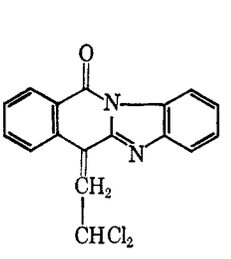


VI

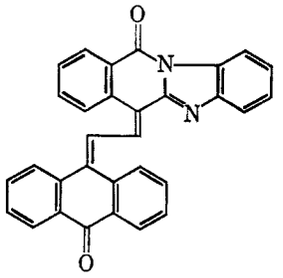


VII

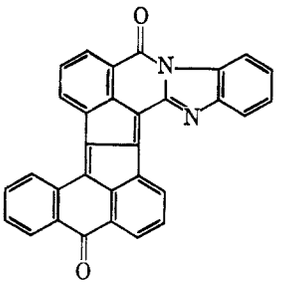
An unsymmetrical pentalene structure is obtained from II and anthrone by first condensing II with dichloroacetaldehyde to form the intermediate dichloro compound (VIII) and then condensing this with anthrone to form IX. Cyclization of this unsymmetrical ethanediylidene compound gives a highly colored product X, which forms a green vat and dyes cotton in violet-brown shades.



VIII



IX



X

Experimental Section

All melting points are uncorrected. Ultraviolet absorption spectra were recorded on a Cary Model 14 spectrophotometer and infrared absorption spectra were taken in Nujol mulls on a Perkin-Elmer Model 21 recording spectrophotometer equipped with sodium chloride optics.

α-(2-Benzimidazolyl)-*o*-toluic Acid (IV).—A stirred suspension of homophthalic acid (135 g) and *o*-phenylenediamine (54 g) in 4 *N* hydrochloric acid (500 ml) brought to reflux over a 30-min period was boiled for 40 min more and filtered hot through a steam-jacketed Büchner funnel, and the filtrate was allowed to cool to room temperature in about 30 min. The collected gray precipitate was slurried in water (2000 ml), made alkaline (pH 8) with dilute ammonium hydroxide, stirred at 50° for 15 min, and filtered hot. Acidification with acetic acid gave 40 g of gray powder (IV), which was further purified by repeating the above in the presence of Darco activated charcoal, filtering, and acidifying the chilled filtrate with dilute acetic acid.

Anal. Calcd for $C_{15}H_{12}N_2O_2$: C, 71.0; H, 4.7; N, 11.1. Found: C, 70.5; H, 4.6; N, 11.0

Benzimidazo[1,2-*b*]isoquinolin-5(12H)-one (II).—A stirred suspension of α-(2-benzimidazolyl)-*o*-toluic acid (12 g) in acetic anhydride (180 ml) was heated to 60° over a 30-min period and the stirring at 60° was continued for an additional hour. The collected yellow precipitate was washed with ethanol, slurried in dilute ammonium hydroxide at 40–50° for 30 min, collected, and washed with water. A 90% yield of crude II was obtained. Crystallization from acetic acid gave bright yellow crystals of mp 324–326°, by immersion of the capillary tube into a melting point bath preheated at 315°. II is soluble in dimethylformamide with a strong blue fluorescence.

Anal. Calcd for $C_{15}H_{10}N_2O$: C, 76.9; H, 4.2; N, 11.9. Found: C, 76.6; H, 4.2; N, 11.6.

Melting point, mixture melting point, and infrared spectral behavior showed II to be identical with material prepared according to Bistrzycki and Fassler[1] by condensing homophthalic

anhydride with *o*-phenylenediamine at 200° and purifying the crude product by crystallization from acetic acid.

Benzimidazo[1,2-*b*]isoquinoline-5,12-dione from II.—A solution of *p*-nitroso-N,N-dimethylaniline hydrochloride (0.33 g) in methanol (12 ml) was made alkaline (pH 8.5) by adding a 2 *N* solution of sodium hydroxide at 5°. Benzimidazo[1,2-*b*]isoquinolin-5(12H)-one (II, 0.55 g) dissolved in dimethylformamide (30 ml) was then added, and the mixture was stirred at room temperature for 10–15 min. The addition of water (50 ml) precipitated the *p*-dimethylaminophenylimino derivative of II (0.35 g) as violet needles, mp 236–238°.

Anal. Calcd for $C_{23}H_{18}N_4O$: C, 75.2; H, 4.9; N, 15.3. Found: C, 75.2; H, 4.9; N, 15.3.

To a slurry of the above *p*-dimethylaminophenylimino derivative (5 g) in acetic acid (125 ml) a 2 *N* solution of hydrochloric acid (20 ml) was added dropwise. After stirring at room temperature for 30 min, water (125 ml) was added, and the stirring was continued for an additional 10 min. The collected dull reddish precipitate (2.5 g) crystallized from acetic acid in the presence of a small amount of chromic acid gave 2 g of benzimidazo-[1,2-*b*]isoquinoline-5,12-dione of mp 267–269°. The infrared spectrum shows a strong carbonyl band at 5.86 and a carboxamide band at 5.98 μ.

Anal. Calcd for $C_{15}H_8N_2O_2$: C, 72.4; H, 3.2; N, 11.2. Found: C, 72.3; H, 3.1; N, 11.2.

Proof of Structure of Benzimidazo[1,2-*b*]isoquinoline-5,12-dione Obtained from II. 2-(*o*-Methylbenzyl)benzimidazole.—A stirred suspension of *o*-tolylacetic acid (21 g) and *o*-phenylenediamine (10.8 g) in 4 *N* hydrochloric acid (100 ml) brought to reflux over a 30-min period was boiled for 24 hr and then allowed to cool to room temperature. The collected precipitate was slurried in water (200 ml), made alkaline with sodium carbonate solution, and stirred for an additional hour. The filtered product recrystallized from ethanol gave white crystals (50% yield), mp 220–221°.

Anal. Calcd for $C_{15}H_{14}N_2$: C, 81.0; H, 6.3; N, 12.6. Found: C, 81.0; H, 6.3; N, 12.2.

Benzimidazo[1,2-*b*]isoquinoline-5,12-dione (V).—Chromic acid (6 g) was added in portions to a solution of 2-(*o*-methylbenzyl)-benzimidazole (2.1 g) in acetic acid (20 ml), maintained at 70°, and the mixture was stirred at this temperature for 20 hr. The precipitate obtained by pouring the reaction mixture into water and stirring for about 0.5 hr was collected, boiled with ethanol for 15 min, and filtered hot. Crystallization from acetic acid gave V as shiny, pale yellow plates (yield 30%) of mp 267–269°.

Anal. Calcd for $C_{15}H_8N_2O_2$: C, 72.4; H, 3.2; N, 11.2. Found: C, 72.3; H, 3.2; N, 11.1.

A mixture of V with the dione obtained from II (mp 267–269°) melted at 267–269°. The identity of the two diones of mp 267–269° was further substantiated by comparison of the infrared spectra.

12,12′-Ethanediylidenebisbenzimidazo[1,2-*b*]isoquinolin-5-(12H)-one (VI).—Eleven grams of glyoxal sulfate was added to a mixture of 24 g of benzimidazoisoquinolinone (II) in 200 ml of dimethylformamide. The temperature rapidly rose 10–15° and a thick precipitate of dark red needles began to form. The suspension was warmed to 80° and filtered hot. The precipitate washed with dimethylformamide and with alcohol and dried gave 22 g (87% yield) of VI, as red needles. VI is insoluble in all common organic solvents. It dissolves readily in sulfuric acid to give a blood red solution.

Bisbenzimidazo[1,2-*b*:1′,2′-*b*′]pentaleno[1,2,3-*d*,*e*:4,5,6-*d*′,*e*′]-diisoquinoline-9,19-dione (VII).—An intimate mixture of 25 g of VI and 25 g of sodium *m*-nitrobenzenesulfonate (Sitol) was added in small portions to a melt of 250 g of aluminum chloride, 65 g of sodium chloride, and 25 g of sodium *m*-nitrobenzenesulfonate held at 120–130°. One-half hour after the addition was complete, the hot mixture was drowned in 1500 ml of ice and water and then filtered. The moist cake was redissolved in 110 ml of sulfuric acid and reprecipitated into 200 ml of water. Sodium dichromate (50 g) was added, and the mixture was heated for 0.5 hr at 60–80°. The collected precipitate washed with water and with alcohol gave 12 g of VII, as a reddish violet solid. VII was purified by dissolving it in about 10 times its weight of concentrated sulfuric acid and then slowly diluting with cooling to a concentration of 85–90% sulfuric acid.

12-(2,2-Dichloroethylidene)benzimidazo[1,2-*b*]isoquinolin-5-(12H)-one (VIII).—Benzimidazoisoquinolinone II (120 g) was added slowly to a mixture of 1500 ml of dimethylformamide and 175 ml of freshly distilled dichloroacetaldehyde, while stirring at 45–50°. A thick mass of transparent needles formed. After 1 hr at 45–50°, the product was filtered and washed with alcohol to give 124 g of VIII. This product could not be recrystallized satisfactorily since it decomposed at elevated temperatures.

Anal. Calcd for $C_{17}H_{10}Cl_2N_2O$: Cl, 21.2; N, 8.4. Found: Cl, 21.6; N, 8.4.

12-(10-Oxo-9-anthrylideneethylidene)benzimidazo[1,2-*b*]isoquinolin-5(12H)-one (IX).—(Dichloroethylidene)benzimidazoisoquinolinone (VIII, 3.3 g) was dissolved in 25 ml of sulfuric acid to give a bright red solution. Anthrone (1.8 g) was then added, and the mixture was warmed to 40–50°. The solution became much bluer and hydrochloric acid was evolved. After 0.5 hr the filtered product was washed acid-free to give 3.6 g of IX as brown powder.

Anal. Calcd for $C_{31}H_{18}N_2O_2$: N, 6.2. Found: N, 6.3.

The product was recrystallized by carefully diluting a solution in sulfuric acid with water.

Anthra[9,1-*a*,*b*]pentaleno[4,5,6-*d*,*e*]benzimidazo[1,2-*b*]isoquinoline-8,18-dione (X).—A mixture of 1 g of IX and 1 g of sodium *m*-nitrobenzenesulfonate was added to a melt of 20 g of aluminum chloride, 5 g of sodium chloride, and 1 g of sodium *m*-nitrobenzenesulfonate maintained at 120–130°. After 0.5 hr at this temperature, the mixture was precipitated into a slurry of 150 ml of ice and 15 ml of hydrochloric acid. The filtered precipitate was washed acid-free to give 0.4 g of X as a dark brown powder.

Synthesis and Reactions of 17β-Acetoxy-5α-androstan-3-yl Isocyanates

EMERSON L. FOSTER AND ROBERT T. BLICKENSTAFF

Medical Research Laboratory, Division of Medicine, Veterans Administration Hospital, and Department of Biochemistry, Indiana University School of Medicine, Indianapolis, Indiana

Received December 2, 1965

17β-Acetoxy-5α-androstan-3α-yl isocyanate (IIIc) and 17β-acetoxy-5α-androstan-3β-yl isocyanate (IVc) were synthesized by the action of phosgene on the corresponding amines. The isocyanates react characteristically with ethanol, mercaptans, and amines to give carbamates, thiocarbamates, and ureas, respectively. Cysteine reacts preferentially at the thiol group.

Recent publication of the preparation of 3α-iodo-2β-cholestanyl isocyanate[1] prompts us to report our own work on the synthesis and reactions of two steroidal isocyanates.[2] Sodium borohydride reduction of 17β-acetoxy-5α-androstan-3-one gave a mixture of epimers predominating in the 3β-ol. Treating the mixture with methanesulfonyl chloride, according to Chang,[3] afforded a mixture of mesylates I and II that was separated by fractional crystallization (Scheme I). Individually the mesylates reacted with sodium azide in dimethyl sulfoxide[4] to give azides IIIa and IVa, which

(1) A. Hassner and C. Heathcock, *J. Org. Chem.*, **30**, 1748 (1965).

(2) Steroidal isocyanates have been prepared as intermediates, which were not characterized: I. G. Farbenind., British Patent 465,960 (1937); French Patent 819,975 (1937); Swiss Patent 225,781 (1943).

(3) F. C. Chang, *J. Chinese Chem. Soc.* (Taiwan), **9**, 53 (1962).

(4) K. Ponsold, *J. Prakt. Chem.*, **25**, 32 (1964).