55. *Lipoid-soluble Alloxazine Derivatives.*

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The preparation of a number of lipoid-soluble derivatives of alloxazine is described. The new compounds have no effect on tumour growth, but several cause pigmentation of the hair of the albino rat.

LETTRÉ and FERNHOLZ (*Ber.*, 1940, **73**, 436) have described the preparation of a number of lipoid-soluble derivatives of alloxazine and have tested them for carcinogenic activity. No such action was found during the period of their experiments, which they admitted were of a preliminary nature. They did, however, claim that one of their flavin derivatives—9-*n*-butyl-5 : 6-benz*iso*alloxazine—had a specific damaging effect on cancer cells *in vitro*. Haddow (unpublished results) has tested a number of the compounds described by Lettré and Fernholz for their action on the growth of the transplanted Walker rat carcinoma, but no inhibitory action has been found. The action of the 9-*n*-butyl derivative on tumour cells growing *in vitro* has also been tested by Ludford by a technique similar to that employed by Lettré and his co-workers, but again with negative results (unpublished work). In this paper we describe the preparation of further lipoid-soluble compounds related to alloxazine which have been made primarily in order to test their effect on tumour growth.

Two general methods have been employed for the synthesis of the alloxazines; the first involves the condensation of a substituted or unsubstituted *o*-diamine with alloxan, and the second makes use of the reaction between an *o*-diketone and a 4 : 5-diaminopyrimidine.

9-*Phenyl*-7 : 8-*benz*iso*alloxazine* (I) was obtained by condensation of alloxan with 2-amino-1-anilinonaphthalene. This diamine does not appear to have been made before by an unambiguous method. Harden (*Annalen*, 1889, **255**, 161) claimed to have obtained it by the action of phenylhydrazine on 1-nitroso-2-naphthylamine, but when Morgan and Godden (*J.*,

1910, **97**, 1721) attempted to repeat this work they obtained a product which did not have properties corresponding to the phenyldiamine. Noelting, Grandmougin, and Freimann (*Ber.*, 1909, **42**, 1381) considered that they had isolated this substance from the reduction products of *O*-alkyl derivatives of 2-benzeneazo-1-naphthol. The properties of the compound, which has now been obtained by the method outlined below, agree well with those of the substance described by Noelting *et al.* and so there is little doubt that these workers were the first to obtain 2-amino-1-anilinonaphthalene. The nitration of α-naphthylamine acetate followed by hydrolysis of the product gives a mixture from which 2-nitro-1-naphthylamine can be isolated by the method of Hodgson and Walker (*J.*, 1933, 1205). When the nitro-amine thus obtained is heated with bromobenzene in the presence of potassium carbonate and cuprous iodide, 2-nitro-1-anilinonaphthalene is formed; this is identical with a compound previously prepared by the action of aniline on 1 : 2-dinitronaphthalene (Vesely and Dvorak, *Bull. Soc. chim.*, 1923, **33**, 319). Reduction of the product with stannous chloride affords 2-amino-1-anilinonaphthalene. It was observed that 9-phenyl-7 : 8-benz*iso*alloxazine and other alloxazines now described tenaciously retain solvents after crystallisation, and it is only after intensive drying at high temperature or in some cases after sublimation in a high vacuum that satisfactory analytical figures can be obtained. Most of the new alloxazines have no true m. p. but decompose with evolution of gas at temperatures which are to some extent dependent on the rate of heating.

3 : 4-Diaminodiphenyl (obtained by the improved method of Bell and Kenyon, *J.*, 1926, 2705) condenses with alloxan to give 6- or 7-*phenylalloxazine* (II, *a* or *b*). 3-Nitro-4-aminodiphenyl (Fichter and Sulzberger, *Ber.*, 1904, **37**, 881) affords 3-*nitro*-4-*anilinodiphenyl* when heated with bromobenzene, potassium carbonate, and cuprous iodide, and the oily reduction product of this nitro-amine readily reacts with alloxan giving 6 : 9-*diphenyl*iso*alloxazine* (III). 3-*Nitro*-4-(1′-*naphthylamino*)*diphenyl* was obtained by heating the corresponding nitro-amine with α-bromonaphthalene in the presence of potassium and copper acetates. Catalytic reduction of the nitro-group gave 3-*amino*-4-(1′-*naphthylamino*)*diphenyl*, which did not react with alloxan under the usual conditions.

2 : 3-Diaminonaphthalene condenses with alloxan in dilute hydrochloric acid solution with the production of 6 : 7-*benzalloxazine* (IV). The compound can be obtained in the form of deep red crystals when sublimed in a high vacuum; it is the most deeply coloured alloxazine of the present series.

Waldman and Beck (*Annalen*, 1940, **545**, 52) have prepared 1-(2′-nitroanilino)naphthalene by heating *o*-bromonitrobenzene with α-naphthylamine in the presence of sodium acetate, and the same product has now been obtained by the action of α-bromonaphthalene on *o*-nitroaniline. They reduced this compound with sodium sulphide in ether or with stannous chloride in ethanolic hydrochloric acid; it is now found that 1-(2′-aminoanilino)naphthalene can be more conveniently prepared by catalytic reduction using Raney nickel. The diamine can be converted into 9-(1′-*naphthyl*)iso*alloxazine* (V) in the usual way. 2-(2′-Nitroanilino)naphthalene has been obtained by Warren and Smiles (*J.*, 1932, 2774) by heating 2-*o*-nitroanilinonaphthyl-1-sulphinic acid with acetic acid or by the reduction of either 2-*o*-nitroanilino-1-naphthyl disulphide or bis-2-*o*-nitroanilino-1-naphthyl sulphoxide with hydriodic acid. It has now been prepared by the more straightforward method of heating *o*-nitroaniline with β-bromonaphthalene in the presence of potassium carbonate and cuprous iodide. Catalytic reduction of the nitroamine affords 2-(2′-*aminoanilino*)*naphthalene* which condenses with alloxan to give 9-(2′-*naphthyl*)iso-*alloxazine* (VI).

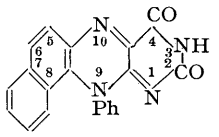
Benzyl-β-naphthylamine couples with the diazonium salt obtained from *p*-toluidine to give 2-*benzylamino*-1-p-*tolueneazonaphthalene*. 1-*Amino*-2-*benzylaminonaphthalene*, obtained by reduction of the azo-compound, can be converted into 9-*benzyl*-5 : 6-*benz*iso*alloxazine* (VII) by the usual procedure.

5 : 6 : 7 : 8-Dibenzalloxazine (VIII, X = O) and its 1 : 3-dimethyl derivative were prepared by the method of Kuhn and Cook (*Ber.*, 1937, **70**, 761), and 2-thio-5 : 6 : 7 : 8-dibenzalloxazine (VIII, X = S) (Robinson and Tomlinson, *J.*, 1935, 467) was also made for testing. The as yet undescribed 2-*imino*-5 : 6 : 7 : 8-*dibenzalloxazine* (VIII, X = NH) has now been obtained by condensation of 2 : 4 : 5-triaminopyrimidine with phenanthraquinone.

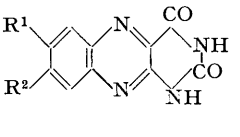
Pyridino(3′ : 2′ : 5 : 6)alloxazine (IX) was prepared by the method of Rudy (*Ber.*, 1938, **71**, 854); this involves the condensation of 5 : 6-diaminoquinoline with alloxan. Rudy admitted that his method of making the diamine, which was similar to that of Kaufman and Zeller (*Ber.*, 1917, **50**, 1626), was somewhat difficult to carry out mainly owing to the variable yields obtained in the nitration of 6-(*p*-toluenesulphonamido)quinoline. We have also experienced difficulty

with this procedure and so a simpler method of preparing the diamine has been developed : this is based on the reduction of 6-*amino*-5-p-*tolueneazoquinoline* with stannous chloride (compare Renshaw, Friedman, and Gajewski, *J. Amer. Chem. Soc.*, 1939, **61**, 3322). There is some confusion in the literature regarding the properties of 5 : 6-diaminoquinoline. Kaufman and Zeller describe it as forming yellow-brown crystals, m. p. 95°; Rudy obtained it as light brown crystals, m. p. 146°, after crystallisation from water in an atmosphere of nitrogen, and after distillation his product had m. p. 150°; and Renshaw *et al.* give the m. p. as 135°. Linkser and Evans have recently (*J. Amer. Chem. Soc.*, 1946, **68**, 874) improved Kaufman and Zeller's method of preparing the diamine dihydrochloride but they did not isolate the free base. As prepared by the method described in this paper, 5 : 6-diaminoquinoline forms golden-yellow, flattened needles, m. p. 149—150°, when crystallised from ether.

None of the alloxazine derivatives described in this paper has any appreciable effect on the growth of the transplanted Walker rat carcinoma. It was, however, observed that several of the compounds caused pigmentation of the hair of albino rats—a property first described by Haddow *et al.* (*Nature*, 1945, **155**, 379). These workers found that 9-phenyl-5 : 6-benz*iso*-alloxazine was the most effective substance in this respect, though several 9-alkyl-substituted alloxazines exhibited this property to a lesser degree. Of the compounds now tested 9-phenyl-7 : 8-benz*iso*alloxazine causes a salmon-coloured pigmentation of the rat's hair and is as effective as the 5 : 6-benz-derivative, whilst 6 : 9-diphenyl*iso*alloxazine produces an orange coloration and is somewhat less effective, though it is still much more active than the 9-alkyl-5 : 6-benz*iso*alloxazines. Substitution in the 3-position does not reduce the pigmenting power, since it has now been shown that 9-phenyl-3-methyl-5 : 6-benz*iso*alloxazine is as effective as the parent compound. It is noteworthy that in all the really active substances as yet tested there is a 9-phenyl substituent. Neither of the 9-naphthyl*iso*alloxazines is effective.



(I.)



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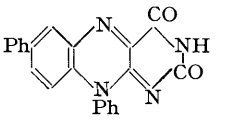
(II*b*, R^1 = H, R^2 = Ph.)



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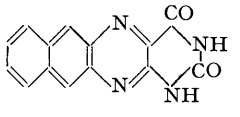

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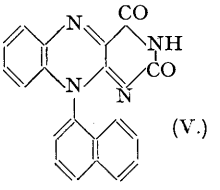

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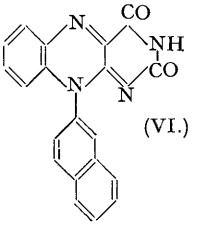

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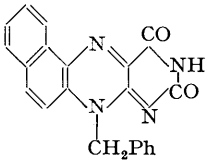

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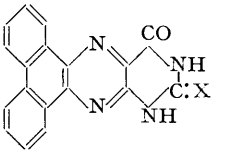
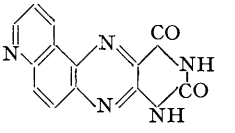


(VIII.)



(IX.)

Experimental.

All m. p.s, which are uncorrected, were determined in sealed capillaries.

2-*Nitro*-1-*anilinonaphthalene*.—50 G. of α-naphthylamine acetate were nitrated as described by Hodgson and Walker (*loc. cit.*) and the product was heated under reflux for one hour with methanolic potassium hydroxide (450 c.c., 5%). The mixture was diluted with water, and the solid which separated was collected and dried. It was then dissolved in glacial acetic acid (1 l.), and concentrated hydrochloric acid (50 c.c.) was added; this caused the precipitation of the hydrochloride of the 4-nitro-compound, which was filtered off and washed with acetic acid. The combined filtrates were diluted with an equal volume of water, whereupon the 2-nitro-amine separated; this was collected by filtration, washed with a little water, and dried. Yield 5 g., m. p. 140—143°. This method of separation was found more convenient than that first described by Hodgson and Walker which involves the solution of the mixed amines in nitrobenzene and subsequent saturation of the solution with hydrogen chloride. 2-Nitro-1-naphthylamine (2 g.), bromobenzene (20 c.c.), potassium carbonate (1·2 g.), and a little cuprous iodide

were refluxed together for 16 hours and then the excess of bromobenzene was removed by steam distillation. The residue was dissolved in benzene, and the solution passed down a short column of activated alumina. When the column was developed by washing with more benzene, a purple band due to the anilino-compound moved slowly down, whilst a red band due to unchanged nitro-amine remained at the top of the column. Evaporation of the benzene eluates gave an orange solid which crystallised from methanol in platelets, m. p. 113—114° (Found: N, 10·2. Calc. for $C_{16}H_{12}O_2N_2$: N, 10·6%). Vesely and Dvorak (*loc. cit.*) give m. p. 110—111°.

2-*Amino*-1-*anilinonaphthalene*.—The above nitro-amine (2 g.) was heated on a steam-bath for 2 hours with a mixture of stannous chloride (8 g.), concentrated hydrochloric acid (40 c.c.), and ethanol (80 c.c.). The solution, which was originally orange-yellow, soon became purple and then slowly decolorised as reduction proceeded. The mixture was diluted with water, and an excess of sodium hydroxide solution was added, after which the product was extracted with ether. The colourless solid obtained by evaporating the dried extract formed small prisms, m. p. 168—169°, from methanol (Found: N, 11·8. Calc. for $C_{16}H_{14}N_2$: N, 11·9%). Harden (*loc. cit.*) gives m. p. 160° and Noelting *et al.* (*loc. cit.*) give 170°. The solution of the base in methanol exhibits a pale blue fluorescence.

9-*Phenyl*-7 : 8-*benz*iso*alloxazine*.—The diamine (1 g.), alloxan (1 g.), boric acid (0·5 g.), and acetic acid (40 c.c.) were heated together on a steam-bath for ½ hour. The solution soon darkened and then deposited an orange-red solid: this solid was collected and recrystallised from acetic acid, giving 9-*phenyl*-7 : 8-*benz*iso*alloxazine* which decomposes at 360—365°. The analytical specimen was dried for 3 hours at 300° (Found: C, 70·8; H, 3·7; N, 16·4. $C_{20}H_{12}O_2N_4$ requires C, 70·6; H, 3·6; N, 16·5%). The alloxazine dissolves in dilute sodium hydroxide to give a light red solution and in acetone to give a yellow solution which exhibits a strong eosin-like fluorescence.

6- *or* 7-*Phenylalloxazine*.—3 : 4-Diaminodiphenyl (1 g.) (Bell and Kenyon, *loc. cit.*) was dissolved in water (25 c.c.) containing concentrated hydrochloric acid (3 c.c.), and the mixture was added to a hot solution of alloxan (1 g.) in water (10 c.c.) which was then heated on a steam-bath for ½ hour. The yellow precipitate of the *alloxazine* was collected, washed with water and then ethanol, and finally dried at 200°/0·001 mm. It decomposed at 335° (Found: N, 19·1. $C_{16}H_{10}O_2N_4$ requires N, 19·3%). The compound dissolves in dilute ammonia solution and to a less extent in acetone: both these solutions show a yellow-green fluorescence.

3-*Nitro*-4-*anilinodiphenyl*.—3-Nitro-4-aminodiphenyl (1·5 g.), potassium carbonate (0·9 g.), bromobenzene (10 c.c.), and a little cuprous iodide were heated together for 10 hours and then the excess of bromobenzene was removed by distillation in steam. A solution of the solid residue in benzene was percolated through a column of activated alumina. Early eluates contained an orange-red solid; this was 3-*nitro*-4-*anilinodiphenyl*, which crystallised from methanol in orange plates, m. p. 137—138° (Found: N, 9·9. $C_{18}H_{14}O_2N_2$ requires N, 9·7%). Further washing of the chromatographic column with benzene gave unchanged nitro-amine, m. p. 165—167°.

6 : 9-*Diphenyl*iso*alloxazine*.—The above nitro-base (1 g.), granulated tin (4 g.), ethanol (30 c.c.), and concentrated hydrochloric acid (15 c.c.) were heated together under reflux for 2 hours. After dilution with water, an excess of sodium hydroxide solution was added and the mixture was extracted with ether. The colourless oil obtained by evaporating this extract did not solidify, and so it was dissolved in glacial acetic acid (30 c.c.) and heated under reflux with alloxan (1 g.) and boric acid (0·5 g.). After one hour, water was added and the brown precipitate was collected by filtration. The *alloxazine* was crystallised from acetic acid, forming orange-brown flattened needles which decomposed at 380° (Found: N, 14·8. $C_{22}H_{14}O_2N_4$ requires N, 15·3%). 6 : 9-Diphenyl*iso*alloxazine is slightly soluble in acetone and the solution exhibits a brilliant yellow-green fluorescence.

3-*Nitro*-4-(1′-*naphthylamino*)*diphenyl*.—3-Nitro-4-aminodiphenyl (3 g.), α-bromonaphthalene (5 g.), fused potassium acetate (3 g.), and cupric acetate (0·2 g.) were heated together at the b. p. of the mixture for 4 hours. The product was boiled with light petroleum (b. p. 60—80°), and the extract passed down a short column of activated alumina. The red gum which remained was then dissolved in benzene, and this solution was poured on to the same column which was then developed by washing with more benzene. The first colourless eluates contained all the unchanged bromonaphthalene, and as a deep purple band moved off the column the eluates became orange-coloured and contained the required *nitro-diphenyl*. This formed scarlet plates, m. p. 146—147°, when crystallised from methanol (Found: N, 8·1. $C_{22}H_{16}O_2N_2$ requires N, 8·2%).

3-*Amino*-4-(1′-*naphthylamino*)*diphenyl*.—3-Nitro-4-(1′-naphthylamino)diphenyl (1 g.), Raney nickel catalyst (1 g.), and ethanol (100 c.c.) were shaken in an atmosphere of hydrogen until no further uptake of gas was observed (about 8 hours). The solution, which was originally orange-coloured, gradually became colourless and deposited a white solid. This solid, which was sparingly soluble in alcohol and ether, was dissolved by adding acetone and then boiling off the ether. The solution was filtered to remove the catalyst and then concentrated under reduced pressure. The *diamine*, crystallised from methanol–chloroform or methanol–acetone, separated in the form of colourless prisms, m. p. 184—185° (Found: N, 8·6. $C_{22}H_{18}N_2$ requires N, 9·0%). When the diamine (2 g.), alloxan (2 g.), and acetic acid (120 c.c.) were heated together for 2 hours the only isolatable product was the unchanged diamine.

6 : 7-*Benzalloxazine*.—2 : 3-Diaminonaphthalene (2 g.) was dissolved in water (75 c.c.) containing concentrated hydrochloric acid (5 c.c.). The solution was boiled with a little charcoal (to remove coloured impurity) and then filtered. Alloxan (2 g.), dissolved in water (25 c.c.), was added to the colourless filtrate, and the mixture was boiled for 10 minutes, a paste of orange-red solid then having formed. The product was collected by filtration, washed well with water and then alcohol, and finally dried in a steam-oven. The compound sublimed at 280°/0·001 mm., forming deep red prisms which decomposed at 385° [Found: (for material dried at 100°) C, 61·2; H, 3·4; (for the sublimed substance) C, 63·8; H, 3·0. $C_{14}H_8O_2N_4$ requires C, 63·6; H, 3·05%). 6 : 7-*Benzalloxazine* dissolves in dilute ammonia solution and in acetone: the former solution shows a reddish-blue fluorescence, and the latter an orange-yellow fluorescence.

1-(2′-*Nitroanilino*)*naphthalene*.—*p*-Nitroaniline (2 g.), α-bromonaphthalene (10 c.c.), potassium

carbonate (2 g.), and a small amount of cuprous iodide were heated together for 3 hours. The product was purified by the chromatographic method described above : it formed orange prismatic needles, m. p. 158—159°, when crystallised from methanol (Found : N, 10·3. Calc. for $C_{16}H_{12}O_2N_2$: N, 10·6%). Waldman and Beck (*loc. cit.*) give m. p. 155° for the product obtained by heating *o*-bromoaniline with α-naphthylamine.

1-(2′-*Aminoanilino*)*naphthalene*.—The above nitro-amine (2·5 g.) was reduced in ethanol solution by aid of a Raney nickel catalyst in the manner already described : reduction was complete in one hour. The oil obtained after evaporating the filtered ethanolic solution solidified on rubbing with light petroleum (b. p. 60—80°). The diamine (1·5 g.) crystallised from benzene–light petroleum in well-defined prismatic tablets, m. p. 136—138° (Found : N, 11·9. Calc. for $C_{16}H_{14}N_2$: N, 11·9%). Waldman and Beck (*loc. cit.*) give m. p. 135°. The colourless solid is quite stable, but solutions of the diamine readily darken in air.

9-(1′-*Naphthyl*)iso*alloxazine*.—The diamine (0·5 g.) was dissolved in acetic acid (20 c.c.) containing concentrated hydrochloric acid (1 c.c.), and after addition of alloxan (0·5 g.) in water (2 c.c.) the mixture was heated on a steam-bath for one hour. The orange-brown crystals which had separated were collected and washed with a little methanol. On recrystallisation from acetic acid the *naphthyl*iso*alloxazine* formed deep yellow needles which decomposed at 265° (Found : N, 15·9. $C_{20}H_{12}O_2N_4$ requires N, 16·4%). The substance dissolved in dry acetone to give a yellow non-fluorescent solution but on addition of water a deep blue fluorescence developed.

2-(2′-*Nitroanilino*)*naphthalene*.—*o*-Nitroaniline (3 g.), β-chloronaphthalene (7 g.), fused potassium acetate (3 g.), and cupric acetate (0·1 g.) were heated together under reflux for 3 hours. The unchanged chloronaphthalene was removed by chromatographic separation, and the required compound was crystallised from methanol; it formed long prismatic orange needles, m. p. 112—113° (Found : N, 10·7. Calc. for $C_{16}H_{12}O_2N_2$: N, 10·6%). Warren and Smiles (*loc. cit.*) give m. p. 110°.

2-(2′-*Aminoanilino*)*naphthalene*.—The nitro-amine was catalytically reduced as previously described, and the residue obtained by evaporation of the ethanol was crystallised from light petroleum (b. p. 60—80°), from which it formed colourless prismatic needles, m. p. 84—85° forming a cloudy melt which cleared at 100° (Found : N, 12·0. $C_{16}H_{14}N_2$ requires N, 11·9%).

9-(2′-*Naphthyl*)iso*alloxazine*.—This *alloxazine* was formed from the above *diamine* in the usual way and was obtained as an orange microcrystalline powder which decomposed at 280° (Found : N, 16·0. $C_{20}H_{12}O_2N_4$ requires N, 16·4%).

Benzyl-β-naphthylamine.—β-Naphthylamine (5 g.), benzaldehyde (5 c.c.), and methanol (75 c.c.) were warmed for ½ hour on a steam-bath by which time all the material had passed into solution. The cooled solution deposited 7 g. of benzylidene-β-naphthylamine, m. p. 102°. This was dissolved in ethanol (100 c.c.), Raney nickel catalyst (2 g.) added, and the mixture shaken in an atmosphere of hydrogen. The theoretical amount of gas was absorbed during 2 hours and then the solution was filtered and evaporated. The colourless residue (6 g.) crystallised from light petroleum (b. p. 60—80°) in prisms, m. p. 68°.

2-*Benzylamino*-1-p-*tolueneazonaphthalene*.—A solution of the diazonium salt obtained by adding sodium nitrite (1·75 g.) to *p*-toluidine (2·7 g.) dissolved in a mixture of concentrated hydrochloric acid (8 c.c.) and water (25 c.c.) was added to a cooled solution of benzyl-β-naphthylamine (6 g.) in ethanol (300 c.c.). After a short time an orange solid began to separate, and the mixture was left in an ice-chest overnight to complete the reaction. The *azo*-compound was collected and recrystallised from methanol, forming long, orange-red needles (8 g.), m. p. 105—106° (Found : C, 82·3; H, 6·2; N, 12·5. $C_{24}H_{21}N_3$ requires C, 82·1; H, 6·0; N, 12·0%).

1-*Amino*-2-*benzylaminonaphthalene*.—A solution of stannous chloride (20 g.) in concentrated hydrochloric acid (30 c.c.) was added to a solution of the azo-compound in ethanol (300 c.c.) and the mixture was warmed on a steam-bath until the colour was discharged (about 30 minutes). After dilution with water, an excess of 10N-sodium hydroxide was added, and the cooled mixture was extracted with ether. The ethereal extract, which showed a strong yellow-green fluorescence, was dried and evaporated. When the residue was crystallised from light petroleum (b. p. 60—80°) the *diamine* was obtained as colourless prisms, m. p. 86—87° (Found : C, 81·9; H, 6·4. $C_{17}H_{16}N_2$ requires C, 82·2; H, 6·5%). Solutions of the diamine rapidly darken in contact with air.

9-*Benzyl*-5 : 6-*benz*iso*alloxazine*.—The benzyldiamine (1 g.), alloxan (1·5 g.), and boric acid (2·0 g.) were dissolved in glacial acetic acid (70 c.c.), and the solution heated under reflux for 30 minutes. Careful dilution of the mixture with water caused precipitation of short orange-brown needles (0·6 g.). 9-*Benzyl*-5 : 6-*benz*iso*alloxazine* formed orange needles, decomposing at 295°, when recrystallised from acetic acid (Found : C, 70·9, 71·0; H, 4·5, 4·5. $C_{21}H_{14}O_2N_4$ requires C, 71·2; H, 4·0%). The alloxazine dissolved in dilute sodium hydroxide solution to give a yellow solution and in acetone or methanol to give a strongly fluorescent solution (yellow-green).

2-*Imino*-5 : 6 : 7 : 8-*dibenzalloxazine*.—The bisulphite compound of 2 : 4 : 5-triaminopyrimidine (prepared by the method of Totter, *J. Biol. Chem.*, 1944, **152**, 147) was suspended in water, and dilute sulphuric acid added. After a short time the insoluble sulphate was collected, and 2 g. of this salt were heated in acetic acid (100 c.c.) with phenanthraquinone (2 g.), the mixture being gently boiled for 15 minutes by which time a buff-coloured solid had separated. This was the required *iminoalloxazine*; it decomposed when heated above 400° (Found : N, 22·2. $C_{18}H_{11}ON_5$ requires N, 22·4%).

6-*Amino*-5-p-*tolueneazoquinoline*.—6-Nitroquinoline (Cook, Heilbron, and Steger, *J.*, 1943, 416) was reduced in ethanol solution in the presence of a palladium–strontium carbonate catalyst (compare Rudy, *Ber.*, 1938, **71**, 854). 6-Aminoquinoline (1·44 g.) was dissolved in ethanol (50 c.c.), and to the ice-cooled solution was added the diazonium salt prepared from *p*-toluidine (1·07 g.) dissolved in water (10 c.c.) containing concentrated hydrochloric acid (3 c.c.), and a solution of sodium nitrite (0·7 g.) in water (5 c.c.). The mass of red crystals which separated overnight was collected by filtration and washed with a little methanol. The *azo*-compound formed red flattened needles, m. p. 245° (decomp.), when crystallised from methanol (Found : N, 21·4. $C_{16}H_{14}N_4$ requires N, 21·4%).

Pyridino(3′ : 2′ : 5 : 6)*alloxazine*.—The azo-compound (0·8 g.) was dissolved in concentrated hydrochloric acid (12 c.c.), and after the addition of stannous chloride (4 g.) the mixture was heated on a steam-bath. The solution at first assumed a deep carmine colour but as reduction proceeded a light orange colour developed and a solid separated. When no further lightening of the colour occurred, more hydrochloric acid (5 c.c.) was added and the mixture was cooled in ice. The crystalline product was filtered off and washed with methanol. The free base was obtained by dissolving the dihydrochloride in water and then covering the solution with a layer of ether; an excess of sodium hydroxide solution was then added, and the mixture well shaken. The dried ethereal extract was concentrated until golden-yellow flattened needles of 5 : 6-diaminoquinoline separated. After a further crystallisation from ether the product had m. p. 149—150°. Pyridinoalloxazine was prepared from the diamine by the method of Rudy (*loc. cit.*); it formed a yellow microcrystalline powder, decomposing above 400°, when crystallised from formic acid.

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