57. *Acridine Syntheses and Reactions. Part III. Synthesis of Amino-acridines from Formic Acid and Amines.*

By Adrien Albert

The mechanism of the condensation of *m*-phenylenediamine with formic acid to give proflavine (2 : 8-diaminoacridine) was investigated in Parts I and II (Albert, *J.*, 1941, 121, 484). A study of the scope of this synthesis, using a variety of amines, now shows it to be a most useful method for preparing symmetrically substituted derivatives of proflavine.

The following examples of the reaction of oxalic (and/or formic) acid with diamines to give yellow dyes are in the patent literature: (i) with *m*-tolylenediamine to give an un-named product (CIBA, F.P. 203,467, 1890); (ii) with *m*-aminodimethylaniline, *m*-aminodiethylaniline, *m*-aminoethylaniline, *m*-aminomethyl-*o*-toluidine, and *m*-aminoethyl-*o*-toluidine to give un-named acridine dyes (Leonhardt, D.R.-P. 67,126, 1890); (iii) with *m*-phenylenediamine to give proflavine (Poulenc, D.R.-P. 347,819, 1921); (iv) with *N*-(diethylaminoethyl)-*m*-phenylenediamine to give 2 : 8-bis(diethylaminoethylamino)acridine (I.G. Farben., D.R.-P. 488,890, 1924); (v) with 2 : 4-diamino-anisole or -phenetole to give 3 : 7-dimethoxy- or -diethoxy-proflavine (B.D. Corp., B.P. 248,282, 1926).

In Parts I and II (*locc. cit.*), conditions for the synthesis of proflavine from *m*-phenylenediamine were explored and the reaction was shown to proceed *via* *m*-aminoformanilide and tetra-aminobenzhydrol (yield, 60%). As no other use of the reaction is recorded in the general literature, a survey of its scope and probable usefulness has been attempted.

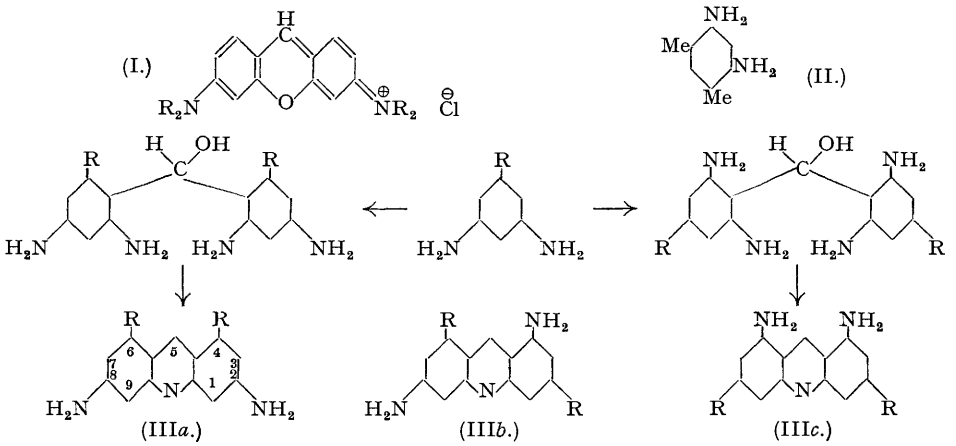
Amines other than m-*Diamines.*—No acridines were obtained when the conditions optimal for *m*-phenylenediamine were applied to the following amines, which were recovered unchanged or in the form of their formyl or oxalyl derivatives: aniline, *p*-phenylenediamine, *p*-toluidine, *m*-toluidine, *m*-anisidine, *m*-chloroaniline, *m*-nitroaniline, *m*-aminobenzoic acid, and metanilic acid. No improvement resulted on increasing the severity of the conditions (4 hours at 200° instead of ¾ hour at 155°) or by varying the amount of hydrogen chloride between 0 and 2 molecules per molecule of amine. *o*-Phenylenediamine gave benziminazole (with formic acid) and a tetracyclic compound, fluoflavine (with oxalic acid, cf. Hinsberg and Pollak, *Ber.*, 1896, 29, 784).

m-Aminophenol could conceivably react in three ways, losing ammonia or water to give (i) 2 : 8-dihydroxyacridine, (ii) 2-amino-8-hydroxyacridine, or (iii) 2 : 8-diaminoxanthylium chloride (I, R = H). Since *N*-acylation takes precedence over *O*-acylation, the first intermediate should be *m*-hydroxyformanilide (cf. Part II), and possibility (iii) is rendered unlikely. Actually, no (I) was formed, the principal product being a *complex* of two molecules of 2-amino-8-hydroxyacridine and one of 2 : 8-dihydroxyacridine. *m*-Diethylaminophenol, which cannot give an acridine, produced Pyronine B (I, R = Et) in good yield; this constitutes a new xanthen synthesis.

The failure of *m*-toluidine and *m*-anisidine to give acridines with formic acid reveals that a high electron density *ortho* to an amino-group is a prerequisite for this reaction; the failure of *p*-toluidine stands in contrast to its ready reaction with formaldehyde and mineral acid to give 3 : 7-dimethylacridan (Ullmann, *Ber.*, 1903, 36, 1018).

These results indicate that the main usefulness of the reaction under consideration will be confined to *m*-diamines.

N-*Substituted* m-*Diamines.*—*m*-Aminodimethylaniline was condensed with oxalic acid to give a 60% yield of acridine-orange (2 : 8-bisdimethylaminoacridine) identified by its having



the same properties as, and not depressing the m. p. of, a specimen prepared by the action in turn of formaldehyde, hydrochloric acid, and ferric chloride on the same amine (Biehringer, *J. pr. Chem.*, 1896, **54**, 241). A cream-coloured by-product (3% yield) proved to be *bis*-m-*dimethylamino-oxanilide*, confirmed by its synthesis from *m*-aminodimethylaniline and oxalyl chloride. That this compound is a by-product and not an intermediate was shown by its failure to give an acridine when heated with zinc chloride and glycerol. As formic acid had proved somewhat superior to oxalic acid up to this point, the use of the latter was abandoned in the remainder of this survey.

3-Aminodiphenylamine gave a 40% yield of 2 : 8-*bisphenylaminoacridine* and a by-product, m. p. 217°, which proved to be 2-aminoacridine.

C-*Substituted* m-*Diamines.*—It was shown (Part II) that *m*-phenylenediamine could conceivably yield three isomerides, *viz.*, 2 : 8-, 2 : 6-, and 4 : 6-diaminoacridine (III*a*, *b*, and *c*; R = H), but only the 2 : 8-isomeride (proflavine) is actually produced. It is now found that 4 : 6-diamino-*m*-xylene (II), which could give only 4 : 6-diamino-1 : 3 : 7 : 9-tetramethylacridine, does not react at all. It seems that there is little or no tendency for the carbon bridge between the two benzene rings to be formed by an attack on the 2-position.

In the case of 1 : 3-diamines with a substituent in the 2-position, the formation of isomerides is physically impossible. 2 : 6-Tolylenediamine gave 2 : 8-*diamino*-1 : 9-*dimethylacridine* (57% yield), characterised as its *monoacetyl* derivative, and some by-products, not obtained pure, resembling the family of diacridyl ethers described in Part I.

In the case of 1 : 3-diamines having a substituent in the 5-position, there are the same possibilities of formation of isomerides as with *m*-phenylenediamine. However, only one product analysing for a diaminodimethylacridine was isolated from 3 : 5-tolylenediamine. It was a fairly strong base ($pK_a = 10{\cdot}1$ in 50% ethanol at 20°) with yellow salts and, for the reason given above, was assigned the constitution 2 : 8-*diamino*-4 : 6-*dimethylacridine* (III*a*; R = Me). Data obtained in the determination of pK_a values for 120 acridines (Albert and Goldacre, *J.*, 1946, 706) enable the following values to be predicted for (III*a*, *b*, and *c*) respectively: 10·0, 9·1, and 6·6 (all $\pm$ 0·5). This confirmatory evidence may be supplemented by the strong likelihood that a 4 : 6-diamine (such as III*c*) would have blue or green salts (Albert and Bird, *J. Soc. Dyers and Col.*, 1943, **59**, 74, and unpublished work). 3 : 5-*Diaminoanisole* was prepared for use in this reaction, but underwent demethylation during the condensation which resulted in an intractable mixture. 3 : 5-Diaminobenzoic acid gave a 65% yield of a diaminoacridinedicarboxylic acid (probably 2 : 8-4 : 6-). This resisted decarboxylation, a process which could have confirmed the orientation.

1 : 3-Diamines having a substituent in the 4-position can conceivably give rise to six isomerides; for example, 2 : 4-tolylenediamine could give (i) 2 : 8-diamino-3 : 7-dimethylacridine, (ii) 2 : 6-diamino-3 : 9- (and 3 : 7-) dimethylacridines, and (iii) 4 : 6-diamino-1 : 7- (and 1 : 9- and 3 : 7-) dimethylacridines. Groups (ii) and (iii) are excluded by the results obtained with 4 : 6-diamino-*m*-xylene (see above). In each of the following cases, only one disubstituted diaminoacridine could be isolated and the pK_a values were consistent with the amino-

groups being in the 2 : 8-positions. 2 : 4-Tolylenediamine gave a 75% yield of acridine-yellow (2 : 8-diamino-3 : 7-dimethylacridine) identical with the product obtained by the action in turn of formaldehyde, hydrochloric acid, and ferric chloride on the same amine (Ullmann and Fitzenkam, *Ber.*, 1905, **38**, 3794). No m. p. has been recorded for acridine-yellow; it was found to be 325° (quite sharp) for specimens prepared by either route and no depression occurred when they were mixed.

Likewise 2 : 4-diaminoanisole, 2 : 4-diaminophenetole and chloro-2 : 4-diaminobenzene gave respectively 2 : 8-*diamino*-3 : 7-*dimethoxy*-, 2 : 8-*diamino*-3 : 7-*diethoxy*-, and 3 : 7-*dichloro*-2 : 8-*diamino-acridine*. The yields were only moderate, and diacridyl ethers were simultaneously formed. 3 : 7 : 3′ : 7′-*Tetrachloro*-2 : 8 : 2′ : 8′-*tetra-amino*-5 : 10-*dihydrodiacridyl* 5 : 5′-*ether* was obtained analytically pure.

When 2-dimethylamino-*p*-toluidine was condensed with formic acid, 2 : 8-*bismethylamino*-3 : 7-*dimethylacridine* was obtained. The loss of two *N*-methyl groups under hot, acid conditions may be connected with the serious clashing of van der Waals radii (*N*-methyl with *C*-methyl) which a scale drawing reveals. Lewis and Bigeleisen (*J. Amer. Chem. Soc.*, 1943, **65**, 1144) draw attention to the considerable steric effect of an *ortho* methyl group upon a dimethylamino-group which is thereby thrown out of the plane of the (aromatic) ring.

Non-symmetrical Condensations.—Unsuccessful attempts were made to condense together two different amines under the above conditions (*m*-aminoformanilide, or *m*-phenylenediamine plus formic acid, with aniline hydrochloride or *p*-phenylenediamine dihydrochloride; also formanilide with *m*-phenylenediamine dihydrochloride). In each case proflavine was the only acridine formed, usually in good yield, thus demonstrating the mobility of the formyl group under the experimental conditions. Similar results were obtained when substantially anhydrous conditions were used.

This led to the investigation of five patents which appeared to produce non-symmetrically substituted acridines from two different diamines, one of them formylated. In D.R.-P. 149,409 and 149,410 (Geigy, 1903) the amines are heated at 210—240° without solvent, using ammonium chloride or aniline hydrochloride as catalysts, and (un-named) acridines are said to be produced. Actually, no non-symmetrical acridine could be obtained by this method, *e.g.*, *m*-tolylenediamine and monoformyl-*m*-phenylenediamine (D.R.-.P. 149,409, example 2) gave only unidentified yellow resinous material of low basic strength with ammonium chloride as catalyst, plus some acridine-yellow when aniline hydrochloride was used. The same result was obtained when glycerol was used as a solvent at 150—190° (as in D.R.-.P. 161,699; Geigy, 1905). However, it seems that conditions may be more favourable when one reactant is a *m*-aminophenol because, on following example 4 from the latter patent (*m*-diethylaminophenol, condensed with diformyl-*m*-tolylenediamine and *m*-tolylenediamine dihydrochloride), a 14% yield of 2-*amino*-8-*diethylamino*-3-*methylacridine* was obtained.

The production of non-symmetrically substituted derivatives of proflavine by condensing equimolecular amounts of a diamine and the dihydrochloride of another diamine, one of them formylated (in glycerol at 185—190°), is claimed in D.R.-P. 292,848 and 346,961 (AGFA, 1915), no extraneous catalyst being used. The most important example in these patents would appear to be the condensation of 2-dimethylamino-*p*-toluidine dihydrochloride with 2-*amino*-4-*formamidotoluene* (from 2-*nitro*-4-*formamidotoluene*). This example claims to give 2-amino-8-dimethylamino-3 : 7-dimethylacridine which can be methylated in the 10-position to give the antibacterial drug, Flavicid (AGFA, D.R.-P. 366,096, 1921) which had already been withdrawn from use when Wagner wrote: "The non-symmetrical structure prevents uniformity in manufacture" (*Z. Immunitätsforsch.*, 1938, **94**, 171). In attempting to reproduce this example, a 35% yield of a symmetrical product (acridine-yellow) and a 20% yield of 2-*amino*-8-*methylamino*-3 : 7-*dimethylacridine* were obtained, a methyl group being lost from 2-dimethylamino-*p*-toluidine as before.

Two patents claiming to condense 2-naphthylamine similarly with diamines to give aminobenzacridines gave only symmetrical products derived from the diamine (AGFA, D.R.-PP. 346,962 and 346,963, 1915).

Conclusions.—The reaction between *m*-diamines and formic acid is the most direct and useful method for producing symmetrically substituted acridines carrying amino-groups in the 2- and 8-positions. Only one operation is involved and the mechanism is now fairly well understood. Yields lie between 20 and 75% and mixtures of isomerides are not formed as in some alternative syntheses. The reaction is not very successful in producing non-symmetrically substituted acridines and fails when acetic or benzoic acid is substituted for formic acid.

The diaminoacridines described in this work have marked anti-bacterial properties (cf. Albert. Rubbo, Goldacre, Davey, and Stone, *Brit. J. Exper. Path.*, 1945, **26**, 160).

Experimental.

Standard Conditions.—Unless otherwise specified, the condensation was carried out by heating the amine (0·1 mol.), formic acid (0·05 mol., anhydrous), hydrogen chloride (0·115 mol., in the form of standardized concentrated acid), and glycerol (three times the weight of the amine) to 155° (internal) during ½ hour and maintaining this temperature a further ½ hour. The best results were obtained in an open vessel of cross-section area 1·5 sq. cm. per g. of amine, with continuous, gentle stirring. Where zinc chloride is specified, an amount equal to 1·5 times the weight of the amine was used.

m-*Aminophenol.*—This was condensed under the standard conditions. The melt was boiled with twice its volume of water, cooled, made neutral with sodium bicarbonate, and filtered (yield, 67% total acridines; colourless filtrate indicates absence of xanthylium salts). The precipitate was boiled with 0·5N-hydrochloric acid and cooled. 2 : 8-Dihydroxyacridine hydrochloride, deposited in 5% yield, was identified by comparison with a sample made from proflavine (Benda, *Ber.*, 1912, **45**, 1794) and conversion to the dimethyl ether, m. p. and mixed m. p. 138° (Cassella, D.R.-P. 392,066, 1923). The material with soluble hydrochloride was neutralised with sodium bicarbonate and extracted with 100 parts of 75% alcohol. On concentrating the solution, orange micro-crystals were deposited in 40% yield, not melting at 350° and sparingly soluble in common solvents (Found: C, 68·6; H, 5·0; N, 10·3. $C_{13}H_9O_2N,2C_{13}H_{10}ON_2,3H_2O$ requires C, 68·3; H, 5·1; N, 10·2%). The substance is soluble in alkali and can be diazotised and coupled with β-naphthol (red), and hence may be designated a trihydrated *complex* of 2 : 8-dihydroxyacridine and 2-amino-8-hydroxyacridine.

m-*Diethylaminophenol.*—Only half the acid given under standard conditions was used, and heating at 155° was prolonged to 1¼ hours. The brilliant magenta-coloured melt was diluted with 50% aqueous zinc chloride solution and the crystals that separated were recrystallised from dilute hydrochloric acid. Yield, 55% of the zincichloride of 2 : 8-bisdiethylaminoxanthylium chloride (Pyronine B), which was compared with a commercial specimen.

m-*Aminodimethylaniline.*—This amine (15 g.) was added to a solution of zinc chloride (20 g.; 1·3 mols.) in glycerol (40 g.; 4 mols.) at 130° and hydrated oxalic acid (14 g.; 1 mol.) added. The temperature was raised to 155° (internal) during ½ hour and kept there for ¾ hour longer. The product was thinned with boiling water (90 ml.) and poured into excess of sodium hydroxide solution. The solid was filtered off, dissolved in dilute acid, and again precipitated with sodium hydroxide. It was dried, powdered, extracted with alcohol, and the extract cooled, depositing 0·62 g. of bis-(*m*-diethylamino)oxanilide, m. p. 198°. The filtrate, on concentration, gave crystals of the base of acridine-orange, m. p. 176°. Crystallisation from 9 parts of 65% alcohol gave solvated orange crystals which were dried at 120°; m. p. 181—182° [184—186° (corr.)]. This substance is less soluble in boiling water than is proflavine. Fluorescences in ultra-violet (Wood's) light: base (in alcohol), yellow-green becoming yellow when dilute; hydrochloride (in water), reddish-orange becoming yellow when dilute.

Oxalyl chloride was mixed at 0° with pyridine (5 mols.) followed by *m*-aminodimethylaniline (2 mols.). Dilution with water gave a 50% yield of *bis*-(m-*dimethylamino*)*oxanilide*, as cream coloured crystals from 300 parts of alcohol, m. p. 198° [203° (corr.)] (Found: C, 65·9; H, 7·0; N, 16·8. $C_{18}H_{22}O_2N_4$ requires C, 66·2; H, 6·8; N, 17·2%). This substance could not be acetylated, and was only slowly hydrolysed by boiling alkalis. Specimens prepared by the two methods did not depress one another's m. ps. The hydrochloride is white and insoluble in alcohol.

3-*Aminodiphenylamine.*—This was condensed with formic acid under the standard conditions, plus ½ hour at 175°. The product was boiled with water and collected. The crude, insoluble *hydrochloride* of 2 : 8-bis(phenylamino)acridine was extracted with 1000 parts of boiling alcohol, and the extract concentrated and then diluted with water. This was repeated several times giving scarlet micro-crystals, not melting at 365°. Soluble in propylene glycol, carbitol, and benzyl alcohol without temperature gradients. The solutions are orange and fluoresce orange in ultra-violet light (Found: C, 75·2; H, 5·3; N, 10·7. $C_{25}H_{19}N_3,HCl$ requires, C, 75·4; H, 5·1; N, 10·6%). The base is sparingly soluble in alcohol and other common solvents. Solutions are yellow and fluoresce green in ultra-violet light. No solvent in which it has any temperature gradient could be found.

2 : 6-*Tolylenediamine.*—This was condensed under the standard conditions, plus zinc chloride, the heating at 155° being prolonged to 5 hours. The method of working up the product has been described (Albert and Magrath, *J. Soc. Chem. Ind.*, 1945, **64**, 30). Yield, 57%. Varying the time of condensation gave 50% (1 hour), or 55% (3 or 7 hours). Varying the molecular proportion of hydrogen chloride (cf. graph for proflavine in Part II) gave 31% (0·55 mol.), 38% (0·85), 50% (1·00), 53% (1·30), and 45% (1·45). Raising the temperature to 175°, even for ½ hour, resinified the batch. 2 : 8-*Diamino*-1 : 9-*dimethylacridine* forms bright yellow solvated crystals from 2 parts of boiling acetone which, when dried in a vacuum and then at 120°, have m. p. 170° [173° (corr.)]. It is soluble in 18 parts of boiling benzene and 14 parts of boiling alcohol (poor gradients), almost insoluble in water and light petroleum. The alcoholic solution has a green fluorescence in daylight and a yellow in Wood's light (Found: C, 75·7; H, 6·4; N, 17·5. $C_{15}H_{15}N_3$ requires C, 75·9; H, 6·4; N, 17·7%). The orange-red hydrochloride is very soluble in water with green fluorescence. It gives a transient violet colour with nitrous acid and the ensuing yellow solution couples with β-naphthol (deep red).

The hydrochloride (2·74 g.) was suspended in acetic acid (50 ml.) and heated with acetic anhydride (2·5 g.; 5 equivs.) to 120° during ½ hour and maintained there for 1 hour longer, refrigerated, and the sparingly soluble hydrochloride of the acetyl derivative collected, washed with water, dried at 120°, and rubbed in a mortar with dilute ammonia until the scarlet colour changed to yellow. The base was filtered off, washed, and dried at 120°. After recrystallisation from 50% alcohol, in which it was more soluble than in absolute alcohol or water, yellow crystals were obtained, m. p. 259—262° (decomp., sealed), almost insoluble in benzene (Found: N, 14·95. $C_{17}H_{17}ON_3$ requires N, 15·05%). This

2-*amino*-8-*acetamido*-1 : 9-*dimethylacridine* is very soluble in water to an orange solution lacking fluorescence. The yellow diazonium solution couples with β-naphthol (dark red).

3 : 5-*Tolylenediamine*.—This (b. p. 184°/34 mm., prepared by the reduction of 3 : 5-dinitrotoluene with Raney nickel and hydrogen in alcohol) was condensed under the standard conditions plus an extra ½ hour at 175°. The reaction mixture was poured into dilute sodium hydroxide. The precipitate was filtered off, dissolved in dilute sulphuric acid, and the solution refrigerated. The sparingly soluble sulphate was recrystallised from dilute sulphuric acid, basified, and the 2 : 8-*diamino*-4 : 6-*dimethylacridine* recrystallised from 15 and then from 35 parts of alcohol. Yield, 20% of brownish-yellow crystals, m. p. 294—295° (sealed), slightly soluble in benzene with a temperature gradient. The solutions are yellow with a green fluorescence (Found : C, 75·4; H, 6·2; N, 17·5. $C_{15}H_{15}N_3$ requires C, 75·9; H, 6·4; N, 17·7%). The hydrochloride is more soluble in water than the isomeric acridine-yellow; the solutions are orange and develop a green fluorescence on dilution. Nitrous acid gives a violet solution which couples with β-naphthol (red).

3 : 5-*Diaminoanisole*.—This was prepared by the reduction (as above) of 3 : 5-dinitroanisole made from trinitrobenzene. Yield, 90% of a white, waxy solid which begins to melt at 66° to a viscous liquid, b. p. 198°/5·5 mm. 3 : 5-Diaminoanisole has not previously been obtained solid (Found : N, 20·45. Calc. for $C_7H_{10}ON_2$: N, 20·3%). When condensed under standard conditions it gave a brown resinous mixture, partly soluble in alkalis with a brilliant green fluorescence.

3 : 5-*Diaminobenzoic Acid Dihydrochloride*.—This (4·5 g.; 0·02 mol.), anhydrous sodium formate (2·04 g.; 0·03 mol.), glycerol (12 g.), and water (2 ml.) were heated to 155° during ½ hour, then to 185° in ½ hour and kept there for ¾ hour longer. The melt was cooled, thinned with water (20 ml.), and refrigerated. The crystals of diaminoacridinedicarboxylic acid hydrochloride were dissolved in sodium carbonate solution, the solution filtered, and the filtrate acidified with acetic acid (yield, 65%). The *substance* was insoluble in all low-boiling solvents and in pyridine, nitrobenzene, carbitol, cellosolve, aniline, chlorobenzene, dioxan, phenol, phenylethyl alcohol, and methyl amyl ketone. It was purified by dissolving the mono-sodium salt in 67% acetone and fractionally precipitating it with acetic acid, which gave an orange, microcrystalline solid, not melting at 365° (Found, N, 14·2. $C_{15}H_{11}O_4N_3$ requires N, 14·1%). The di-sodium salt forms a gel with cold water. The acid could not be esterified with methanolic hydrogen chloride. It did not decarboxylate on heating to 365° nor on refluxing with copper in quinoline or diphenyl ether.

2 : 4-*Tolylenediamine*.—This was condensed as under standard conditions, plus ½ hour at 175° which materially improved the yield (72%). The latter was raised only to 75% by the use of zinc chloride. Oxalic acid (2 hours at 155°, plus zinc chloride) gave a 70% yield. The melt was diluted with twice its volume of water and precipitated with sodium hydroxide. The precipitate was dissolved in water (40 ml.) containing formic acid (1 ml., anhydrous) and ammonia was added until litmus was just blued. A small precipitate of a weak base was filtered off; this substance (3% yield, if a diacridyl ether) forms a very soluble hydrochloride and, if not removed, can prevent acridine-yellow base from crystallising from alcohol. The strong base was precipitated with sodium hydroxide and recrystallised from 180 parts of alcohol, giving yellow crystals of 2 : 8-diamino-3 : 7-dimethylacridine, m. p. 325° (sealed or open tube). The alcoholic solution has an intense green fluorescence (Found : C, 76·1; H, 6·4; N, 17·5. Calc. for $C_{15}H_{15}N_3$: C, 75·9; H, 6·4; N, 17·7%). It is soluble in 20 parts of N/4-lactic acid. The sparingly soluble hydrochloride gives an intense purple colour with nitrous acid which fades on standing for a short time and couples with β-naphthol (red).

2 : 4-*Diaminoanisole*.—This was prepared by suspending an equimolecular amount of 4-nitro-2-aminoanisole in alcohol and hydrogenating at atmospheric temperature and pressure in the presence of Raney nickel. The filtered solution was rapidly treated with the requisite amount of hydrochloric acid, the alcohol driven off, and the residue treated as in the standard reaction (plus ½ hour at 175° which doubled the yield). The diluted melt was made strongly alkaline with sodium hydroxide, boiled, cooled, and filtered. The precipitate was extracted with N/20-hydrochloric acid, and the filtered extract was adjusted with hydrochloric acid until 3% was present, and refrigerated. The hydrochloride was collected and basified, giving 2 : 8-*diamino*-3 : 7-*dimethoxyacridine*, which was purified by dissolution in 60 parts of alcohol and concentration to one third volume (supersaturation technique). Solvated hygroscopic yellow needles, m. p. 244° (sealed), were obtained in 20% yield based on the nitroanisidine. They were sparingly soluble in benzene, moderately in water with a temperature gradient, moderately in acetone with bluish fluorescence. The alcoholic solution is brown with an intense green fluoreseencc (Found, for material dried at 120° in air : C, 65·5; H, 5·7; N, 15·2. $C_{15}H_{15}O_2N_3,\frac{1}{3}H_2O$ requires C, 65·4; H, 5·7; N, 15·3%). The orange hydrochloride is sparingly soluble in water (intense green fluorescence) and is completely precipitated by chloride ions. It gives a red solution with nitrous acid which couples with β-naphthol (scarlet). The addition of zinc chloride to this condensation did not affect the yield. Heating at 200°, even for ½ hour, completely spoilt the condensation.

2 : 4-*Diaminophenetole*.—This was prepared from 2 : 4-dinitrophenetole similarly to the previous example and, without isolation, condensed and worked up in the same way. 2 : 8-*Diamino*-3 : 7-*diethoxyacridine* formed pale yellow crystals from 150 parts of benzene, m. p. 238° (not raised by further recrystallisation from benzene, alcohol, or pyridine). Yield (based on dinitrophenetole), 20%. The pale yellow solution in alcohol has an intense green fluorescence (Found : C, 68·7; H, 6·5; N, 15·8 (the nitrometer contained ethane). $C_{17}H_{19}O_2N_3$ requires C, 68·6; H, 6·5; N, 14·1%). This is evidently not the brown substance, m. p. 281°, obtained by the action of formic acid on 2 : 4-diaminophenetole in B.P. 248,182. The hydrochloride produces orange solutions in water with a green fluorescence, when very dilute, and foaming properties which persist on dilution. It is more soluble than the dimethoxy-analogue but is precipitated by N/2-hydrochloric acid. It gives no colour with nitrous acid and couples with β-naphthol (scarlet).

4-*Chloro*-1 : 3-*diaminobenzene*.—This was condensed as in the standard method using zinc chloride and prolonging the heating at 155° for 3 hours. Heating at 175° caused resinification; omission of zinc chloride severely lowered the yield. The melt was poured into N-sodium hydroxide and filtered. The precipitate was extracted with cold N-acetic acid, filtered from resinous material, and reprecipitated.

The precipitate was dissolved in boiling 0·5N-hydrochloric acid and chilled overnight. The hydrochloride was filtered off, heated with excess of ammonia solution, and dried at 120°. The 3 : 7-*dichloro*-2 : 8-*diaminoacridine* was recrystallised by extracting it with 150 parts of alcohol and concentration to 40 parts (supersaturation). It formed orange-yellow crystals (35% yield) which decomposed above 300° (sealed tube). It is only sparingly soluble in common solvents and the solutions have a green fluorescence (Found: C, 56·2; H, 3·6; N, 14·8. $C_{13}H_9N_3Cl_2$ requires C, 56·1; H, 3·3; N, 15·1%). The hydrochloride fluoresces green in dilute solution. It gives no colour with nitrous acid but couples with β-naphthol (scarlet).

The above reaction gave a by-product soluble in 0·5N- (but insoluble in 2N-) hydrochloric acid. The free base, which contains chlorine, was obtained as yellow crystals from dilute alcohol (Found: C, 54·5; H, 3·3; N, 14·6. $C_{26}H_{18}ON_6Cl_4$ requires C, 54·5; H, 3·2; N, 14·7%). This substance is apparently 3 : 7 : 3′ : 7′-*tetrachloro*-2 : 8 : 2′ : 8′-*tetra-amino*-5 : 10-*dihydrodiacridyl* 5 : 5′-*ether* and not an isomeride of 3 : 7-dichloroproflavine as was previously suggested (Albert, *J.*, 1939, 920). Neither base nor salts fluoresce in solution. It diazotises and couples with β-naphthol (red).

2-*Dimethylamino*-p-*toluidine*.—This was prepared from pure dimethyl-*o*-toluidine and carefully fractionated. It was condensed under the standard conditions (plus ½ hour at 175°), cooled, diluted with water, and made alkaline with sodium hydroxide. The pasty precipitate was recrystallised from dilute alcohol and then from 10 parts of absolute alcohol. 2 : 8-*Bismethylamino*-3 : 7-*dimethylacridine* was obtained in 20% yield as yellow crystals, m. p. 308—309° (sealed), moderately soluble in acetone and sparingly in toluene but with good gradient. The alcoholic solutions fluoresce intense green (Found: C, 76·4; H, 7·2; N, 15·6. $C_{17}H_{19}N_3$ requires C, 76·9; H, 7·2; N, 15·8. 2 : 8-Bisdimethylamino-3 : 7-dimethylacridine, $C_{19}H_{23}N_3$, requires C, 77·8; H, 7·9; N, 14·3%). The hydrochloride is orange and readily precipitated by chloride ions. The solution in water has a green fluorescence when dilute. It could not be diazotised and coupled. No other pure product was isolated.

Non-symmetrical Condensations.—All the examples cited in D.R.-PP. 149,409, 149,410, 346,962, and 346,963 were attempted with the results stated in the Introduction, and no modification of the conditions produced non-symmetrical products. The following was more successful (D.R.-P. 161,699; Example 4). 2 : 4-Tolylenediamine dihydrochloride (1·95 g.; 0·01 mol.), diformyl-1 : 3-tolylenediamine (1·78 g.; 0·01 mol.), *m*-diethylaminophenol (1·67 g.; 0·01 mol.), and anhydrous glycerol (10 g.) were heated to 155° during ½ hour and kept there for 1½ hours. The melt was diluted with water (20 ml.) and treated with sodium hydroxide until Orange II paper was reddened, and the precipitate was collected, washed, and dissolved in N-acetic acid. The filtered solution was made 0·5N with hydrochloric acid and chilled overnight. The yellow precipitate, on basification, gave pure acridine-yellow base, m. p. and mixed m. p. 325° (yield, 25%). The filtrate was basified, and the solid was extracted with toluene, filtered from some red material, and concentrated until it crystallised. Further recrystallisation from a little acetone gave orange crystals of 2-*amino*-8-*diethylamino*-3-*methylacridine*, m. p. 216—217° (sealed) in 14% yield (Found: C, 77·2; H, 7·5; N, 15·0. $C_{18}H_{21}N_3$ requires, C, 77·4; H, 7·6; N, 15·0%). Identical results were obtained when this condensation was carried out on 1 : 3-tolylenediamine and *m*-diethylaminophenol under the standard conditions, the heating at 155° being prolonged to 1½ hours. The orange hydrochloride is soluble in hydrochloric acid of all concentrations and its solutions have only a slight fluorescence in daylight. With nitrous acid it gives an intense blue, stable solution that couples with β-naphthol (scarlet). These highly coloured (usually blue or violet) colours with nitrous acid are given by 2 : 8-diaminoacridine and several of its derivatives (see above) and also by 2 : 6- (but not by 2 : 7-, or 3 : 7-) diaminoacridine (Albert and Linnell, *J.*, 1936, 1614) and not by any of the monoaminoacridines. The present example and the next make it clear that only one *primary* amino-group is required for the production of the colour.

Attempt to produce 2-*Amino*-8-*dimethylamino*-3 : 7-*dimethylacridine* (D.R.-P. 292,848; Ex. 2).—2-Dimethylamino-*p*-toluidine dihydrochloride (2·33 g.; 0·01 mol.), 2-amino-4-formamidotoluene (1·5 g.; 0·01 mol.) and glycerol (8 g.) were heated to 155° during ½ hour, kept there for ½ hour, and kept at 175° for ½ hour. (On another occasion heating was continued for 6 hours at 185° causing greater decomposition.) The melt was diluted with 0·5N-hydrochloric acid and chilled; it then deposited crystals of pure acridine-yellow (m. p. and mixed m. p. of amine 325°) in 35% yield based on the 2-amino-4-formamidotoluene. The filtrate was basified and the precipitate taken up in N-acetic acid, filtered from weak bases, and basified. The precipitate was crystallised from 50% acetone and then from toluene. Yellow crystals of 2-*amino*-8-*methylamino*-3 : 7-*dimethylacridine*, m. p. 264° (sealed), very soluble in alcohol with intense green fluorescence, were obtained in 20% yield (Found: C, 76·3; H, 6·7; N, 16·95. $C_{16}H_{17}N_3$ requires C, 76·4; H, 6·8; N, 16·7%). The acetate is orange and very soluble in water. The dihydrochloride is soluble in 3N-hydrochloric acid from which it is partly precipitated as monohydrochloride on dilution to 0·5N; however, 0·1N-acid does not contain a sufficient concentration of chloride ion to precipitate the latter. Solutions of the salts fluoresce intensely green when dilute and give violet solutions with nitrous acid which couple with β-naphthol (red). No other pure product could be isolated. The same results were obtained (with unformylated amine) under the standard conditions plus ½ hour at 175°.

2-*Amino*-4-*formamidotoluene*.—2-Nitro-*p*-toluidine (15·0 g.; 0·1 mol.), formic acid (10 ml.), and toluene (50 ml.) were heated so that the vapours slowly distilled through a Widmer (spiral) column while the temperature of the bath was gradually raised to 170°. When no more liquid distilled below 105° (about 2 hours), the flask contents were poured into a dish, cooled, powdered, and dried at 120°. Yield, 17·75 g. (quantitative) of buff crystals of 2-*nitro*-4-*formamidotoluene*, m. p. 133—134°; not raised by recrystallisation from toluene, or from alcohol (3 parts) (Found: N, 15·4. $C_8H_8O_3N_2$ requires N, 15·55%). When this compound (8·9 g.) was suspended in alcohol (50 ml.) and hydrogenated with Raney nickel and hydrogen at atmospheric temperature and pressure, and the solution filtered, concentrated to 12 ml., and refrigerated, buff crystals of 2-*amino*-4-*formamidotoluene* were obtained in 85% yield, m. p. 112—113°, raised by repeated recrystallisation from toluene to 113—114° (Found: N, 18·8. $C_8H_{10}ON_2$ requires N, 18·7%). The only previous reference to these two substances is in D.R.-P. 138,839.

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