Some Newer Aspects of the Organic Chemistry of Nitrogen.

HUGO MÜLLER LECTURE, DELIVERED BEFORE THE CHEMICAL SOCIETY AT BURLINGTON HOUSE ON THURSDAY, DECEMBER 16TH, 1954.

By G. R. CLEMO.

I SHOULD like to introduce this Lecture with a few points from H. E. Armstrong's characteristic obituary notice (1917) of the man who endowed it. Hugo Müller worked with Wohler (1852) and was an assistant to Liebig in Munich when he was invited by Hofmann in 1853 to become private assistant to Warren De la Rue. Later, Hugo Muller became a partner in the latter's firm and as "the prototype of the modern industrial chemist . . . he ultimately became a very wealthy man." He was President of the Chemical Society in 1885—1886 and retired from business in 1902 and was thereafter happy in his garden and "particularly in the last dozen years of his life in the laboratory seeking to contribute to the solution of the problems which Nature ever held before him." Armstrong states that he died of a broken heart in 1917 and his widow and daughter made handsome gifts to the Society. This Lecture is given every 3 years and alternates between a topic linking chemistry with mineralogy and one linking chemistry with botany. My subject will, I hope, fulfill the latter requirement and allow us to take a glance at a few of the problems which Nature holds before us.

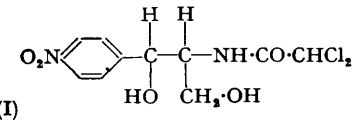
The three basic processes in Nature are photosynthesis, respiration, and nitrogen fixation; and as the latter is the basis of all natural organic nitrogen compounds—the amino-acids, the pre-eminently important proteins, the alkaloids, etc.—it is fitting to start by summarising the present knowledge of nitrogen fixation.

The sources of nitrogen for plant syntheses are free nitrogen, ammonium salts, and nitrates. Some years ago a crop analysis over a vast land area of the U.S.A. showed that two-thirds of all organic plant nitrogen was fixed by bacteria, one-quarter came from rainfall, and only one-twelfth from added fertilizers. It is stated, however, that 90% of all plant synthesis occurs at or near the surface of the seas, and no claim has been made for nitrogen fixation in this case. The soil has a colossal microbiological population (estimated at 5×10^9 bacteria per g. !) and of these the *Rhizobium* root bacteria of nodules and the two kinds of *Azotobacter* in the soil are responsible for nitrogen fixation by enzymes. That a definite relation exists between photosynthesis and nitrogen fixation appears to have been first established when Kamen showed in 1949 that illumination increased the assimilation of nitrogen by the photosynthetic bacterium *R. rubrum*. Wall and his co-workers later showed ammonia to be the key intermediate in the nitrogen fixation of the three types of photosynthetic bacteria, *Chromatium*, *Chlorobacterium*, and *R. rubrum*. The mechanism whereby nitrogen is thus enzymically converted into ammonia is, however, chemically still obscure. The dissociation energy of molecular nitrogen is so large (170 kcal./mole) that fission into atoms seems unlikely and possibly both nitrogen and oxygen molecules are brought into reaction by enzyme catalysts through the aid of some of the metals cobalt, iron, nickel, vanadium and particularly manganese and molybdenum. Virtanen has been the chief worker on the mode of action of *Rhizobium* root-nodule bacteria and, having detected oxime-nitrogen in some cases, suggested hydroxylamine as an intermediate ($N_2 \longrightarrow N_2O \longrightarrow H_2N_2O_2 \longrightarrow NH_2{\cdot}OH$); but this hypothesis has not been accepted and Virtanen himself stated at the 6th International Congress of Microbiology in 1953 that whilst there is evidence of the participation of hydroxylamine in amino-acid formation, ammonia appears to be the central compound in their synthesis. Newton, Wilson, and Burris (*J. Biol. Chem.*, 1953, **204**, 445), using ^{15}N, claim to have demonstrated the production of ammonia by *Azotobacter* by finding a high ^{15}N level in the glutamic acid; the chemistry invoked was: conversion of nitrogen into ammonia by various unknown steps involving energy; utilisation of ammonia for synethesis of glutamic acid by reductive amination; and formation of other amino-acids by transamination.

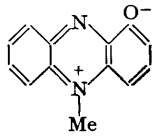
Burk (1934) claimed, however, that either ammonia or nitrate ion inhibited nitrogen fixation by *Azotobacter*.

Recently, Virtanen, using peas, provided evidence that all effective nitrogen-fixing nodules contained a red hæmoglobin pigment, leghæmoglobin, with a molecular weight of

acid could form α-alanine from pyruvic acid, and either α- or β-alanine by simple loss of carbon dioxide). In spite of the large amount of work on amino-acids by those mentioned and by others, such as Chibnall, the mechanism of the fundamental fixation of nitrogen by *Rhizobium* and particularly by *Azotobacter* remains unexplained. As for the utilisation of nitrates, so important for marine synthesis, it is claimed that nitrate is first reduced to ammonia, and although Quastel has done much valuable work on the reaction its mechanism is also unknown. In this connection the striking discovery in 1948 of the first natural product with a nitro-group present in the soil, namely, chloromycetin (I) (note the C_6C_3 unit so common in natural products),

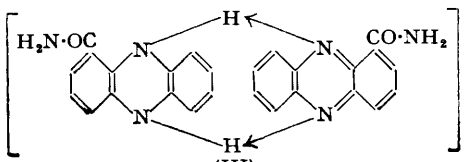


(I)

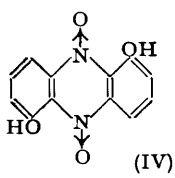


(II)

doubtless has a significance apart from the fact that it is the best-known remedy for the dirt-borne disease typhoid fever! Our lack of precise knowledge as to how nitrogen, free or fixed, is utilised in nitrogen fixation is in marked contrast with the striking story of the actual transmutation of nitrogen into carbon ($^{14}_{7}N + n \longrightarrow {}^{14}_{6}C + p$); and, since the isotopes, particularly of carbon, hydrogen, oxygen, nitrogen, and sulphur, are confidently expected to help greatly in the elucidation of biological processes, we will now take note of some of the help thus given already. First, however, a brief account of my own limited incursion into the field of bacterial nitrogen fixation.



(III)



(IV)

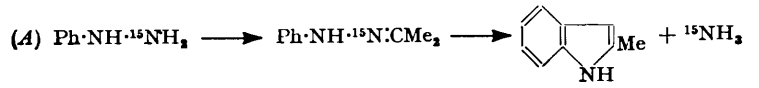
The phenazine ring systems in pyocyanine (II), chlororaphin (III), and chromo-iodinum (IV) are produced by bacteria, the first by the common *B. pyocyaneus* of pus. This usually needs organic nitrogen for its growth and pigmentation, but thanks to the kindness of Professor Dunlop of the Department of Bacteriology of King's College (Newcastle-on-Tyne) and the skill of Mr. T. K. Miller, B.Sc., of that Department a strain was found, after over 100 varieties from hospital patients had been tested, which grew and became pigmented on a medium containing the " AnalaR " reagents, magnesium sulphate, potassium dihydrogen phosphate, sodium acetate and citrate, and ferrous sulphate (the last is not essential but it inhibits the concurrent formation of a yellow pigment), to which combined inorganic nitrogen was added. If hydroxylamine was added no growth occurred, but with ammonium nitrate good growth and pigmentation took place whilst with potassium nitrate colour production was poor. In order to determine whether one or both of the nitrogen atoms in the phenazine ring of pyocyanine came from the ammonium or nitrate ions, $^{15}NH_4NO_3$ (32·5% enriched) was added to the medium. The resulting blue pigment was isolated by extraction with chloroform and recrystallised from water to give lovely deep blue needles, m. p. 133°. Analyses for carbon, hydrogen, and nitrogen were, however, no more successful than with the earlier workers, but the ultraviolet spectrum and melting point leave no doubt that it was pure pyocyanine. My best thanks are also offered to Mr. P. Kennedy for assistance with the preparative work involved and especially to Mr. P. Kelly, B.Sc., who in the absence of published data on the mass-spectrographic determination of ^{15}N in small samples had to devise the technique for its assay. In view of the cost * of the $^{15}NH_4NO_3$ used, only a few mg. of the pigment were obtained in each experiment, but it was found that admixture with unlabelled pyocyanine enabled reproducible ^{15}N figures to be obtained. The enrichment values thus found for the pyocyanine were 29·4, 28·9, and 32·7 ± 1%, thus proving that almost all the phenazine nitrogen came from the ammonium and *not* the nitrate ion. The poor result from potassium nitrate suggests that the bacteria cannot effectively reduce the nitrate ion to ammonia, and that the nitrate is not

* The only suitable isotope of nitrogen is the stable ^{15}N available as $^{15}NH_4NO_3$ (£90 approx. for 1 g. of ^{15}N) and as phthalimide. Although the natural abundance of ^{15}N is 0·38%, the price is an indication of the elaborate fractionation involved in the enrichment up to even 30%.

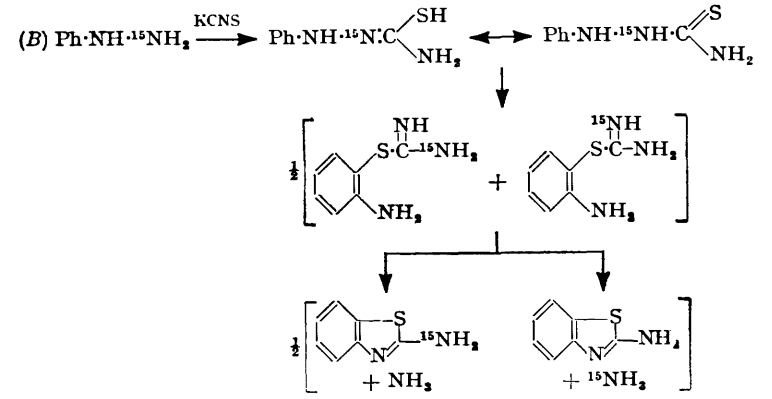
really needed for the bacterial utilisation of the ammonia is shown by the good result obtained by using ammonium chloride as the sole source of nitrogen present.

The following are some of the main uses of ^{15}N with a few examples, from the many, of the application of ^{14}C to problems in chemistry. In view of the fact that the main application of organic chemistry in recent years has been to biochemical problems it is not surprising that the chief use of the isotopes has been to them. A striking result of this application has been to reveal the surprising dynamic condition of many molecules. Few chemists, I think, expected to find all the hydrogen atoms of phenol so easily replaceable by deuterium and Schoenheimer's work has proved the rapid dynamic condition of even such very complicated molecules as make up the protein content of animals. Thus, using labelled amino-acids, he showed the rate of protein synthesis in the rat to be 11 g. of nitrogen per kg. body weight per day and when [^{15}N]leucine or [^{15}N]glycine was fed the ^{15}N appeared in almost all the amino-acids.

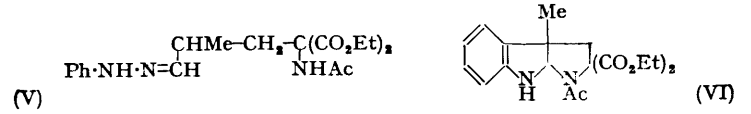
Of purely chemical interest was the following use of ^{15}N to confirm Robinson's views of the mechanism of the Fischer indole reaction, as set out in the annexed formulæ (*A*).



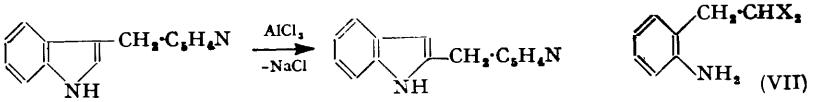
A related reaction is represented in scheme (*B*).



An interesting recent application of the Fischer indole reaction is by H. Kissmans, Ark, and B. Witkop (*Experientia*, 1952, **8**, 36) in which the compound (V) yields the ring systems of physostigmine (VI). Recently, certain 3-substituted indoles, *e.g.*, skatole and 3-2′-pyridyl-

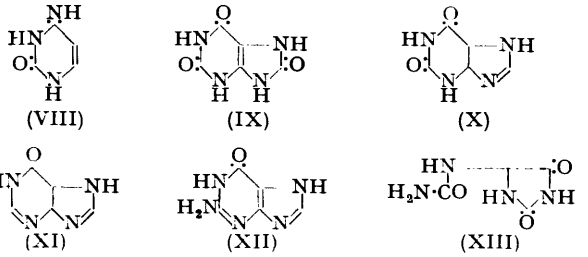


methylindole, have been shown to give the 2-substituted isomers when heated with aluminium chloride and sodium chloride (Clemo and Seaton, *J.*, 1954, 2582). This change does not take place when the indole-NH is acetylated.

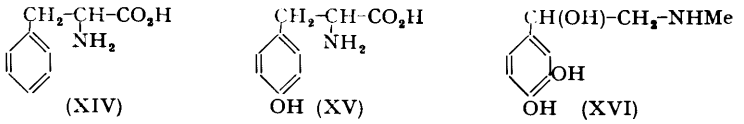


The simple di- and tri-meric indoles have excited interest for many years and in a recent study (G. F. Smith, *Chem. and Ind.*, 1954, 1451) good reasons are given for structure (VII; X = 3-indolyl) for the trimer. The linear structure of the azides was confirmed by using ^{15}N (Clusius and Weisser, *Helv. Chim. Acta*, 1952, **35**, 1958). ^{15}N has been used successfully in the purine field. It is of interest to note that whilst this ring system is of the greatest importance the parent purine itself has only recently been found in Nature in a mushroom (Lofgrin and Lüning, *Acta Chem. Scand.*, 1953, **7**, 225). The easy preparation of ^{15}N-labelled urea and guanidine has led to the syntheses of cytosine (VIII), uric acid (IX), xanthine (X), hypoxanthine (XI), and guanine (XII), all 1 : 3-labelled with ^{15}N; and use of ^{13}C, ^{14}C, and ^{18}N in

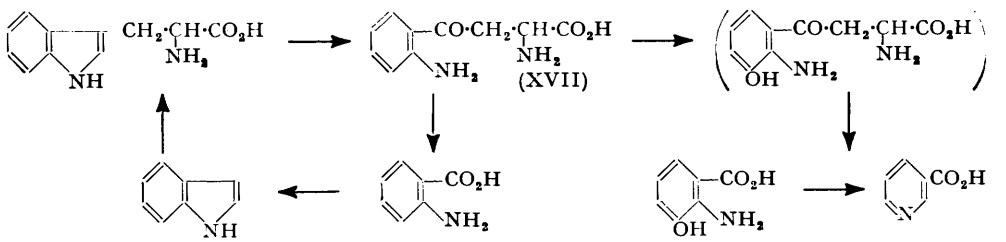
combination in uric acid has led to the elucidation of both its biosynthesis and its degradation. In the latter case the surprising result has emerged that in the oxidative degradation the five-membered ring of allantoin (XIII) comes equally from the five- and six-membered rings of uric acid.



From the large number of isotopic studies already carried out in the biochemical field it is possible to glance at only a few. If phenylalanine (XIV) is given in food, even in the presence of sufficient tyrosine, it is converted into tyrosine (XV) in the liver and is then decarboxylated by kidney tissues to tyramine, the postulated precursor of adrenaline (XVI):



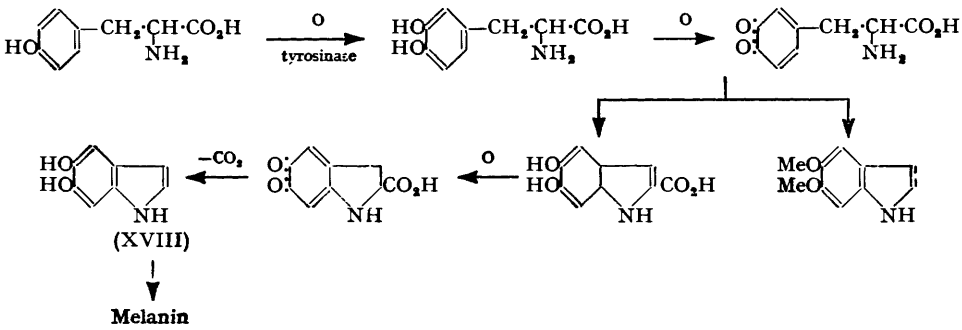
Very striking transformations of tryptophan *via* anthranilic acid into nicotinic acid have also been demonstrated. Thus several organisms such as *Claviceps purpurea* (ergot) can convert indole into tryptophan by interaction with serine in the presence of the coenzyme pyridoxal phosphate, the indole being derived from anthranilic acid. Certain moulds easily convert tryptophan into kynurenine (XVII), and *Neurospora* can convert the latter *via* anthranilic acid and indole back into tryptophan. 3-Hydroxykynurenine, which occurs in Nature, can replace nicotinic acid in the growth of some moulds and is thus claimed as an intermediate in the conversion of tryptophan into nicotinic acid. In this connection 3 : 4-fission of 3-hydroxy-



anthranilic acid has been claimed, and the recent work of Wass (*Z. Naturforsch.*, 1954, **9b**, 740) on the enzymic oxidation of 3-hydroxyanthranilic acid strongly supports it. Leifer, Langham, Nye, and Mitchell (*J. Biol. Chem.*, 1950, **184**, 589), growing *Neurospora* with $^{15}NH_4Cl$, furthermore claimed a diamino-compound as an intermediate in the conversion of 3-hydroxyanthranilic acid into nicotinic acid since half of the nicotinic acid nitrogen was ^{15}N, but the interpretation of this work appears to have been questioned. The earlier work on this important but involved cycle was discussed by Dalgliesh (*Quart. Rev.*, 1951, **5**, 227).

The use of ^{14}C has shed some light on the still unknown mechanism whereby tyrosine is converted into the black melanin pigment. Although such pigments are widespread in hair, some skins, and potatoes, and in certain pathological conditions, they are probably not all identical. Raper (*Biochem. J.*, 1927, **21**, 87) showed that tyrosinase converted tyrosine into melanin with the uptake of 5·2 oxygen atoms per molecule and the loss of carbon dioxide and when he isolated 5 : 6-dimethoxyindole by methylation at an intermediate stage he postulated 5 : 6-dihydroxyindole (XVIII) as the intermediate in melanin formation (see annexed scheme). This point was first proved rigidly by Robertson's synthesis of this dihydroxyindole which in alkaline solution rapidly changed to melanin (*J.*, 1948, 223). In spite of this proof and of other work of Robertson, Burton, and Harley-Mason in this country and of Mason in the U.S.A., little

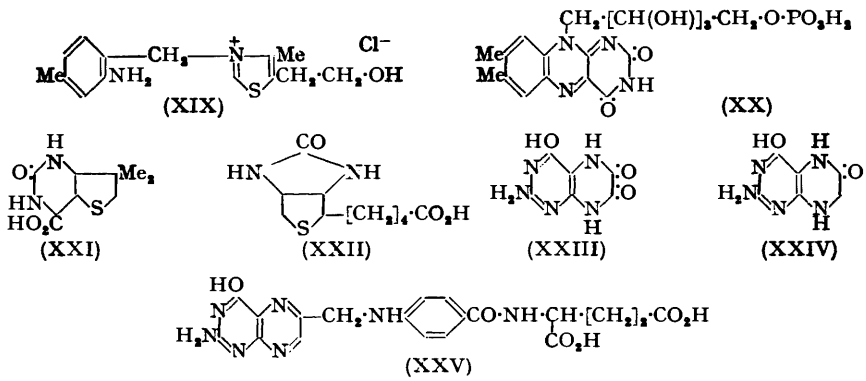
is known today of the stages in the conversion of the indole into melanin. In Raper's scheme it was assumed that the carbon dioxide evolved came from the carboxyl group of the tyrosine but when this group was labelled as $^{14}CO_2H$, the Lecturer (with Drs. Swan and Duxbury) found



that half of the carbon dioxide evolved came from some other carbon atom. It had been shown earlier that 3 : 4-dihydroxyphenylethylamine easily formed a melanin and when we labelled its α- and its β-carbon atom severally, the carbon dioxide evolved was found to be nearly inactive and in fact 96% of it came from the carbon of the benzene ring. Robertson has shown however that in the conversion of 3 : 4-dihydroxyindole into melanin (non-enzymically) 0·46 mol. of hydrogen peroxide is evolved (*J.*, 1954, 1947); and we showed that if catalase is present to decompose the hydrogen peroxide the evolution of carbon dioxide is much reduced. Thus in the absence of catalase we found that one carbon atom from the ring of each two molecules of the indole was evolved as carbon dioxide but only one atom from four molecules of the indole was so evolved when catalase was present. Fission of the catechol ring has been demonstrated (Clemo and Duxbury, *Chem. and Ind.*, 1953, 1364) and it seems possible that this ring in the dihydroxyindole may split—although, as mentioned above for the tryptophan cycle the pyrrole ring can also be opened under biological conditions—and that some such fission products are involved in the condensation leading to the complex polymeric melanin. Nature fortunately still confronts us with great unsolved problems.

Isotopes are also being pressed into service in our attack on the ills afflicting the animal kingdom. Thus in the fight against a major scourge, carcinogens have been labelled including the nitrogen in 2-acetamidofluorene but so far there are not many definite results to report.

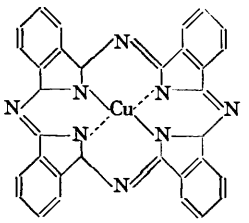
In marked contrast the advances in structural organic chemistry are so numerous and striking that only a few will be mentioned with the object of showing something of the wide range of ring types formed in Nature, before turning to a review of one particular system.



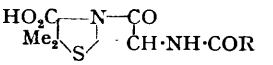
Examples to illustrate the point are many of the B vitamins such as B_1 (XIX), B_2 (XX), the biotins a and b (XXI and XXII), and the linking of the colouring matters of certain butterflies and wasps [leucopterin (XXIII) and xanthopterin (XXIV), respectively] through pteroylglutamic acid (XXV) and B_{12} with the vital subject of nutrition. The fundamental biological importance of *p*-aminobenzoic acid is well illustrated in these compounds.

Porphyrin chemistry is exciting new interest and linked to it are the valuable phthalocyanin

pigments such as Monastral Blue (XXVI) whose chemistry was elucidated by Linstead. Recognition of the key function of nucleic acids in cell syntheses adds greatly to the value of the synthetic work being done in this field at Cambridge. The antibiotics such as the penicillins (XXVII) also illustrate the remarkable range of cell syntheses.

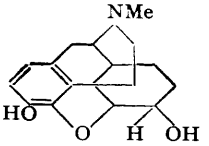


(XXVI)

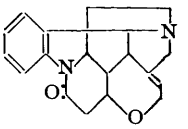


(XXVII)

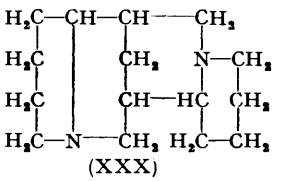
There is intensive activity today in the alkaloid field, both in structural determination and in synthesis : the verification of Robinson's 1926 morphine structure (XXVIII) by Gates and Tschudi (*J. Amer. Chem. Soc.*, 1950, **72**, 228, 4839) and the recent total synthesis of strychnine (XXIX) by Woodward (*J. Amer. Chem. Soc.*, 1954, **76**, 4749, 4751) are remarkable achievements. A glimpse at the structural formulæ of such diverse types as sparteine (XXX), colchicine (XXXI) (whose seven-membered ring structure was largely worked out by J. W. Cook), emetine (XXXII), strychnine, and morphine prompts the remark that all these structures were unravelled by the classical methods without recourse to the newer mechanical aids.



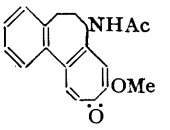
(XXVIII)



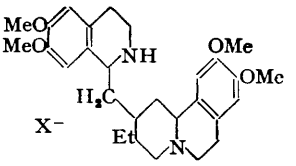
(XXIX)



(XXX)

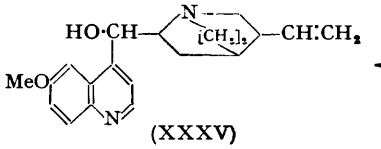


(XXXI)

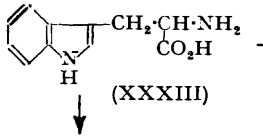


(XXXII)

The very diverse structural types found in these few alkaloids would appear to render very remote any understanding of their biosynthesis were it not for the fact that Robinson's original essay on this subject (*J.*, **1917**, **111**, 876) has now evolved into such a great and powerful unifying conception that, to take one example, such diverse structural types as the yohimbine, *Strychnos*, and *Cinchona* groups of alkaloids can all be based on tryptophan as a common precursor (cf. XXXIII—XXXV).

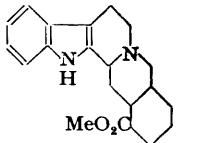


(XXXV)



(XXXIII)

(XXIX)

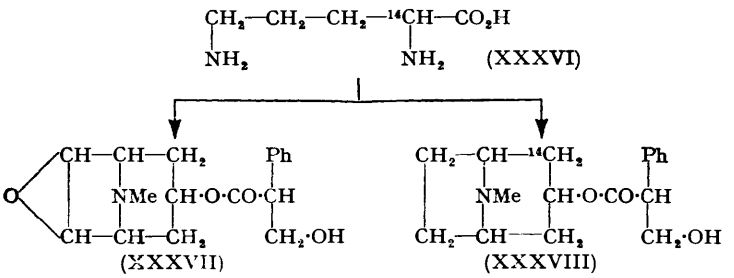


(XXXIV)

Of much interest is the recent work of Leete, Marion, and Spenser (*Nature*, 1954, **174**, 650) who gave a [^{14}C]ornithine (XXXVI) to *Datura* and obtained non-radioactive hyoscine (XXXVII) and radioactive hyoscyamine (XXXVIII) which on degradation was found to have all the ^{14}C in a bridgehead of the tropine system.

The newer developments in the chemistry of pyrrole, indole, quinoline, and *iso*quinoline are so numerous and have been convered so fully by others that it is not proposed to mention them further here. We may however refer to a number of heterocyclic ring systems, which cannot be

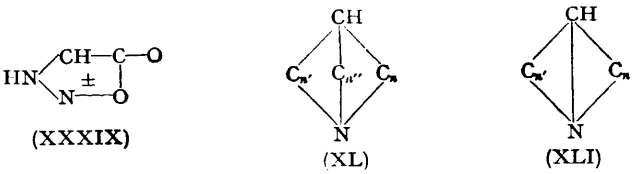
satisfactorily represented by a single formula of the usual valency-bond type, and for which W. Baker and his collaborators (*J.*, 1945, 267; 1949, 307; 1950, 1542; 1951, 289) have proposed the term " mesoionic ". Sydnone (XXXIX) is a typical example.



(XXXVII) (XXXVIII)

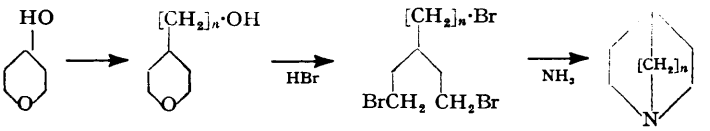
There has been no notable advance in the stereochemistry of organic nitrogen compounds since the classical work of Mills although azo-compounds have been prepared in *cis*- and *trans*-forms. The mechanism of the nitration reaction has been elucidated by Ingold's penetrating work.

We turn now to summarise that less-known chapter in which one or more nitrogen atoms are common to two rings, as in the 1-aza*bicyclo*-[$n : n' : n''$]- and -[$n : n' : 0$]-systems (XL) and (XLI). Much of this work is new and a considerable part of it has the merit of being simple and will deserve to be as well-known as is the chemistry of quinoline.



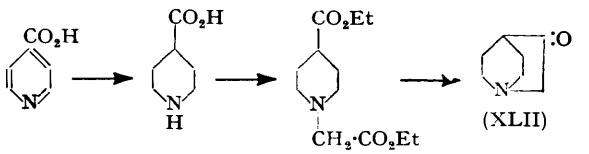
(XXXIX) (XL) (XLI)

The best known example of the former type is quinuclidine (XL; $n = n' = n'' = 2$) which has only been found in the *Cinchona* alkaloids and a good account of whose chemistry is given by Turner and Woodward in Manske's " Alkaloids," Vol. III; one recalls too the brilliant synthesis of quinine itself by Woodward and Doering (*J. Amer. Chem. Soc.*, 1944, **66**, 879; 1945, **67**, 860), and the curious fact that quinuclidine, although first prepared by Löffler and Stirtzel (*Ber.*, 1909, **42**, 124), was only obtained crystalline in 1920 by Meisenheimer. A more convenient synthesis was effected by Prelog (*Annalen*, 1937, **532**, 69) and this has been adapted as a general method for compounds of this type (see attached scheme).



($n = 1$, 2, or 3)

In the same year we prepared 3-oxoquinuclidine (XLII), a compound of evident synthetic possibilities, and converted this into the fully reduced base. The stereochemistry of this system prevents unsaturation at the bridgehead, thus excluding aromatic character, in marked contrast to what is found in the second type (XLI) where fully aromatic compounds are not uncommon. Even here, however, the saturated members of the group are more stable and



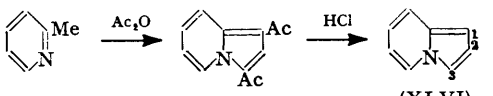
(XLII)

more frequently encountered. Neither pyrrolizine (XLIII) nor its quaternary dehydro-compounds (XLIV) have as yet been prepared; although the keto-derivative (XLV*b*) was easily

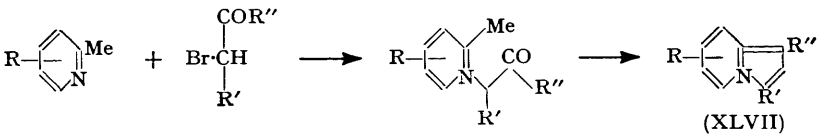
obtained as a stable crystalline solid (Clemo and Ramage, *J.*, **1931**, **49**), all attempts to prepare 1-methylpyrrolizidine [shown by Menschikov to be (±)-heliotridane] by the Grignard reaction and reduction gave only indefinite gums.

(XLIII) (XLIV) (XLV)

Pyrrocoline (XLVI) on the other hand was prepared by Scholtz (*Ber.*, **1912**, **45**, **734**) by heating α-picoline with acetic anhydride and hydrolysing the resulting 1 : 3-diacetyl derivative with hydrogen chloride. A general synthesis by Chitschibabin (*Ber.*, **1927**, **60**, 1607 and later papers) has been used extensively by Borrows and Holland (*J.*, **1946**, **1069**; **1947**, 670) to obtain derivatives (XLVII) and to study the chemistry, including substitution, of the system.

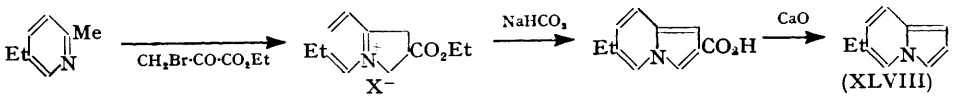


(XLVI)



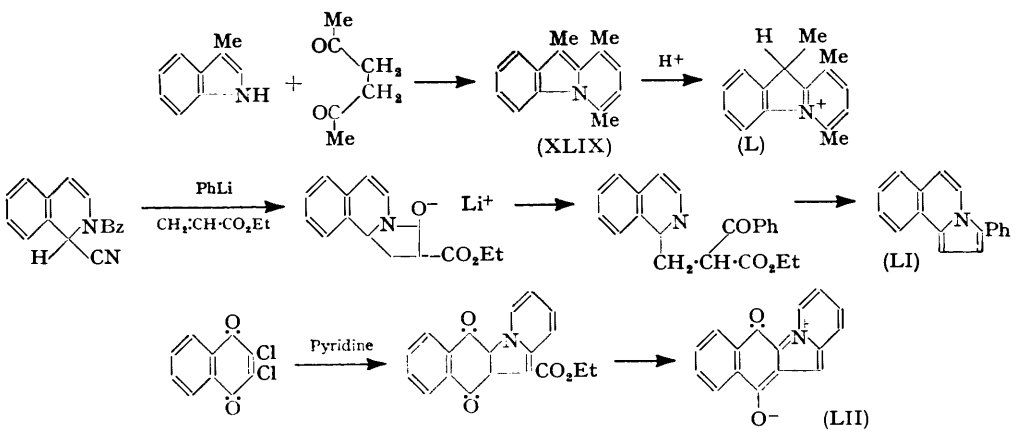
(XLVII)

Pyrrocoline itself is a typical aromatic compound, easily nitrated and halogenated, reaction occurring first at the 3- and then at the 1-position. Treatment with methyl iodide results in nuclear methylation (cf. pyrrole). Hydrogenation first reduces the six-membered ring, and the resulting pyrrole is further hydrogenated only with difficulty. Generally pyrrocolines are yellow and are weak bases.



(XLVIII)

Clemo, Fox, and Raper recently (*J.*, **1953**, **4173**) prepared 6-ethylpyrrocoline (XLVIII) in the still unsuccessful effort to identify the base $C_{10}H_{11}N$ obtained from strychnine. Various octahydropyrrocolines such as the 2-ethyl and the 1 : 7-dimethyl derivative have been made in the same quest.

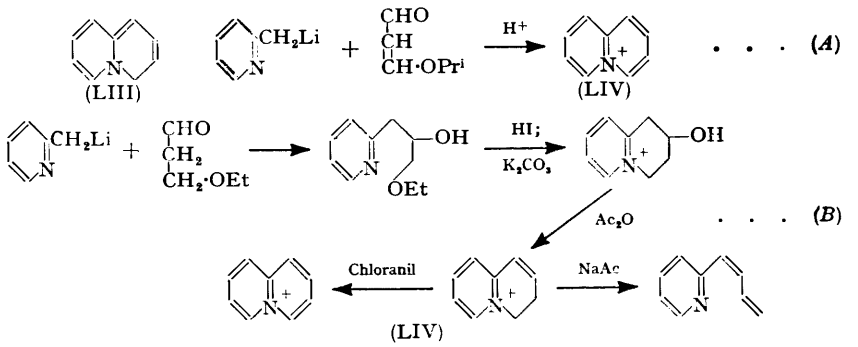


(XLIX) (L) (LI) (LII)

Robinson and Saxton (*J.*, **1950**, **3136**; **1952**, 976) have shown 2 : 3-benzopyrrocolines, *e.g.*, (XLIX), to be weak *pseudo*-bases : salt formation is slow but the salt, *e.g.*, (L) of the much stronger pyridinium base is formed. A recent synthesis of 7 : 8-benzopyrrocolines, *e.g.*, (LI), is that of Boekelheide and Godfrey (*J. Amer. Chem. Soc.*, **1953**, **75**, **3679**), and an unusual one of

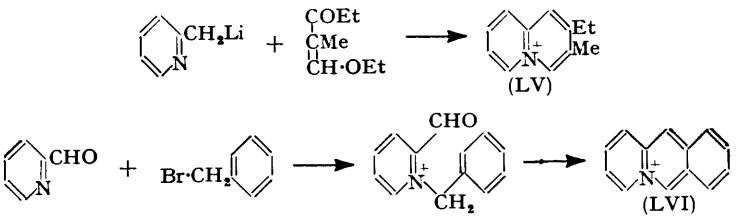
more complex pyrrocolines, *e.g.*, (LII), is described by Pratt, Luckenbaugh, and Erickson (*J. Org. Chem.*, 1954, **19**, 176).

The 6 : 6 ring-system of pyridocoline (LIII) on the other hand cannot be fully aromatic, and all attempts to prepare it have failed, but the dehydropyridocolinium ion (LIV) is aromatic and has been made in two ways—by Woodward and Beaman (1951, quoted in following ref.) (cf. *A*) and by Boekelheide and Gall (*J. Amer. Chem. Soc.*, 1954, **76**, 1832) (cf. *B*). The last stage in (*B*)

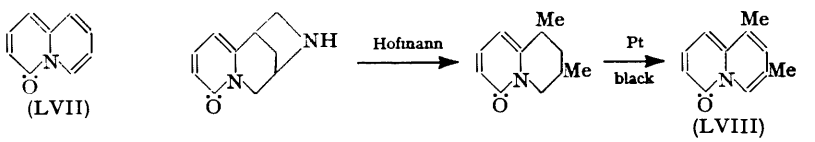


is the nearest approach to a preparation of pyridocoline so far, but this substance seems to be so unstable that it rearranges to 1-2′-pyridylbutadiene.

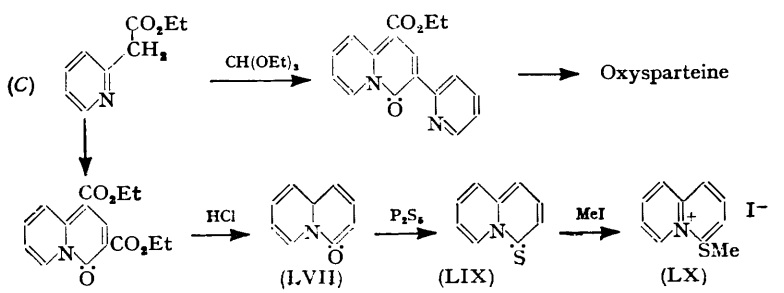
More recently Richards and Stevens (*Chem. and Ind.*, 1954, 905) have extended the reaction to the 2-ethyl-3-methyl derivative (LV), and Bradsher and Beavers have prepared the 1 : 2- and the 2 : 3-benzo-derivative (LVI) (*Chem. and Ind.*, 1954, 1394).



4-Oxopyridocoline (LVII) is also fully aromatic and was obtained in low yield by Späth and Galinowsky (*Ber.*, 1936, **69**, 761) in connection with their work on cytosine in which 1 : 3-dimethyl-5-oxopyridocoline (LVIII) was obtained. A similar dehydrogenation of 4-oxonorlupinane gave the parent base (LVII). 4-Oxopyridocoline was prepared in much better yield by

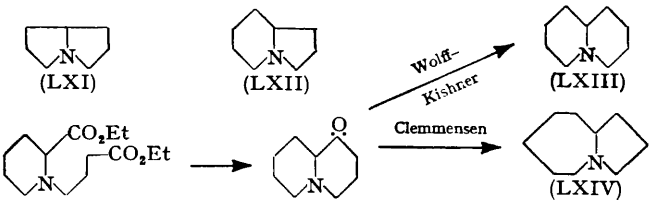


Boekelheide and Lodge (*J. Amer. Chem. Soc.*, 1951, **73**, 3681) by exploiting the reaction first used by Clemo, Raper, and Morgan (*J.*, 1936, 1025) in the synthesis of oxysparteine (scheme *C*).

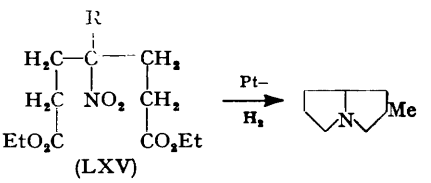


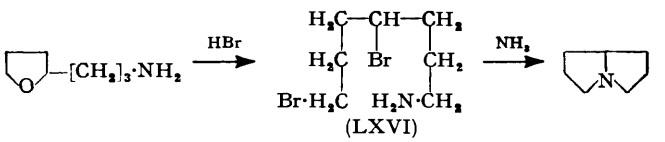
The keto-compound, like its derivatives, forms yellow crystals and shows a marked blue fluorescence in solution. It is extremely soluble in water, and is stable to Grignard-type reagents—an enol-type complex is formed which on acidification liberates the unchanged pyridocoline. With phosphorus pentasulphide the 4-thione (LIX) is formed and this with methyl iodide gives a reactive methylthio-iodide (LX) which readily condenses with malonic ester and with lepidine gives a cyanine dye The presence of substituents greatly changes the reactivity of the system: thus the 6-methyl ketone does not give a thione. Little is yet known about substitution in the system but hydrogenation gives the fully reduced octahydro-4-oxo-pyridocoline.

The fully reduced systems of pyrrolizidine (LXI), octahydropyrrocoline (LXII), and octahydropyridocoline (LXIII) have been made in three main ways. (1) With Dr. Raper and

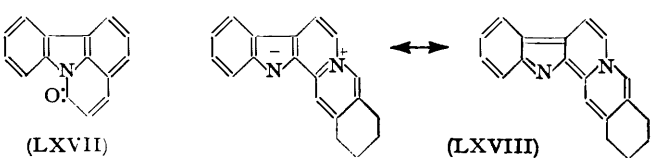


many colleagues we have used the Dieckmann reaction to give a ketone which on reduction by either the Clemmensen or the Wolff–Kishner method usually give the required base. However, we found (*J.*, 1931, 3190) that whilst reduction of octahydro-1-oxopyridocoline by the latter method gave the desired norlupinane, the former method gave an isomeric base, which Prelog subsequently showed to be the *bicyclo*[5 : 3 : 0]-base (LXIV); this is a case of the rare structural change in a Clemmensen reaction. (2) Leonard and his co-workers used reductive cyclisation of the nitro-ester (LXV), and (3) and Šorm *et al.* (1947) used the cyclisation of halogeno-amines such as (LXVI).





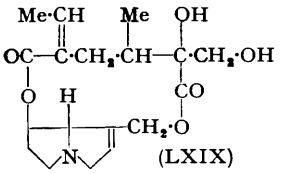
Although the ring systems mentioned above are frequently found in Nature in the reduced form they rarely occur in their aromatic condition. Two examples of the latter, however, are the alkaloids canthin-6-one (LXVII) and sempervirine (LXVIII). The reduced pyrrolizidine



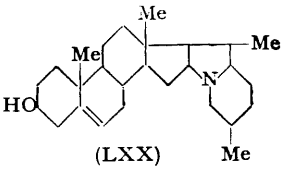
system is the basis of the numerous and interesting group from the Compositæ family (*e.g.*, the common ragwort) comprising the *Senecio* alkaloids whose chemistry was largely developed by Roger Adams and has latterly been extended by Warren in South Africa, with the result that the structures of at least five have been shown to be derivatives of active 1-methyl-pyrrolizidine (heliotridane) [*e.g.*, retrorsine (LXIX)]. Ragwort poisoning of grazing animals causes cirrhosis of the liver and a few months ago the owner of a herd in this country was awarded £4000 for the loss of stock through eating the material in a powdered grass mixture.

The pyrrocoline system is present in the important steroid group of *Solanum* and *Veratrum*

alkaloids whose chemistry has advanced greatly in recent years the potato-shoot alkaloid solanidine (LXX) is a well-known member.



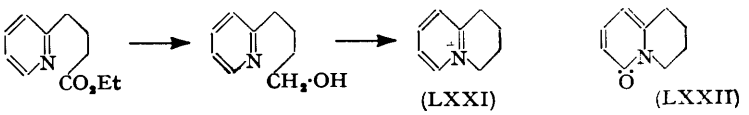
(LXIX)



(LXX)

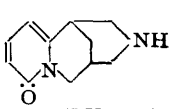
The pyridocoline system occurs as the dehydro-ion or as the corresponding pseudo-base in the well-known berberine group; the best synthesis of this system is still the classical one of Haworth, Koepfli, and Perkin (*J.*, 1927, 548).

The dihydropyridocolines include the ions of the type (LXXI) and the corresponding " pyridones " (LXXII). The former has been prepared recently by Bohlmann, Ottawa, and

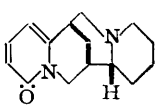


(LXXI) (LXXII)

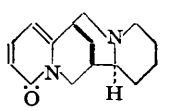
Keller (*Annalen*, 1954, **587**, 162) in the manner shown, but attempts to convert it into the " pyridone " by oxidation under alkaline conditions led to degradation. The pyridone system however is present in the laburnum and gorse alkaloids, cytosine (LXXIII), thermopsine (LXXIV), and anagyrine (LXXV), none of which has yet been synthesised in spite of many



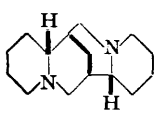
(LXXIII)



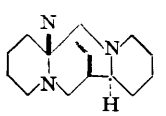
(LXXIV)



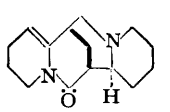
(LXXV)



(LXXXVI)

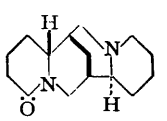


(LXXVII)

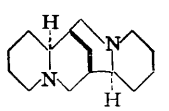


(LXXVIII)

attempts. The hexahydro-derivatives, hexahydrocytosine, α-*iso*sparteine (LXXVI) and sparteine (LXXVII), have however all been synthesised. Aphillidine (LXXVIII) has an oxidation state between anagyrine and sparteine, and reduction gives aphilline isomeric with lupanine (LXXIX).

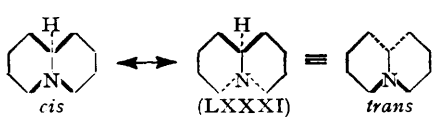


(LXXIX)



(LXXX)

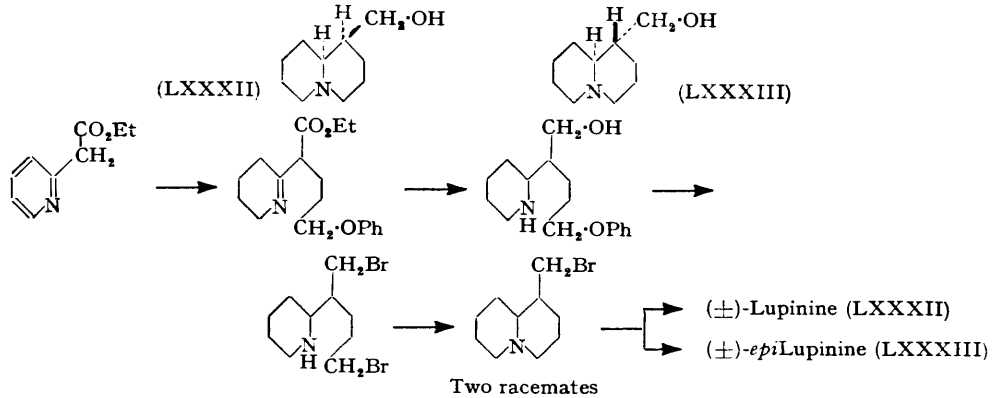
The simplest reduced pyridocoline is norlupinane (LXXXI), easily obtained from lupinine (LXXXII). It is not resolvable since the *cis*- and *trans*-forms are interconvertible owing to the oscillation of the nitrogen valencies as in ammonia. In the case of lupinine (LXXXII), how-



cis (LXXXI) *trans*

ever, with its second asymmetric centre two racemates are possible. This alkaloid, with sparteine (LXXVII) and lupanine (LXXIX), occurs in various lupin seeds; and sparteine occurs also in gorse. These fully reduced systems have two fused pyridocoline systems but their stereochemistry is such that only *cis*-fusion is possible so that three forms of sparteine (LXXVI, LXXVII, and LXXX) are possible. Many workers, since Willstätter's early attack on lupinine (*Ber.*, 1902, **35**, 1914), have contributed to the chemistry of this group, particularly Karrer on

lupinine, and Ing on cytosine; and with Drs. Raper and Morgan we synthesised lupinine and sparteine (the former is illustrated below), and I had hoped to add lupanine.



In this brief review of recent advances many perforce have gone unmentioned, such as Pauling's great contribution to protein chemistry, the unravelling of the structure of insulin at Cambridge, and so on.

A separate lecture would be necessary to deal with other recent applications of organic nitrogen compounds, of which the use of *iso*nicotinic hydrazide as a tuberculostatic drug is a striking example.

My grateful thanks are offered to many who have helped in literature searches and in other ways, but particularly to Drs. Swan, Vipond, and Cockburn, and to Miss N. Brown, B.Sc., who also made the slides.