415. *The Alkaloids of* Picralima Klaineana, *Pierre. Part II.*

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THOUGH the seeds of *Picralima klaineana*, Pierre, known as "akuamma" in the Gold Coast Colony, have a wide-spread reputation among natives in tropical Africa as a remedy for malaria, the tree yielding them is nowhere abundant and supplies of seed are difficult to obtain. It was hoped that if native belief in the therapeutic value of the seeds were confirmed, cultivation of the tree might be undertaken, but the results of trials of the total alkaloids and of the

principal alkaloid, akuammine, in bird malaria, for which the author is indebted to Dr. J. W. S. Macfie, have shown that these alkaloids are without remedial action in this disease and presumably also in human malaria. It is an interesting fact that the reputation of no native remedy for malaria has so far survived careful pharmacological or clinical experiment, with the exception of cinchona, and it is a disputed point whether the anti-malarial value of cinchona was known before the advent of Europeans to South America.

In the course of the present investigation six samples of akuamma seed have been examined, four from the Gold Coast Colony, one from the Belgian Congo, and one from Uganda. In these the percentage of total alkaloids has varied from 3·5 to 4·8.

The components of the total alkaloids so far known (Henry and Sharp, J., 1927, 1950) are (A) a weakly basic, asphaltic substance; (B) a strongly basic, amorphous alkaloid, readily distinguished by the ultramarine-blue colour which aqueous solutions of its salts give with ferric chloride; (C) the crystalline alkaloid akuammine, $C_{22}H_{28}O_4N_2$, which forms from 11 to 16% of the total alkaloids; and (D) a second crystalline alkaloid, for which the name *akuammicine* is now proposed, and which has not hitherto been available in sufficient quantity for detailed description.

The present paper describes (1) a better method for the isolation of akuammicine; (2) the isolation and characterisation of six new alkaloids, of which five are crystalline and the sixth has been obtained as a well-crystallised scarlet *picrate*; and (3) the occurrence of akuammine hydrate in residues from the purification of akuammine, in which special precautions had been taken to avoid conversion of akuammine into the hydrate (*loc. cit.*, p. 1958), so the latter must now be regarded as a natural constituent of the seeds.

The alkaloids are isolated from the seeds in two main groups, the first group (a) being removed, along with fat and wax, by light petroleum and the second group (b) by extraction of the fat-free seeds with alcohol. Group (a) is almost entirely soluble in benzene and from such a solution three alkaloids can be separated in the following order, by fractional extraction with *N*-hydrochloric acid : amorphous alkaloid B (limit of fraction indicated by the colour reaction with ferric chloride already mentioned), pseudakuammicine, $C_{19}H_{20}O_2N_2$, and its isomeride akuammicine, the hydrochlorides of the last two being readily separable owing to the great difference in their solubility in hot water. Group (b) is mainly composed of the asphaltic base A, from which the other bases are separated by extraction, in a Soxhlet apparatus, with ether containing a little alcohol. Akuammine and akuammidine, $C_{21}H_{24}O_3N_2$, crystallise together as extraction proceeds and are finally separated by treatment with

boiling alcohol, in which akuammidine is easily soluble and akuammine almost insoluble. The alkaloids remaining in solution in the ether are recovered, dissolved as far as possible in benzene, and isolated by fractional extraction with *N*-hydrochloric acid, when the hydrochlorides are removed in the following order: amorphous alkaloid B (ferric chloride test), akuammiline, $C_{22}H_{24}O_4N_2$, pseudakuammigine, $C_{22}H_{26}O_3N_2$, and akuammenine, $C_{20}H_{22}O_4N_2$, the last two being isolated as picrates partly from the ultimate acid extracts and partly from the residual benzene solution. Throughout this fractionation of both groups of alkaloids tar separates, and from this by treatment with acetone, akuammigine, $C_{22}H_{26}O_3N_2$, is obtained as the hydrochloride.

The names, extended linear formulæ, yields, and typical colour reactions of the nine crystalline alkaloids so far obtained from the seeds are given in the following table.

Name.	Extended linear formula.	Yield on seeds, %.	Colour reactions. Conc. HNO_3.	Colour reactions. Piperonal + HCl.
Akuammicine*	$C_{18}H_{17}ON_2(OMe)$	0·0064	Bright green, becoming blue on dilution with water	Magenta, changing to ultramarine blue on standing
Pseudakuammicine	$C_{18}H_{17}ON_2(OMe)$	0·0037		
Akuammenine	$C_{19}H_{19}O_3N_2(OMe)$	0·0006		
Akuammidine*	$C_{20}H_{21}O_2N_2(OMe)$	0·0340	Yellow	Pink, changing to amethyst or standing
Akuammigine*	$C_{21}H_{23}O_2N_2(OMe)$	0·0100	Bright yellow	
Pseudakuammigine*	$C_{20}H_{20}O_2N(OMe)(NMe)$	0·0170	Brown, changing to yellow	
Akuammiline*	$C_{21}H_{21}O_3N_2(OMe)$	0·0107	None	
Akuammine*	$C_{20}H_{21}O_2N(OH)(OMe)(NMe)$	0·5600	Blood-red	
Akuammine hydrate*	$C_{20}H_{23}O_3N(OH)(OMe)(NMe)$	—	Blood-red	

The samples of akuamma seed worked up were usually small and the yields recorded in the table were those obtained from the only large sample, 21 kilog., received from the Gold Coast. With the exception of akuammine, of which about 120 grams have now been accumulated for further work, only small quantities of these alkaloids, ranging from 0·2 gram of akuammenine to 7·5 grams of akuammidine, were available and consequently nothing but their characterisation and a few exploratory experiments could be attempted with them.

The alkaloids whose names are marked with an asterisk in the table all yield crystalline monomethiodides except akuammigine, in which case it is still amorphous. All these methiodides behave as quaternary iodides, indicating the presence of at least one tertiary nitrogen atom in the parent alkaloid. No evidence that the second nitrogen atom in akuammine is present as an imino-group has been obtained. This base gives a benzoyl derivative, which behaves as a normal *O*-benzoyl ester, and with nitrous acid it yields nitroakuammine. Akuammidine also yields a monobenzoyl derivative, but this is neutral in reaction and insoluble in acids and the entering acyl group is apparently attached to a nitrogen atom; this, however,

does not necessarily imply the presence of an imino-group in akuammidine, and may be another of the now numerous instances of acylation being preceded by the scission of a heterocyclic nucleus. A nitroso-derivative of akuammidine has not been obtained, nitrous acid converting the base into an intractable amorphous product. Akuammigine, pseudakuammigine, and akuammiline also yield neutral benzoyl derivatives, apparently similar in type to benzoylakuammidine, but they crystallise badly and have not yet been obtained in a satisfactory condition for analysis. Benzoyl chloride converts akuammicine into a basic substance, which is very soluble in water, cannot be extracted from its aqueous solution by immiscible solvents, and has so far only been isolated as an impure picrate.

In the hope of securing a further source of supply of these alkaloids the bark and leaves of *Picralima klaineana* have also been examined, but from them only amorphous alkaloids could be isolated; with possibly a trace of akuammidine from the bark.

Among the non-alkaloidal constituents of the seeds are sucrose and ammonia with a trace of an alkylamine. There are also present a semi-solid fat, wax, and a substance resembling inulin. None of these has been examined so far, and they may prove of considerable biochemical interest, since *Picralima klaineana* is the only known member of its genus and little is known regarding the non-alkaloidal constituents of the botanical family, *Apocynaceæ*, to which this genus belongs.

The author is indebted to the authorities of the Imperial Institute for the supplies of seed, bark, and leaves used, to Messrs. L. Barnett and A. C. Camfield for much assistance in experimental work, and to Messrs. A. Bennett and H. C. Clarke for numerous micro-analyses.

Experimental.

Petroleum Extract.—As the seeds contain wax and fat which produce difficulties in the later operations, it is convenient to remove these components first by extraction with light petroleum, b. p. 60—80° (Soxhlet). This extract contains alkaloids, which are recovered by shaking it with portions of *N*-HCl until the acid is no longer neutralised. From this aq. solution the alkaloids are pptd. by means of sat. Na_2CO_3 aq. The washed ppt. is dried at room temp. in vac., weighed, dissolved as far as possible in C_6H_6, and fractionally extracted with *N*-HCl as described below. The fractions giving no colour with $FeCl_3$ (p. 2760), on slow evaporation deposit in the following order *pseudakuammicine hydrochloride*, m. p. 215°, *akuammicine hydrochloride*, m. p. 175°, and a little *akuammiline hydrochloride*, m. p. 198°, sometimes accompanied by traces of *akuammigine* and *pseudakuammigine hydrochlorides*. The latter is always present in the final residues, but it is not possible to isolate it as the picrate (p. 2763) owing to the persistently gelatinous character of the picrates of the other bases remaining in this residue.

Alcoholic Extract.—The alc. extract of the fat-free seeds is worked up, as

described already, to the point at which the total alkaloids are pptd. with Na_2CO_3 aq. (*loc. cit.*, p. 1952). The washed dried ppt. is then exhausted (Soxhlet) with Et_2O containing 3% EtOH. The crude akuammine, which accumulates in the extraction flask, is filtered out and purified as described (*loc. cit.*, p. 1953). The ether–alcohol solution on concn. deposits a mixture of akuammine and *akuammidine*, separable into its components by boiling EtOH, in which the first is sparingly, and the second readily, sol. The ether–alcohol filtrate is evaporated to dryness, and the residue exhausted with boiling C_6H_6. The weight of alkaloid in the C_6H_6 solution is determined on an aliquot part and the whole solution, which usually deposits a little more akuammidine as it cools, is fractionally extracted with *N*-HCl, aliquot parts of acid, each equiv. to about 1/10th of the amount of *N*-acid required to extract the whole of the alkaloid present (1 g. of alkaloid = 2·25 c.c. of *N*-acid, approx.), being used. The earlier fractions of acid take out alkaloid B, as indicated by the ultramarine colour produced by a drop of $FeCl_3$ aq. The middle fractions give a slate colour with $FeCl_3$ and these on concn. deposit some akuammigine hydrochloride in small hard prisms, or gelatinise owing to the separation of this alkaloidal salt in colloidal form. The later fractions give no colour with $FeCl_3$ aq. and on concn. deposit akuammiline hydrochloride, and eventually leave residues from which pseudakuammigine can be isolated as the picrate (see below), or as the hydriodide by the addition of hot 40% KI aq. to a filtered boiling aq. solution of the residue. When the portions of *N*-acid cease to be neutralised, the C_6H_6 solution is evaporated to dryness, the residue dissolved in a little EtOH, and a boiling EtOH solution of picric acid added. Pseudakuammigine picrate is pptd. as the solution cools and can be crystallised by the addition of Me_2CO drop by drop to its suspension in boiling EtOH.

The mother-liquors from the crystn. of crude pseudakuammigine picrate are almost black and in closed vessels gradually deposit more of this salt as a brilliant scarlet ppt. When the latter is boiled with MeOH the clean, bright yellow picrate is left undissolved and the filtrate on cooling deposits brilliant scarlet flakes of crude *akuammenine picrate*.

If the preliminary extraction of the seed with light petroleum is omitted or is incomplete, akuammicine and pseudakuammicine are found in the C_6H_6 solution and a complex mixture of their hydrochlorides with those of pseudakuammigine and akuammiline separates in place of crude akuammiline hydrochloride. Such a mixture is best separated by repeated fractional crystn., alternately from boiling EtOH and boiling H_2O; the four then separate in the order just named, the colour reactions of the first three with conc. HNO_3 being used as a rough guide to the progress of the separation.

During the fractional extraction of the C_6H_6 solutions with acid, tar separates and gradually covers the sides of the separator with a thin, hard, black crust. The separator is finally emptied, allowed to drain, and Me_2CO run down the sides drop by drop. The tar dissolves and leaves behind minute crystals of akuammigine hydrochloride. The latter is also present in traces in the crude hydrochloride fractions of the other alkaloids (see above) and is recovered in the course of their purification.

Description of the Alkaloids.—In the following account the m. p.'s are also decomp. points except where two are given, in which case the lower is a true m. p.; the temp. are corrected. The combustion results and the determinations of methoxyl, methylimino-groups and halogens are from micro-analyses. The

methiodides were all prepared in the cold by allowing the base to stand with MeI: if necessary to secure solution, MeOH was added. In preparing acyl derivatives 0·2 to 0·4 g. of the base was boiled for a few min. with enough BzCl (or Ac_2O in two cases) to dissolve it. After 24 hr. the mixture was shaken with H_2O until the odour of the acylating agent had disappeared; $CHCl_3$ was then added, and after thorough agitation the aq. acid liquid containing any *O*-benzoyl derivative separated. The $CHCl_3$ solution was shaken with small quantities of sat. Na_2CO_3 aq. until BzOH was no longer removed, then dried, and the solvent removed, leaving any neutral and presumably *N*-benzoyl derivative.

Akuammine (*loc. cit.*, p. 1954). When 0·25 g. of the base, dissolved in 10 c.c. of HCl (2%) and cooled to 0°, is mixed with 0·07 g. of $NaNO_2$ in 1 c.c. of H_2O, a scarlet ppt. is formed, which can be recryst. from boiling H_2O in minute prisms. It darkens at 230°, decomposes at 246° without melting, does not give Liebermann's nitrosoamine reaction, and is readily sol. in Na_2CO_3 aq., from which the corresponding base can be slowly extracted by Et_2O and more readily by $CHCl_3$. The substance has the composition of a *nitroakuammine hydrochloride* [Found: (1) loss at 120° in vac., 7·35. $C_{22}H_{27}O_4N_2(NO_2),HCl,2H_2O$ requires H_2O, 7·2%. (2) for dry salt: C, 56·7; H, 5·8; N, 9·0; Cl, 7·35. $C_{22}H_{27}O_4N_2(NO_2),HCl$ requires C, 56·7; H, 6·05; N, 9·0; Cl, 7·6%]. *Benzoylakuammine*, prepared by the general method and recovered from the crude hydrochloride, crystallises from hot EtOH in anhyd. needles, m. p. 245° (Found: C, 70·9; H, 6·4; N, 5·9. $C_{22}H_{27}O_4N_2Bz$ requires C, 71·2; H, 6·6; N, 5·7%).

When boiled with HCl (*d* 1·1), akuammine is converted into an asphaltic substance, which has all the physical properties of alkaloid A found naturally in akuamma seeds. The product could not be crystallised, but after boiling with EtOH, until nothing more dissolved, it formed a granular reddish-brown powder, the composition of which indicates that it is formed by demethylation and subsequent condensation of two mols. of akuammine, followed by loss of one Me from a NMe group (Found: C, 69·3; H, 6·5; N, 7·8; OMe, nil; NMe, 4·4. $C_{41}H_{48}O_7N_4$ requires C, 69·45; H, 6·8; N, 7·9; OMe, nil; NMe, 4·12 or 8·24% for 2NMe).

Akuammine hydrate methiodide. When akuammine hydrate (*loc. cit.*, p. 1958), suspended in MeOH, is treated with MeI, a dark green solution is formed, which rapidly decolourises and deposits a quant. yield of the *methiodide* in minute, pale brown needles. This substance has no m. p. up to 300°, and no satisfactory solvent has been found for recrystallising it without decomp. (Found for dry substance: C, 51·5; H, 5·3; N, 5·15; OMe, 6·6; NMe, 11·3; I, 22·5. $C_{22}H_{30}O_5N_2,MeI$ requires C, 50·7; H, 6·1; N, 5·1; OMe, 5·5; 2NMe, 10·6; I, 23·3%). The analytical results are, as was to be expected, not very good, but they leave no doubt that the substance is a simple methiodide of akuammine hydrate.

Akuammidine. The base, isolated as described, separates from boiling EtOH in lustrous, thick, translucent needles, containing about 12% H_2O ($3H_2O$ requires 13·3%): on exposure to air the crystals gradually become chalk-white and finally contain about 4·5% H_2O. The substance has m. p. 248·5°, $[\alpha]_D^{16°}$ + 21° (c = 1 in EtOH) or + 0·44° (c = 1·25 in Me_2CO). In spite of the fall in rotation in Me_2CO, the base is recovered unchanged from that solvent. It is sparingly sol. in $CHCl_3$, EtOH, or Et_2O and almost insol. in C_6H_6 [Found: (1) in air-dry base: loss at 120° in vac., 4·3. $C_{21}H_{24}O_3N_2,H_2O$ requires H_2O, 4·9%.

(2) in dry base : C, 71·3; H, 6·8; N, 8·3; OMe, 8·7; NMe, 1·6. $C_{21}H_{24}O_3N_2$ requires C, 71·5; H, 6·8; N, 7·9; OMe, 8·8; NMe, 8·2%]. The hydrochloride separates as an oil from H_2O or EtOH and has not been crystallised. A solution of the base in *N*/10-HCl has $[\alpha]_D^{16°}$ + 70·23° (*c* = 2·74). The *hydriodide*, pptd. from a hot aq. solution of the oily hydrochloride by KI, separates as an oil, which on standing hardens and on addition of EtOH to its suspension in H_2O gradually crystallises in transparent prisms, m. p. 90° or m. p. 238° (anhyd.) [Found : (1) loss in vac. first at room temp. and finally at 120°, 10·1. $C_{21}H_{24}O_3N_2,HI,3H_2O$ requires H_2O, 10·1. (2) for dry salt : C, 52·5; H, 5·45; N, 6·7; I, 26·2; OMe, 5·8; NMe, 2·3. $C_{21}H_{24}O_3N_2,HI$ requires C, 52·4; H, 5·2; N, 5·8; I, 26·4; OMe, 6·4; NMe, 6·0%]. The perchlorate forms translucent hexagonal prisms, m. p. 70° and 110°. The freshly-pptd. picrate, dissolved in hot EtOH, separates on cooling in hard, dull yellow spheroids, m. p. 215°, and is then insol. in ordinary solvents. The *methiodide* crystallises from 50% MeOH in transparent prisms, m. p. 195° and 233°, $[\alpha]_D^{18°}$ + 16·3° (*c* = 2·512 in MeOH) [Found : (1) loss at 120° in vac., 4·0. $C_{21}H_{24}O_3N_2,CH_3I,H_2O$ requires H_2O, 3·6. (2) for dry salt : C, 53·2; H, 5·5; N, 5·6; OMe, 6·6; NMe, 6·2. $C_{21}H_{24}O_3N_2,CH_3I$ requires C, 53·4; H, 5·5; N, 5·6; OMe, 6·2; NMe, 5·8%]. The methiodide is isomeric, but not identical, with pseudakuammigine hydriodide.

On acylation by the general method (see above) the acyl derivative of the base remains in the $CHCl_3$, from which it is not extracted by acids or alkalis. On isolation, it is insol. in acids, but can be converted into a picrate or a methiodide : when boiled with either acids or alkalis, it regenerates akuammidine and the appropriate acid.

Acetylakuammidine is best crystallised by adding EtOH to its solution in $CHCl_3$ and slowly distilling off the latter. It forms minute, anhyd., colourless prisms, m. p. 272° (Found : C, 69·5; H, 6·5. $C_{21}H_{23}O_3N_2Ac$ requires C, 70·1; H, 6·6%). *Benzoylakuammidine* crystallises from boiling EtOH in colourless needles, m. p. 219° (Found for substance dried at 120° in vac. : C, 73·35; H, 5·9; N, 6·4; OMe, 6·3; NMe, 1·2. $C_{21}H_{23}O_3N_2Bz$ requires C, 73·6; H, 6·2; N, 6·1; OMe, 6·8; NMe, 6·3%). *Benzoylakuammidine methiodide* crystallises from boiling MeOH in minute colourless prisms, m. p. 240° (Found for substance dried at 120° in vac. : C, 58·05; H, 5·7; N, 4·7; OMe, 4·7; NMe, 6·4. $C_{21}H_{23}O_3N_2Bz,CH_3I$ requires C, 58·2; H, 5·2; N, 4·6; OMe, 5·2; NMe, 4·8%).

Akuammiline. The crude hydrochloride, cryst. until of constant m. p. and rotation, gives on analysis results which cannot be reconciled with those of the pure base and is therefore purified through the latter, a wasteful process, since the base only crystallises well from EtOH, in which it is readily sol. Akuammiline forms translucent anhyd. prisms with a faintly yellow tinge, m. p. 160°, $[\alpha]_D^{20°}$ + 47·9° (*c* = 4·5 in EtOH), and is readily sol. in Et_2O, $CHCl_3$ or warm C_6H_6 (Found : C, 69·8; H, 6·6; N, 7·2; OMe, 7·7; NMe, 1·45. $C_{22}H_{24}O_4N_2$ requires C, 69·4; H, 6·3; N, 7·3; OMe, 8·1; NMe, 7·6%). The *hydrochloride*, prepared by neutralisation of the pure base, crystallises from H_2O or EtOH in colourless needles, m. p. 196°, $[\alpha]_D^{20°}$ − 29·6° (*c* = 3·84 in H_2O) [Found : (1) loss at 120° in vac., 3·9. $C_{22}H_{24}O_4N_2,HCl,H_2O$ requires H_2O, 4·1. (2) for dry salt : C, 63·6; H, 6·4; N, 7·2; Cl, 9·0; OMe, 6·8; NMe, 1·1. $C_{22}H_{24}O_4N_2,HCl$ requires C, 63·3; H, 6·0; N, 6·7; Cl, 8·5; OMe, 7·4; NMe, 6·9%]. The *hydriodide* is unstable and apt to resinify on crystn. from hot H_2O, but separates from hot 25% EtOH in colourless hair-like needles, m. p.

210° (Found for salt dried at 120° in vac. : C, 52·1; H, 4·8; N, 5·9; I, 25·5; OMe, 5·25. $C_{22}H_{24}O_4N_2$,HI requires C, 51·95; H, 4·95; N, 5·5; I, 24·9; OMe, 6·1%). The *nitrate* crystallises from boiling H_2O in stellate groups of prisms, m. p. 204° (Found : N, 9·1. $C_{22}H_{24}O_4N_2$,HNO_3 requires N, 9·5%). The *methiodide* separates from hot H_2O in anhyd. rosettes of colourless needles, m. p. 233°, $[\alpha]_D^{20°}$ — 83·3° (c = 1·36 in EtOH). Its aq. solution is not pptd. on addition of KHO aq. (Found : C, 53·6; H, 5·5; N, 5·4; I, 24·7; OMe, 6·0; NMe, 6·9. $C_{22}H_{24}O_4N_2$,CH_3I requires C, 52·85; H, 5·2; N, 5·4; I, 24·3; OMe, 5·9; NMe, 5·6%). The benzoyl derivative is of the same type as that yielded by akuammidine, but has not been obtained cryst. : it yields a cryst. methiodide, which has not been obtained pure, and an amorphous picrate.

Akuammigine. The crude hydrochloride can be crystallised with difficulty from boiling H_2O, EtOH or 50% MeOH, owing to its sparing solubility, and then forms minute, anhyd., colourless prisms, m. p. 287°, $[\alpha]_D^{20°}$ — 37·8° (c = 0·835 in MeOH) (Found : C, 65·5; H, 6·4; N, 7·4; OMe, 7·8; NMe, 0·8. $C_{22}H_{26}O_3N_2$,HCl requires C, 65·5; H, 6·75; N, 6·95; OMe, 7·7; NMe, 7·1%). The *nitrate* crystallises from boiling 50% EtOH in anhyd., lustrous, flattened prisms, m. p. 261° (Found : C, 61·5; H, 6·3; N, 9·9. $C_{22}H_{26}O_3N_2$,HNO_3 requires C, 61·5; H, 6·3; N, 9·8%). The picrate forms garnet-coloured prisms from boiling EtOH and has m. p. 240°. The *base* crystallises from EtOH, on slight addition of H_2O, in flat, square, faintly yellow tablets, m. p. 125° $[\alpha]_D^{20°}$ — 44·4° (c = 2·775, air-dry substance in EtOH). All attempts to dry the base without decomp. failed. Even at room temp. in a vac. desiccator, it gradually turns orange and finally brown. Analyses of the air-dried substance indicate that it contains one mol. H_2O (Found : C, 68·3; H, 6·75; N, 7·8; OMe, 9·1; NMe, 0·7. $C_{22}H_{26}O_3N_2$,H_2O requires C, 68·7; H, 7·3; N, 7·3; OMe, 8·8; NMe, 8·2%). The methiodide has not been obtained cryst., nor has the benzoyl derivative, which is insol. in acids and similar in type to that of akuammidine (p. 2765).

Pseudakuammigine. The base is not readily recoverable from the picrate first isolated (p. 2763) and is best obtained by pouring an Me_2CO solution of the salt on to CaO, allowing the solvent to evaporate, mixing the residue into a thin paste with EtOH, leaving this to dry, and finally drying thoroughly in a vac. desiccator. The dry powder is then exhausted (Soxhlet) with Et_2O. The cryst. residue left on removal of the solvent is dissolved in a little hot EtOH, filtered, and H_2O carefully added. Anhyd. colourless prisms separate, m. p. 165°, $[\alpha]_D^{20°}$ — 53·8° (c = 3·42 in EtOH) (Found : C, 71·9; H, 7·0; N, 7·9; OMe, 8·4; NMe, 6·9. $C_{22}H_{26}O_3N_2$ requires C, 72·1; H, 7·1; N, 7·6; OMe, 8·4; NMe, 7·9%). The *hydrochloride* crystallises from hot H_2O in slender colourless prisms, m. p. 183° (air-dry) or 218° (anhyd.), $[\alpha]_D^{20°}$ — 15·4° (c = 1·065 in EtOH) [Found : (1) loss on drying at 120° in vac., 4·35. $C_{22}H_{26}O_3N_2$,HCl,H_2O requires H_2O, 4·2%. (2) in dry substance : C, 64·9; H, 6·8; N, 6·8; Cl, 8·2. $C_{22}H_{26}O_3N_2$,HCl requires C, 65·5; H, 6·7; N, 6·9; Cl, 8·8%]. The *hydriodide* separates from boiling H_2O in lustrous hair-like needles, m. p. 215°, is sparingly sol. in H_2O or EtOH, more sol. in Me_2CO, and has $[\alpha]_D^{20°}$ — 1·43° (c = 2·84 in Me_2CO) [Found : (1) loss on drying at 120° in vac., 3·5. $C_{22}H_{26}O_3N_2$,HI,H_2O requires H_2O, 3·5%. (2) for dry substance : C, 53·2; H, 5·2; N, 5·6; I, 25·6; OMe, 6·0; NMe, 9·8. $C_{22}H_{26}O_3N_2$,HI requires C, 53·4; H, 5·5; N, 5·6; I, 25·6; OMe, 6·2; NMe, 5·8; 2NMe, 11·6%]. No explanation of the high methylimino-figure can be given. In three determinations the results were 9·8, 9·85 (micro-analyses), and 9·78 (macro-analysis). The results obtained

with the base and the methiodide (see below) leave little doubt that only one methylimino-group is present in the alkaloid and that the hydriodide gives abnormal results for this component. The *picrate* crystallises from boiling Me_2CO in anhyd. yellow rosettes of needles, m. p. 223°, and is almost insol. in boiling EtOH [Found : C, 57·0; H, 4·8; N, 11·9. $C_{22}H_{26}O_3N_2,C_6H_2(OH)(NO_2)_3$ requires C, 56·4; H, 4·9; N, 11·7%]. The *methiodide* separates from EtOH in flat, transparent, anhyd. tablets, m. p. 275° (Found : C, 54·4; H, 5·7; N, 5·7; I, 25·2; OMe, 7·1; NMe, 11·0. $C_{22}H_{26}O_3N_2,CH_3I$ requires C, 54·3; H, 5·75; N, 5·5; I, 24·9; OMe, 6·1; 2NMe, 11·4%). The benzoyl derivative resembles that of akuammidine in type, but has not been crystallised or made to yield a cryst. derivative.

Akuammenine. The minute amount of the *picrate* of this base obtained was recrystallised to const. m. p., 225°, from boiling MeOH and was thus obtained in scarlet flakes [Found : C, 53·2; H, 4·45; N, 11·65; OMe, 5·3; NMe, nil. $C_{20}H_{22}O_4N_2,C_6H_2(OH)(NO_2)_3$ requires C, 53·4; H, 4·3; N, 12·0; OMe, 5·3%].

Akuammicine. The preliminary description of this base already published (base D; *loc. cit.*, p. 1958) may now be supplemented as follows. The crude hydrochloride as isolated (p. 2762) may contain some pseudakuammicine hydrochloride, which may be removed by keeping solutions of the crude fractions in 10 times their weight of boiling H_2O for about 1 hr. and pouring off the mother-liquors from the characteristic stumpy needles of the less sol. isomeride. The combined mother-liquors on slow concn. in a vac. desiccator deposit colourless prisms of *akuammicine hydrochloride*, which can be recrystallised from boiling H_2O or EtOH until of const. m. p. 144° (air-dry) or 171° (dry), $[\alpha]_D^{16°}$ — 626·2° (c = 1·22 in EtOH) [Found : (1) loss in vac. at 100°, 8·8. $C_{19}H_{20}O_2N_2,HCl,2H_2O$ requires H_2O, 9·5. (2) for dry salt : C, 66·1; H, 6·05; N, 7·9; OMe, 7·8; NMe, trace. $C_{19}H_{20}O_2N_2,HCl$ requires C, 66·15; H, 6·1; N, 8·1; OMe, 9·0%]. The sulphate (*loc. cit.*) forms colourless cubes from H_2O, m. p. 161° (air-dry) [Found : (1) loss in vac. at 100°, 8·9. Calc. for $(C_{19}H_{20}O_2N_2)_2,H_2SO_4,4H_2O$: H_2O, 9·2%. (2) for dry salt : S, 4·5. Calc. : S, 4·5%]. The nitrate crystallises from boiling H_2O in brilliant spangles, m. p. 182·5°. The *base* crystallises from hot EtOH in brilliant colourless leaflets, m. p. 177·5° (Found for substance dried at 100° in vac. : C, 74·0; H, 6·3; N, 8·9; OMe, 9·45; NMe, 1·6. $C_{19}H_{20}O_2N_2$ requires C, 73·9; H, 6·5; N, 9·1; OMe, 10·1; NMe, 9·4%). The base and all its salts on melting decompose into a bright red froth. The *methiodide* crystallises from boiling H_2O in minute, faintly yellow, anhyd. prisms, m. p. 252° (Found : N, 6·2; OMe, 6·9; NMe, 6·75. $C_{19}H_{20}O_2N_2,CH_3I$ requires N, 6·2; OMe, 6·8; NMe, 6·45%). On acylation by the general method, akuammicine yields a salt of a new base, which is not recoverable by the addition of alkali and extraction with any immiscible solvent tried, but can be pptd. with picric acid, yielding a picrate which crystallises badly and has not been obtained analytically pure.

Pseudakuammicine. The *hydrochloride* isolated as described (pp. 2762, 2767) is readily purified by crystn. from boiling EtOH or boiling H_2O and forms minute stumpy needles, m. p. 216° [Found : (1) loss in vac. at 120°, 5·6. $C_{19}H_{20}O_2N_2,HCl,H_2O$ requires H_2O, 5·0. (2) for dry salt : C, 65·7; H, 6·1; N, 7·7; Cl, 9·8; OMe, 8·5; NMe, 1·0. $C_{19}H_{20}O_2N_2,HCl$ requires C, 66·15; H, 6·1; N, 8·1; Cl, 10·3; OMe, 9·0; NMe, 8·4%]. The *base*, pptd. as a gum, rapidly crystallises and can be recrystallised from boiling EtOH in anhyd., colourless, square plates, m. p. 187·5° (Found : C, 74·6; H, 7·0; N, 8·8; OMe, 9·7; NMe, 2·2. $C_{19}H_{20}O_2N_2$ requires C, 73·9; H, 6·5; N, 9·1;

OMe, 10·1; NMe, 9·4%). The base persistently gave high carbon results on analysis, due it is believed to slight decomp., which occurs on drying at 100° in vac., the dried product having a faintly brown tinge. On melting, the base and all its salts decompose into a red froth. The picrate forms dark olive-green prisms, m. p. 196°.

Non-alkaloidal constituents. As the preliminary extraction of the seeds with light petroleum (p. 2762) proceeds, there accumulates in the flask a greyish-yellow ppt. consisting mainly of wax. This on solution in $CHCl_3$ leaves undissolved a small amount of a water-sol. product, which reduces Fehling's solution on boiling and resembles crude inulin in appearance. The light petroleum extract after removal of the alkaloids (p. 2762) leaves on evaporation of the solvent a dark greyish-green, semi-solid fat. None of these products has yet been examined in detail.

From one sample of seeds from the Belgian Congo, the crude alc. extract, which had been kept for a few days before being worked up, deposited a partly cryst. ppt. This on repeated extraction with EtOH was separated into two portions, the wax already referred to and a sugar. The latter after repeated washing with $CHCl_3$ to remove traces of wax was crystallised from aq. EtOH until of const. m. p. 187°. It reduced Fehling's solution very slightly before, and copiously after, hydrolysis with boiling HCl. It had $[\alpha]_D^{22°}$ + 64·84° (c = 3·2 in H_2O) (Found : C, 41·8; H, 6·45. Calc. for $C_{12}H_{22}O_{11}$: C, 42·1; H, 6·43%). The sugar is therefore sucrose and it showed no depression of m. p. on admixture with that substance.

Volatile bases. The residual aq. liquors remaining after pptn. of the total alkaloids developed a slight fishy odour on addition of NaOH. They were therefore steam-distilled after the addition of NaOH, the alkaline steam collected in dilute HCl, and the still acid distillate evaporated to dryness. The residue was recrystallised from H_2O in three fractions, which contained respectively 66·25, 66·2, and 65·1% Cl : the portion which did not crystallise from H_2O was separated into two more cryst. fractions by trituration first with dry EtOH and finally with Me_2CO. These contained 66·1 and 64·2% Cl. In a second instance the crude residue of hydrochloride was fractionated by Goodson's method (Sharp and Solomon, J., 1931, 1477). The three fractions obtained, representing 25% of the whole and the richest in alkylamine, contained 66·1, 65·0, and 61·65% Cl (Calc. for NH_4Cl : Cl, 66·3. Calc. for CH_3NH_2,HCl : Cl, 52·5%). There can be no doubt, therefore, that only a minute amount of alkylamine is present and that NH_3 is the chief constituent of the volatile bases.

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