Some Semi-synthetic Derivatives of Pyrrolizidine Alkaloids

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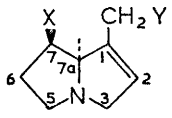
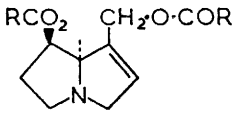
Diesters (VII—XXIII) of retronecine (I), and acyl derivatives of the alkaloids indicine (XXIV), monocrotaline (XXVII), and retrorsine (XXIX), have been prepared for toxicological studies. Dichlororetronecine (III) is described, and also retronamine (V), an amino-analogue of retronecine, which provide a simple route to amide analogues of pyrrolizidine esters.

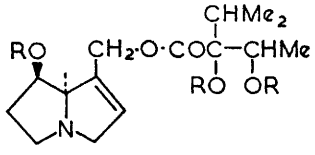
SEMI-SYNTHETIC esters and other analogues and derivatives of pyrrolizidine alkaloids were prepared for comparison with the natural alkaloids, many of which are toxic.[1,2] A preliminary account has appeared.[3]



(I) X = Y = OH
(II) X = OH, Y = Cl
(III) X = Y = Cl
(IV) X = OH, Y = N_3
(V) X = OH, Y = NH_2
(VI) X = OH, Y = NHAc

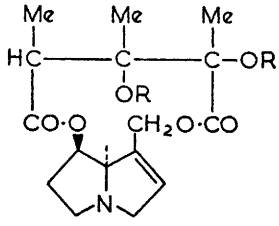


(VII)—(XXIII) (see Table)

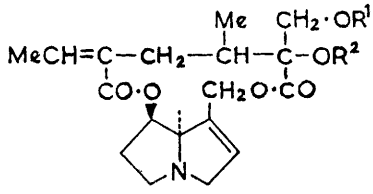


(XXIV) R = H
(XXV) R = Ac
(XXVI) R = Me_2CHCH_2CO

prepared by heating retronecine hydrochloride with acid chlorides (see Table).



(XXVII) R = H
(XXVIII) R = Ac



(XXIX) $R^1 = R^2 = H$
(XXX) R^1 = Ac, R^2 = H
(XXXI) $R^1 = R^2$ = Ac

The esters were reasonably stable in acid solution, but some were rapidly hydrolysed by alkali. Thus, diacetylretronecine (VII) was extracted from aqueous solution at pH 8, but above pH 9 it was hydrolysed before extraction was possible. Since the basic esters were unstable oils they were isolated, purified, and characterised as their picrates (Table). On regeneration from the picrates with anion exchange resin, the bases gave single spots on paper chromatograms. Their high R_F

Retronecine diesters and derivatives

Ester (base)			Picrate								Hydrochloride									
					Found (%)			Required (%)						Found (%)				Required (%)			
R	$[\alpha]_D^{20}$ (abs. EtOH)	R_F	M.p.	Formula	C	H	N	C	H	N	M.p.	$[\alpha]_D^{20}$ (H_2O)	Formula	C	H	Cl	N	C	H	Cl	N
(VII) Me	+17·4° [a]	0·56	146° [c,g]	$C_{18}H_{20}N_4O_{11}$							114—115° [b]	−17·1°	$C_{12}H_{18}ClNO_4$				5·3				5·1
(VIII) $MeCH_2$	+16·75	0·81	107 [d]	$C_{20}H_{24}N_4O_{11}$	48·2	4·7	11·6	48·4	4·8	11·3	124 [e,h]	−14·2	$C_{14}H_{22}ClNO_4$								
(IX) $Me[CH_2]_2$		0·90	83 [d]	$C_{22}H_{28}N_4O_{11}$	50·5	5·6	10·6	50·4	5·3	10·7	66—67 [b]		$C_{16}H_{26}ClNO_4$			10·9	4·4			10·7	4·2
(X) $Me[CH_2]_3$	+13·7	0·92	67 [d]	$C_{24}H_{32}N_4O_{11}$	52·0	5·7	10·5	52·2	5·8	10·1	82—83 [b,e]	−13·9	$C_{18}H_{30}ClNO_4$	59·9	8·4	10·1	4·2	60·0	8·35	9·9	3·9
(XI) $Me[CH_2]_4$		0·95	30—32 [d]	$C_{26}H_{36}N_4O_{11}$	54·0	6·3	9·7	53·8	6·2	9·7	113 [b,e]	−8·0	$C_{20}H_{34}ClNO_4$	61·9	8·7	9·5	3·7	61·9	8·8	9·2	3·6
											127 [d]	−16·5	$C_{16}H_{26}ClNO_4$	58·0	7·9	11·1	4·3	57·9	7·85	10·7	4·2
(XII) Me_2CH		0·93	111 [d]	$C_{22}H_{28}N_4O_{11}$	50·4	5·5	10·8	50·4	5·3	10·7											
(XIII) Me_3C	+8·2	0·95	132—133 [d]	$C_{24}H_{32}N_4O_{11}$	52·2	5·9	10·2	52·2	5·8	10·1	142 [e]	−22·6	$C_{18}H_{30}ClNO_4$, H_2O	58·8	8·45	9·9	3·9	58·8	8·7	9·7	3·8
(XIV) $MeCH_2{\cdot}C(Me_2)$	+2·4	0·95	135 [e]	$C_{26}H_{36}N_4O_{11}$	53·9	6·35	9·7	53·8	6·2	9·7	[f]	−21·4									
(XV) $Me[CH_2]_2{\cdot}C(Me_2)$		0·94	102 [c]	$C_{28}H_{40}N_4O_{11}$	55·3	6·6	9·25	55·2	6·6	9·2	[f]	−19·0									
(XVI) $Me_2CH{\cdot}CH_2$	+7·0	0·90	114 [d]	$C_{24}H_{32}N_4O_{11}$	52·5	6·0	10·0	52·2	5·8	10·1	113—114 [e]	−16·6	$C_{18}H_{30}ClNO_4$	59·8	8·2		4·3	60·0	8·35		3·9
(XVII) Me_3CCH_2	+5·6°	0·92	140° [c]	$C_{26}H_{36}N_4O_{11}$	54·0	6·3	9·5	53·8	6·2	9·7	103	−16·0	$C_{20}H_{34}ClNO_4$	61·4	8·7	9·2	3·85	61·9	8·8	9·2	3·6
(XVIII) $Me_2C{=}CH$	+7·5	0·88	115 [d,j]	$C_{24}H_{28}N_4O_{11}$	52·9	5·2	10·3	52·6	5·1	10·2	126 [d,j]	−18·2	$C_{18}H_{26}ClNO_4$	60·5	7·4		4·3	60·7	7·3		3·9
(XIX) $MeCH{=}C(Me)$	+2·15	0·865	117 [d]	$C_{24}H_{28}N_4O_{11}$	52·4	5·2	10·0	52·6	5·1	10·2	[f]	−21·8									
(XX) $CH_2[CH_2]_2CH$	+12·0	0·90	142 [c]	$C_{26}H_{34}N_4O_{11}$	54·4	5·6	9·4	54·0	5·9	9·7	85 [e]	−12·7	$C_{20}H_{32}ClNO_4$			9·7	3·35			9·2	3·6
(XXI) $Me_2CH{\cdot}[CH_2]_4$	+7·35	0·90	69—70 [d]	$C_{30}H_{44}N_4O_{11}$	56·5	7·0	8·6	56·6	6·9	8·8	[f]	−9·5									
(XXII) $Me_2CH{\cdot}OCH_2$		0·79	110 [e]	$C_{24}H_{32}N_4O_{13}$	49·3	5·7	9·6	49·3	5·5	9·6	75 [b]		$C_{18}H_{30}ClNO_6$				3·8				3·6
(XXIII) Ph	−8·4		136 [d,k]	$C_{28}H_{24}N_4O_{11}$	56·7	4·0	9·6	66·8	4·1	9·5	162 [d]	−45	$C_{22}H_{22}ClNO_4$	66·65	5·6	9·1	3·7	66·1	5·5	8·9	3·5

[a] Lit.[4] +15·8°. [b] Deliquescent. [c] Blades. [d] Needles. [e] Leaflets. [f] Gum. [g] Lit.[5,6] 146°. [h] Lit.[4] 114·5—115·5°. [j] Reported in preliminary communication.[3] [k] Lit.[7] 134°.

For comparison, retronecine (I), indicine (XXIV), monocrotaline (XXVII), and diacetylmonocrotaline (XXVIII) had R_F values 0·21, 0·58, 0·50, and 0·84 respectively.

Diesters of retronecine (5,6,7,7aα-tetrahydro-7β-hydroxy-1-hydroxymethyl-3*H*-pyrrolizine) (I) have been

[1] R. Schoental, *Israel J. Med. Sci.*, 1968, **4**, 1133.
[2] A. R. Mattocks, *Nature*, 1968, **217**, 723.
[3] R. Schoental and A. R. Mattocks, *Nature*, 1960, **185**, 842.
[4] Ming-Chien Chiang and Pang Li, *J. Chinese Chem. Soc.*, 1951, **18**, 184 (*Chem. Abs.*, 1952, **46**, 8632).
[5] G. Barger, T. R. Seshadri, H. E. Watt, and T. Yabuta, *J. Chem. Soc.*, 1935, 11.
[6] R. Adams and E. F. Rogers, *J. Amer. Chem. Soc.*, 1939, **61**, 2815.
[7] S. H. Eggers, M.Sc. Thesis, University of Natal, 1961.

values (Table) reflected their high oil : water distribution coefficients compared with most natural alkaloids. Although the hydrochlorides, made from the freshly prepared bases, generally crystallised readily, some were hygroscopic gums which failed to crystallise (Table).

Diacetylretronecine (VII) has been reported to show green fluorescence after distillation under reduced pressure at 125—134°.[4,5] None of our retronecine esters showed this fluorescence. A labile yellow impurity with intense green fluorescence was formed, however, when retrorsine pyrrole or monocrotaline pyrrole [8] was heated with aqueous alkali and it is likely that the fluorescent constituent of distilled diacetylretronecine arises from the effects of oxidation and heat on the ester under basic conditions.

The free hydroxy-groups in the alkaloids monocrotaline (XXVII) and retrorsine (XXIX) were acetylated by conventional methods. Under mild conditions retrorsine formed the monacetyl derivative (XXX), and prolonged treatment gave diacetylretrorsine (XXXI). Tri-isovalerylindicine (XXVI) was prepared in the same way as the previously reported triacetylindicine (XXV).[9] These acyl derivatives were, in general, more hepatotoxic than the parent alkaloids.[1,9]

Retronecine (I) was converted through the monochloro-derivative (II) into retronazide (IV). This was reduced by zinc and acid or by lithium aluminium hydride to retronamine (V), the acetyl amide (VI) of which was prepared by alkaline hydrolysis of the crude *ON*-diacetyl derivative or, better, using acetic acid and dicyclohexylcarbodi-imide, the last being a general method suitable for preparing a series of acylamino-analogues of pyrrolizidine monoesters. Prolonged treatment of retronecine (I) with thionyl chloride gave the dichloro-derivative (III), which was isolated as its picrate.

EXPERIMENTAL

M.p.s are corrected. Extracts were dried with anhydrous sodium sulphate. 'Ether' refers to diethyl ether. I.r. spectra were recorded with Perkin-Elmer 137 or 457 spectrophotometers.

Paper Chromatography.—Descending chromatograms were run on Whatman No. 1 paper, buffered with 0·1M-sodium acetate [10] with butanol–acetic acid–water.[11] The dried papers were sprayed with platinic iodide reagent.[12]

Isopropoxyacetic Acid and Isopropoxyacetyl Chloride.—To a stirred solution of sodium isopropoxide [from sodium (16 g.) and isopropyl alcohol (350 ml.)], heated under reflux, was added ethyl bromoacetate (66 ml.) during 45 min. After being heated for a further 30 min., 30% aqueous sodium hydroxide (500 ml.) was added, and the mixture was heated for a further 4·5 hr. under reflux. Water (600 ml.) was added and 500 ml. of liquor was distilled off. The cooled residue was acidified with 50% sulphuric acid (240 ml.), and extracted with ether (×3). The combined extracts were dried, the solvent removed, and the product fractionated to give isopropoxyacetic acid, b.p. 66°/0·6 mm. The acid was characterised as its *anilide* which formed needles, m.p. 29—30° from light petroleum (b.p. 30—40°) (Found: C, 68·0; H, 7·8; N, 7·3. $C_{11}H_{15}NO_2$ requires C, 68·4; H, 7·8; N, 7·25%). The acid chloride had b.p. 138—140°.

Retronecine Diesters (VII—XXIII).—The general method is illustrated by the preparation of di-n-valeryl retronecine (X). Retronecine hydrochloride (0·5 g.), prepared by hydrolysing monocrotaline,[6] was heated with n-valeryl chloride (2 ml.) on a steam-bath for 2 hr. Excess of acid chloride was decomposed with water, and the acidic solution was washed twice with ether and basified (cooling) to pH 8—9 with disodium hydrogen phosphate. The ester was extracted immediately with ether (×3) and the combined extracts were dried and concentrated to give an oil. This was neutralised with ethanolic picric acid to give *di-n-valerylretronecine picrate* (1·4 g., 95%), m.p. 60—64°. Five recrystallisations from ethanol furnished pale yellow needles, m.p. 67°.

The amino-ester was regenerated (82·5%) by passing the picrate, in chloroform, through Dowex 1 resin (analytical grade, OH form, washed with methanol and chloroform), and concentrating the eluate.

The amino-ester, in ethanol, was neutralised with anhydrous ethanolic hydrogen chloride. The solution was concentrated, ether was added, and after the solution had been cooled below 0° the deliquescent *hydrochloride* was collected by centrifugation. It formed leaflets, m.p. 82—83° from ethanol–ether.

Tri-isovalerylindicine (XXVI).—This was prepared from indicine [13] and isovaleryl chloride by the method described for triacetylindicine.[9] The *picrate* formed yellow blades, m.p. 104° (Found: C, 55·1; H, 6·7; N, 7·4. $C_{30}H_{49}NO_8,C_6H_3N_3O_7$ requires C, 55·3; H, 6·7; N, 7·2%). The base, recovered from the picrate by using anion exchange resin, was a viscous oil, $[\alpha]_D^{20}$ −3·35° (*c* 5·37 in absolute ethanol). The hydrochloride formed a glass, $[\alpha]_D$—18·6° (*c* 5·2 in H_2O).

Diacetylmonocrotaline (XXVIII).—Monocrotaline (2·5 g.), acetyl chloride (16 ml.), and acetic anhydride (4 ml.) were heated under reflux for 7·5 hr. Excess of reagents were removed under reduced pressure, and the residue was worked up to give *diacetylmonocrotaline picrate*, m.p. 275° (decomp.) after two recrystallisations from ethanol (Found: C, 49·0; H, 4·8; N, 8·4. $C_{20}H_{27}NO_8,C_6H_3N_3O_7$ requires C, 48·9; H, 4·7; N, 8·8%). The base, obtained by passing the picrate, in chloroform, through alumina (B.D.H. chromatographic grade), was a gum (77%), $[\alpha]_D^{19}$ 137·5° (*c* 2·46 in chloroform). The *hydrochloride* formed needles (from aqueous acetone), m.p. 178° (decomp.) (Found: C, 52·0; H, 6·4; Cl, 7·6; N, 3·2. $C_{20}H_{27}NO_8,HCl,H_2O$ requires C, 51·7; H, 6·5; Cl, 7·65; N, 3·0%).

Diacetyl Retrorsine (XXXI).—Retrorsine, acetylated in the way described for monocrotaline, gave *diacetylretrorsine hydrochloride* (84%), as prisms (from ethyl acetate), m.p. 123° (Found: C, 54·5; H, 6·9. $C_{22}H_{29}NO_8,HCl,H_2O$ requires C, 54·0; H, 6·5%).

The *base* formed prisms (from ethyl acetate–ether), m.p. 134°, $[\alpha]_D^{20}$ −9·8° (*c* 7·0 in chloroform) (Found: C, 60·9;

[8] A. R. Mattocks, *J. Chem. Soc.* (*C*), 1969, 1155.
[9] A. R. Mattocks, *J. Chem. Soc.* (*C*), 1967, 329.
[10] R. Munier, M. Macheboeuf, and N. Cherrier, *Bull. Soc. chim. biol.*, 1952, **34**, 204.
[11] H. C. Crowley and C. C. J. Culvenor, *Austral. J. Chem.*, 1959, **12**, 694.
[12] A. R. Mattocks, *J. Chem. Soc.*, 1964, 1974.
[13] A. R. Mattocks, R. Schoental, H. C. Crowley, and C. C. J. Culvenor, *J. Chem. Soc.*, 1961, 5400.

H, 6·8; N, 3·35. $C_{22}H_{29}NO_8$ requires C, 60·7; H, 6·7; N, 3·2%). The *picrate* formed needles (from ethanol), m.p. 184° (Found: C, 50·6; H, 4·9. $C_{22}H_{29}NO_8,C_6H_3N_3O_7$ requires C, 50·6; H, 4·8%).

Monoacetyl Retrorsine (XXX).—Retrorsine (3 g.) and acetyl chloride (20 ml.) were heated under reflux for 5 min. The reagent was removed under reduced pressure and the residue was dissolved in water, washed with ether, and basified with ammonia. Extraction with ether provided *acetylretrorsine* (2·4 g., 71%) which formed blades (from benzene–light petroleum), m.p. 84—85° (Found: C, 75·6; H, 7·1; N, 3·0. $C_{20}H_{27}NO_7,C_6H_6$ requires C, 66·1; H, 7·0; N, 3·0%). The *picrate* formed leaflets (from ethanol), m.p. 117° (Found: C, 50·3; H, 5·2; N, 8·5. $C_{20}H_{27}NO_7,C_6H_3N_3O_7$ requires C, 50·15; H, 4·8; N, 9·0%).

5,6,7,7aα-*Tetrahydro*-7β-*chloro*-1-*chloromethyl*-3H-*pyrrolizine* (III).—Retronecine hydrochloride (0·8 g.) and thionyl chloride (5 ml.) were heated under reflux for 4 hr., and then the excess of reagent was distilled off. Residual reagent was removed by addition of benzene which was then removed under reduced pressure; this was repeated several times. The brown crystals were dissolved in water, the solution washed with ether, basified with sodium hydroxide to pH *ca.* 10 and immediately extracted with ether (×4). The combined extracts were neutralised at once with picric acid and the *picrate* (1·4 g., 80%) was recrystallised from acetone–ethanol to give needles, m.p. 161° (Found: C, 40·2; H, 3·55; Cl, 16·9; N, 13·55. $C_8H_{11}Cl_2N,C_6H_3N_3O_7$ requires C, 39·9; H, 3·3; Cl, 16·85; N, 13·3%). The free base, obtained from the picrate by using an anion exchange resin, polymerised rapidly, but if neutralised at once with hydrochloric acid it gave a hydrochloride, m.p. 145—146°.

Retronazide (IV).—Monochlororetronecine hydrochloride [14] (3 g.), and sodium azide (4 g.), in water (20 ml.) were heated at 90—100° for 2 hr. The solution was cooled, basified with sodium hydroxide, and extracted with chloroform (×5). The combined extracts were dried and concentrated to give the *azide* (2·38 g., 85%), which crystallised from ether–light petroleum as blades, m.p. 67° (Found: C, 53·3; H, 6·8; N, 31·1. $C_8H_{12}N_4O$ requires C, 53·3; H, 6·7; N, 31·1%). The i.r. spectrum (KBr) showed hydroxy and azide bands at 3330m and 2075s cm.$^{-1}$ respectively.

5,6,7,7aα-*Tetrahydro*-7β-*hydroxy*-1-*aminomethyl*-3H-*pyrrolizine* (*Retronamine*) (V).—Retronazide (2 g.) was stirred with a suspension of lithium aluminium hydride (1 g.) in ether (80 ml.) at room temp., for 2 hr. Excess of reagent was destroyed with water (20 ml.) and the aqueous layer was washed with ether (×2), basified with sodium hydroxide, and evaporated to dryness. The residue was extracted with chloroform (×4), and the combined extracts were dried and concentrated to give a gum (1·29 g., 75%) which crystallised on trituration with ether. Recrystallisation from ethyl acetate gave *retronamine* as hygroscopic prisms, m.p. 82° (Found: C, 62·1; H, 9·0; N, 17·7. $C_8H_{14}N_2O$ requires C, 62·3; H, 9·1; N, 18·2%).

The azide was also reduced to the amine by zinc and diluted hydrochloric acid during 1 hr. at room temp. *Retronamine dipicrate* formed blades (from ethanol), m.p. 182° (Found: C, 39·5; H, 3·55; N, 17·8. $C_{20}H_{20}N_8O_{15}$ requires C, 39·2; H, 3·3; N, 18·3%).

Retronamine dipicrolonate recrystallised from acetone–ethanol, had m.p. 241° (decomp.) (Found: C, 48·9; H, 5·0; N, 19·7. $C_{28}H_{30}N_{10}O_{11},C_2H_5OH$ requires C, 49·4; H, 4·9; N, 19·2%).

N-*Acetylretronamine* (VI).—(*a*) Solutions of acetic acid (60 mg.) and dicyclohexylcarbodi-imide (206 mg.) in acetonitrile (1 ml. each) were mixed and then immediately added to a solution of retronamine (55 mg.) in acetonitrile (2 ml.). After 2 hr. at room temp., dicyclohexylurea (140 mg.) was collected and the liquor was evaporated to dryness. The residue was shaken with 0·5N-hydrochloric acid (10 ml.) and the solution was filtered, washed with ether (×3), basified with sodium hydroxide, and evaporated to dryness. The residue was extracted with chloroform (×5), and the combined extracts were dried and concentrated to give the *acetyl derivative* (60 mg., 86%). The compound crystallised from ethanol–ethyl acetate as prisms, m.p. 184° (Found: N, 13·9. $C_{10}H_{16}N_2O_2$ requires N, 14·3%). The i.r. spectrum (KBr) included bands at 3275s (NH), 3080w (unsaturated ring), and 1640s; 1585s cm.$^{-1}$ (amide).

(*b*) Retronamine (150 mg.) was heated under reflux with acetyl chloride (7 ml.) for 20 min. Excess of reagent was removed under reduced pressure and the residue was dissolved in water, made strongly alkaline with sodium hydroxide, and washed with chloroform (×3). The combined chloroform extracts were dried and concentrated to give a gum (60 mg., 26%) which contained an *ON*-diacetyl derivative (ester and amide bands in i.r.). The aqueous liquor was concentrated to dryness and the residue was extracted with chloroform to give *N*-acetylretronamine (93 mg., 49%), identical with that described above.

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