Pyrrolizidine Alkaloid Analogues. Synthesis of 11-Membered Macrocyclic Diesters of (±)-Synthanecine A

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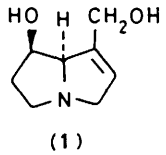
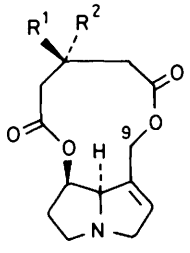
Treatment of (±)-synthanecine A (**4**) with various glutaric anhydride derivatives [(**5a**—**f**) and (**7**)] yielded the corresponding 6-monoesters of (±)-synthanecine A. Lactonisation of these monoesters was carried out by the Corey–Nicolaou method to give a range of 11-membered macrocyclic diesters [(**6a**—**f**), (**8**), and (**9**)] of synthanecine A. The macrocyclic nature of these new pyrrolizidine alkaloid analogues was established by spectroscopic data and by comparison of their ^{1}H n.m.r. and mass spectra with those of macrocyclic pyrrolizidine alkaloids.

Pyrrolizidine alkaloids often occur as macrocyclic dilactones, in which a pyrrolizidine diol (necine), such as (+)-retronecine (**1**), is combined with a diacid to produce 11-, 12-, or 13-membered rings.[1] (14-Membered rings containing retronecine together with three ester linkages are also known.) These alkaloids are widespread and exhibit a range of biological activities, particularly hepatotoxicity. The most important structural features necessary for toxicity are the presence of 1,2-unsaturation in the pyrrolizidine nucleus and esterification at C-7 and C-9 of the necine.[2] The most toxic pyrrolizidine alkaloids combine these features and contain a macrocyclic dilactone system as in dicrotaline (**2**). The toxic action is believed to involve dehydrogenation of the 1,2-unsaturated alkaloids to give the corresponding pyrrole derivatives by hepatic microsomal oxidase enzymes. These pyrrole metabolites act as bifunctional alkylating agents through activation of the two ester groups by conjugation with the pyrrole nitrogen.[2]

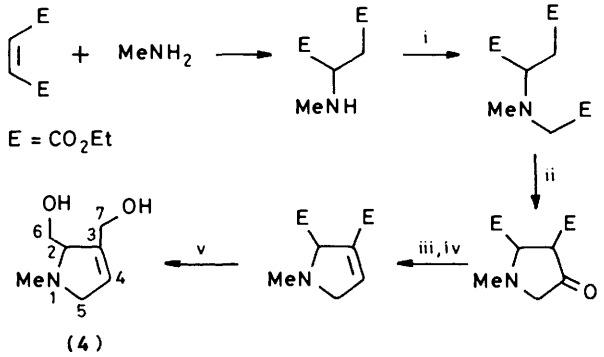


(**1**)



(**2**) R^1 = OH, R^2 = Me
(**3**) R^1 = R^2 = Me

In order to develop our understanding of structure–activity relationships in this area, much effort has been directed towards the synthesis of macrocyclic pyrrolizidine alkaloids and analogues. The initial success in this area was our preparation of a series of 11-membered macrocyclic diesters of (+)-retronecine.[3] One of these pyrrolizidine alkaloid analogues, (3,3-dimethylglutaryl)retronecine (**3**), was shown to produce hepatotoxic effects similar to those of monocrotaline, a common macrocyclic pyrrolizidine alkaloid.[4] The first synthesis of macrocyclic pyrrolizidine alkaloids was only achieved recently, and has been limited to (+)-dicrotaline (**2**),[5] (±)-integerrimine,[6] (±)-fulvine and (±)-crispatine,[7] and the *O*-acetyl derivative of crobarbatine.[8] Recently, a series of 10-membered macrocyclic diesters of (+)-retronecine was prepared.[9] The use of (+)-retronecine as the base component of these analogues is dependent on its extraction from natural sources or its preparation by one of the fairly lengthy synthetic routes reported.[10] The amounts of these analogues available are therefore limited, but synthetic analogues of the most toxic macrocyclic alkaloids are required in reasonable quantities for further metabolic and toxicological studies. A solution to this problem was apparent when Mattocks demonstrated that the saturated ring in the pyrrolizidine nucleus is not involved in the toxic process. He prepared a series of monocyclic analogues of necines, called synthanecines.[11] A monocyclic analogue of retronecine is 2,3-bis(hydroxymethyl)-1-methyl-2,5-dihydropyrrole (**4**) (synthanecine A). Diester derivatives of synthanecine A were shown to produce damage to the livers of animals, similar to that caused by pyrrolizidine alkaloids.[12] We therefore decided to prepare 11-membered macrocyclic diesters of synthanecine A in order to see if increased toxicity is observed with these analogues.



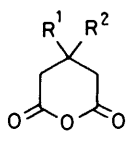
(**4**)

Scheme. *Reagents:* i, $BrCH_2CO_2Et$, K_2CO_3; ii, NaH, toluene; iii, $NaBH_4$, 2% NaOH; iv, 4-$MeC_6H_4SO_2Cl$, pyridine; v, Bu^i_2AlH

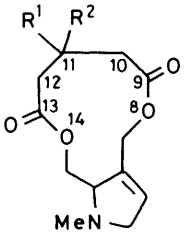
Results and Discussion

(±)-Synthanecine A (**4**) was prepared as outlined in the Scheme, using a combination of the procedures described in two publications by Mattocks.[11] For the diacid component, we first chose to use symmetrically substituted glutaric anhydride derivatives to avoid the problem of the formation of diastereoisomers. Thus, treatment of equimolar amounts of a series of 3,3-disubstituted glutaric anhydride derivatives (**5a**—**d**) with synthanecine A (**4**) in dry 1,2-dimethoxyethane (DME) gave a quantitative yield of the corresponding 6-monoesters of synthanecine A. This regioselective esterification was indicated by a downfield shift of *ca.* 0.4 p.p.m. for the protons at C-6 in the ^{1}H n.m.r. spectrum of the monoesters compared with the signal for synthanecine A. The chemical shifts of the C-7 protons of the monoesters remained unchanged when compared with those in

synthanecine and no evidence could be obtained for the formation of any 7-monoesters. No 6,7-diester formation was observed (t.l.c., ^{1}H n.m.r. and mass spectral data), probably because the 6-monoesters formed initially are zwitterionic and precipitate from the reaction mixture. The extent of this reaction was therefore monitored by the disappearance of synthanecine A (**4**) (t.l.c.) It was generally found that reasonable yields for the sequence of reactions were obtained only when the completeness of each step was monitored carefully by t.l.c. (silica gel; chloroform–methanol–conc. ammonia 85:14:1).



(**5**)



(**6**)

a; $R^1 = R^2 = H$
b; $R^1 = R^2 = Me$
c; $R^1R^2 = [CH_2]_4$
d; $R^1R^2 = [CH_2]_5$
e; $R^1 = Me, R^2 = H$
f; $R^1 = Me, R^2 = OH$

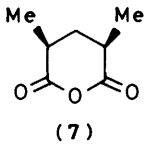
The Corey–Nicolaou double-activation method was selected for the lactonisation step.[13] The pyridine-2-thiol esters were prepared by addition of 2,2′-dithiodipyridine and triphenylphosphine to a suspension of the 6-monoesters of synthanecine A. Vigorous stirring gradually effected dissolution, and the mixture was stirred until formation of the pyridine-2-thiol esters was complete (R_F *ca.* 0.3). The pyridine-2-thiol esters underwent lactonisation when heated at reflux in DME (macrocyclic compounds appeared at R_F 0.5—0.6). Purification of the compounds was achieved by column chromatography on basic alumina. The 11-membered macrocyclic diester (**6a**) was obtained as an oil, and characterised as the picrolonate, while the three other macrocyclic products (**6b—d**) were crystallised (*ca.* 30% overall yield). Correct accurate mass measurements were obtained for the four racemic macrocyclic products (**6a—d**), and similar fragmentation patterns were observed in their mass spectra with main peaks at *m/z* 123, 107, 94, 82, and 80. It is known[14] that the characteristic fragmentation pattern for retronecine diesters is *m/z* 136, 120, 94, 82 and 80. These fragments probably arise by cleavage of the allylic ester and loss of the diacid portion. The two fragmentation patterns are very similar, with the peaks at *m/z* 136 and 120 containing an additional CH in the base fragment compared with the analogous peaks in the mass spectra of the macrocyclic synthanecine dilactones.

In the ^{1}H n.m.r. spectra of the cyclised products (**6a—d**) recorded in deuteriochloroform, the protons assigned to C-7 had shifted downfield by *ca.* 0.45 p.p.m. relative to the signal in synthanecine A, and appeared as an AB quartet. The chemical-shift difference between these protons was small [(0.10—0.19 p.p.m. for (**6b—d**), and *ca.* 0 p.p.m. for (**6a**)]. Values of 0—1.24 p.p.m. for the chemical-shift difference between the protons at C-9 [as in (**2**)] have been observed for 11-membered macrocyclic diesters of retronecine.[3,5] The two protons at C-6 of the macrocyclic compounds (**6a—d**) are also non-equivalent in their ^{1}H n.m.r. spectra, and chemical-shift differences of 0.07—0.18 p.p.m. were observed for these protons. The distinctive mass spectra of compounds (**6a—d**) and the non-equivalence of the protons at C-6 and C-7 in their ^{1}H n.m.r. spectra indicate that 11-membered macrocyclic diesters of synthanecine A have been produced.

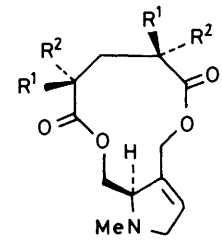
Reaction of 3-methylglutaric anhydride (**5e**) with synthanecine A (**4**) and subsequent lactonisation produced two macrocyclic diastereoisomeric racemates (**6e**) in *ca.* 1:1 ratio. This was deduced from the doubling of some of the signals in the ^{13}C n.m.r. spectrum of the mixture. The diastereoisomers could not be separated by t.l.c. in a variety of solvent systems.

A pyrrolizidine alkaloid analogue with the same diacid portion as dicrotaline (**2**) was also prepared. Treatment of synthanecine A (**4**) with 3-hydroxy-3-methylglutaric anhydride (**5f**) followed by lactonisation of the monoesters again produced two diastereoisomeric racemates (**6f**) in *ca.* 1:1 ratio as judged by their ^{13}C n.m.r. spectra. Separation of these compounds could not be effected by t.l.c.

Finally, efforts were made to prepare macrocyclic dilactones with more steric crowding at the α-positions of the diacid, since steric hindrance around the ester groups is believed to enhance toxicity in the alkaloids by reducing their susceptibility to



(**7**)



(**8**) $R^1 = H, R^2 = Me$
(**9**) $R^1 = Me, R^2 = H$
} One enantiomer is shown

detoxification by hydrolysis.[15] Therefore, 2,4-*meso*-dimethylglutaric anhydride (**7**) was treated with synthanecine A (**4**), and the monoesters were lactonised by means of their pyridine-2-thiol esters to give two diastereoisomeric racemates (**8**) and (**9**). These racemates were partially purified by column chromatography, and complete separation was achieved by p.l.c. Both racemates displayed fragmentation patterns in their mass spectra similar to those shown by the analogues (**6a—f**). An *X*-ray structure determination on the more polar base confirmed its macrocyclic nature and established the relative configuration of the three chiral centres to be as shown in structure (**9**).[16] For the enantiomer drawn (**9**), the ester carbonyl groups are *syn*-parallel and directed above the plane of the macro-ring, whereas most 11-membered macrocyclic diesters of retronecine have ester carbonyl groups that are *syn*-parallel and directed below the plane of the ring.

The ^{1}H n.m.r. spectra of the two racemates (**8**) and (**9**) showed some differences from the analogous (**6a—f**). In particular, higher chemical-shift differences were observed for the protons at C-6 [0.48 for (**8**) and 0.51 p.p.m. for (**9**)] and for those at C-7 [0.20 for (**8**) and 0.40 p.p.m. for (**9**)]. This suggests that the conformations of the macrocyclic rings in compounds (**8**) and (**9**) in deuteriochloroform solution may be different from those of the analogues (**6a—f**), due to the presence of the methyl groups at C-10 and C-12 in (**8**) and (**9**). The conformations of these analogues may be an important factor in determining their relative toxicity. A favourable conformation may allow preferential oxidation of the pyrroline ring to the toxic pyrrole metabolite, rather than detoxification by *N*-oxidation.[4]

The toxicity of the new macrocyclic 11-membered pyrrolizidine alkaloid analogues [(**6a—f**), (**8**), and (**9**)] can now be established.

Experimental

M.p.s were measured with a Kofler hot-stage apparatus. Organic solutions were dried with anhydrous $MgSO_4$, and solvents were evaporated off under reduced pressure below 50 °C. N.m.r. spectra were recorded with a Varian XL-100 spectrometer operating at 100 MHz or with a Bruker WP-200 SY spectrometer operating at 200 MHz. Spectra were recorded for solutions in deuteriochloroform unless otherwise stated, with tetramethylsilane as internal standard. Mass spectra were obtained with A.E.I. MS 12 or 902 spectrometers. T.l.c. of the bases was carried out on Kieselgel G plates of 0.25 mm thickness developed with chloroform–methanol–conc. ammonia (85:14:1), and the bases were located by oxidation with *o*-chloranil, followed by treatment with Ehrlich's reagent.[17] DME was dried by distillation from potassium hydroxide and then from sodium–benzophenone under argon immediately prior to use.

(±)-*Synthanecine A* (**4**).—This was prepared as detailed by Mattocks,[11] and had δ_C (25 MHz) 40.9 (NMe), 59.2 (C-6), 61.0 (C-5 and -7), 73.8 (C-2), 124.2 (C-4), and 141.7 (C-3); *m/z* 143 (M^+), 112, 94, 82, 67, 53, and 42.

General Procedure for Synthesis of Synthanecine A Dilactones (**6a**—**f**), (**8**), *and* (**9**).—The anhydride (**5**) or (**7**) (1 mmol) was added to a solution of (±)-synthanecine A (**4**) (1 mmol) in dry DME (20 ml) under argon. The reaction mixture was stirred at room temperature until all the synthanecine A had reacted to form the zwitterionic monoester (t.l.c., R_F 0.0; 12—18 h). 2,2′-Dithiodipyridine (1.2 mmol) and triphenylphosphine (1.2 mmol) were added and the mixture was stirred vigorously until thiolester formation was complete (t.l.c., R_F *ca.* 0.3; 12—18 h). The clear yellow solution was diluted with DME (20 ml), and the reaction mixture was heated at reflux under argon until lactonisation was complete (t.l.c., R_F 0.5—0.6; *ca.* 6 h). The cooled solution was concentrated under reduced pressure to afford an oil, and the residue was extracted with 1M-aqueous citric acid (3 × 4 ml). The combined acidic extracts were washed with chloroform (6 × 12 ml), then basified with conc. ammonia (pH > 10), and extracted with chloroform (4 × 15 ml). The basic chloroform extracts were dried, filtered, and concentrated to give crude cyclised products as yellow oils. Purification was achieved by column chromatography on basic alumina and elution with increasing proportions of chloroform in dichloromethane. Isolated yields of 20—30% of macrocyclic dilactones (**6a**—**f**) and (**8**) + (**9**) were obtained.

(±)-6,7-0,0-(*Glutaryl*)*synthanecine A* (**6a**) was obtained as an oil, R_F 0.55; ν_{max} (CCl_4) 2 950, 2 850, 2 790, 1 745, 1 455, 1 275, 1 245, and 1 025 cm^{-1}; δ_H (200 MHz) 1.98 (2 H, m, 11-H_2), 2.33 (4 H, m, 10- and 12-H_2), 2.42 (3 H, s, NMe), 3.16 (1 H, m, 5-H), 3.45 (1 H, m, 2-H), 3.77 (1 H, m, 5-H), 4.05 (1 H, dd, J_{gem} 12, J_{vic} 5 Hz, 6-H), 4.12 (1 H, dd, J_{gem} 12, J_{vic} 3 Hz, 6-H), 4.63 (2 H, br s, 7-H_2), and 5.86 (1 H, br s, 4-H); δ_C (25 MHz) 20.7 (C-11), 33.7 and 34.1 (C-10 and -12), 40.8 (NMe), 59.8 and 60.7 (C-5 and C-6), 63.6 (C-7), 71.0 (C-2), 130.0 (C-4), 137.0 (C-3), and 172.3 p.p.m. (C-9 and -13); *m/z* 239 (M^+), 123, 107, 94, 82, 67, 53, and 42 (Found: M^+, 239.1154. $C_{12}H_{17}NO_4$ requires *M*, 239.1158). The *picrolonate* had m.p. 192—194 °C (decomp.) (Found: C, 52.45; H, 4.9; N 13.6. $C_{22}H_{25}N_5O_9$ requires C, 52.48; H, 5.01; N, 13.91%).

(±)-6,7-0,0-(3,3-*Dimethylglutaryl*)*synthanecine A* (**6b**) was obtained as prisms, m.p. 93—94 °C [from benzene–light petroleum (b.p. 60—80 °C)]; R_F 0.6; $\nu_{max.}$ (CCl_4) 2 960, 2 950, 2 880, 2 790, 1 740, 1 330, 1 180, and 1 150 cm^{-1}; δ_H (90 MHz) 1.20 (6 H, s, 15- and 16-H_3), 2.21 and 2.25 (both 2 H, s, together 10- and 12-H_2), 2.48 (3 H, s, NMe), 3.22 (1 H, m, 5-H), 3.59 (1 H, m, 2-H), 3.84 (1 H, m, 5-H), 3.96 (1 H, dd, J_{gem} 11, J_{vic} 7 Hz, 6-H), 4.13 (1 H, dd, J_{gem} 11, J_{vic} 3 Hz, 6-H), 4.58 (1 H, d, J_{gem} 12 Hz, 7-H), 4.74 (1 H, d, J_{gem} 12 Hz, 7-H), and 5.87 (1 H, br s, 4-H); δ_C (25 MHz) 30.1 (C-15 and -16), 34.0 (C-11), 41.4 (NMe), 44.2 and 44.3 (C-10 and -12), 60.9 and 61.1 (C-5 and -6), 65.4 (C-7), 71.3 (C-2), 129.9 (C-4), 137.0 (C-3), and 171.3 and 171.8 p.p.m. (C-9 and -13); *m/z* 267 (M^+), 153, 123, 107, 94, 80, 55, and 41 (Found: M^+, 267.1474; C, 62.9; H, 7.78; N, 5.2. $C_{14}H_{21}NO_4$ requires *M*, 267.1471; C, 62.90; H, 7.92; N, 5.24%). The *picrolonate* had m.p. 207—208 °C (decomp.) (Found: C, 54.2; H, 5.4; N, 13.1. $C_{24}H_{29}N_5O_9$ requires C, 54.23; H, 5.50; N, 13.18%).

(±)-6,7-0,0-(3,3-*Tetramethyleneglutaryl*)*synthanecine A* (**6c**) was obtained as prisms, m.p. 93—96 °C [from benzene–light petroleum (b.p. 60—80 °C)]; R_F 0.6; $\nu_{max.}$ ($CHCl_3$) 2 950, 2 880, 2 790, 1 733, 1 460, 1 328, and 1 160 cm^{-1}; δ_H (360 MHz) 1.66 (8 H, m, 15-, 16-, 17-, and 18-H_2), 2.31 (4 H, m, 10- and 12-H_2), 2.49 (3 H, s, NMe), 3.22 (1 H, m, 5-H), 3.55 (1 H, m, 2-H), 3.86 (1 H, m, 5-H), 4.03 (1 H, dd, J_{gem} 11, J_{vic} 7 Hz, 6-H), 4.10 (1 H, dd, J_{gem} 11 Hz, J_{vic} 3 Hz, 6-H), 4.62 (1 H, br d, J_{gem} 12 Hz, 7-H), 4.72 (1 H, br d, J_{gem} 12 Hz, 7-H), and 5.87 (1 H, br s, 4-H); δ_C (25 MHz) 23.4 (C-16 and -17), 39.3 and 39.5 (C-15 and -18), 41.4 (NMe), 42.7 (C-10 and -12), 44.7 (C-11), 60.9 and 61.1 (C-5 and -6), 65.4 (C-7), 71.4 (C-2), 129.8 (C-4), 137.2 (C-3), and 171.8 and 172.2 p.p.m. (C-9 and -13); *m/z* 293 (M^+), 123, 107, 94, 82, 67, 53, and 42 (Found: M^+, 293.1633; C, 65.55; H, 8.0; N, 4.65. $C_{16}H_{23}NO_4$ requires *M*, 293.1627; C, 65.51, H, 7.90; N, 4.75%). The *picrolonate* had m.p. 217—220 °C (decomp.) (Found: C, 56.1; H, 5.7; N, 12.6. $C_{26}H_{31}N_5O_9$ requires C, 56.01; H, 5.60; N, 12.56%).

(±)-6,7-0,0-(3,3-*Pentamethyleneglutaryl*)*synthanecine A* (**6d**) was obtained as prisms, m.p. 101—102 °C [from benzene–light petroleum (b.p. 60—80 °C)]; R_F 0.6; $\nu_{max.}$ ($CHCl_3$) 2 930, 2 860, 2 790, 1 730, 1 455, 1 330, and 1 165 cm^{-1}; δ_H (90 MHz) 1.55 (10 H, m, 15-, 16-, 17-, 18-, and 19-H_2), 2.28 (2 H, s, 10- or 12-H_2), 2.30 (2 H, s, 12- or 10-H_2), 2.46 (3 H, s, NMe), 3.20 (1 H, m, 5-H), 3.58 (1 H, m, 2-H), 3.85 (1 H, m, 5-H), 3.98 (1 H, dd, J_{gem} 13, J_{vic} 7 Hz, 6-H), 4.11 (1 H, dd, J_{gem} 13, J_{vic} 4 Hz, 6-H), 4.58 (1 H, d, J_{gem} 13 Hz, 7-H), 4.71 (1 H, d, J_{gem} 13 Hz, 7-H), and 5.86 (1 H, br s, 4-H); δ_C (25 MHz) 21.6 (C-16 and -18), 25.8 (C-17), 36.7 (C-11), 37.4 and 37.6 (C-15 and -19), 41.4 (NMe), 41.8 (C-10 and -12), 60.9 and 61.1 (C-5 and -6), 65.5 (C-7), 71.2 (C-2), 129.8 (C-4), 137.0 (C-3), and 171.5 and 171.9 p.p.m. (C-9 and -13); *m/z* 307 (M^+) 167, 123, 108, 107, 94, 81, 67, 53, and 42 (Found: M^+, 307.1795; C, 66.6; H, 8.3; N, 4.6. $C_{17}H_{25}NO_4$ requires *M*, 307.1784; C, 66.42; H, 8.20; N, 4.57%). The *picrolonate* had m.p. 210—214 °C (decomp.) (Found: C, 56.8; H, 5.75; N, 12.0. $C_{27}H_{33}N_5O_9$ requires C, 56.73; H, 5.82; N, 12.25%).

6,7-0,0-(3-*Methylglutaryl*)*synthanecine A* (**6e**) (as a 1:1 mixture of two diastereoisomeric racemates) was obtained as an oil, R_F 0.6; $\nu_{max.}$ ($CHCl_3$) 2 970, 2 950, 2 880, 2 785, 1 740, 1 460, 1 379, 1 300, 1 257, and 1 185 cm^{-1}; δ_H (90 MHz) 0.98 and 1.05 (3 H, d, 15-H_3), 2.25 (5 H, m, 10-H_2, 11-H, and 12-H_2) 3.14 (1 H, m, 5-H), 3.45 (1 H, m, 2-H), 3.80 (1 H, m, 5-H), 4.07 (2 H, m, 6-H_2), 4.62 (2 H, m, 7-H_2), and 5.85 (1 H, br s, 4-H); δ_C (25 MHz) 22.6 (C-15), 28.6 and 29.0 (C-11), 40.8, 41.0, 41.2, 41.8, and 42.5 (C-10, C-12, and NMe), 59.6, 60.0, and 60.8 (C-5 and -6), 63.4 and 63.8 (C-7), 71.3 and 71.5 (C-2), 129.6 and 130.5 (C-4), 137.0 and 137.4 (C-3), and 171.7, 172.0, and 172.5 p.p.m. (C-9 and -13); *m/z* 253 (M^+), 123, 107, 94, 82, 69, 53, and 41 (Found: M^+, 253.1304. $C_{13}H_{19}NO_4$ requires *M*, 253.1314). The picrolonate of the mixture (**6e**) had m.p. 172—174 °C (Found: C, 53.4; H, 5.3; N, 13.4. $C_{23}H_{27}N_5O_9$ requires C, 53.38; H, 5.26; N, 13.54%).

6,7-0,0-(3-*Hydroxy*-3-*methylglutaryl*)*synthanecine A* (**6f**) (as a 1:1 mixture of two diastereoisomeric racemates) was obtained as an oil, R_F 0.5; $\nu_{max.}$ (CCl_4) 3 530, 2 980, 2 950, 2 850, 2 790, 1 740, 1 725, 1 455, 1 375, 1 325, 1 260, and 1 165 cm^{-1}; δ_H (90 MHz) 3.92 (1 H, br s, OH), 1.39 (3 H, s, 15-H_3), 2.47 (3 H, s, NMe), 2.50 (2 H, s, 10- or 12-H_2), 2.56 (2 H, s, 12- or 10-H_2), 3.23 (1 H, m, 5-H), 3.58 (1 H, m, 2-H), 3.85 (1 H, m, 5-H), 4.37 (2 H, m, 6-H_2), 4.55 (1 H, d, J_{gem} 12 Hz, 7-H), 4.72 (1 H, d, J_{gem} 12 Hz, 7-H), and 4.91 (1 H, br s, 4-H); δ_C (25 MHz) 29.5 (C-15), 41.2

(NMe), 45.5, 45.6, and 45.8 (C-10, -11, and -12), 60.7, 60.9, and 61.6 (C-5 and -6), 65.4 and 65.5 (C-7), 70.9 and 71.1 (C-2), 130.6 and 131.2 (C-4), 136.2 and 136.4 (C-3), and 171.1, 171.4, and 171.8 p.p.m. (C-9 and -13); *m/z* 269 (M^+), 107, 94, 82, 60, 51, and 43 (Found: M^+, 269.1261. $C_{13}H_{19}NO_5$ requires *M*, 269.1263).

6,7-0,0-(meso-2,4-*Dimethylglutaryl*)*synthanecine A* was obtained as a mixture of two diastereoisomeric racemates, R_F 0.55 and 0.6. The picrolonate had m.p. 192—196 °C (decomp.) (Found: C, 54.35; H, 5.3; N, 13.2. $C_{24}H_{29}N_5O_9$ requires C, 54.23; H, 5.50; N, 13.14%).

The mixture (55 mg) was partially separated by column chromatography on basic alumina. Pure samples of each diastereoisomeric racemate were obtained by preparative layer chromatography.

(2R,10S,12R)- and (2S,10R,12S)-6,7-0,0-(meso-2,4-*Dimethylglutaryl*)*synthanecine A* (**8**) (11 mg) was obtained as an oil; R_F 0.6; $\nu_{max.}$ (CCl_4) 2 975, 2 940, 2 880, 2 790, 1 745, 1 463, 1 380, and 1 255 cm^{-1}; δ_H (90 MHz) 1.12 (6 H, d, J_{vic} 7 Hz, 15- and 16-H_3), 1.16—2.50 (4 H, m, 10-H, 11-H_2 and 12-H), 2.48 (3 H, s, NMe), 3.24 (1 H, m, 5-H), 3.50 (1 H, m, 2-H), 3.82 (1 H, m, 5-H), 4.00 (1 H, dd, J_{gem} 12, J_{vic} 4 Hz, 6-H), 4.48 (1 H, dd, J_{gem} 12, J_{vic} 2 Hz, 6-H), 4.56 (1 H, d, J_{gem} 12 Hz, 7-H), 4.76 (1 H, d, J_{gem} 12 Hz, 7-H), and 5.94 (1 H, br s, 4-H); δ_C (50 MHz) 18.7 (C-15 and -16), 38.8 (C-10 or -12), 40.0 (C-11), 40.6 (NMe and C-12 or -10), 58.8 and 60.8 (C-5 and C-6), 62.4 (C-7), 71.1 (C-2), 130.7 (C-4), 137.2 (C-3), and 175.3 and 176.8 p.p.m. (C-9 and -13); *m/z* 267 (M^+), 123, 107, 94, 82, 67, 55, and 42 (Found: M^+, 267.1471. $C_{14}H_{21}NO_4$ requires *M*, 267.1470).

(2R,10R,12S)- and (2S,10S,12R)-6,7-0,0-(meso-2,4-*Dimethylglutaryl*)*synthanecine A* (**9**) (16 mg) was obtained as prisms [from benzene–light petroleum (b.p. 60—80 °C)], m.p. 53 °C; R_F 0.55; $\nu_{max.}$(CCl_4) 2 980, 2 940, 2 880, 2 785, 1 750, 1 462, 1 258, 1 180, and 1 155 cm^{-1}; δ_H (90 MHz) 1.13 (3 H, d, J_{vic} 7 Hz, 15- or 16-H_3), 1.14 (3 H, d, J_{vic} 7 Hz, 16- or 15-H_3), 1.17—2.50 (4 H, m, 10-H, 11-H_2, and 12-H), 3.24 (1 H, m, 5-H), 3.52 (1 H, m, 2-H), 3.77 (1 H, m, 5-H), 3.90 (1 H, dd, J_{gem} 13, J_{vic} 6 Hz, 6-H), 4.41 (1 H, dd, J_{gem} 12, J_{vic} 6 Hz, 6-H), 4.53 (1 H, d, J_{gem} 14 Hz, 7-H), 4.93 (1 H, d, J_{gem} 14 Hz, 7-H), and 5.82 (1 H, br s, 4-H); δ_C (25 MHz) 18.9 (C-15 and -16), 39.2 (C-10 or -12), 39.7 (C-11), 40.5 (C-12 or -10), 40.9 (NMe), 60.3 and 60.7 (C-5 and - 6), 63.3 (C-7), 71.8 (C-2), 127.4 (C-4), 138.0 (C-3), and 175.5 and 176.1 (C-9 and -13) p.p.m.; *m/z* 267 (M^+), 181, 123, 107, 94, 82, 56, and 42 (Found: M^+, 267.1470. $C_{14}H_{21}NO_4$ requires *M*, 267.1470).

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References

1 D. J. Robins, *Fortschr. Chem. Org. Naturst.*, 1982, **41**, 115; 'The Alkaloids,' Specialist Periodical Reports, The Royal Society of Chemistry, London, 1978—1983, vols. 8—13; *Nat. Prod. Rep.*, 1984, **1**, 235.
2 A. R. Mattocks, 'Phytochemical Ecology,' ed. J. B. Harborne, Academic Press, London and New York, 1972, p. 179.
3 D. J. Robins and S. Sakdarat, *J. Chem. Soc., Chem. Commun.*, 1980, 282; J. A. Devlin, D. J. Robins, and S. Sakdarat, *J. Chem. Soc., Perkin Trans. 1*, 1982, 1117.
4 A. R. Mattocks, *Chem.-Biol. Interact.*, 1981, **35**, 301.
5 J. A. Devlin and D. J. Robins, *J. Chem. Soc., Chem. Commun.*, 1981, 1272; K. Brown, J. A. Devlin, and D. J. Robins, *J. Chem. Soc., Perkin Trans. 1*, 1983, 1819.
6 K. Narasaka, T. Sakakura, T. Uchimaru, and D. Guedin-Vuong, *J. Am. Chem. Soc.*, 1984, **106**, 2954.
7 E. Vedejs and S. D. Larsen, *J. Am. Chem. Soc.*, 1984, **106**, 3030.
8 J. Huang and J. Meinwald, *J. Am. Chem. Soc.*, 1981, **103**, 861.
9 M. Burton and D. J. Robins, *J. Chem. Soc., Perkin Trans. 1*, 1985, 611.
10 T. A. Geissman and A. C. Waiss, *J. Org. Chem.*, 1962, **27**, 139; J. J. Tufariello and G. E. Lee, *J. Am. Chem. Soc.*, 1980, **102**, 373; G. E. Keck and D. G. Nickell, *ibid.*, p. 3632; E. Vedejs and G. R. Martinez, *ibid.*, 7993; T. Ohsawa, M. Ihara, K. Fukumoto, and T. Kametani, *Heterocycles*, 1982, **19**, 2075; H. Niwa, A. Kuroda, and K. Yamada, *Chem.* Lett., 1983, 125; H. Rüeger and M. Benn, *Heterocycles*, 1983, **20**, 1331; J. G. Buchanan, G. Singh, and R. H. Wightman, *J. Chem. Soc., Chem. Commun.*, 1984, 1299.
11 A. R. Mattocks, *J. Chem. Soc., Perkin Trans. 1*, 1974, 707; 1978, 896.
12 H. E. Driver and A. R. Mattocks, *Chem.-Biol. Interact.*, 1984, **51**, 201, and refs. cited therein.
13 E. J. Corey and K. C. Nicolaou, *J. Am. Chem. Soc.*, 1974, **96**, 5614.
14 L. B. Bull, C. C. J. Culvenor, and A. T. Dick, 'The Pyrrolizidine Alkaloids,' North-Holland, Amsterdam, 1968.
15 A. R. Mattocks, *Toxicol. Lett.*, 1982, **14**, 111.
16 G. A. Sim, unpublished results.
17 H. J. Huizing, F. DeBoer, and T. M. Malingré, *J. Chromatogr.*, 1980, **195**, 407; R. J. Molyneux and J. N. Roitman, *ibid.*, p. 142.

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