# The Stereochemistry of the β-Hydroxyleucine Unit of Frangulanine

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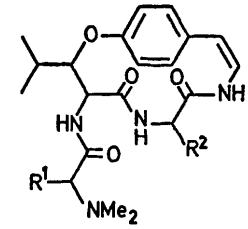
*Summary* $^1$H n.m.r. spectral analysis shows that the β-hydroxyleucine portion of the peptide alkaloids frangulanine and discarine A and B possess an *erythro*-configuration; stereochemical analysis of frangulanine by enzymic oxidation of its product of reduction and subsequent hydrolysis reveals the subunit to be of the L-*erythro*-β-hydroxyleucine-type.

NUMEROUS peptide alkaloids contain a 14-membered heterocycle[1,2] which has been shown in most cases to be composed of an α-amino-acid, β-amino-*p*-hydroxystyrene, and β-alkyl- or β-aryl-serine. The last-named have been considered to possess a *threo*-configuration since *threo*-α-amino-β-hydroxy-acids were isolated from the acid hydrolysates of some of the alkaloids.[2] Since, however, the serine unit exists as an α-amino-β-aryloxyamide moiety in the alkaloids and since acid hydrolysis of the β-aryloxy-function can occur only by solvolytic β-CO bond cleavage or by a β-elimination/β-addition path, the isolation of a *threo*-acid may be of no relevance to the stereochemistry of the β-substituted serine portion of the alkaloids.[3] As a consequence the stereochemistry of this all-important subunit was reinvestigated and frangulanine (**1**) and discarine A (**2**) and B (**3**), the major alkaloids of *Discaria longispina*,[4] utilized for this purpose.




(1) $R^1$ = Bu$^s$, $R^2$ = Bu$^t$
(2) $R^1$ = β-indolylmethyl, $R^2$ = Bu$^s$
(3) $R^1$ = Bu$^s$, $R^2$ = β-indolylmethyl

The 220 MHz $^1$H n.m.r. spectra of $(CD_3)_2SO$ solutions of the three bases (frangulanine at 80°) exhibit α- and β-methine signals of the common β-oxyleucine unit at 4·40 ± 0·02 and 4·77 ± 0·04 p.p.m.; respectively. Further-

more, the α-methine signal appears in form of a doublet of $J = 8$ Hz and that of the β-methine as a doublet of doublets of $J = 8$ and 2 Hz in $(CD_3)_2SO$ solutions of the alkaloids whose amido-hydrogens have been replaced by deuteriums. Since coupling of 8 Hz reflects an α-H/β-H dihedral bond relationship[5] of 0—20° or 150—180° and the fairly rigid 14-membered ring of (**1**)—(**3**) permits only the latter geometry, an *anti*, *i.e. erythro*-configuration can be assigned to the β-oxyleucine portion of the alkaloids. Similarly, the 7·5 and 8 Hz α-H/β-H coupling reported for the β-oxyphenylalanine unit of debenzoylaralionine A[6] and canthiumine,[7] respectively, favours an *erythro*-form for this part of their 14-membered rings. Thus, on the basis of $^1$N n.m.r. data, the natural bases aralionine A,[6] canthiumine,[7] discarine A[4] and B,[4] frangulanine,[4,8] and lasiodine B[3] have an *erythro*-β-substituted serine moiety in common.

The stereochemistry of the β-oxyleucine unit of frangulanine (**1**) was investigated also by chemical means. For this purpose authentic *threo*- and *erythro*-β-hydroxyleucine were prepared[9] and their differentiability determined on an amino-acid analyzer and by gas chromatography of the methyl esters of their *N*-trifluoroacetyl derivatives. These analytical methods were utilized to show that hydrolysis of dihydrofrangulanine in 6N-hydrochloric acid at 120° for 12 h yielded β-hydroxyleucine of only *threo*-configuration, in accordance with the findings of Tschesche.[8] However reduction of dihydrofrangulanine by lithium in liquid methylamine and acid hydrolysis of the product yielded solely *erythro*-β-hydroxyleucine. Since the reduction transforms the aromatic nucleus into an enol ether and since hydrolysis of the latter liberates the hydroxy-group of the β-oxyleucine unit without affecting the chirality of the carbon centre to which it is attached, frangulanine (**1**) contains an *erythro*-β-aryloxyleucine moiety. Finally, the latter was shown to possess the L-configuration as follows. A part of the hydrolysate of the reduction product of dihydrofrangulanine was subjected to the action of hog kidney D-amino-acid oxidase, another to rattlesnake venom L-amino-acid oxidase, and a third left untreated. Automatic amino-acid analysis of the three samples showed *erythro*-β-hydroxyleucine to be absent from the solution treated with the L-amino-acid oxidase, but present in the other two samples. Since *threo*-β-hydroxyleucine, as threonine,[10] is inert to the two oxidases, the configuration of the Tschesche hydrolysis product[8] remained undetermined

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