In conclusion, we have described a new class of compounds based on quinoline-2-carboxylic acid bearing an acetic acid side chain linked to the nucleus through a heteroatom at C-4. These compounds, especially **5**, have been shown to be potent and selective antagonists acting at the glycine binding site on the NMDA receptor complex. The potential utility of **5** for the treatment of neurological disorders characterized by excessive receptor activation (e.g., epilepsy, stroke) is currently under active investigation.

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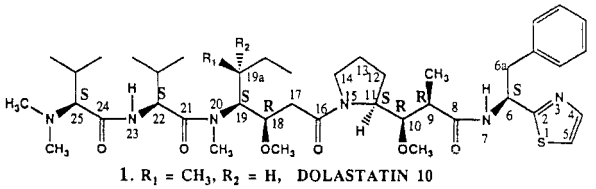
Chiral Modifications of Dolastatin 10: The Potent Cytostatic Peptide (19a*R*)-Isodolastatin 10[1]

When opisthobranch mollusca (sea hares) are threatened, they secrete complex mixtures of defensive substances from the genital pore, purple gland, and body surface. The latter secretion is believed to be the most potent.[2] Some estimation of consequences from higher than therapeutic levels can be obtained from the applications of Locusta,[3] who murdered[4] Caesar Agustus and Claudius Britonnicus, among others, with potions from a *Dolabella* sp. believed to be *auricularia*.[5] Our 17-year investigation of the potent antineoplastic constituents of this sea hare culminated in the isolation and total synthesis of the powerful dolastatin 10 (**1**).[6] In order to probe the chiral and other structural parameters for such remarkable antineoplastic activity, we have first explored the synthesis and biological effects of 18 (of 128 possible) chiral isomers of dolastatin 10 when one to five asymmetric carbons in the Dil-Dap sequence were reversed. We now report that isodolastatins **3-19** (Table I) are less cytostatic than the parent pentapeptide (**1**, P388 ED_{50} 10^{-4} μg/mL) against cell growth of the P388 lymphocytic leukemia (PS system) with ED_{50} values near the 10^{-3} μg/mL level and that (19a*R*)-isodolastatin 10 (**2**) was consistently found to be up to 10-fold more cell growth inhibitory, affording an ED_{50} of 4.9×10^{-5} μg/mL.



1. $R_1 = CH_3$, $R_2 = H$, DOLASTATIN 10
2. $R_1 = H$, $R_2 = CH_3$, 19aR-ISODOLASTATIN 10

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Table I. Dolastatin 10 Chiral Isomers

no.	isodolastatin 10	mp,[a] °C	$[\alpha]^{28}_D$, deg (*c*, $CHCl_3$)	P388 ED_{50}, μg/mL
2	19a*R*	69-71	-57 (0.54)	4.9×10^{-5}
3	9*S*	73-75	-34 (0.85)	1.6×10^{-3}
4	10*S*	72-74	-49 (0.33)	3.9×10^{-3}
5	18*S*,19*R*	82-85	-47 (0.68)	3.2×10^{-3}
6	9*S*,19a*R*	85-89	-82 (0.5)	5.6×10^{-3}
7	18*S*,19*R*,19a*R*	78-80	-44 (0.34)	3.2×10^{-3}
8	9*S*,10*S*	75-77	-45 (0.40)	3.9×10^{-2}
9	18*S*	112-14	-97 (1.3)	6.0×10^{-1}
10	9*S*,18*S*	87-88	-115 (0.65)	6.2×10^{-1}
11	9*S*,18*S*,19*R*	82-84	-77 (0.5)	2.7×10^{-1}
12	9*S*,18*S*,19a*R*	88-91	-131 (0.26)	4.1×10^{-1}
13	9*S*,18*S*,19*R*,19a*R*	78-80	-93 (0.27)	1.5×10^{-1}
14	10*S*,18*S*,19*R*,19a*R*	65-69	-34 (0.18)	2.0×10^{-1}
15	9*S*,10*S*,18*S*,19*R*,19a*R*	76-79	-55 (0.48)	6.3×10^{-1}
16	9*S*,19*R*	108-12	-29 (0.34)	$>1.0 \times 10^{0}$
17	9*S*,19*R*,19a*R*	90-93	-16 (0.31)	$>1.0 \times 10^{0}$
18	9*S*,10*S*,18*S*	82-85	-55 (0.3)	$>1.0 \times 10^{0}$
19	9*S*,10*S*,19*R*,19a*R*	70-74	+19 (0.16)	$>1.0 \times 10^{0}$

[a] All recrystallized from acetone-hexane.

Elucidation of the absolute configurations of dolastatin 10 (**1**) and its total synthesis[6] allowed us to begin evaluating subtle chiral changes that were expected to profoundly affect the conformation and thereby certain biological activities. That expectation was amply realized in the case of (19a*R*)-isodolastatin 10 (**2**). The total synthesis of (19a*R*)-isodolastatin 10 followed the general reaction sequence (Table II)[7] employed for obtaining dolastatin 10.[6] Physical and PS cytostatic properties of the isodolastatins (**2-19**) are given in Table I. Detailed syntheses of the isodolastatins, their synthetic precursors and minimum-energy conformations will be summarized in a future detailed report.

Presently PS cell line results indicate that epimerization at 19a (allo-iso-ile series) strongly increases the cytostatic effects and can compensate, in part, for more profound inversions of configuration elsewhere (cf. **6** and **7**). Inversion at C-19 does not markedly reduce the cytostatic effects (see **5** and **7**) while at C-18 alone and in general for C-9, inhibition of cell growth is greatly moderated to ED_{50} $10^{-1\text{ to }0}$ values. Furthermore, multiple inversions, as expected, greatly diminished potency. For example, (9*S*)-, (10*S*)-, (18*S*)-, (19*R*)-, (19a*R*)-isodolastatin 10 gave PS ED_{50} 6.3×10^{-1} μg/mL. Any change in the stereochemistry of one or more stereocenters at C-9, C-10, C-18, and C-19 reduced or eliminated the cytostatic activity in the series of isodolastatins (see **11-19**). More significantly, the diastereomeric 18*S*,19*R* pair (refer to **1-8, 11, 13, 14**) was found to have better cytostatic activity than the 18*S*,19*S* pair (e.g., **16-19**). A 10-fold reduction of cytostatic activity was observed by inversion of C-9 or C-10 (**3, 4**). Inversion of C-9 and C-10 in the same molecule diminished the activity 100-fold (**18, 19**). A simple inversion of the chiral center at C-18 (**9**) reduced the in vitro activity by 1000-fold. Similar effects were observed by changing the chirality at C-9 and C-18 (**10**) or multiple inversions at C-9, C-10, C-18, and C-19a in any combination (**11-15**). Inversion of C-9 or C-10 with the C-18*S**,C-19*R** combination generally produced isodolastatins with relatively better cytostatic activity than using the diastereomeric pair 18*S*,19*S*. Comparison of the two sets (19a*S* and 19a*R*) of isodolastatins containing 18*S*,19*R* diastereomeric pairs revealed that the 18*S*,19*R* pair with 19a*S* (**5**) was 10 times less effective in inhibiting cell growth than the 18*R*,19*S* of dolastatin 10 (**1**). In the 18*S*,19*R* combination with 19a*R*

(7) All the isodolastatins and reported intermediates gave satisfactory elemental analyses or HREIMS (or HRFABMS) and 400-MHz NMR spectra.

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Table II. Chiral Modification of the Dolastatin 10 Amino Acid Dil Carbon 19a

no.	19a*R*-Dil sequence product	recryst solvent	mp, °C	$[\alpha]^{28}_D$, deg (*c*, $CHCl_3$)	yield, %
20[a]	*N*-Z-(*S*,*R*)-Ile		oil	−12 (4.9)	84
21[b]	*N*-Z-*N*-Me-(*S*,*R*)-Ile	ether	85–87	−64 (4.9)	92
22[c]	*N*-Z-*N*-Me-(*S*,*R*)-isoleucinol		oil	−1 (5.3)	93
23[d]	*N*-Z-*N*-Me-(*S*,*R*)-isoleucinal		oil	−63 (2.4)	80
24[e]	*tert*-butyl 3-hydroxy-4(*S*)-(*N*-Z-*N*-Me)-5(*R*)-methylheptanoate		oil	−37 (3.7)	76 (*R* and *S* isomers)
25[f]	*N*-Z-(3*R*,4*S*,5*R*)-dolaisoleucine-OBu^t		oil	−15 (2.8)	91
26[g]	(5*R*)-dolaisoleucine-OBu^t·HCl	ether	178–80	−2 (5.5)	79
27[h]	*N*-Z-*S*-Val-(3*S*,4*S*,5*R*)-Dil-OBu^t		glass	−27 (3.7)	69
28[i]	*S*-Dov-*S*-Val-(3*S*,4*S*,5*R*)-Dil-OBu^t		glass	−55 (2.2)	72
29[j,k]	*N*-Boc-(2*S*,2′*R*,3′*R*)-Dap-(*S*)-Doe	acetone–hexane	120–21	−68 (1.5)	69[l]

[a] *S*,*R*-Ile, benzyl chloroformate, 1.5 N NaOH. [b] NaH, CH_3I, THF. [c] BH_3·THF, THF. [d] SO_3–pyridine, DMSO, TEA. [e] *tert*-Butyl acetate, LDA, −78 °C. [f] $(CH_3O)_3OBF_4$, Proton Sponge, CH_2Cl_2. [g] Pd/C, H_2, Et_2O–HCl. [h] N-*Z*-(*S*)-Val, $(CH_3)_3CCOCl$, NMM, $CHCl_3$. [i] (*S*)-Dov-Opfp, H_2, Pd/C, dioxane. [j] TFA, CH_2Cl_2. [k] **28**·TFA + **29**·TFA, DEPC, TEA, DME, 2 (90%). [l] Coupling of *N*-Boc-(2*S*,2′*R*,3′*R*)-Dap, Doe·TFA, DEPC, TEA, DME.

(cf. **7**) this substance was 100-fold less active compared to the 18*R*,19*S* (cf. **2**).

For isodolastatins so far studied, modifications in the C-18 and C-19 (followed by C-10) stereochemistry results in profound effects on cytostatic activity. Conversion from the *S*-amino acid series to the *R* series at one to five chiral centers and substitution of amino acid units in the natural dolastatin 10 sequence are currently under study.

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Articles

Flexible *N*-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine Analogues: Synthesis and Monoamine Oxidase Catalyzed Bioactivation

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Eighteen analogues of *N*-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) were synthesized and evaluated as substrates of monoamine oxidase. In general, the flexible analogues, characterized by the presence of a methylene (or ethylene) bridge between the aryl/heteroaryl and tetrahydropyridyl moieties, were better substrates of the enzyme than the conformationally restricted MPTP. It is suggested that the increased oxidative activity of these flexible analogues reflects enhanced binding due to the ability of the C-4-aryl/heteroaryl substituent to gain access to a hydrophobic pocket within the substrate binding site.

Introduction

N-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine, MPTP, is oxidized in vivo by monoamine oxidase (MAO; EC 1.4.3.4) to yield the cationic metabolite *N*-methyl-4-phenylpyridinium, MPP^+.[1-3] The latter selectively accumulates within catecholaminergic neurons via the catecholamine reuptake system[4-6] and subsequently induces a Parkinsonian syndrome in humans and nonhuman primates.[7-9] The discovery that MAO can convert relatively nontoxic molecules into potent neurotoxins has generated interest in the role of this enzyme in the bioactivation of potential neurotoxins present in the environment.

During the last few years, several MPTP analogues have been synthesized and tested for oxidative activity in MAO

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