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MARINE BIOTECHNOLOGY

Marine Pharmacology in 2000: Marine Compounds with Antibacterial, Anticoagulant, Antifungal, Anti-inflammatory, Antimalarial, Antiplatelet, Antituberculosis, and Antiviral Activities; Affecting the Cardiovascular, Immune, and Nervous Systems and Other Miscellaneous Mechanisms of Action

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Abstract: During 2000 research on the pharmacology of marine chemicals involved investigators from Australia, Brazil, Canada, Egypt, France, Germany, India, Indonesia, Israel, Italy, Japan, the Netherlands, New Zealand, Phillipines, Singapore, Slovenia, South Korea, Spain, Sweden, Switzerland, United Kingdom, and the United States. This current review, a sequel to the authors' 1998 and 1999 reviews, classifies 68 peer-reviewed articles on the basis of the reported preclinical pharmacologic properties of marine chemicals derived from a diverse group of marine animals, algae, fungi, and bacteria. Antibacterial, anticoagulant, antifungal, antimalarial, antiplatelet, antituberculosis, or antiviral activity was reported for 35 marine chemicals. An additional 20 marine compounds were shown to have significant effects on the cardiovascular and nervous system, and to possess anti-inflammatory or immunosuppressant properties. Finally, 23 marine compounds were reported to act on a variety of molecular targets and thus could potentially contribute to several pharmacologic classes. Thus, as in 1998 and 1999, during 2000 pharmacologic research with marine chemicals continued to contribute potentially novel chemical leads to the ongoing global search for therapeutic agents in the treatment of multiple disease categories.

Keywords: marine pharmacology, 2000, toxicology, review, secondary metabolites.

INTRODUCTION

The purpose of this article is to review the 2000 primary literature on pharmacologic and toxicologic studies of marine natural products using a format similar to the one used in our previous 1998 and 1999 reviews of the marine pharmacology peer-reviewed literature (Mayer and Leh-

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mann, 2000; Mayer and Hamann, 2002). The 2000 review of reports on marine-derived compounds with antitumor and cytotoxic activity has been published elsewhere (Mayer and Gustafson, 2003). Consistent with our 1998 and 1999 reviews, the present review includes only those articles reporting on the bioactivity or pharmacology of marine chemicals whose structures have been established. We have used Schmitz's chemical classification (Schmitz et al., 1993) to assign each marine compound to a major chemical class: namely, polyketides, terpenes, nitrogen-containing compounds, or polysaccharides. Publications reporting on antibacterial, anticoagulant, antifungal, antimalarial, antiplatelet, antituberculosis, or antiviral properties of marine chemicals have been tabulated (Table 1) and corresponding structures are shown in Figure 1. The articles reporting on marine compounds affecting the cardiovascular and nervous system, as well as those with anti-inflammatory and immunosuppressant effects, are grouped in Table 2, and the structures are presented in Figure 2. Finally, marine compounds targeting a number of distinct cellular and molecular targets and mechanisms are shown in Table 3, and their structures are depicted in Figure 3. Publications on the biological or pharmacological activity of marine extracts or as yet structurally uncharacterized marine compounds are not included in the present review, though several reports were published during 2000 (Khudyakova et al., 2000; Matsubara et al., 2000).

Marine Compounds with Antibacterial, Anticoagulant, Antifungal, Antimalarial, Antiplatelet, Antiituberculosis, and Antiviral Activities

Table 1 summarizes the main findings of 17 papers that reported on the preclinical antibacterial, anticoagulant, antifungal, antimalarial, antiplatelet, antituberculosis, and antiviral pharmacology of the 35 marine natural products shown in Figure 1.

Antibacterial Compounds

During 2000 only 2 studies contributed to the antibacterial pharmacology of marine natural products. A novel C_{14} *acetylenic acid* with antibacterial activity was isolated from the marine sponge *Oceanapia* sp. (Matsunaga et al., 2000). This fatty acid is the first reported midchain acetylenic acid without a bromine atom that inhibited growth of the Gram-negative bacteria *Escherichia coli* and *Pseudomonas aeruginosa* and the Gram-positive bacteria *Bacillus subtillis* and *Staphylococcus aureus. Discorhabdin R*, a novel antibacterial pyrroloiminoquinone, was isolated from the southern Australian sponges *Negombata* sp. and an Antarctic *Latrunculia* sp. (Ford and Capon, 2000). Although discorhabdin R appeared to have been partially responsible for the antibacterial activity against both Gram-positive bacteria (*Staphylococcus aureus* and *Micrococcus luteus*) and Gram-negative bacteria (*Serratia marcescens* and *Escherichia coli*), the pertinent data were not reported. Furthermore, the authors reported neither a comparison with established clinically used antibiotics nor an investigation on the mechanism of action of either marine product.

Anticoagulant Compounds

Three papers were published during 2000 on the anticoagulant properties of marine polysaccharides. An investigation on the *in vivo* anticoagulant pharmacology of the sulfated polysaccharide *fucoidan* was completed during 2000 by Thorlacius et al. (2000). These researchers determined the effect of fucoidan on the function of P- and L-selectin, two members of the selectin family of adhesion molecules. The fact that fucoidan inhibited thrombus formation in arterioles and venules *in vivo* with no effect on P- and L-selectin function suggested that the anticoagulant effect of fucoidan was mainly responsible for its powerful antithrombotic property *in vivo*.

Matsubara et al. reported a novel *sulfated proteoglycan* isolated from the marine green alga *Codium pugniformis* collected in Japan, which helped extend current knowledge on the anticoagulants isolated from the genus *Codium* (Matsubara, 2000). Although the novel proteoglycan inhibited both the intrinsic and common pathways of coagulation, it showed a weaker anticoagulant activity than heparin and involved a mechanism of direct inhibition of thrombin as well as the potentiation of antithrombin III.

Farias et al. (2000) characterized a unique *sulfated* D-*galactan* from the red algae *Botryocladia occidentalis.* The algal sulfated D-galactan had potent anticoagulant activity, similar to that of unfractionated heparin, owing to enhanced inhibition of thrombin and factor Xa by antithrombin or heparin cofactor II. The presence of 2,3-di-*O*-sulfated galactose residues probably resulted in an ''amplifying effect'' of the anticoagulant activity of the algal sulfated galactans when compared to sulfated

Table 1. Marine Compounds with Antibacterial, Anticoagulant, Antifungal, Antimalarial, Antiplatelet, Antituberculosis, and Antiviral Activities

Drug class	Compound/organism[a]	Chemistry	Pharmacologic activity	MMOA[b]	Country[c]	References
Antibacterial	Acetylenic acid/sponge	Fatty acid[d]	Gram-positive and negative inhibition	Undetermined	JPN, NETH	Matsunaga et al., 2000
Antibacterial	Discorhabdin R/sponge	Alkaloid[f]	Gram-positive and negative inhibition	Undetermined	AUS	Ford and Capon et al. 2000
Anticoagulant	Fucoidan/alga	Sulfated Polysaccharide[g]	Inhibition of microvascular thrombus	No effect on P- and L-selectin	SWE,GER	Thorlacius et al., 2000
Anticoagulant	Proteoglycan/alga	Polysaccharide[g]	Anticoagulant	Inhibition of thrombin and potentiation of antithrombin III	JPN	Matsubara, 2000
Anticoagulant	Sulfated D-galactan/alga	Sulfated Polysaccharide[g]	Anticoagulant	Inhibition of thrombin and factor Xa	BRA	Farias et al., 2000
Antifungal	Lyngbyabellin B/bacterium	Depsipeptide[f]	*C. albicans inhibition*	Undetermined	USA	Milligan et al., 2000
Antifungal	Naamine D/sponge	Alkaloid[f]	*C. neoformans* inhibition	Nitric oxide inhibition	USA, NZ	Dunbar et al., 2000
Antimalarial	Ascosalipyrrolidin one A/fungus	Polyketide[d]	*P. falciparum* inhibition	$p56^{lck}$ tyrosine kinase inhibition	GER, SWI	Osterhage et al., 2000
Antimalarial	Homofascaplysin A & fascaplysin/sponge	Sesterterpene[e]	*P. falciparum* inhibition	Undetermined	GER, SWZ	Kirsch et al., 2000
Antimalarial	Manzamine A/sponge	Alkaloid[f]	In vivo *P. berghei* inhibition	Undetermined	SING, JPN, USA	Ang et al., 2000
Antiplatelet	Eryloside F/sponge	Sterol glycoside[e]	Platelet aggregation inhibition	Thrombin receptor antagonist	USA, UK	Stead et al., 2000
Antituberculosis	Axisonitrile-3/sponge	Sesquiterpene[e]	*M. tuberculosis* inhibition	Undetermined	GER	Konig et al., 2000
Antituberculosis	Elisapterosin B/soft coral	Diterpene[e]	*M. tuberculosis* inhibition	Undetermined	USA	Rodriguez et al., 2000
Antiviral	Cyclodidemniserinol trisulfate/ascidian	Polyketide[d]	In vitro HIV infection inhibition	HIV-1 integrase inhibition	USA	Mitchell et al., 2000
Antiviral	Dragmacidin F/sponge	Alkaloid[f]	In vitro HSV-1 and HIV-1 inhibition	Undetermined	ITA	Cutignano et al., 2000
Antiviral	Lobohedleolide, 17-dimethylamino/soft coral	Diterpene[e]	In vitro HIV infection inhibition	Undetermined	USA	Rashid et al., 2000
Antiviral	Mololipids/sponge	Alkaloid[f]	In vitro HIV-1 infection inhibition	Undetermined	USA	Ross et al., 2000

[a] *Kingdom Animalia*: brittle star and cucumber (phylum Echinodermata), clam and mussel (phylum Mollusca), sponge (phylum Porifera), tunicate (phylum Chordata). *Kingdom Fungi*: fungus; *Kingdom Plantae*: alga; *Kingdom Monera*: bacterium (phylum Cyanobacteria).

[b] MMOA is molecular mechanism of action.

[c] AUS is Australia; BRA, Brazil; GER, Germany; ITA, Italy; JPN, Japan; NETH, The Netherlands; NZ, NewZealand; SING, Singapore; SWE, Sweden; SWZ, Switzerland; UK, United Kingdom.

[d] Polyketides; [e] Terpene; [f] Nitrogen-containing compound; [g] Polysaccharide.

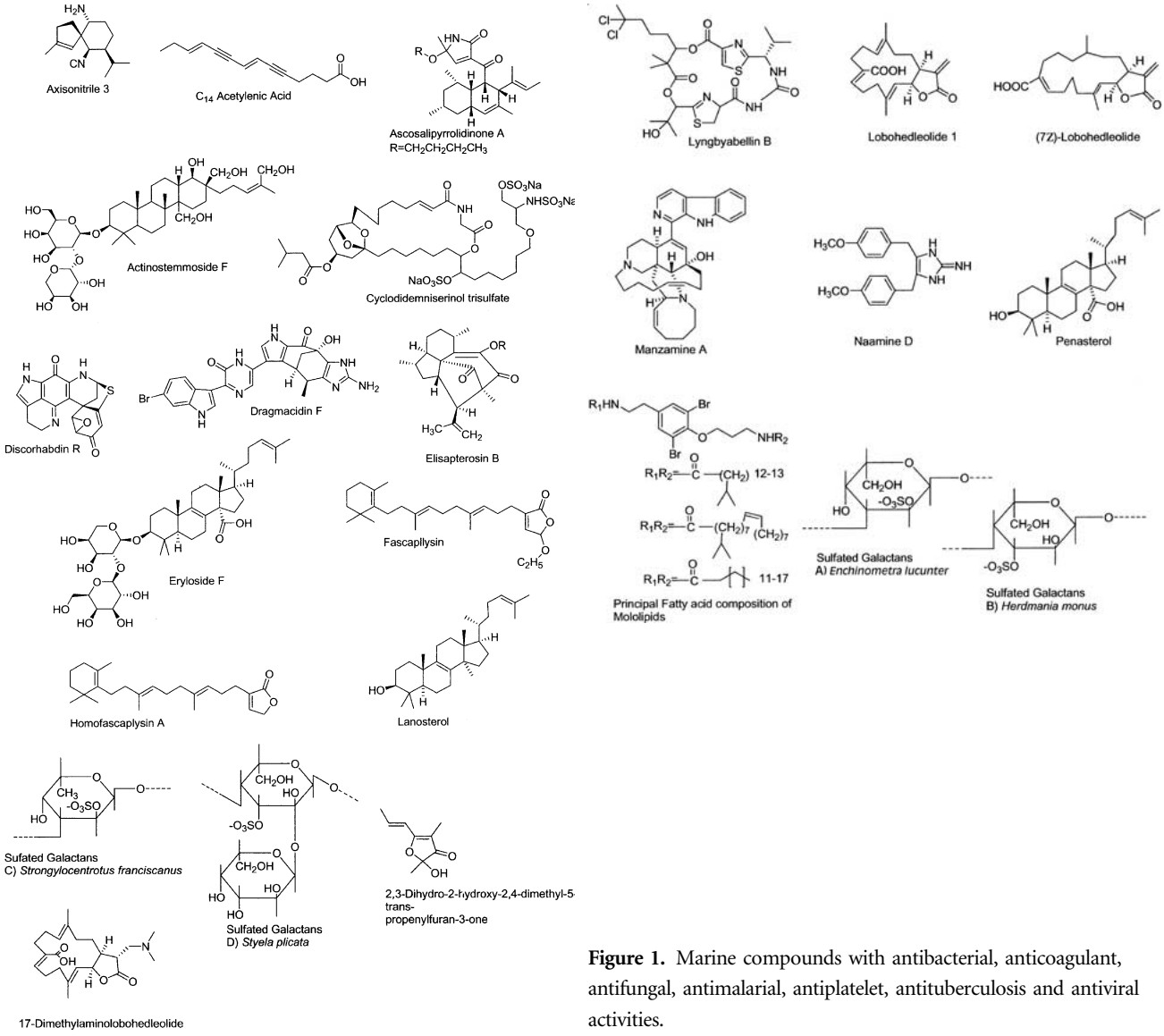

Figure 1. Marine compounds with antibacterial, anticoagulant, antifungal, antimalarial, antiplatelet, antituberculosis and antiviral activities.

L-galactans from marine invertebrates, which have well-defined structures and were also studied in the authors' experiments. The authors proposed that "sulfated galactans from *B. occidentalis* are natural candidate molecules for testing in experimental thrombosis."

Antifungal Compounds

Two studies were published during 2000 on the antifungal properties of two novel marine natural products. *Lyngbyabellin B*, an antifungal depsipeptide from the marine cyanobacterium *Lynbya majuscula*, proved active against *Candida albicans* in a disk diffusion assay (Milligan et al., 2000). A novel imidazole alkaloid, *naamine D*, was isolated from the Red Sea sponge *Leucetta* cf. *chagosensis* along with 4 known alkaloids: namely, naamidine A, B, D, and G (Dunbar et al., 2000). Although no extensive studies on mechanism of action were completed, naamine D had a MIC of 6.25 μg/ml against *Cryptococcus neoformans*. Although a comparison with established antifungal agents was not included in these studies, thus precluding a comparative assessment of this compound's antifungal potency, interestingly naamine D at 1 mM competitively inhibited murine macrophage-inducible nitric oxide synthase by 50%.

Antimalarial, Antiplatelet, and Antituberculosis Compounds

During 2000, 6 studies were reported in the area of antimalarial, antiplatelet, and antituberculosis pharmacology

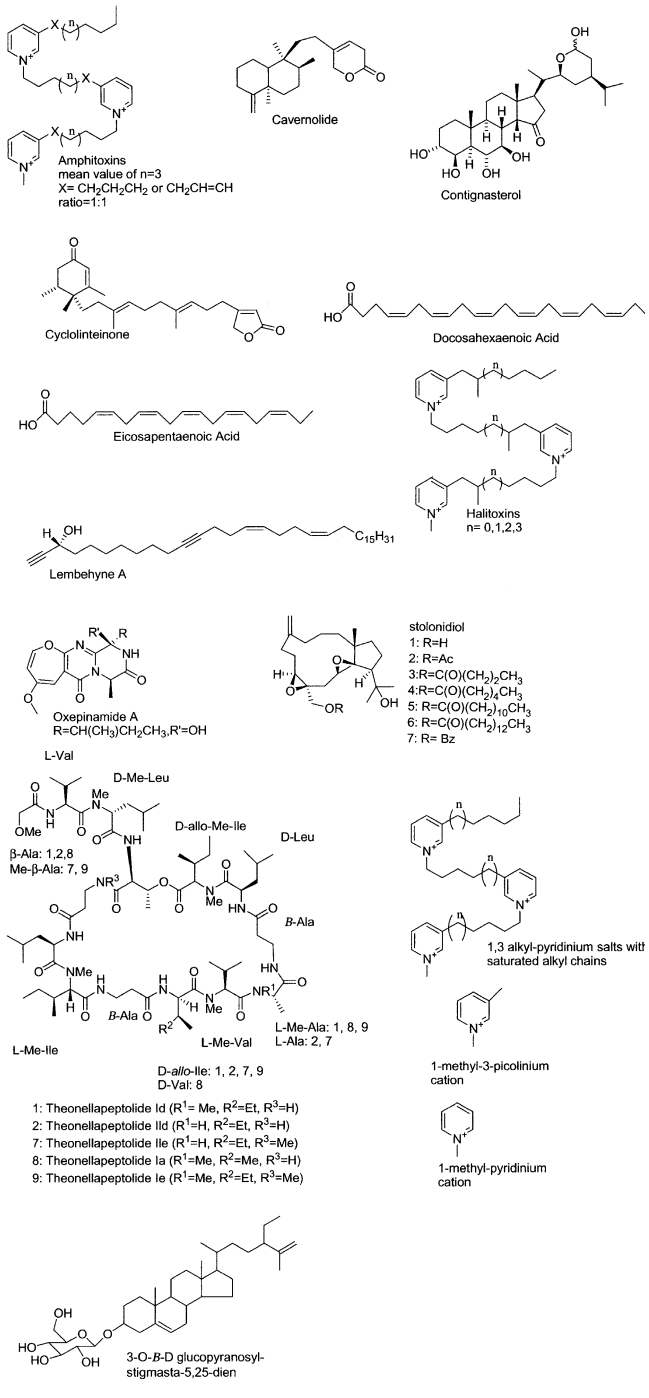

Figure 2. Marine compounds with anti-inflammatory and immunosuppressant properties and effects on the cardiovascular and nervous systems.

of structurally characterized marine natural products. Three compounds were shown to possess antimalarial activity. As a result of studies with fungal strains associated with marine algae, Osterhage et al. (2000) isolated the novel polyketide *ascosalipyrrolidinone A* from the obligate marine fungus *Ascochyta salicorniae*. Clearly significant is the fact that ascosalipyrrolidinone A was shown to have some antiplasmodial activity toward both *Plasmodium falciparum* strain K1, a strain resistant to chloroquine, and strain NF 54, a strain susceptible to standard antimalarials. In addition, the compound demonstrated limited antimicrobial activity and tyrosine kinase $p56^{lck}$ inhibition. *Homofascaplysin A* and *fascaplysin*, sesterterpenes isolated from the Fijian marine sponge *Hyrtios* cf. *erecta*, demonstrated potent activity *in vitro* against both chloroquine-susceptible (NF-54) and chloroquine-resistant (K-1) *Plasmodium falciparum* strains (IC_{50} = 14–24 ng/ml) (Kirsch et al., 2000). Their reduced cytotoxicity against muscle myoblast cells and mouse peritoneal macrophages suggests that these compounds might constitute potential leads for novel antimalarial compounds. Finally, *manzamine A*, a β-carboline alkaloid, was shown to inhibit the growth of the rodent malaria parasite *Plasmodium berghei*, not only *in vitro* but also *in vivo* (Ang et al., 2000). Remarkably, manzamine A caused reduction of parasitemia by either the oral or intraperitoneal route of administration, suggesting it is a potentially promising antimalarial agent.

Only a single paper reported on antiplatelet pharmacology of marine natural products during 2000. *Eryloside F*, a penasterol disaccharide isolated form the marine sponge *Erylus formosus*, demonstrated potent and relatively selective thrombin receptor antagonist activity (Stead et al., 2000). Eryloside F was identified upon completion of a study that involved screening over 50,000 compounds and natural product samples using a thrombin receptor antagonist high-throughput screen. Evaluation in a platelet aggregation assay revealed that eryloside F inhibited thrombin-induced human platelet aggregation *in vitro* in a concentration-dependent manner.

Two papers reported work on antituberculosis pharmacology with marine natural products. In a systematic study involving more than 30 selected marine compounds belonging to several structural classes and derived from both marine algae and invertebrates, antimycobaterial activities against either *Mycobacterium tuberculosis* or *Mycobacterium avium* were observed in approximately one third of the screened compounds (Konig et al., 2000). Of interest was the observation that molecules with an isonitrile group were the most active. In particular, *axisonitrile-3*, a compound isolated from the sponge *Acanthella kletra*, was shown to be extremely active against *M. tuberculosis*, with a concomitant low cytotoxicity, and constituting in the

Table 2. Marine Compounds with Anti-inflammatory and Immunosuppressant Effects and Affecting the Cardiovascular and Nervous Systems

Drug class	Compound/organism[a]	Chemistry	Pharmacological Activity	MMOA[b]	Country[c]	References
Anti-inflammatory	Cavernolide/coral	Terpene[e]	In vitro TNF-α, NO and PGE_2 inhibition	$sPLA_2$, iNOS and COX_2 inhibition	ITA, SPA	Posadas et al., 2000
Anti-inflammatory	Contignasterol/sponge	Sterol[e]	In vivo allergen-induced plasma protein exhudation inhibition	Undetermined	AUS	Coulson et al. 2000
Anti-inflammatory	Cyclolinteinone/sponge	Sesterterpene[e]	In vitro NO and PGE_2 inhibition	NF-κB binding, iNOS and COX_2 expression inhibition	ITA	D'Acquisto et al., 2000
Anti-inflammatory	Oxepinamide A/fungus	Alkaloid[f]	In vivo neurogenic inflammation assay	Undetermined	GER, USA	Belofsky et al., 2000
Anti-inflammatory	Sterol/alga	Sterol glycoside[e]	In vivo inflammation assay	Undetermined	EGYP	Awad 2000
Cardiovascular	Docosahexaenoic acid/fish	Fatty acid[d]	In vivo vascular reactivity assays and in vitro biochemical assays	Undetermined	AUS	Mori et al., 2000
Immunosuppressant	Theonellapeptolides/sponge	Peptide[f]	In vitro mixed lymphocyte reaction assay	Undetermined	JPN, IND	Roy et al., 2000
Nervous system	Conantokin G, T/snail	Peptide[f]	In vivo Parkinson's disease model	Alterations of striatal efferent neurons function	USA	Adams et al., 2000
Nervous system	Conantokin-G/snail	Peptide[f]	In vivo block of dopamine-enhancing drug methamphetamine	NMDA receptor antagonism	USA	Bush et al., 2000
Nervous system	Conantokin-G/snail	Peptide[f]	In vitro neuronal and oocyte whole-cell electrophysiology	Competitive antagonist of NR2B NMDA receptors	USA	Donevan et al., 2000
Nervous system	Conantokin-G/snail	Peptide[f]	In vivo and in vitro neuroprotective assays	Decrease Ca^{2+} responses to NMDA	USA	Williams et al., 2000
Nervous system	Conantokin-R/snail	Peptide[f]	In vitro NMDA receptor antagonist and in vivo anticonvulsant assays	NR2 NMDA receptor selectivity	PHIL, USA	White et al., 2000
Nervous system	Conantokin-R/snail	Peptide[f]	In vitro binding assays, spectroscopy, and NMR	Disulfide loop not essential for receptor and cation binding	USA	Blandl et al., 2000

Nervous system	*C. marmoreus* conotoxin/snail	Peptide[f]	In vitro and in vivo electrophysiology & binding assays	Undetermined	PHIL, USA	McIntosh et al., 2000a
Nervous system	α-Conotoxin MII/snail	Peptide[f]	In vitro nicotinic, receptor-transfected, oocyte electrophysiology	Blocks β-3 nicotinic receptor subunit	USA	McIntosh et al., 2000b
Nervous system	Halitoxins/bacterium	Alkaloids[f]	In vitro neuronal electrophysiology and calcium imaging	Pore formation in biological membranes	UK	Scott et al., 2000
Nervous system	Lembehyne A/sponge	Fatty acid[d]	In vitro neuritogenic assay	Actin polymerization and protein synthesis dependent	JPN, INDO	Aoki et al., 2000
Nervous system	Stolonidiol/soft coral	Diterpene[e]	In vitro choline acetyltransferase activity assay	Undetermined	JPN	Yabe et al., 2000

[a] *Kingdom Animalia*: coral (phyllum Cnidaria); snail (phylum Mollusca); sponge (phylum Porifera); seastar and sea cucumber (phylum Echinodermata); seal (phylum Chordata). *Kingdom Plantae*: dinoflagellate and alga. *Kingdom Monera*: bacterium (phylum Cyanobacteria).
[b] MMOA is molecular mechanism of action.
[c] AUS is Australia; EGYP, Egypt; GER, Germany; IND, India; INDO, Indonesia; ITA, Italy; JPN, Japan; PHIL, Phillipines; SPA, Spain; UK, United Kingdom.
[d] Polyketides; [e] Terpenes; [f] Nitrogen-containing compounds.

authors' view, "a very good antimycobacterial lead compound for the first time." Rodriguez and his collaborators (2000) isolated the novel diterpene *elisapterosin B* from the West Indian gorgonian *Pseudopterogorgia elisabethae*. Although no mechanistic studies were reported, the elisapterosin B demonstrated inhibitory activity against *M. tuberculosis* at a concentration of 12.5 μg/ml.

Antiviral Compounds

Interest in the antiviral pharmacology of marine natural products remained high during 2000 as evidenced by the 4 papers published, a number similar to those reported in our corresponding 1998 and 1999 reviews (Mayer and Lehmann, 2000; Mayer and Hamann, 2002). As a result of an ongoing program focused on the discovery of new nonsteroidal inhibitors of the human immunodeficiency virus (HIV)-1 integrase from marine invertebrates, Mitchell et al. (2000) reported on the isolation of *cyclodidemniserinol trisulfate* from the Palauan ascidian *Didemnum guttatum.* Although cyclodidemniserinol trisulfate inhibited the purified HIV-1 integrase (IC_{50} = 60 μg/ml), it also inhibited the topoisomerase enzyme of the *Molluscum contagiosum* virus (IC_{50} = 60 μg/ml) with a similar potency, thus demonstrating a limited selectivity for the HIV-1 integrase. Cutignano and collaborators (2000) reported on a new antiviral bromoindole alkaloid, *dragmacidin F*, isolated from the Mediterranean sponge *Halicortex* sp. In collaboration with French research groups, these investigators determined that dragmacidin F weakly inhibited herpes simplex virus (HSV)-1–infected cells from HSV-induced destruction (IC_{50} = 95.8 μM) and furthermore delayed syncytia formation by HIV-2 (IC_{50} = 0.91 μM). Bioassay-guided fractionation of aqueous extracts from the Phillipine soft coral *Lobophytum* sp. yielded 2 known cembranoid diterpenes, lobohedleolide and *(7Z)-lobohedleolide*, and a new compound, *17-dimethylaminolobohedleolide*, with moderate HIV-inhibitory activity (IC_{50} = 9–10.2 μg/ml) in a cell-based *in vitro* anti-HIV assay (Rashid et al., 2000). Ross et al. (2000) reported on a new series of anti-HIV bromotyrosine-derived lipids, namely the *mololipids*, identified in an Hawaiian sponge of the order Verongida. Interestingly the mololipids appeared to be selectively but weakly active against HIV-1 (EC_{50} = 52.2 μM), with low cytotoxicity against human peripheral blood mononuclear cells, thus suggesting to the authors that this series had some potential "for future studies."

Table 3. Marine Compounds with Miscellaneous Mechanisms of Action

Compound/organism[a]	Chemistry	Pharmacologic activity	MMOA[b]	Country[c]	References
Adociasulfate 10/sponge	Sulfated triterpene[e]	Undetermined	Kinesin motor inhibition	USA	Blackburn et al., 2000
Agelasine F/sponge	Alkaloid[f]	Antituberculosis activity	Undetermined	PHIL, USA	Mangalindan et al., 2000
Anthraquinone/betaenone/fungus	Polyketide[d]	Undetermined	Protein kinase inhibition	GER	Brauers et al., 2000
B-5354c/bacterium	Alkaloid[f]	Undetermined	Sphingosine kinase inhibition	JPN	Kono et al., 2000
Bisprasin/sponge	Alkaloid[f]	In vitro sketeletal muscle sarcoplasmic reticulum assay	Ryanodine receptor Ca^{2+} release induction	JPN	Suzuki et al., 2000
Ceratospongamide/alga-sponge	Depsipeptide[f]	Undetermined	Secretory Phospholipase A_2 expression inhibition	USA	Tan et al., 2000
5α-Cholest-7-en-3β-ol/starfish	Sterol[e]	Antigenotoxic and mutagenic	Undetermined	S.KOR	Han et al., 2000
Clavosines A & B/sponge	Polyketide[d]	Undetermined	Protein phosphatase-1 inhibition	USA, CAN	McCready et al., 2000
Dysidotronic acid/sponge	Diterpene[e]	Undetermined	Phospholipase A_2 inhibition	ITA, FRA, SPA	Giannini et al., 2000
Equistatin/sea anemone	Protein[f]	In vitro inhibition of papain and cathepsin D	Characterization of cysteine proteinase inhibitory domains	SLOV, NETH	Strukelj et al., 2000
Fascaplysin/sponge	Alkaloid[f]	In vitro cyclin-dependent kinase assays	Cyclin-dependent kinase 4 catalytic subunit inhibition	SWZ	Soni et al., 2000
Hymenialdisine/sponge	Alkaloid[f]	In vitro and In vivo inhibition of protein phosphorylation	Cyclin-dependent kinase 5 inhibition	FRA, GER, UK, USA	Meijer et al., 2000
Jasplakinolide/sponge	Peptide[f]	Apoptosis	Caspase-3-like protease dependent pathway	JPN, USA	Odaka et al., 2000
Latrunculin A/sponge	Polyketide[d]	Antiprion effect	Actin cytoskeleton inhibition	USA	Bailleul-Winslett et al., 2000
Latrunculin-B/sponge	Polyketide[d]	In vivo antiglaucoma assay	G-actin sequestration	ISR, USA	Peterson et al., 2000
Microginins/bacterium	Peptide[f]	Undetermined	Zinc metalloproteases inhibition	JPN	Ishida et al., 2000
Miraziridine A/sponge	Peptide[f]	Undetermined	Cathepsin B inhibition	JPN	Nakao et al., 2000a
Penarolide sulfates A_1 & A_2 sponge	Lipopeptide[f]	Undetermined	A-glucosidase inhibition	JPN, NETH	Nakao et al., 2000b
Phomopsidin/fungus	Polyketide[d]	Undetermined	Microtubule assembly inhibition	JPN	Namikoshi et al., 2000
Pyridinium alkaloids/sponge	Alkaloid[f]	Undetermined	Phospholipase A_2 inhibition	ITA, FRA, SPA	De Marino et al., 2000
Scalarane & homoscalarane/nudibranchs	Sesquiterpenes[e]	Undetermined	Phospholipase A_2 inhibition	ITA, FRA, SPA	Fontana et al., 2000
Sphingosines/tunicate	Fatty acid[d]	Undetermined	Phospholipase A_2 inhibition	FRA	Loukaci et al., 2000

Stelliferins/sponge	Triterpenes[e]	In vitro cytotoxicity and morphology assays	Undetermined	JPN	Oku et al., 2000
Sterols/sponge	Sterols[e]	Undetermined	Interleukin 8 receptor antagonist	AUS	Leone et al., 2000
Turbotoxins A & B/gastropod	Alkaloid[f]	Undetermined	Acetylcholinesterase inhibition	JPN	Kigoshi et al., 2000

[a] *Kingdom Animalia*: anemones, corals and hydroids (phylum Cnidaria), mollusk (phylum Mollusca), sea cucumber (phylum Echinodermata), sponge (phylum Porifera), *Kingdom Fungi*: fungus. *Kingdom Plantae*: alga; *Kingdom Monera*: bacterium (phylum Cyanobacteria).
[b] MMOA is molecular mechanism of action.
[c] AUS is Australia; CAN, Canada; FRA, France; GER, Germany; ISR, Israel; ITA, Italy; JPN, Japan; NETH, The Netherlands; PHIL, Phillipines; S.KOR, South Korea; SLO, Slovenia; SPA, Spain; SWZ, Switzerland; UK, United Kingdom.
[d] Polyketides.
[e] Terpenes.
[f] Nitrogen-containing compounds.

Compounds with Anti-inflammatory and Immunosuppressant Effects and Affecting the Cardiovascular and Nervous Systems

Table 2 summarizes preclinical pharmacologic research completed on 20 marine chemicals shown in Figure 2 that were shown to affect the cardiovascular and nervous systems and to possess anti-inflammatory and immunosuppressant activities.

Anti-inflammatory Compounds

The anti-inflammatory pharmacology of carvernolide, contignasterol, cyclolinteinone, oxenamide A, and an algal sterol glycoside was reported during 2000, an increase of one compound over 1999 (Mayer and Hamann 2002). Posadas et al. (2000) reported on the mechanism of action of *cavernolide*, a novel C_{21} terpene lactone isolated from the sponge *Fasciospongia cavernosa*. Their results suggested that cavernolide's potent inhibition of tumor necrosis factor-α, nitric oxide, and prostaglandin E_2 *in vitro* was the result of both human synovial phospholipase A_2 (IC_{50} = 8.8 μM) inhibition, and inhibition of inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2) gene expression in intact cells.

Coulson and O'Donnell (2000) extended the *in vivo* pharmacology of *contignasterol*, a highly oxygenated sterol isolated from the sponge *Petrosia contignata* that appears to be as potent as nedocromil in inhibiting allergen-induced bronchoconstriction *in vivo*. The study demonstrated that contignasterol dose-dependently inhibited plasma exudation *in vivo* in response to ovalbumin, indicating that this is a potential anti-inflammatory compound.

D'Acquisto et al. (2000) determined the effect of *cyclolinteinone*, a sesterterpene isolated from the sponge *Cacospongia linteiformis*, on iNOS synthase and COX-2 enzyme *in vitro*. Cyclolinteinone's reported anti-inflammatory properties included inhibition of nuclear transcription factor-κB binding activity, a decrease of both iNOS and COX-2 expression, and the concomitant weak *in vitro* inhibition of both prostaglandin E_2 (IC_{50} = 50 μM). and nitric oxide (IC_{50} = 50 μM).

Belofsky et al. (2000) reported on new oxepinamides and two fumiquinazolines, alkaloids isolated from cul-

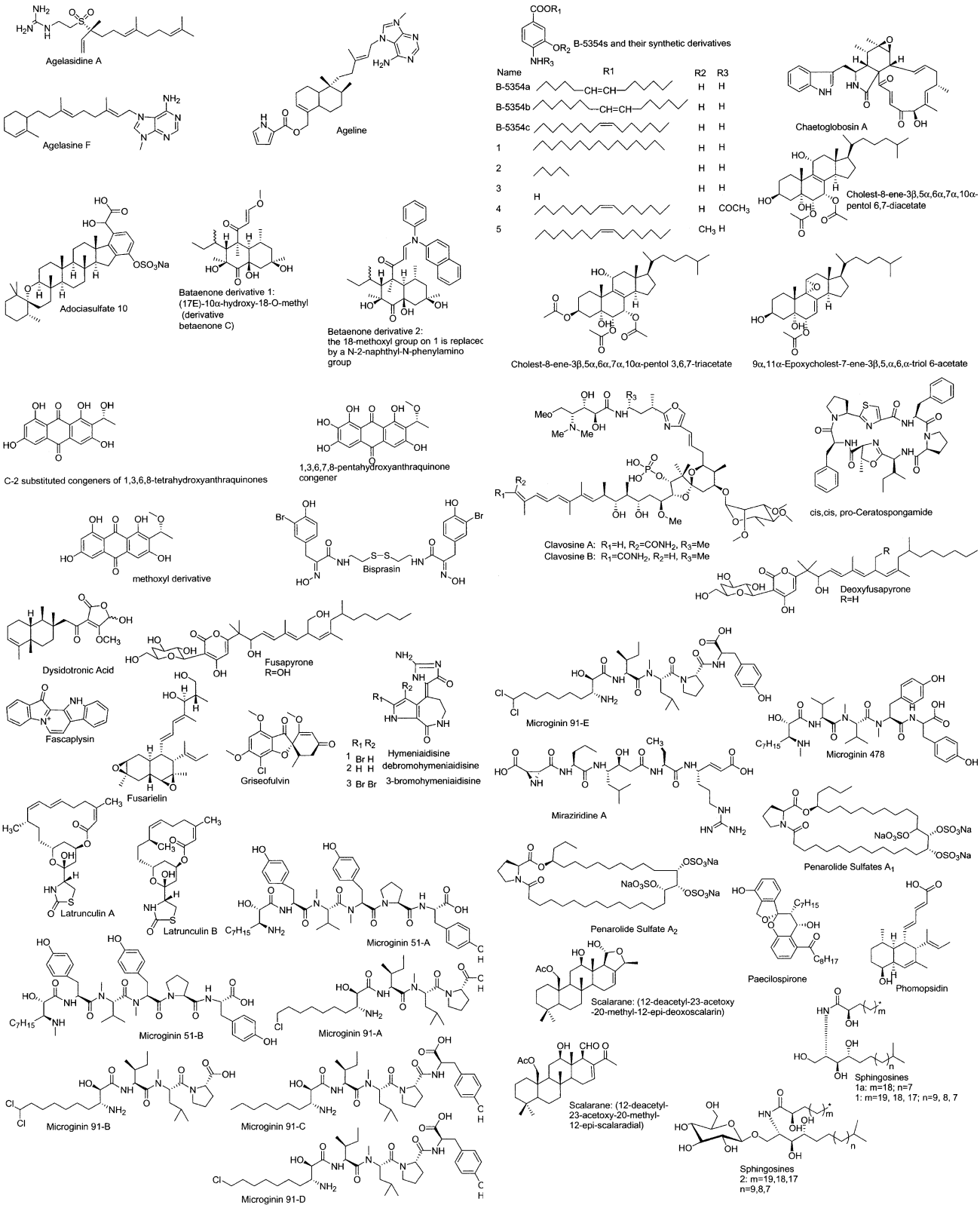

Figure 3. Marine compounds with miscellaneous mechanisms of action.

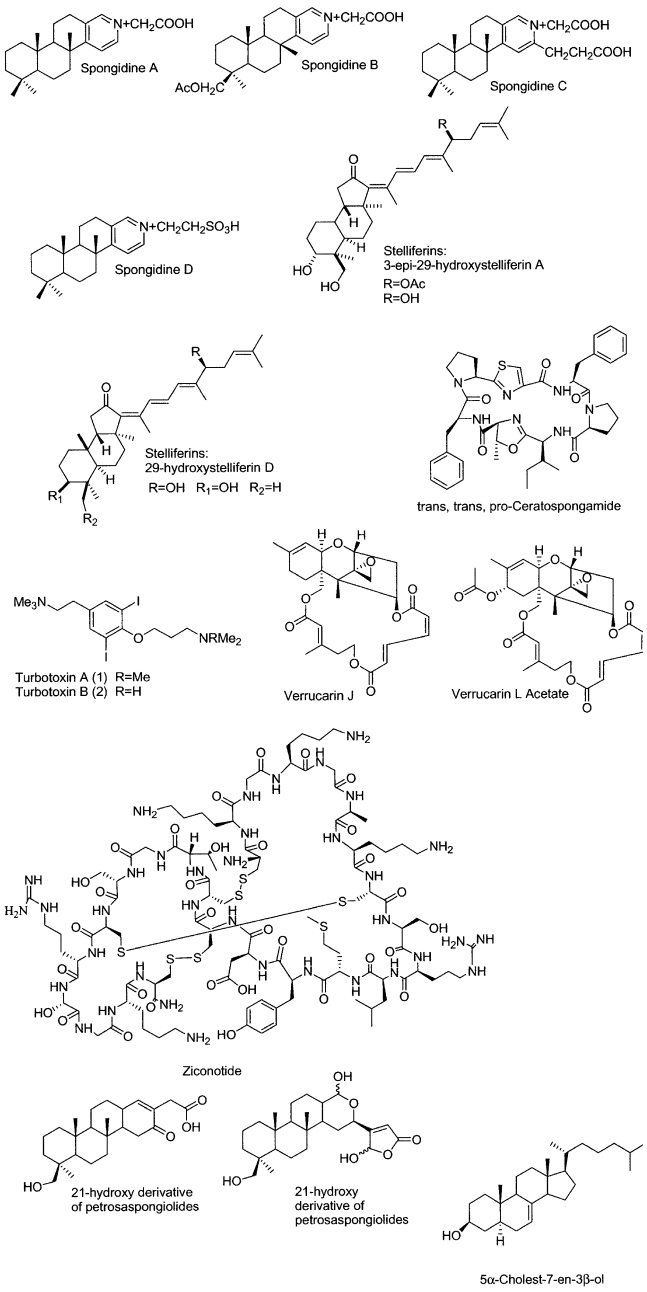

Figure 3. Continued.

tures of the marine fungus *Acremonium* sp. Only *oxepinamide A* exhibited good *in vivo*anti-inflammatory activity in the resiniferatoxin-induced topical mouse ear edema assay, a test for neurogenic inflammation. Awad (2000) reported on a novel *sterol glycoside* from *Ulva lactuca* collected from Alexandrian shores in Egypt. The glycoside exhibited good *in vivo* anti-inflammatory activity in the phorbol-ester-induced topical mouse ear edema assay.

Immunosuppressant Compounds; Cardiovascular Pharmacology

Only 2 reports during 2000 contributed to the immunosuppressant and cardiovascular pharmacology of marine natural products. Roy et al. (2000) presented novel information on potential immunosuppresant activity of cyclic peptides isolated from the marine sponge *Theonella swinhoei*, which have some common structural features with the cyclosporins, agents currently used clinically as immunosuppressants. The tridecapeptide lactones *theonellapeptolides Ia*, *Id*, and *IId* were strongly immunosuppressive, possibly as a result of ''their cytotoxic effect.'' In a contribution to the cardiovascular pharmacology of marine-derived *eicosapentaenoic* and *docosahexanoic acid*, Mori et al. (2000) investigated the effect of these purified omega-3 fatty acids on vascular reactivity of the forearm circulation in hyperlipidemic and overweight men. The results of a double-blind, placebo-controlled trial involving 59 overweight, mildly hyperlipidemic men demonstrated that docosahexanenoic acid, but not eicosapentaenoic acid, enhanced the vasodilator mechanisms and constrictor responses in forearm circulation, thus contributing to selective blood-pressure-lowering effects. The authors proposed that these observations could be relevant to the food industry with respect to the possible inclusion of omega-3 fatty acids into foodstuffs and animal feeds, and ''hence the human food chain.''

Nervous System Pharmacology

Reports on both central and autonomic nervous system pharmacology of marine natural products increased slightly over 1998 and 1999 (Mayer and Lehmann, 2000; Mayer and Hamann, 2002), with the 2000 studies involving the conantokins-G, T, and R, the marine *C. marmoreus* conotoxin and α-Conotoxin MII, and the halitoxins, lembehyne A and stolonidiol.

Several studies extended the pharmacology of the conantokins and conotoxins, a family of small peptide toxins derived from the venom of marine snails of the genus *Conus* that have been shown to bind to excitable tissue (Olivera, 1997). Adams et al. (2000) examined whether 2 peptides derived from the marine cone snail *Conus geographus*, and potent *N*-Methyl-D-asparate (NMDA) ionotropic glutamate receptor antagonists, *conantokin-G* and *conantokin-T(G)*, would potentiate

contralateral rotation induced by L-3,4-dihydroxyphenylalanine (L-DOPA) in 6-hydroxydopamine-treated rats, an animal model used to search for antiparkinsonian compounds. The 2 conantokins potentiated the behavioral effects of L-DOPA, probably by alterations in the function of striatal efferent neurons, suggesting that further research with the conantokins may lead to useful adjuncts for the treatment of Parkinson's disease.

Bush et al. (2000) reported that *conantokin-G* enhanced the behavioral effects of methamphetamine, a potent central nervous system stimulant that causes dopamine release, as well as attenuated tissue levels of 2 neuropeptides found in the striatum and substantia nigra: namely, neurotensin and dynorphin A. Donevan and McCabe (2000) contributed to the mechanism of action of conantokin G by demonstrating that it is a NR2B-selective competitive antagonist of the NMDA receptor. The unique subunit selectivity may explain its favorable *in vivo* profile compared to those of other nonselective NMDA antagonists. Williams et al. (2000) evaluated the *in vivo* and *in vitro* neuroprotective properties of conantokin-G. While *in vivo* conantokin-G reduced infarct volume and increased both neurologic recovery and electroencephalogram power scores when administered even 4 hours after occlusion of the middle cerebral artery, *in vitro* conantokin-G demonstrated neuroprotective properties against multiple forms of neuronal injury by blocking NMDA-induced calcium signaling. The authors concluded this extensive and detailed investigation by proposing that ''conantokin-G may represent an excellent adjunct treatment with drug therapy targeting the penumbral tissue.''

Blandl et al. (2000) completed a study to determine the bioactivity and conformation properties of the 7-amino acid peptide *conantokin-R*, a selective NMDA receptor antagonist isolated from the venom of the fish-hunting snail *Conus radiatus*. The study provided experimental evidence that the structural elements common to conantokin-R and other conantokins are the primary determinants for receptor function and cation binding or secondary structure stability. White et al. (2000) completed an extensive *in vitro* and *in vivo* characterization of conantokin-R. In *in vitro* studies conantokin-R showed selectivity for NR2 NMDA receptor subunits: NR2B $\sim$ NR2A $>$ NR2C $>>$ NR2D, but no effect on AMPA and kainate ionotropic glutamate receptors. In a battery of *in vivo* seizure models, conantokin-R demonstrated potent anticonvulsant activity at doses devoid of behavioral toxicity. Thus, because conantokin-R could be used as a pharmacologic agent to differentiate between the anticonvulsant and toxic effects of NMDA antagonists, it might contribute to the ongoing search for novel subunit-selective NMDA antagonists to treat human epilepsy.

A new *conotoxin* was isolated and characterized from the venom of *Conus marmoreus* during 2000 (McIntosh et al., 2000a). The peptide sequence has a novel disulfide bond connectivity that appears highly divergent from all other known conotoxins, and it targets a yet undetermined antinociceptive receptor. In a short communication McIntosh and collaborators (2000b) extended the pharmacology of *α-conotoxins*, widely used to probe neuronal nicotinic receptors. The investigators demonstrated that α-conotoxin MII blocked $\alpha_3\beta_2\beta_3$ human nicotinic acetylcholine receptors expressed in *Xenopus* oocytes.

Three interesting papers appeared on the halitoxins, lembehyne A and stolonidiol, compounds that appear to affect nervous tissue in different ways. Analysis of structure and electrophysiologic actions of *halitoxins*, marine 1,3 alkyl-pyridinium salts isolated from the marine sponge *Callyspongia ridleyi*, were reported by Scott et al. (2000). Most notable is that because halitoxins form ion-permeable pores in biological and artificial membranes, allowing flux of monovalent (K^+, Na^+) and divalent (Ca^{2+}) cations, they could be useful in a number of applications, including the development of novel cytotoxic molecules and possibly as agents for intracellular drug delivery. Aoki et al. (2000) reported on *lembehyne A*, a novel fatty acid derived from the sponge *Haliclona* sp., which induces neurite outgrowth. Although the molecular target of lembehyne A is unknown at this time, this is the first report that a linear polyacetylene can induce neurite outgrowth *in vitro*, a biological response that appears to be dependent on actin polymerization and de novo protein synthesis. A significant contribution to the search for choline acetyltransferase inducers as potential agents to improve cognitive function in persons with diseases exhibiting cholinergic deficits, such as Alzheimer's disease, was reported by Yabe et al. (2000). The fact that a structure-activity relationship study with *stolonidiol*, a marine diterpenoid isolated from the soft coral *Clavularia* sp., demonstrated potent choline-acetyltransferase-inducible activity in neuronal cultures *in vitro* suggests that stolonidiol or some of its derivates may serve as leads in the discovery of more useful agents with potential benefit to the cholinergic nervous system.

Marine Compounds with Miscellaneous Mechanisms of Action

Table 3 lists 23 marine compounds with miscellaneous mechanisms of action, and their structures are shown in Figure 3. Interestingly, and in contrast with the chemicals included in Tables 1 and 2, this third group of marine compounds includes nitrogen-containing compounds (i.e. proteins, peptides), terpenes, and polyketides, but not polysaccharides.

For some of these marine chemicals—namely, bisprasin, equistatin, fascaplysin, hymenialdisine, jasplakinolide, and latrunculin-B—both pharmacologic activity and a molecular mechanism of action have been investigated. For agelasine F, 5α-cholest-7-en-3β-ol, and stelliferins, only the pharmacologic activity has been investigated so far, and little is known about their molecular mechanism of action. Finally, although adociasulfate 10, B-5354c, ceratospongamide, clavosines A and B, dysidotronic acid, microginins, miraziridine A, penarolide sulfates, phomopsidin, pyridinium alkaloids, scalarane, and homoscalarane, sphingosines, sterols, and turbotoxins have been explored at the molecular level, they have not been assigned to a particular pharmacologic class at this time.

Reviews on Marine Pharmacology

Several reviews covering selected aspects of marine pharmacology were published during 2000: structure-function studies of anticoagulant marine sulfated polysaccharides (Mulloy et al., 2000); heparinoid-active sulfated polysaccharides from marine algae as potential blood anticoagulant agents (Shanmugam and Mody, 2000); an evaluation of intrathecal ziconotide for the treatment of chronic pain (Jain, 2000); natural product leads for the chemotherapy of HIV infection (De Clercq, 2000); different approaches to the pharmaceutical application of marine lipids (Masson et al., 2000); and the chemistry of marine natural products (Faulkner, 2000).

Conclusions

Although during 2000 no new marine natural product was approved for patient care by the U.S. Food and Drug Administration, during 2000 preclinical pharmacologic research with marine chemicals continued to proceed at a very active pace, involving both natural product chemists and pharmacologists from 21 foreign countries and the United States.

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References

Adams, A.C., Layer, R.T., McCabe, R.T., and Keefe, K.A. (2000). Effects of conantokins on L-3,4-dihydroxyphenylalanine-induced behavior and immediate early gene expression. *Eur J Pharmacol* 404:303–313.

Ang, K.K.H., Holmes, M.J., Higa, T., Hamann, M.T., and Kara, U.A.K. (2000). In vivo antimalarial activity of the beta-carboline alkaloid manzamine A. *Antimicrob Agents Chemother* 44:1645–1649.

Aoki, S., Matsui, K., Tanaka, K., Satari, R., and Kobayashi, M. (2000). Lembehyne a, a novel neuritogenic polyacetylene, from a marine sponge of *Haliclona* sp. *Tetrahedron* 56:9945–9948.

Awad, N.E. (2000). Biologically active steroid from the green alga Ulva lactuca. *Phytother Res* 14:641–643.

Bailleul-Winslett, P.A., Newman, G.P., Wegrzyn, R.D., and Chernoff, Y.O. (2000). An antiprion effect of the anticytoskeletal drug Latrunculin A in yeast. *Gene Expression* 9:145–156.

Belofsky, G.N., Anguera, M., Jensen, P.R., Fenical, W., and Kock, M. (2000). Oxepinamides A-C and fumiquinazolines H–I: bioactive metabolites from a marine isolate of a fungus of the genus *Acremonium*. *Chem Eur J* 6:1355–1360.

Blackburn, C.L., and Faulkner, D.J. (2000). Adociasulfate 10, a new merohexaprenoid sulfate from the sponge *Haliclona* (aka *Adocia*) sp. *Tetrahedron* 56:8429–8432.

Blandl, T., Warder, S.E., Prorok, M., and Castellino, F.J. (2000). Structure-function relationships of the NMDA receptor antagonist peptide, conantokin-R. *FEBS Lett* 470:139–146.

Brauers, G., Edrada, R.A., Ebel, R., Proksch, P., Wray, V., Berg, A., Grafe, U., Schachtele, C., Totzke, F., Finkenzeller, G., Marme, D., Kraus, J., Munchbach, M., Michel, M., Bringmann, G., and Schaumann, K. (2000). Anthraquinones and betaenone derivatives from the sponge-associated fungus *Microsphaeropsis* species: novel inhibitors of protein kinases. *J Nat Prod* 63:739–745.

Bush, L., McCabe, T., and Hanson, G.R. (2000). Selective antagonism of nigral neuropeptide responses to methamphetamine by conantokin G, a naturally occurring conopeptide. *Eur J Pharmacol* 387:55–58.

Coulson, F.R., and O'Donnell, S.R. (2000). The effects of contignasterol (IZP-94,005) on allergen-induced plasma protein exudation in the tracheobronchial airways of sensitized guinea-pigs in vivo. *Inflamm Res* 49:123–127.

Cutignano, A., Bifulco, G., Bruno, I., Casapullo, A., Gomez-Paloma, L., and Riccio, R. (2000). Dragmacidin F: a new antiviral bromoindole alkaloid from the Mediterranean sponge *Halicortex* sp. *Tetrahedron* 56:3743–3748.

D'Acquisto, F., Lanzotti, V., and Carnuccio, R. (2000). Cyclolinteinone, a sesterterpene from sponge *Cacospongia linteiformis*, prevents inducible nitric oxide synthase and inducible cyclooxygenase protein expression by blocking nuclear factor-kappaB activation in J774 macrophages. *Biochem J* 346(Pt 3):793–798.

De Clercq, E. (2000). Current lead natural products for the chemotherapy of human immunodefiency virus (HIV) infection. *Med Res Rev* 20:323–349.

De Marino, S., lorizzi, M., Zollo, F., Debitus, C., Menou, J.L., Ospina, LF., Alcaraz, M.J., and Paya, M. (2000). New pyridinium alkaloids from a marine sponge of the genus *Spongia* with human phospholipase A(2) inhibitor profile. *J Nat Prod* 63:322–326.

Donevan, S.D., and McCabe, R.T. (2000). Conantokin G is an NR2B-selective competitive antagonist of *N*-methyl-D-aspartate receptors. *Mol Pharmacol* 58:614–623.

Dunbar, D.C., Rimoldi, J.M., Clark, A.M., Kelly, M., and Hamann, M.T. (2000). Anti-cryptococcal and nitric oxide synthase inhibitory imidazole alkaloids from the calcareous sponge *Leucetta* cf *chagosensis. Tetrahedron* 56:8795–8798.

Farias, W.R.L., Valente, A.P., Pereira, M.S., and Mourao, P.A.S. (2000). Structure and anticoagulant activity of sulfated galactans—isolation of a unique sulfated galactan from the red algae *Botryocladia occidentalis* and comparison of its anticoagulant action with that of sulfated galactans from invertebrates. *J Biol Chem* 275:29299–29307.

Faulkner, D.J. (2000). Marine natural products. *Nat Prod Rep* 17:7–55.

Fontana, A., Mollo, E., Ortea, J., Gavagnin, M., and Cimino, G. (2000). Scalarane and homoscalarane compounds from the nudibranchs *Glossodoris sedna* and *Glossodoris dalli*: chemical and biological properties. *J Nat Prod* 63:527–530.

Ford, J., and Capon, R.J. (2000). Discorhabdin R: a new antibacterial Pyrroloiminoquinone from two latrunculiid marine sponges, *Latrunculia* sp. and Negombata sp. *J Nat Prod* 63:1527–1528.

Giannini, C., Debitus, C., Posadas, I., Paya, M., and D'Auria, M.V. (2000). Dysidotronic acid, a new and selective human phospholipase A(2) inhibitor from the sponge *Dysidea* sp. *Tetrahedron Lett* 41:3257–3260.

Han, Y.H., Ham, J.H., Lee, N.J., Park, C.H., Shin, Y.H., and Lee, D.U. (2000). Antimutagenic activity of 5alpha-cholest-7-en-3beta-ol, a new component from the starfish *Asterina pectinifera. Biol Pharm Bull* 23:1247–1249.

Ishida, K., Kato, T., Murakami, M., Watanabe, M., and Watanabe, M.F. (2000). Microginins, zinc metalloproteases inhibitors from the cyanobacterium *Microcystis aeruginosa*. *Tetrahedron* 56:8643–8656.

Jain, K.K. (2000). An evaluation of intrathecal ziconotide for the treatment of chronic pain. *Expert Opin Investig Drugs* 9:2403–2410.

Khudyakova, Y.V., Pivkin, M.V., Kuznetsova, T.A., and Svetashev, V.I. (2000). Fungi in sediments of the sea of Japan and their biologically active metabolites. *Microbiology* 69:608–611.

Klgoshi, H., Kanematsu, K., Yokota, K., and Uemura, D. (2000). Turbotoxins A and B, novel diiodotyramine derivatives from the Japanese gastropod *Turbo marmorata. Tetrahedron* 56:9063–9070.

Kirsch, G., Kong, G.M., Wright, A.D., and Kaminsky, R. (2000). A new bioactive sesterterpene and antiplasmodial alkaloids from the marine sponge *Hyrtios* cf. *erecta. J Nat Prod* 63:825–829.

Konig, G.M., Wright, A.D., and Franzblau, S.G. (2000). Assessment of antimycobacterial activity of a series of mainly marine derived natural products. *Planta Med* 66:337–342.

Kono, K., Tanaka, M., Mizuno, T., Kodama, K., Ogita, T., and Kohama, T. (2000). B-5354a, b and c, new sphingosine kinase inhibitors, produced by a marine bacterium; taxonomy, fermentation, isolation, physico-chemical properties and structure determination. *J Antibiot (Tokyo)* 53:753–758.

Leone, P.D., Redburn, J., Hooper, J.N.A., and Quinn, R.J. (2000). Polyoxygenated *Dysidea* sterols that inhibit the binding of [I^{125}] IL-8 to the human recombinant IL-8 receptor type A. *J Nat Prod* 63:694–697.

Loukaci, A., Bultel-Ponce, V., Longeon, A., and Guyot, M. (2000). New lipids from the tunicate *Cystodytes* cf. *dellechiajei*, as PLA_2 inhibitors. *J Nat Prod* 63:799–802.

Mangalindan, G.C., Talaue, M.T., Cruz, L.J., Franzblau, S.G., Adams, L.B., Richardson, A.D., Ireland, C.M., and Concepcion, G.P. (2000). Agelasine F from a Philippine *Agelas* sp. sponge exhibits in vitro antituberculosis activity. *Planta Med* 66:364–365.

Masson, W., Loftsson, T., and Haraldsson, G.G. (2000). Marine lipids for prodrugs, soft compounds and other pharmaceutical applications. *Pharmazie* 55:172–177.

Matsubara, K. (2000). An anticoagulant proteogylcan from the marine green alga, *Codium pungniformis*. *J Appl Phycol* 12:9–14.

Matsubara, K., Hori, K., Matsura, Y., and Miyazawa, K. (2000). Purification and characterization of a fibrinolytic enzyme and identification of fibrinogen clotting enzyme in a marine green alga, *Codium divaricatum*. *Comp Biochem Physiol B Biochem Mol Biol* 125:137–143.

Matsunaga, S., Okada, Y., Fusetani, N., and van Soest, R.W.M. (2000). An antimicrobial C_{14} acetylenic acid from a marine sponge *Oceanapia* species. *J Nat Prod* 63:690–691.

Mayer, A.M., and Gustafson, K.R. (2003). Marine pharmacology in 2000: antitumor and cytotoxic compounds. *Int J Cancer* 105:291–299.

Mayer, A.M.S., and Hamann, M.T. (2002). Marine pharmacology in 1999: compounds with antibacterial, anticoagulant, antifungal, anti-inflammatory, anthelmintic, anti-inflammatory, antiplatelet, antiprotozoal and antiviral activities; affecting the cardiovascular, endocrine, immune, and nervous systems; and other miscellaneous mechanisms of action. *Comp Biochem Physiol C Pharmacol Toxicol Endocrinol* 132:315–339.

Mayer, A.M.S., and Lehmann, V.K.B. (2000). Marine pharmacology in 1998: marine compounds with antibacterial, anticoagulant, antifungal, anti-inflammatory, anthelmintic, antiplatelet, antiprotozoal, and antiviral activities; with actions on the cardiovascular, endocrine, immune, and nervous systems; and other miscellaneous mechanisms of action. *Pharmacologist* 42: 62–69.

Mayer, A.M.S., and Lehmann, V.K.B. (2001). Marine pharmacology in 1999: antitumor and cytotoxic compounds. *Anticancer Res* 21:2489–2500.

McCready, T.L., Islam, B.F., Schmitz, F.J., Luu, H.A., Dawson, J.F., and Holmes, C.F. (2000). Inhibition of protein phosphatase-1 by clavosines A and B: novel members of the calyculin family of toxins. *J Biol Chem* 275:4192–4198.

McIntosh, J.M., Corpuz, G.O., Layer, R.T., Garrett, J.E., Wagstaff, J.D., Bulaj, G., Vyazovkina, A., Yoshikami, D., Cruz, L.J., and Olivera, B.M. (2000a). Isolation and characterization of a novel *Conus* peptide with apparent antinociceptive activity. *J Biol Chem* 275:32391–32397.

McIntosh, J.M., Gardner, S., Luo, S.Q., Garrett, J.E., and Yoshikami, D. (2000b). Conus peptides: novel probes for nicotinic acetylcholine receptor structure and function. *Eur J Pharmacol* 393:205–208.

Meijer, L., Thunnissen, A.M.W.H., White, A.W., Garnier, M., Nikolic, M., Tsai, L.H., Walter, J., Cleverley, K.E., Salinas, P.C., Wu, Y.Z., Biernat, J., Mandelkow, E.M., Kim, S.H., and Pettit, G.R. (2000). Inhibition of cyclin-dependent kinases, GSK-3 beta and CK1 by hymenialdisine, a marine sponge constituent. *Chem Biol* 7:51–63.

Milligan, K.E., Marquez, B.L., Williamson, R.T., and Gerwick, W.H. (2000). Lyngbyabellin B, a toxic and antifungal secondary metabolite from the marine cyanobacterium *Lyngbya majuscula*. *J Nat Prod* 63:1440–1443.

Mitchell, S.S., Rhodes, D., Bushman, F.D., and Faulkner, D.J. (2000). Cyclodidemniserinol trisulfate, a sulfated serinolipid from the Palauan ascidian *Didemnum guttatum* that inhibits HIV-1 integrase. *Org Lett* 2:1605–1607.

Mori, T.A., Watts, G.F., Burke, V., Hilme, E., Puddey, I.B., and Bellin, L.J. (2000). Differential effects of eicosapentaenoic acid and docosahexaenoic acid on vascular reactivity of the forearm microcirculation in hyperlipidemic, overweight men. *Circulation* 102:1264–1269.

Mulloy, B., Mourao, P.A.S., and Gray, E. (2000). Structure/function studies of anticoagulant sulphated polysaccharides using NMR. *J Biotechnol* 77:123–135.

Nakao, Y., Fujita, M., Warabi, K., Matsunaga, S., and Fusetani, N. (2000a). Miraziridine A, a novel cysteine protease inhibitor from the marine sponge *Theonella* aff. *mirabilis*. *J Am Chem Soc* 122:10462–10463.

Nakao, Y., Maki, T., Matsunaga, S., van Soest, R.W.M., and Fusetani, N. (2000b). Penarolide sulfates A(1) and A(2) new alpha-glucosidase inhibitors from a marine sponge *Penares* sp. *Tetrahedron* 56:8977–8987.

Namikoshi, M., Kobayashi, H., Yoshimoto, T., Meguro, S., and Akano, K. (2000). Isolation and characterization of bioactive metabolites from marine-derived filamentous fungi collected from tropical and sub-tropical coral reefs. *Chem Pharm Bull (Tokyo)* 48:1452–1457.

Odaka, C., Sanders, M.L., and Crews, P. (2000). Jasplakinolide induces apoptosis in various transformed cell lines by a caspase-3-like protease-dependent pathway. *Clin Diagn Lab Immunol* 7:947–952.

Oku, N., Matsunaga, S., Wada, S., Watabe, S., and Fusetani, N. (2000). New isomalabaricane triterpenes from the marine sponge *Stelletta globostellata* that induce morphological changes in rat fibroblasts. *J Nat Prod* 63:205–209.

Olivera, B.M. (1997). E.E. Just Lecture (1996) Conus venom peptides, receptor and ion channel targets, and drug design: 50 million years of neuropharmacology. *Mol Biol Cell* 8:2101–2109.

Osterhage, C., Kaminsky, R., Konig, G.M., and Wright, A.D. (2000). Ascosalipyrrolidinone A, an antimicrobial alkaloid, from the obligate marine fungus *Ascochyta salicorniae. J Org Chem* 65:6412–6417.

Peterson, J.A., Tian, B.H., Geiger, B., and Kaufman, P.L. (2000). Effect of latrunculin-B on outflow facility in monkeys. *Exp Eye Res* 70:307–313.

Posadas, I., Terencio, M.C., De Rosa, S., and Paya, M. (2000). Cavernolide: a new inhibitor of human $sPLA_2$ sharing unusual chemical features. *Life Sci* 67:3007–3014.

Rashid, M.A., Gustafson, K.R., and Boyd, M.R. (2000). HIV-inhibitory cembrane derivatives from a Philippines collection of the soft coral *Lobophytum* species. *J Nat Prod* 63:531–533.

Rodriguez, A.D., Ramirez, C., Rodriguez, I.I., and Barnes, C.L. (2000). Novel terpenoids from the West Indian sea whip *Pseudopterogorgia elisabethae* (Bayer). Elisapterosins A and B: rearranged diterpenes possessing an unprecedented cagelike framework. *J Org Chem* 65:1390–1398.

Ross, S.A., Weete, J.D., Schinazi, R.F., Wirtz, S.S., Thamish, P., Scheuer, P.J., and Hamann, M.T. (2000). Mololipids, a new series of anti-HIV bromotyramine-derived compounds from a sponge of the order Verongida. *J Nat Prod* 63:501–503.

Roy, M.C., Ohtani, I.I., Ichiba, T., Tanaka, J., Satari, R., and Higa, T. (2000). New cyclic peptides from the Indonesian sponge *Theonella swinhoei. Tetrahedron* 56:9079–9092.

Schmitz, F.J., Bowden, B.F., and Toth, S.I. (1993). Antitumor and cytotoxic compounds from marine organisms. In: *Marine Biotechnology*, Attaway, D.H., and Zaborsky, O.R. (Eds.). New York, N.Y.: Plenum Press, pp 197–308.

Scott, R.H., Whyment, A.D., Foster, A., Gordon, K.H., Milne, B.F., and Jaspars, M. (2000.). Analysis of the structure and electrophysiological actions of halitoxins: 1,3 alkyl-pyridinium salts from *Callyspongia ridleyi. J Membr Biol* 176:119–131.

Shanmugam, M., and Mody, K.H. (2000). Heparinoid-active sulphated polysaccharides from marine algae as potential blood anticoagulant agents [Review]. *Curr Sci* 79:1672–1683.

Soni, R., Muller, L., Furet, P., Schoepfer, J., Stephan, C., Zumstein-Mecker, S., Fretz, H., and Chaudhuri, B. (2000). Inhibition of cyclin-dependent kinase 4 (Cdk4) by fascaplysin, a marine natural product. *Biochem Biophys Res Commun* 275:877–884.

Stead, P., Hiscox, S., Robinson, P.S., Pike, N.B., Sidebottom, P.J., Roberts, A.D., Taylor, N.L., Wright, A.E., Pomponi, S.A., and Langley, D. (2000). Eryloside F, a novel penasterol disaccharide possessing potent thrombin receptor antagonist activity. *Bioorg Med Chem Lett* 10:661–664.

Struket, B., Lenarcic, B., Gruden, K., Pungercar, J., Rogelj, B., Turk, V., Bosch, D., and Jongsma, M.A. (2000). Equistatin, a protease inhibitor from the sea anemone *Actinia equina*, is composed of three structural and functional domains. *Biochem Biophys Res Commun* 269:732–736.

Suzuki, A., Matsunaga, K., Shin, H., Tabudrav, J., Shizuri, Y., and Ohizumi, Y. (2000). Bisprasin, a novel $Ca^{(2+)}$ releaser with caffeine-like properties from a marine sponge, *Dysidea* spp. acts on $Ca^{(2+)}$-induced $Ca^{(2+)}$ release channels of skeletal muscle sarcoplasmic reticulum. *J Pharmacol Exp Ther* 292:725–730.

Tan, L.T., Williamson, R.T., Gerwick, W.H., Watts, K.S., McGough, K., and Jacobs, R. (2000). *Cis,cis*- and *trans,trans*-ceratospongamide, new bioactive cyclic heptapeptides from the Indonesian red alga *Ceratodictyon spongiosum* and symbiotic sponge *Sigmadocia symbiotica. J Org Chem* 65:419–425.

Thorlacius, H., Vollmar, B., Seyfert, U.T., Vestweber, D., and Menger, M.D. (2000). The polysaccharide fucoidan inhibits microvascular thrombus formation independently from P- and L-selectin function in vivo. *Eur J Clin Invest* 30:804–810.

White, H.S., McCabe, R.T., Armstrong, H., Donevan, S.D., Cruz, L.J., Abogadie, F.C., Torres, J., Rivier, J.E., Paarmann, I., Hollmann, M., and Olivera, B.M. (2000). In vitro and in vivo characterization of conantokin-R, a selective NMDA receptor antagonist isolated from the venom of the fish-hunting snail *Conus radiatus. J Pharmacol Exp Ther* 292:425–432.

Williams, A.J., Dave, J.R., Phillips, J.B., Lin, Y., McCabe, R.T., and Tortella, F.C. (2000). Neuroprotective efficacy and therapeutic window of the high-affinity *N*-methyl-D-aspartate antagonist conantokin-G: in vitro (primary cerebellar neurons) and in vivo (rat model of transient focal brain ischemia) studies. *J Pharmacol Exp Ther* 294:378–386.

Yabe, T., Yamada, H., Shimomura, M., Miyaoka, H., and Yamada, Y. (2000). Induction of choline acetyltransferase activity in cholinergic neurons by stolonidiol: structure-activity relationship. *J Nat Prod* 63:433–435.