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Current Trends Review

Marine Products as a Source of Antiviral Drug Leads

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ABSTRACT

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A survey is presented of the occurrence of organic compounds from aquatic organisms that have been reported to have antiviral activities. Studies of the chemical structures and antiviral properties of unusual metabolic products of aquatic life have demonstrated that marine organisms offer excellent prospects in the search for antiviral drugs.

Key words: marine natural products, active principles, antiviral activity

INTRODUCTION

Antiviral chemotherapy began to evolve 3 decades ago following the successful treatment of herpes simplex keratitis with idoxuridine [Kaufman, 1962]. Since then, the progress

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of antiviral drug development has been slow, and until now the few U.S. Food and Drug Administration-approved antiviral drugs (acyclovir, amantadine, idoxuridine, ribavirin, trifluridine, vidarabine, and zidovudine) have only limited applications in a few viral diseases. The demand for new antiviral therapies is thus great. In the last few years the development of antiviral drugs has been revolutionized by two events. First, the clinical success of acyclovir has demonstrated that selective inhibition of virus-specific processes is a realistic target; second, the discovery of zidovudine soon after the identification of the human immunodeficiency virus (HIV) as a causative agent of the acquired immunodeficiency syndrome (AIDS) has initiated international concerns and interests in the search for anti-HIV agents.

There are generally two approaches in the search for pharmaceutical leads; namely, activity screening and rational drug design. For antiviral drug discovery, the screening approach remains the most common and productive method. Candidates from different sources, synthetic or natural, have been evaluated in a variety of test systems to determine their biological activities. Apart from the synthetic compounds, such as the purine-pyrimidine antimetabolites, many natural products from terrestrial plants and microorganisms have demonstrated antiviral potentials [for reviews, see Hudson, 1989; Che, 1991a,b]. There is also sufficient basis for the belief that aquatic organisms offer great promise as a source of prospective antiviral substances. Recent years have witnessed growing attention to marine organisms for their biomedical applications, and they have become the subject of many chemical and pharmaceutical investigations. Consequently, a large number of novel chemical structures have been discovered from aquatic animals, plants, and microbes [for reviews, see Scheuer, 1978–1983; Okuda et al., 1982; Faulkner, 1984a,b, 1986, 1987, 1988]. Attention to pharmacologically active substances from marine flora and fauna was first drawn by Emerson and Taft (1945), and subsequently many marine products were found to have interesting pharmacological actions [for reviews, see Nigrelli et al., 1967; Burkholder and Sharma, 1969; Der Marderosian, 1969; Ruggieri, 1976; Grant and Mackie, 1977; Kaul, 1981; Rinehart et al., 1981c; Harnden and Planterose, 1985; Braekman and Daloze, 1986; Cardellina, 1986; Kaul and Daftari, 1986; Krebs, 1986; Okami, 1986; Munro et al., 1987; Kitagawa, 1988; Scheuer, 1989].

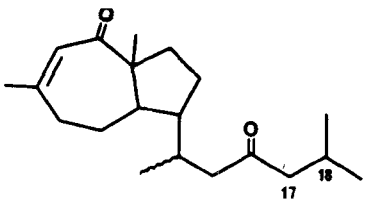
Although marine natural products have never been intensely studied for their antiviral properties, available literature data clearly indicate that a number of secondary metabolites from aquatic organisms display in vitro antiviral activities in bioassay systems. The present review summarizes these results, with the objective of demonstrating that aquatic organisms can provide an array of leads with diverse chemical structures that may have potential for being developed into antiviral drugs.

TERPENOIDS

From the deepwater sponge *Epipolasis reiswigi,* reiswigins A (1) and B (2) were isolated and found to have potent in vitro antiviral activity [Kashman et al., 1987]. Being noncytotoxic at 2 μg levels, both compounds completely inhibited herpes simplex virus type 1 (HSV-1) and vesicular stomatitis virus (VSV). Antiviral activity was also observed at 20 μg levels against coronavirus A59.

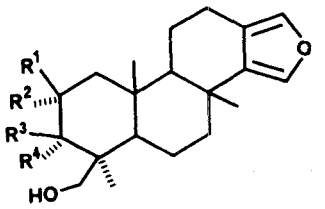
Spongiadiol (3), epispongiadiol (4), and isospongiadiol (5) are furanoditerpenes obtained from a Caribbean deepwater sponge of the genus *Spongia* [Kohmoto et al., 1987]. They exhibited antiviral activity against HSV-1, with IC_{50} values of 0.25, 12.5, and 2 μg/ml, respectively. These compounds were also cytotoxic to P388 cells.

Groweiss et al. [1988] isolated six novel diterpene lactones, solenolides A–F, from a new species of marine octocoral *Solenopodium.* When tested in antiviral bioassays, solenolide A (6) inhibited rhinovirus (IC_{50} 0.39 μg/ml), poliovirus III, herpes virus, Ann Arbor virus, and Maryland virus; solenolide D (7) inhibited Semliki forest virus and Ann Arbor virus;



1

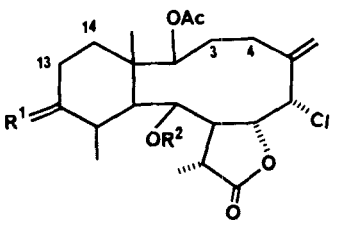
2 17,18-Dehydro-



3 $R^1 + R^2 = O$; $R^3 = H$; $R^4 = OH$

4 $R^1 + R^2 = O$; $R^3 = OH$; $R^4 = H$

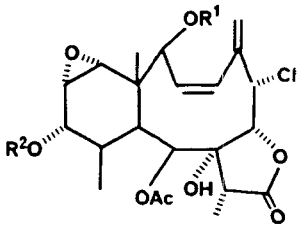
5 $R^1 = H$; $R^2 = OH$; $R^3 + R^4 = O$



6 $R^1 = \alpha\text{-}C_5H_{11}COO\text{-}$, β-H;
$R^2 = H$;
13,14-Epoxy-

7 $R^1 = \alpha\text{-}OAc$, β-H;
$R^2 = Ac$;
3,4;13,14-Diepoxy-

8 $R^1 = O$; $R^2 = H$;
13,14-Dehydro-



9 $R^1 = R^2 = COC_3H_7$

10 $R^1 = COC_3H_7$; $R^2 = Ac$

11 $R^1 = R^2 = Ac$

Fig. 1. Structures of compounds 1–11.

and solenolide E (8) suppressed rhinovirus (IC_{50} 12.5 μg/ml), herpes virus, and Ann Arbor virus.

Other diterpene lactones, such as briantheins V (9), Y (10) and Z (11) from the gorgonian coral *Briareum asbestinum*, were found to have weak antiviral activity [Coval et al., 1988]. They exhibited IC_{50} values of 400, 80, and 50 μg/ml, respectively, against coronavirus. Brianthein Z (11) was also active against HSV-1 at 80 μg/ml.

Avarol (12) and avarone (13), two antimitotic and antimutagenic sesquiterpenes from

the sea sponge *Dysidea avara,* inhibited in vitro HIV replication without showing any cytotoxic effects to noninfected cells at a concentration of 0.1 μg/ml [Sarin et al., 1987]. Derivatives of these sesquiterpenes were also claimed to be active and have been patented for their antiviral properties [Mueller, 1988]. These compounds were further demonstrated to cause T-lymphotrophic cytostatic effect in murine and human lymphocytes and have low in vivo toxicity in mice [Muller et al., 1986]. Avarol (12) was able to induce γ-interferon in human peripheral blood lymphocytes [Voth et al., 1988].

Snader and Higa [1986] have patented the chamigrene derivatives (14), obtained from the digestive gland of the sea hare *Aplysia dactylomela,* as antiviral agents against HSV and VSV.

Carter and Rinehart [1978a] reported the chemical structure of aplidiasphingosine (15) from a marine tunicate (*Aplidium* sp.). In addition to the antimicrobial and cytotoxic activities, aplidiasphingosine was antiviral against HSV-1. The compound has been synthesized to define the stereochemistry [Mori and Umemura, 1981; Umemura and Mori, 1987].

From an extract of *Laurencia venusta,* a red alga collected in Okinawan waters, three active compounds were isolated by Sakemi et al. [1986] following activity-guided fractionation procedures. The compounds were determined to be thyrsiferol (16), thyrsiferyl 23-acetate (17), and venustatriol (18), all of which are tetracyclic ethers of triterpenoid origin active against HSV-1 and VSV.

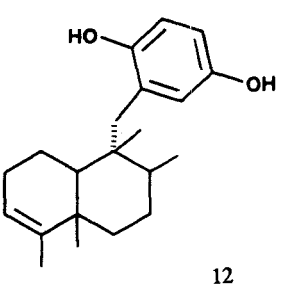
Andersson et al. [1989] evaluated 24 saponins and saponin-like substances from starfish and brittle-stars for antibacterial, cytotoxic, and antiviral activities. Of the 18 compounds tested for antiviral effects, only two reduced about 25% of the pseudorabies virus plaque formation at 10 μg/ml levels. They were crossasterosides B (19) and D (20), obtained from *Crossaster papposus.*

NUCLEOSIDES

Among the few licenced antiviral drugs in the United States, vidarabine (adenine arabinoside; Ara-A) (21) is the only naturally occurring compound, and the first to be given systemically for the treatment of herpes simplex keratitis and herpes simplex encephalitis [De Clercq, 1984]. Ara-A has been obtained from both marine and microbial sources, including the Mediterranean gorgonian *Eunicella cavolini* [Cimino et al., 1984] and the jellyfish *Stomolophus meleagris* [Betz and Der Marderosian, 1987]. Inhibition of viral DNA synthesis seems to be its major target of antiviral action, although other less specific mechanisms, such as inhibition of nucleic acid methylation by blocking the S-adenosyl homocysteine hydrolase, may contribute to the overall antiviral action. The pharmacological properties and clinical applications of ara-A have been reviewed by Muller [1979]; North and Cohen [1984]; and Buchanan and Hess [1985].

Thymine arabinoside (spongothymidine, Ara-T) (22), originally isolated from the sponge *Cryptoethya crypta* [Bergmann and Feeney, 1951] and more recently from the jellyfish *Stomolophus meleagris* [Betz and Der Marderosian, 1987], has also been well documented as an antiviral agent [North and Cohen, 1984]. It is phosphorylated by the herpes virus-induced thymidine kinase, and in its triphosphate form it inhibits the DNA polymerase reaction.

A number of nucleoside analogs have been identified for their potent antiviral activities [North and Cohen, 1984; Robins and Revankar, 1988]. Despite their antiviral efficacy, the therapeutic value of many nucleosides is limited because of the mutagenic, teratogenic, carcinogenic, or cytotoxic side effects. In recent years, however, the selectivity of this class of compounds has been enhanced through structure-activity relationship studies. The success of acyclovir [9-(2-hydroxyethoxymethyl)guanine] in clinical trials demonstrates not only that structural modifications can improve the therapeutic value of lead compounds but, more importantly, that selective inhibition of viral replication is a valid approach to antiviral therapy.



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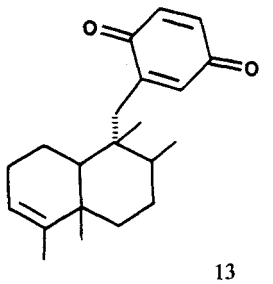

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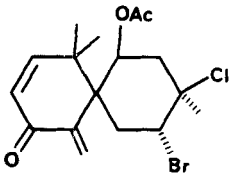

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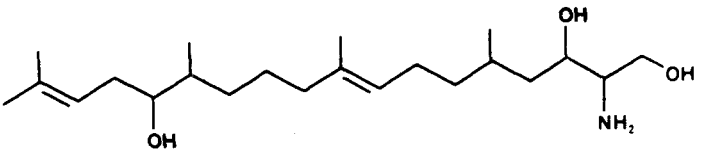

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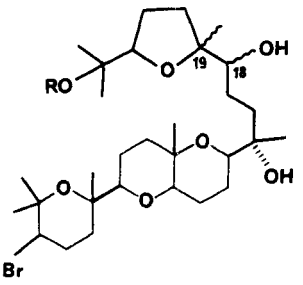


16 R = H; 18β-OH; 19β-Me

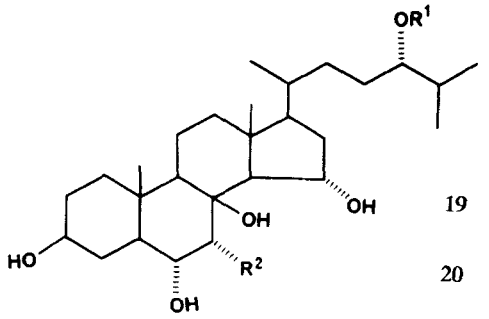
17 R = Ac; 18β-OH; 19β-Me

18 R = H; 18α-OH; 19α-Me

Fig. 2. Structures of compounds 12–18.

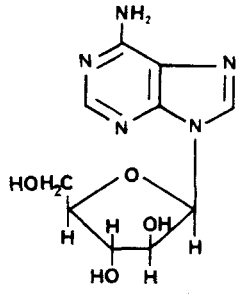
ALKALOIDS AND OTHER NITROGEN-CONTAINING COMPOUNDS

From a Caribbean tunicate, *Eudistoma olivaceum*, seventeen β-carboline alkaloids (eudistomins) have been isolated [Kobayashi et al., 1984; Rinehart et al., 1984, 1987; Rinehart, 1989]. They belong to four structural groups, including simple β-carbolines, pyrrolyl β-carbolines, pyrrolinyl β-carbolines, and tetrahydro-β-carbolines containing an oxathiazepine ring. Among these metabolites, eudistomins C (23) and E (24), bearing the oxathiazepine



19 R^1 = 4-OMe-Xyl-(1→2)-3-OMe-Xyl-
R^2 = H

20 R^1 = Xyl-(1→2)-3-OMe-Xyl-
R^2 = OH



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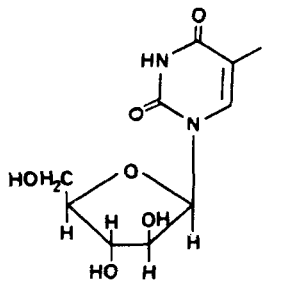

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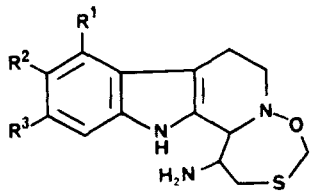
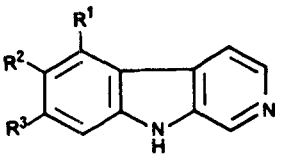


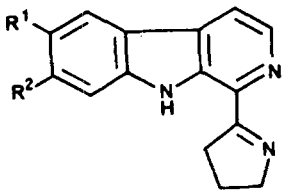
	R^1	R^2	R^3	
23	H	OH	Br	
24	Br	OH	H	
25	H	H	Br	
26	H	H	Br	S→O
27	H	H	H	
28	H	Br	H	

Fig. 3. Structures of compounds 19–28.

moiety, were most active against HSV-1 (5–10 ng/disk) and HSV-2 (25 ng/disk). Eudistomins D (29), H (33), K (25), L (28), N (30), and P (34) were less active (100–500 ng/disk). Metabolites of the same class have also been obtained from the unrelated New Zealand ascidian *Ritterella sigillinoides* [Blunt et al., 1987; Lake et al., 1988, 1989]; they included eudistomin C (23), eudistomin K (25), eudistomin K sulfoxide (26), debromoeudistomin K (27), eudistomin O (31), and β-carboline (32). Debromoeudistomin K (27) and eudistomin K sulfoxide (26) showed antiviral activity against HSV-1 at concentration levels of 400 ng/disk, while eudistomin O (31) and the β-carboline (32) were less active (500–2,000 ng/disk). The antiviral applications of eudistomins A (35) and M (36), or their pharmaceutically acceptable salts, have also been patented [Rinehart, 1985].

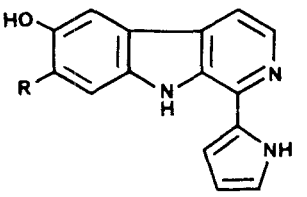


	R^1	R^2	R^3
29	Br	OH	H
30	H	Br	H
31	H	H	Br
32	H	H	H



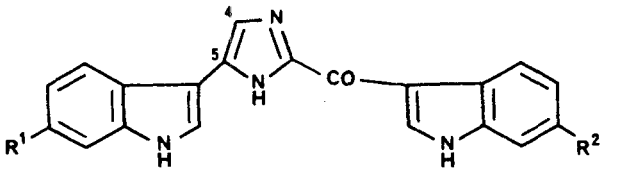
33 R^1 = Br; R^2 = H

34 R^1 = OH; R^2 = Br



35 R = Br

36 R = H



37 R^1 = H; R^2 = OH

38 R^1 = Br; R^2 = OH

39 R^1 = Br; R^2 = H; 4,5-Dihydro-

Fig. 4. Structures of compounds 29–39.

A series of bis-indole alkaloids including topsentin (37), bromotopsentin (38), and 4,5-dihydro-6″-deoxybromotopsentin (39), were isolated from a Mediterranean shallow water sponge *Topsentia genitrix* by Bartik et al. [1987] as defense chemicals. Tsujii et al. [1988] obtained the same compounds from a Caribbean deep sea sponge of the genus *Spongosorites* and reported the antiviral activity of these metabolites. Topsentin (37) showed activity against HSV-1 (50 μg/disk), coronavirus A59 (2 μg/disk), and VSV; bromotopsentin (38) was active against HSV-1 (200 μg/disk) and coronavirus A59 (10 μg/disk), whereas dihydrodeoxybromotopsentin (39) was active only against coronavirus A59 (2 μg/disk). These compounds also had varying degrees of cytotoxicity against P388 cells.

A pentacyclic aromatic alkaloid dercitin (40) was isolated from a deepsea sponge (*Dercitus* sp.) and found to be anti-HIV at concentrations of 5 μg/well [Gunawardana et al., 1988]. The compound also exhibited cytotoxic and immunosuppressive activities. Sakai and Higa [1987] isolated tubastrine (41) from *Tubastrea aurea* at the coral reefs of Okinawa. The compound was active against HSV-1 and VSV.

Acarnidines a–c (42–44), containing substituted homospermidine skeleton, were isolated from the sponge *Acarnus erithacus* from the Gulf of California [Carter and Rinehart, 1978b]. They exhibited antiviral activity against HSV-1 at 80 μg/disk levels.

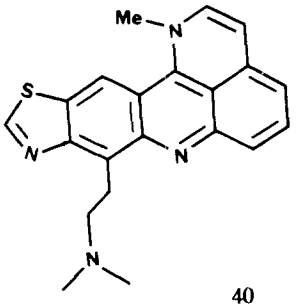
From a colonial tunicate (*Polyandrocarpa* sp.), Cheng and Rinehart [1978] reported two antibacterial, cytotoxic and antiviral substances called polyandrocarpidines I and II. Later, Carte and Faulkner [1982] demonstrated that these compounds were isomeric mixtures of four components, polyandrocarpidines A–D (45–48). A mixture of these compounds displayed weak antiviral activity against HSV-1 [Cheng and Rinehart, 1978].

Didemnins are a group of cyclic depsipeptides isolated from a Caribbean tunicate (*Tridemnum* sp.). In addition to didemnins A (49), B (50), and C (51), initially obtained by Rinehart et al. [1981a–c], more than ten members of this class have now been isolated or chemically prepared [Rinehart et al., 1990]. These compounds not only possess antiviral properties but also have antitumor and immunosuppressive activities. Didemnins A (49) and B (50) inhibited the replication of HSV-1 and HSV-2, at concentrations of 1 and 0.05 μM, respectively. Similar efficacy was demonstrated against cosxackie virus A21, equine rhinovirus, and parainfluenza virus 3 [Rinehart et al., 1981a,b]. When the study was extended to other RNA viruses, including the highly virulent human pathogens Rift Valley fever virus, Venezuelan equine encephalomyelitis virus, and yellow fever virus, inhibition was observed at concentrations ranging from 0.08 to 1.4 μg/ml. Doses of 1.25–5.0 mg/kg didemnin A and 0.25 mg/kg didemnin B administered subcutaneously for 5 days to mice infected with a lethal dose of Rift Valley fever virus significantly increased the numbers of survivors, although drug-related toxicity was also observed [Canonico et al., 1982]. Additionally, when didemnin A was applied intravaginally to HSV-2-infected mice at a dose of 1 mg/ml, three times per day for 3 days, 60% of the animals were protected. Didemnin B was as effective, even at lower doses (0.23 mg/ml) [Rinehart et al., 1983]. Despite the significant antiviral properties, didemnins are cytotoxic and inhibitory to cellular DNA, RNA, and protein synthesis at concentrations close to those at which viral growth was affected [Rinehart et al., 1983]. Consequently, they have both a low antiviral selectivity and therapeutic index. Nevertheless, some improvement in antiviral/cytotoxicity ratios have been achieved through structural modifications, and further investigations are warranted to yield better results.

Rinehart and his associates have reported a series of dimeric bromopyrroles from the sponges *Agelas coniferin* and *A.* cf. *mauritiana* [Rinehart, 1988, 1989; Rinehart et al., 1990], including the previously known sceptrin (52) [Walker et al., 1981], its debromo (53)- and dibromo (54)- derivatives, oxysceptrins (55, 56), and ageliferins (57–59). These compounds all showed antiviral activity against HSV-1 at concentrations as low as 20 μg/disk.

POLYSACCHARIDES

The antiviral activity of polysaccharides has been recognized for a number of years. As early as 1947, polysaccharides from diverse sources were found to inhibit viral growth [Ginsberg et al., 1947; Green and Wooley, 1947; Horsfall and McCarty, 1947]. Gerber et al. [1958] then reported the antiviral effects of a polysaccharide extract from seaweeds against influenza B virus and mumps virus in embryonated eggs. Later, agar polysaccharides were also demonstrated to inhibit replications of encephalomyocarditis virus, poliovirus, and herpes virus [Schulze, 1964; Takemoto and Fabisch, 1964; Takemoto and Spicer, 1965]. Kathan [1965] examined extracts of kelp and reported inhibitory activities against the multiplication of influenza virus and viral neuraminidases.



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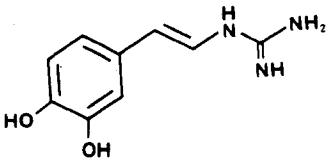

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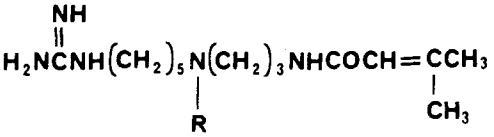
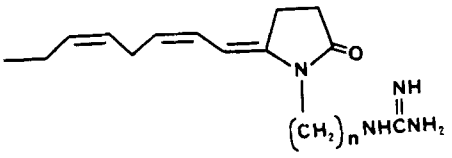


42 R = $-COC_{11}H_{23}$

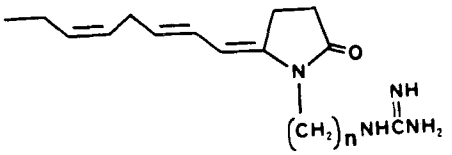
43 R = $-CO(CH_2)_3CH{=}CH(CH_2)_5CH_3$ (cis)

44 R = $-COC_{13}H_{21}$



45 n = 5

47 n = 4



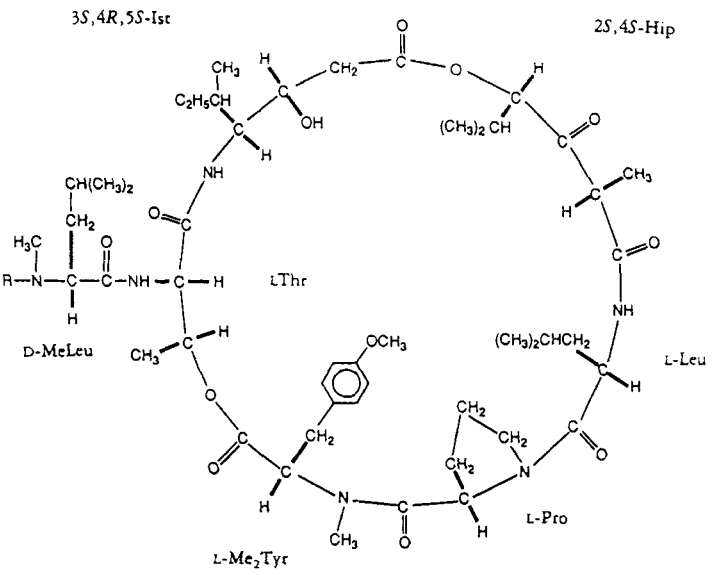
46 n = 5

48 n = 4

Fig. 5. Structures of compounds 40–48.

The polysaccharide-rich fractions from two marine red algae, *Cryptosyphonia woodii* and *Farlowia mollis*, were found to exhibit in vitro antiviral activity against HSV-1 and HSV-2, vaccinia virus, and VSV [Deig et al., 1974; Richards et al., 1978]. The extracts were also shown to protect mice from HSV-2 infection but had no therapeutic effects when applied postinfectiously.

In recent years, sulfated polysaccharides have been demonstrated to inhibit in vitro replication of many enveloped viruses, including the HIV [Baba et al., 1988; Sugawara et al.,



49 R = H

50 R = CH_3CHOHC-N(-C(=O)-)-C(=O)-

51 R = $CH_3CH(OH)CO$-

Fig. 6. Structures of compounds 49–51.

1989]. A sulfated polysaccharide called SAE, isolated from the sea alga *Schizymenia pacifica,* also displayed inhibitory effects on HIV replication and on reverse transcriptase activity [Nakashima et al., 1987a,b].

Sulfated polysaccharides are an interesting class of compounds showing potent in vitro antiviral activity, presumably due to the interference with the viral adsorption process. Although many of them have become useful probes for antiviral research, cytotoxicity, and other undesirable side effects (such as anticoagulation) may preclude any clinical application. Low oral bioavailability of these compounds also constitutes a hindrance in drug development.

MISCELLANEOUS COMPOUNDS

In a screen of New Zealand marine invertebrates, a sponge of the genus *Mycale* showed significant in vitro and in vivo antiviral activities [Perry et al., 1988]. The extract protected coronavirus-infected mice at a dose of 0.1 mg/kg. Subsequently, two active metabolites were determined to be mycalamides A (60) and B (61) [Perry et al., 1988, 1990]. Both compounds were very active against HSV-1 and poliovirus I (MICs 3.5–5 ng/disk and 1–2 ng/disk, respectively, for mycalamides A and B). No in vivo results have been reported for these compounds.

From a sponge of the genus *Theonella* collected in Okinawan waters, an antiviral

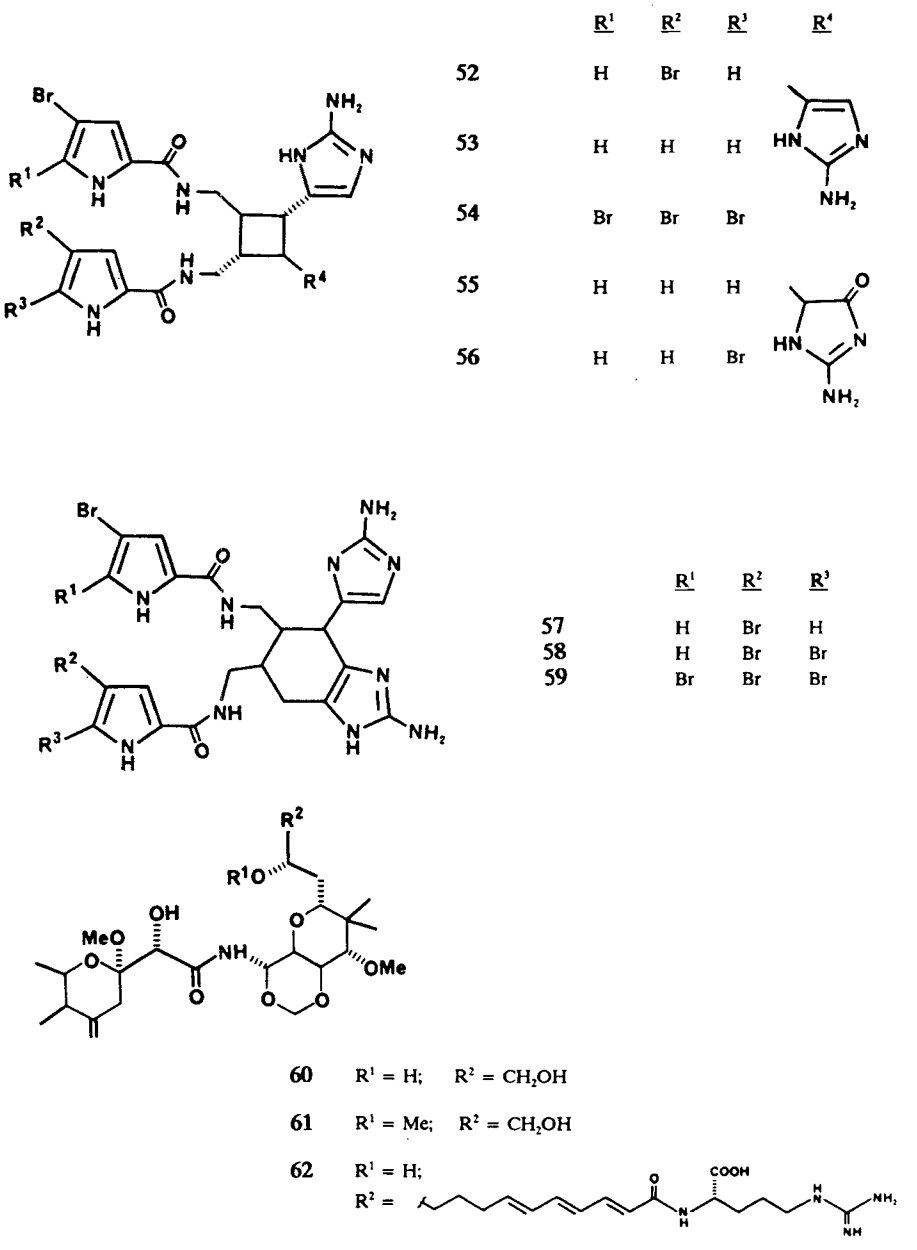


Fig. 7. Structures of compounds 52–62.

metabolite onnamide A (62) was isolated by Sakemi et al. [1988]. It displayed potent in vitro antiviral activity against HSV-1, VSV, and coronavirus A59.

From an unidentified marine bacterium, Gustafson et al. [1989b] reported eight new secondary metabolites. One of them, macrolactin A (63), showed significant inhibition of

HSV-1 and HSV-2 (IC_{50}s 5.0 and 8.3 μg/ml, respectively). More importantly, it protected T-lymphoblast cells against HIV replication, at concentrations of 10 μg/ml.

Gustafson et al. [1989a] isolated a group of sulfonic acid-containing glycolipids (64–67) from anti-HIV extracts of the cyanobacteria (blue-green algae) *Lyngbya lagerheimii* and *Phormidium tenue*. These sulfolipids represent a new structural class of anti-HIV compounds currently under preclinical investigation at the National Cancer Institute. Several other cyanobacterial culture extracts showing positive test results for the presence of sulfolipids were also active in the in vitro anti-HIV assay.

CRUDE EXTRACTS SHOWING ANTIVIRAL ACTIVITY

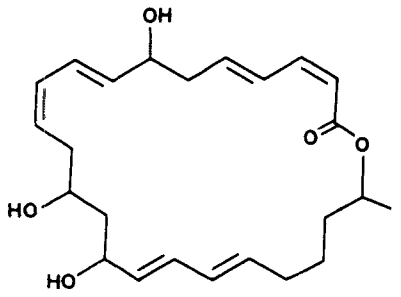
Li and his associates have studied a number of extracts from marine animals for antibacterial and antiviral activities. Their results indicated that some macromolecules (probably glycoproteins or mucoproteins) present in abalone (*Haliotis rufescens*), oyster (*Crassostrea virginica*), clam (*Mercenaria mercenaria*), queen conch (*Strombus gigas*), squid (*Loligo pealii*), and sea snail (*Tegula gallina*) possessed significant antibacterial and antiviral activities both in vitro and in vivo [Li, 1960; Li et al., 1962a,b; Prescott et al., 1964]. The antiviral fraction, designated as Paolin 2, from abalone and oyster extracts inhibited the replication of polyoma virus, influenza A virus, and poliovirus in tissue cultures, and protected mice infected with poliovirus and influenza virus [Li et al., 1962a,b]. Additionally, ammonium sulfate extracts prepared from the common clam have been demonstrated to suppress in vitro replication of HSV and adenovirus type 12 and reduce tumor formation in hamsters induced by the adenovirus [Li et al., 1965].

Andersson et al. [1983] examined extracts from 25 marine organisms collected along the Swedish coast for biological activities, including the inhibition of plaque formation with HSV-1 and influenza virus (WSN strain). Only four extracts exhibited suppressive effects on HSV-1 at concentrations of 100 μg/ml without showing toxicity. They were, namely, chloroform extract of *Fucus serratus* (brown seaweed), petroleum ether extract of *Laminaria digitata* (brown seaweed), petroleum ether extract of *Geodia baretti* (sponge), and petroleum ether extract of *Echinus esculentus* (sea urchin). Organisms showing both antiviral and toxic effects included *Halidrys siliquosa* (brown seaweed), *Laminaria saccharina* (brown seaweed), *Ceramium rubrum* (red seaweed), *Chondrus crispus* (red seaweed), *Phakellia ventilabrum* (sponge), *Halichondria bowerbanki* (sponge), *Flustra foliacea* (bryozoan), *Alcyonidium hirsutum* (bryozoan), *Macandrevia cranium* (brachiopoda), *Asterias rubens* (common starfish), *Ascidia mentula* (tunicate), and *Ascidia obliqua* (tunicate).

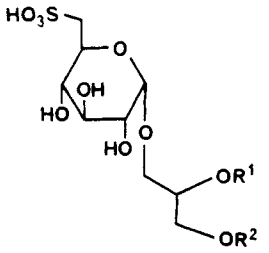
Girre et al. [1987] reported that an extract of the marine alga *Ascophyllum nodosum* inhibited the replication of HSV-1. The MeOH/toluene extracts of two sponge specimens, identified as *Sarcotragus* sp. and *Ircinia* sp., displayed significant in vitro antiviral activity against HSV-1 and poliovirus I [Barrow et al., 1988a,b]. A group of furanosesterterpene tetronic acids, of which variabilin (68) was the major component, were isolated, but none of them displayed significant antiviral activity. The active principle(s) present in these extracts remain unknown.

CONCLUDING REMARKS

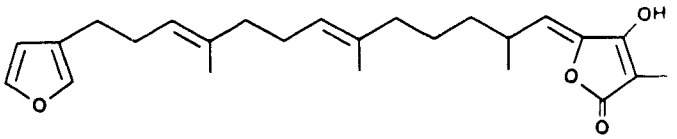
Recent years have witnessed great advances in antiviral chemotherapy. The discovery of acyclovir and zidovudine has produced a marked impact on antiviral drug development. For many years, antiviral research was hampered by inherent problems such as inconsistency of testing assays, lack of appropriate animal models, and incomplete understanding of viral pathogenesis. The biggest problem lies in the fact that viruses share many common cellular mechanisms with infected cells, and it is thus difficult to interfere selectively with viral growth without affecting normal cell growth. As our knowledge of the molecular details of virion



63



	R¹	R²
64	16:0 Fatty acid ester	18:3 Fatty acid ester
65	16:0	18:2
66	16:0	18:1
67	16:0	16:1



68

Fig. 8. Structures of compounds 63–68.

structure and viral replication expands, it will be possible to design target-oriented assay systems to detect chemicals that can selectively interfere with the viral life cycle. It seems also possible to design compounds that are activated only by virus-induced enzymes in the infected cells, so that normal cells will not be affected.

Despite the fact that most antiviral studies have focused on synthetic compounds such as the nucleoside analogs, results presented in this review clearly indicate that a variety of marine natural products possess antiviral potential. It may be true that many of these compounds may never be developed into therapeutic preparations for different reasons (e.g., toxicity, unfavorable pharmacological properties, or bioavailability problems), but it is likely that among them there are several selectively toxic substances that may eventually become

useful in the clinic. It has also to be realized that, until now, only a small number of marine products have been tested for antiviral activity; most have never been evaluated.

There are several reasons to believe that marine products are an interesting group of compounds for antiviral investigation. First, for many years marine organisms have been relatively unexplored from a chemical and pharmaceutical point of view. Major problems were mostly technical ones, such as unavailability of marine specimens, uncertainty of taxonomic classification of marine organisms, difficulties in recollection of the same species, and scarity of isolated compounds for structural and biological determinations. The past few decades have witnessed substantial progress in oceanography and separation chemistry for overcoming these problems. With a close collaboration between marine biologists, organic chemists, and virologists, the abundant marine flora and fauna would become an important source of materials for pharmaceutical research, including antiviral drug development. Second, natural products have been known to be a rich source of enzyme inhibitors, many of which possess significant selectivity against specific enzymes. It is therefore reasonable to believe that, among the enormous variety of marine products, there are selective inhibitors of virus-coded enzymes that may possess the potency and selectivity required for an antiviral agent. Third, difficulties in finding effective antiviral drugs have been partly due to the frequent mutation of the virus into drug-resistant strains. Consequently, combination therapy using drugs of different mechanisms of action may be of benefit. Therefore, a strategy in the search for antiviral compounds is to obtain leads of diverse chemical structures with different action mechanisms. The aquatic habitat enables living organisms to biosynthesize a variety of secondary metabolites whose structures are unlike any compound found in terrestrial species or chemically synthesized. These unique structures may interact with biological systems in novel manners and thus can serve as models in the development of new drugs.

In summary, literature data have demonstrated that aquatic organisms are producing a variety of unique chemical compounds displaying antiviral activity. It is reasonable to postulate that some of these metabolites, or their derivatives, can be further developed into antiviral drugs. Moreover, as a class of natural products that has been relatively unexplored, marine products certainly deserve further attention from chemists and pharmacologists.

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REFERENCES

Andersson, L., Bohlin, L., Iorizzi, M., Riccio, R., Minale, L. and Moreno-Lopez, W.: Biological activity of saponins and saponin-like compounds from starfish and brittle-stars. Toxicon **27:**179–188, 1989.

Andersson, L., Lidgren, G., Bohlin, L., Magni, L., Ogren, S. and Afzelius, L.: Studies of Swedish marine organisms. I. Screening of biological activity. Acta Pharmacol. Suecica **20:**401–414, 1983.

Baba, M., Snoeck, R., Pauwels, R. and De Clercq, E.: Sulfated polysaccharides are potent and selective inhibitors of various enveloped viruses, including simplex virus, cytomegalovirus, vesicular stomatitis virus, and human immunodificiency virus. Antimicrob. Agent Chemother. **32:**1742–1745, 1988.

Barrow, C.J., Blunt, J.W., Munro, M.H.G. and Perry, N.B.: Variabilin and related compounds from a sponge of the genus *Sarcotragus*. J. Nat. Prod. **51:**275–281, 1988a.

Barrow, C.J., Blunt, J.W., Munro, M.H.G. and Perry, N.B.: Oxygenated furanosesterterpene tetronic acids from a sponge of the genus *Ircinia*. J. Nat. Prod. **51:**1294–1298, 1988b.

Bartik, K., Braekman, J.C., Daloze, D., Stoller, C., Huysecom, J., Vandevyver, G. and Ottinger, R.: Topsentins, new toxic bis-indole alklaoids from the marine sponge *Topsentia genitrix*. Can. J. Chem. **65:**2118–2121, 1987.

Bergmann, W. and Feeney, R.: Contribution to the study of marine products. XXXII. The nucleosides of sponges. J. Org. Chem. **16:**981–987, 1951.

Betz, J.M. and Der Marderosian, A.H.: Antiviral/antineoplastic agents from a jellyfish (*Stomolophus meleagris* L. Agassiz 1860). Abstract No. 113, twenty-eighth Annual Meeting of the American Society of Pharmacognosy, Kingston, Rhode Island, July 19–22, 1987.

Blunt, J.W., Lake, R.J., Munro, M.H.G. and Toyokuni, T.: The stereochemistry of eudistomins C, K, E, F and L. Tetrahedron Lett. **28:**1825–1826, 1987.

Braekman, J.C. and Daloze, D.: Chemical defence in sponges. Pure Appl. Chem. **58:**357–364, 1986.

Buchanan, R.A. and Hess, F.: Vidarabine (Vira-A), pharmacology and clinical experience. In Shugar, D. (ed): "Viral Chemotherapy, Vol. 2." New York: Pergamon Press, 1985.

Burkholder, P.R. and Sharma, G.M.: Antimicrobial agents from the sea. Lloydia **32:**466–483, 1969.

Canonico, P.G., Pannier, W.L., Huggins, J.W. and Rinehart, K.L.: Inhibition of RNA viruses in vitro and in Rift Valley fever infected mice by didemnins A and B. Antimicrob. Agents Chemother. **22:**696–697, 1982.

Cardellina, J.H., II: Marine natural products as lead to new pharmaceutical and agrochemical agents. Pure Appl. Chem. **58:**365–374, 1986.

Carte, B. and Faulkner, D.J.: Revised structures for the polyandrocarpidines. Tetrahedron Lett. **23:** 3863–3866, 1982.

Carter, G.T. and Rinehart, K.L. Jr.: Aplidiasphingosine, an antimicrobial and antitumor terpenoid from an *Aplidium* sp. (marine tunicate). J. Am. Chem. Soc. **100:**7441–7442, 1978a.

Carter, G.T. and Rinehart, K.L. Jr.: Acarnidines, novel antiviral and antimicrobial compounds from the sponge *Acarnus erithacus*. J. Am. Chem. Soc. **100:**4302–4304, 1978b.

Che, C.T.: Plants as a source of potential antiviral agents. In Wagner, H. and Farnsworth N.R. (eds.): "Economic and Medicinal Plant Research, Vol. 5." New York: Academic Press, 1991a.

Che, C.T.: Potential antiviral agents from microbial organisms. (In preparation).

Cheng, M.T. and Rinehart, K.L. Jr.: Polyandrocarpidines: antimicrobial and cytotoxic agents from a marine tunicate (*Polyandrocarpa* sp.) from the Gulf of California. J. Am. Chem. Soc. **100:** 7409–7411, 1978.

Cimino, G., De Rosa, S. and De Stefano, S.: Antiviral agents from a gorgonian, *Eunicella cavolini*. Experientia **40:**339–340, 1984.

Coval, S.J., Cross, S., Bernardinelli, G. and Jefford, C.W.: Brianthein V, a new cytotoxic and antiviral diterpene isolated from *Briareum asbestinum*. J. Nat. Prod. **51:**981–984, 1988.

De Clercq, E.: Antiherpes drugs: Promise and pitfalls. Eur. J. Clin. Microbiol. **3:**96–107, 1984.

Deig, E.F., Ehresmann, D.W., Hatch, M.T. and Riedlinger, D.J: Inhibition of herpesvirus replication by marine algae extracts. Antimicrob. Agents Chemother. **6:**524–525, 1974.

Der Marderosian, A.: Marine pharmaceuticals. J. Pharmacol. Sci. **58:**1–33, 1969.

Emerson, G.A. and Taft, C.H.: Pharmacologically active agents from the sea. Texas Rep. Biol. Med. **3:**302–338, 1945.

Faulkner, D.J.: Marine natural products: Metabolites of marine invertebrates. Nat. Prod. Rep. **1:**551–598, 1984a.

Faulkner, D.J.: Marine natural products: metabolites of marine algae and herbivorous marine molluscs. Nat. Prod. Rep. **1:**251–280, 1984b.

Faulkner, D.J.: Marine natural products. Nat. Prod. Rep. **3:**1–33, 1986.

Faulkner, D.J.: Marine natural products. Nat. Prod. Rep. **4:**539–576. 1987.

Faulkner, D.J.: Marine natural products. Nat. Prod. Rep. **5:**613–663, 1988.

Gerber, P., Dutcher, J.D., Adams, E.V. and Sherman, J.H.: Protective effect of seaweed extracts for chicken embryos infected with influenza B of mumps virus. Proc. Soc. Exp. Biol. Med. **99:** 590–593, 1958.

Ginsberg, H.S., Goebel, W.F. and Horsfall, F.L. Jr.: Inhibition of mumps virus multiplication by a polysaccharide. Proc. Soc. Exp. Biol. Med. **66:**99–100, 1947.

Girre, L., Amoros, M., Conan, M., Percehais, S., Yvin, J.C. and Zerial, A.: Sur l'activite antiherpetique d'extraits de vegetaux d'origine marine our terrestre et la standardisation de l'etude des proprietes antivirales. Fitoterapia **58:**371–378, 1987.

Grant, P.T. and Mackie, A.M.: Drugs from the sea—Fact of fantasy? Nature **267:**786–788, 1977.

Green, R.H. and Wooley, D.W.: Inhibition by certain polysaccharides of hemagglutination and multiplication of influenza virus. J. Exp. Med. **86:**55–64, 1947.

Groweiss, A., Look, S.A. and Fenical, W.: Solenolides, new antiinflammatory and antiviral diterpenoids from a marine octocoral of the genus *Solenopodium*. J. Org. Chem. **53:**2401–2406, 1988.

Gunawardana, G.P., Kohmoto, S., Gunasekera, S.P., McConnell, O.J. and Koehn, F.E.: Dercitin, a new biologically active acridine alkaloid from a deep water marine sponge, *Dercitus* sp. J. Am. Chem. Soc. **110:**4856–4858, 1988.

Gustafson, K.R., Cardellina, J.H. II, Fuller, R.W., Weislow, O.S., Kisers, R.F., Snader, K.M., Patterson, G.M.L. and Boyd, M.R.: AIDS-antiviral sulfolipids from cyanobacteria (blue-green algae). JNCI **81:**1254–1258, 1989a.

Gustafson, K., Roman, M. and Fenical, W.: The macrolactins, a novel class of antiviral and cytotoxic macrolides from a deepsea marine bacterium. J. Am. Chem. Soc. **111:**7519–7524, 1989b.

Harnden, M.R. and Planterose, D.N.: Screening leads—Natural products. In Harnden, M.R. (ed): "Approaches to Antiviral Agents", Weinheim, Germany: VCH Press, 1985.

Horsfall, F.L. Jr. and McCarty, M.: The modifying effects of certain substances of bacterial origin on the course of infection with pneumonia virus of mice (PVM). J. Exp. Med. **85:**623–646, 1947.

Hudson, J.B.: "Antiviral Compounds From Plants." Boca Raton, FL: CRC Press, 1989.

Kashman, Y., Hirsch, S., Koehn, F. and Cross, S.: Reiswigins A and B, novel antiviral diterpenes from a deepwater sponge. Tetrahedron Lett. **28:**5461–5464, 1987.

Kathan, R.H.: Kelp extracts as antiviral substances. Ann. N.Y. Acad. Sci. **130:**390–397, 1965.

Kaufman, H.E.: Clinical cure of herpes simplex keratitis by 5-iodo-2′-deoxyuridine. Proc. Soc. Exp. Biol. Med. **109:**251–252, 1962.

Kaul, P.N.: Compounds from the sea with actions on the cardiovascular and central nervous systems. Fed. Proc. **40:**10–14, 1981.

Kaul, P.N. and Daftari, P.: Marine pharmacology: bioactive molecules from the sea. Annu. Rev. Pharmacol. Toxicol. **26:**117–142, 1986.

Kitagawa, I.: Bioactive marine natural products. Yakugaku Zasshi **108:**398–416, 1988.

Kobayashi, J., Harbour, G.C., Gilmore, J. and Rinehart, K.L. Jr.: Eudistomins A, D, G,H, I, J, M, N,O, P, and Q, bromo-, hydroxy-, pyrrolyl-, and 1-pyrrolinyl-β-carbolines from the antiviral Caribbean tunicate *Eudistoma olivaceum*. J. Am. Chem. Soc. **106:**1526–1528, 1984.

Kohmoto, S., McConnell, O.J., Wright, A. and Cross, S.: Isospongiadiol, a cytotoxic and antiviral diterpene from a Caribbean deep water marine sponge, *Spongia* sp. Chem. Lett. 1687–1690, 1987.

Krebs, H.C.: Recent developments in the field of marine natural products with emphasis on biologically active compounds. In Herz, W., Grisebach, H., Kirby, G.W. and Tamm, C. (eds): "Progress in the Chemistry of Organic Natural Products," Vol. 49, New York: Springer-Verlag, 1986.

Lake, R.J., Blunt, J.W. and Munro, M.H.G.: Eudistomins from the New Zealand ascidian *Ritterella sigillinoides*. Aust. J. Chem. **42:**1201–1206, 1989.

Lake, R.J., Brennan, M.M., Blunt, J.W., Munro, M.H.G. and Pannell, L.K.: Eudistomin K sulfoxide, an antiviral sulfoxide from the New Zealand ascidian *Ritterella sigillinoides*. Tetrahedron Lett. **29:**2255–2256, 1988.

Li, C.P.: Antimicrobial effect of abalone juice. Proc. Soc. Exp. Biol. Med. **103:**522–524, 1960.

Li, C.P., Prescott, B., Eddy, B., Caldes, G., Green, W.R., Martino, E.C. and Young A.M.: Antiviral activity of paolins from clams. Ann. N.Y. Acad. Sci. **130:**374–382, 1965.

Li, C.P., Prescott, B. and Jahnes, W.G.: Antiviral activity of a fraction of abalone juice. Proc. Soc. Exp. Biol. Med. **109:**534–538, 1962a.

Li, C.P., Prescott, B., Jahnes, W.G., and Martino, E.C.: Antimicrobial agents from mollusks. Trans. N.Y. Acad. Sci., Series II, **24:**504–509, 1962b.

Mori, K. and Umemura, T.: Stereochemistry of aplidiasphingosine as proposed by the asymmetric synthesis and ^{13}C-NMR study of sphingosine relatives. Tetrahedron Lett. **22:**4433–4436, 1981.

Mueller, W.E.G.: Pharmaceuticals containing avarone or avarol for the treatment of AIDS and AIDS-related complex. Ger. Offen. DE 3,619,202 10 Dec. 1987. Chem. Abstr. **110:**29094j, 1988.

Muller, W.E.G.: Mechanisms of action and pharmacology: Chemical agents. In Glasso, G.J., et al. (eds): "Antiviral Agents and Viral Diseases of Man." New York: Raven Press, 1979.

Muller, W.E.G., Sobel, C., Sachsse, W., Diehl-Seifert, B., Zahn, R.K., Eich, E., Kljajic, Z. and Schroder, H.C.: Biphasic and differential effects of the cytostatic agents avarone and avarol on DNA metabolism of human and murine T and B lymphocytes. Eur. J. Cancer Clin. Oncol. **22:**473–476, 1986.

Munro, M.H.G., Leibrand, R.T. and Blunt, J.W.: The search for antiviral and anticancer compounds from marine organisms. In Scheuer, P.J., (ed): "Bioorganic Marine Chemistry, Vol. 1." Berlin: Springer-Verlag, 1987.

Nakashima, H., Kido, Y., Kobayashi, N., Motoki, Y., Neushul, M. and Yamamoto, N.: Antiretroviral activity in a marine red alga: reverse transcriptase inhibition by an aqueous extract of *Schizymenia pacifica*. J. Cancer Res. Clin. Oncol. **113:**413–416, 1987a.

Nakashima, H., Kido, Y., Kobayashi, N., Motoki, Y., Neushul. M. and Yamamoto, N.: Purification and characterization of an avian myeloblastosis and human immunodeficiency virus reverse transcriptase inhibitor, sulfated polysaccharides extracts from sea algae. Antimicrob. Agent Chemother. **31:**1524–1528, 1987b.

Nigrelli, R.F., Stempien, M.F. Jr., Ruggieri, G.D., Liguori, V.R. and Cecil, J.T.: Substances of potential biomedical importance from marine organisms. Fed. Proc. **26:**1197–1205, 1967.

North, T.W. and Cohen, S.S.: Aranucleosides and aranucleotides in viral chemotherapy. In Shugar, D. (ed): "Viral Chemotherapy, Vol. 1." New York: Pergamon Press, 1984.

Okami, Y.: Marine microorganisms as a source of bioactive agents. Microb. Ecol. **12:**65–78, 1986.

Okuka, R.K., Klein, D., Kinnel, R.B., Li, M. and Scheuer, P.J.: Marine natural products, the past twenty years and beyond. Pure Appl. Chem. **54:**1907–1914, 1982.

Perry, N.B., Blunt, J.W., Munro, M.H.G. and Pannell, L.K.: Mycalamide A, an antiviral compound from a New Zealand sponge of the genus *Mycale*. J. Am. Chem. Soc. **110:**4850–4851, 1988.

Perry, N.B., Blunt, J.W., Munro, M.H.G. and Thompson, A.M.: Antiviral and antitumor agents from a New Zealand sponge, *Mycale* sp. 2. Structures and solution conformations of mycalamides A and B. J. Org. Chem. **55:**223–227, 1990.

Prescott, B., Li, C.P., Martino, E.C. and Caldes, G.: Isolation and characterization of antiviral substances from marine animals. Fed. Proc. **23:**508, 1964.

Richards, J.T., Kern, E.R., Glasgow, L.A. Overall, J.C. Jr., Deig, E.F. and Hatch, M.T.: Antiviral activity of extracts from marine algae. Antimicrob. Agents Chemother. **14:**24–30, 1978.

Rinehart, K.L. Jr.: Antibiotics eudistomins. Eur. Pat. Appl. EP 133,000, 13 Feb. 1985. Chem. Abstr. **102:**226023w, 1985.

Rinehart, K.L. Jr.: Antiviral and antibacterial bromopyrroles from *Agelas coniferin* and halo derivatives thereof. U.S. Patent No. 4,737,510, 12 Apr. 1988. Chem. Abstr. **109:**216002u, 1988.

Rinehart, K.L.: Biologically active marine natural products. Pure Appl. Chem. **61:**525–528, 1989.

Rinehart, K.L. Jr., Gloer, J.B., Cook J.C. Jr., Mizsak, S.A. and Scahill, T.A.: Structures of the didemnins, antiviral and cytotoxic depsipeptides from a Caribbean tunicate. J. Am. Chem. Soc. **103:**1857–1859, 1981a.

Rinehart, K.L. Jr., Gloer, J.B., Hughes, R.G. Jr., Renis, H.E., McGovren, J.P., Swynenberg, E.B., Stringfellow, D.A., Kuentzel, S.L. and Li, L.H.: Didemnins, antiviral and antitumor depsipeptides from a Caribbean tunicate. Science **212:**933–935, 1981b.

Rinehart, K.L. Jr., Gloer, J.B., Wilson, G.R., Hughes, Jr., R.G., Li, L.H., Renis, H.E. and McGovren, J.P.: Antiviral and antitumor compounds from tunicates. Fed. Proc. **42:**87–90, 1983.

Rinehart, K.L., Holt, T.G., Fregeau, N.L., Keifer, P.A., Wilson, G.R., Perun, T.J., Jr., Sakai, R., Thompson, A.G., Stroh, J.G., Shield, L.S., Seigler, D.S., Li, L.H., Martin, D.G., Grimmelikhuijzen, C.J.P. and Gade, G.: Bioactive compounds from aquatic and terrestrial sources. J. Nat. Prod. **53:**771–792, 1990.

Rinehart, K.L. Jr., Kobayashi, J., Harbour, G.C., Gilmore, J., Mascal, M., Holt, T.G., Shield, L.S. and Lafargue, F.: Eudistomins A–Q, β-carbolines from the antiviral Caribbean tunicate *Eudistoma olivaceum*. J. Am. Chem. Soc. **109:**3378–3387, 1987.

Rinehart, K.L. Jr., Kobayashi, J., Harbour, G.C., Hughes, R.G. Jr., Mizsak, S.A. and Scahill, T.A.: Eudistomins C, E, K, and L, potent antiviral compounds containing a novel oxathiazepine ring from the Caribbean tunicate *Eudistoma olivaceum*. J. Am. Chem. Soc. **106:**1524–1526, 1984.

Rinehart, K.L. Jr., Shaw, P.D., Shield, L.S., Gloer, J.B., Harbour, G.C., Koker, M.E.S., Samain, D., Schwartz, R.E., Tymiak, A.A., Weller, D.L., Carter, G.T., Munro, M.H.G., Hughes, R.G., Renis, H.E., Swynenberg, E.B., Stringfellow, D.A., Vavra, J.J., Coats, J.H., Zurenko, G.E., Kuentzel, S.L., Li, L.H., Bakus, G.J., Brusca, R.C., Craft, L.L., Young, D.N. and Connor, J.L.: Marine natural products as sources of antiviral, antimicrobial, and antineoplastic agents. Pure Appl. Chem. **53:**795–817, 1981c.

Robins, R.K. and Revankar, G.R.: Design of nucleoside analogs as potential antiviral agents. In De

Clercq, E., and Walker, R.T. (eds): "Antiviral Drug Development, A Multidisciplinary Approach." New York: Plenum Press, 1988.

Ruggieri, G.D.: Drugs from the sea. Science **194:**491–497, 1976.

Sakai, R. and Higa, T.: Tubastrine, a new guanidinostyrene from the coral *Tubastrea aurea*. Chem. Lett. 127–128, 1987.

Sakemi, S., Higa, T., Jefford, C.W. and Bernardinelli, G.: Venustatriol, a new antiviral triterpene tetracyclic ether from *Laurencia venusta*. Tetrahedron Lett. **27:**4287–4290, 1986.

Sakemi, S., Ichiba, T., Kohmoto, S., Saucy, G. and Higa, T.: Isolation and structure elucidation of onnamide A, a new bioactive metabolite of a marine sponge, *Theonella* sp. J. Am. Chem. Soc. **110:**4851–4853, 1988.

Sarin, P.S., Sun, D., Thornton, A. and Muller, W.E.G.: Inhibition of replication of the etiologic agent of acquired immune deficiency syndrome (human T-lymphotropic retrovirus/lymphadenopathy-associated virus) by avarol and avarone. JNCI **78:**663–666, 1987.

Scheuer, P.J. (ed): "Marine Natural Products, Vol. I–V." New York: Academic Press, 1978–1983.

Scheuer, P.J.: Marine natural products and biomedicine. Med. Res. Rev. **9:**535–545, 1989.

Schulze, I.T.: Reversible inhibition of type 2 dengue virus by agar polysaccharide. Virology **22:**79–90, 1964.

Snader, K.M. and Higa, T.: Antiviral chemigrene derivative. PCT Int. Appl. WO 86 03,739, 03 Jul. 1986. Chem. Abstr. **106:**12959q, 1987.

Sugawara. I., Itoh, W., Kimura, S., Mori, S. and Shimada, K.: Further characterization of sulfated homopolysaccharides as anti-HIV agents. Experientia **45:**996–998, 1989.

Takemoto, K.K. and Fabisch, P.: Inhibition of herpes virus by natural and synthetic acid polysaccharides. Proc. Soc. Exp. Biol. Med. **116:**140–144, 1964.

Takemoto, K.K. and Spicer, S.S.: Effects of natural and synthetic sulfated polysaccharides on viruses and cells. Ann. N.Y. Acad. Sci. **130:**365–373, 1965.

Tsujii, S., Rinehart, K.L., Gunasekera, S.P., Kashman, Y., Cross, S.S., Lui, M.S., Pomponi, S.A. and Diaz, M.C.: Topsentin, bromotopsentin, and dihydrodeoxybromotopsentin, antiviral and antitumor bis(indolyl)imidazoles from Caribbean deepsea sponges of the family Halichondriidae. Structural and synthetic studies. J. Org. Chem. **53:**5446–5453, 1988.

Umemura, T. and Mori, K.: Synthesis of both 2,3-*erythro*- and 2,3-*threo*-isomers of aplidiasphingosine, a marine terpenoid. Agric. Biol. Chem. **51:**217–224, 1987.

Voth, R., Rossol, S., Hess, G., Laubenstein, H.P., Buschenfelde, K.H.M., Schroder, H.C., Bachmann, M., Reuter, P. and Muller, W.E.G.: Induction of γ-interferon by avarol in human peripheral blood lymphocytes. Jap. J. Cancer Res. (Gann) **79:**647–655, 1988.

Walker, R.P., Faulkner, D.J., van Engen, D. and Clardy, J.: Sceptrin, an antimicrobial agent from the sponge *Agelas sceptrum*. J. Am. Chem. Soc. **103:**6772–6773, 1981.