Review

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Progress in the development and acquisition of anticancer agents from marine sources

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Introduction

Since ancient times, nature has been an important source of medicines: a fact illustrated by the large number of natural products currently in use in medical practice. These products have been identified and developed through folklore knowledge of the medicinal properties of plants, animal extracts and minerals. Microorganisms are also a prolific source of novel agents. They have yielded some of the most important pharmaceutical products, such as the antibiotics, penicillin and aminoglycosides, which represent landmarks in the history of medicine.

Almost 60% of drugs approved for cancer treatment are of natural origin. Vincristine, irinotecan, etoposide, taxanes and camptothecines are all examples of plant-derived compounds. Dactinomicine, anthracyclines, mitomycin and bleomycin are anticancer agents derived from microbial sources [1].

Although marine compounds are under-represented in current pharmacopoeia, it is anticipated that the aquatic environment will become an invaluable source of novel compounds in the future. The marine ecosystem represents 95% of the biosphere, and all except one of the 33 animal phyla are represented in aquatic environments [2]. Most sessile marine invertebrates contain a primitive immune system and produce toxic chemicals as a form of defense. Many of these products act as regulators of specific biological functions. Some of them have pharmacological activity due to their specific interactions with receptors and enzymes. Because these substances become immediately diluted by large volumes of seawater, they need to be highly potent on a molar basis, and also have to retain a relatively low solubility [3].

The development of marine compounds as therapeutic agents is still in its infancy due to the lack of an analogous ethno-medical history as compared with terrestrial habitats, together with the relative technical difficulties in collecting marine organisms. Over the last few decades significant efforts have been made, by both pharmaceutical companies and academic institutions, to isolate and identify new marine-derived, natural products. These initiatives have been accompanied by funding support from governmental agencies. Specific programs directed towards the collection and characterization of marine natural products and evaluation of their biological activity have been established [4]. This systematic investigation of marine environments is reflected in the large number of novel compounds reported in the literature over the past decade [5]. Some of these agents have entered preclinical and clinical trials, and it may be expected that this number will increase in the future. This article will review some of the technical strategies that are being employed to collect and identify novel marine products with potential antitumoral properties, and will also provide a summary of the available clinical trial information of agents with promising activity.

Sources, collection, screening and supply of marine anticancer agents

The isolation of new anticancer agents derived from marine sources has been based on the collection of marine macroorganisms, such as algae, sponges, tunicates and bryozoans (Table 1). The progress in scuba-diving techniques and deep-water collection instruments has been pivotal in the collection programs implemented by academic and pharmaceutical groups. While 40 years ago, the collection of marine organisms was limited to those found in intertidal and shallow subtidal environments, the advent of scuba diving has enabled investigators to explore shallow subtidal environments to a depth of 40 m for 15 min with no decompression stops. Depths of up to 200 m are now accessible using closed-circuit computerized mixed gas rebreathers [6]. Deep-water collections can be made by dredging or trawling, and by the use of manned and unmanned submersibles, or remotely operated vehicles (ROVs). Although dredging and trawling are cost-effective methods, they suffer from several disadvantages such as: the limitations on taking photographs; the inability to collect organisms that grow in niches difficult to access; the environmental damage; and the non-selective nature of the sampling. On the other hand, the high cost of ROVs precludes their extensive use in routine collection operations.

The collection of organisms from deep water for pharmacological studies has mainly been performed with the use of manned

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Table 1. Marine organism producers of anticancer drugs

Phylums	Major subgroups	Habitat	Selected cytotoxic compounds	Comments
Algae and marine phanerogams	Chlorophyta (blue algae), Phaeophyta (brown algae), Rhodophyta (red algae)	Ubiquitous	Halomon	Very abundant, easy to collect
Porifera	Calcarea, Hexactinellida, Demospongiae	Ubiquitous	Arabinosyl cytosine, halichondrin B, spongistatin	Highly prevalent, easy to collect; sponges are one of the dominant sources of biologically active products
Bryozoa		Tropical reef habitats	Bryostatin 1	Small size, filter-feeding organism. Contains symbiotic microorganisms
Mollusca	Sea hares	Indian ocean	Dolastatins	Concentrate metabolites obtained from their highly specialized diets, which are based on other marine living organisms (seaweed, sponges) and incorporate them as their own defense mechanism.
Chordata (Tunicates)		Regions with freely flowing water	Didemnins, ectenaiscidins	Sessile, filter-feeding organism; life either solitary or in colonies

submersibles. These vehicles can accommodate up to four passengers, operate at depths of 1000 m and are equipped with storage containers and cameras for documentation [7]. Using these techniques, investigators have implemented collection programs in waters from marine regions spanning the whole globe. Although the marine ecosystem is extremely rich and diverse, resources are limited. The exploration of collectable marine organisms is likely to be almost complete within the next 20 years. The collection of wild organisms needs to take into account a detailed assessment of species abundance and distribution in order to avoid their extinction. Re-collection must be considered only for early screening purposes. Identification and development of synthetic or semi-synthetic approaches as the ultimate sources of supply should be the final goal. This area requires governmental supervision and regulation, as well as international cooperation. The appropriate supply methods must be determined carefully prior to considering a marine chemical entity suitable for clinical development [8].

The expected limitations in the supply of marine macroorganisms, as well as the realization that there is a tremendous biological reserve of marine microorganisms, has resulted in increasing interest in the exploitation of the latter in the search for new chemical entities. Several features make marine microorganisms an attractive source when searching for pharmaceutical compounds. The complex microbiological adaptations needed to grow in the ocean are completely different from those of land-based organisms. Nutrients are scarce and microbial symbiosis is common. Furthermore, competition for resources at the microscopic level is intense. This has resulted in a variety of chemical substances produced by microorganisms for their own defense, a range of compounds that has the potential to be a major source of new drugs [9].

Although largely still unexplored, the complexities of marine microbial growth and cultivation can be solved. It has been demonstrated that marine bacteria are uniquely adapted to the saline environment. They can be selectively isolated and mass cultured in media that uses natural nutrients and growth factors derived from marine sources. In culture, marine microbes have the potential to provide large quantities of natural products. This approach, however, also has its own caveats, which include the difficulties in isolating and culturing marine microbes, the lack of stable production and the fact that the majority of marine microbes are still unknown [10]. New programs are emerging to exploit marine microorganisms and the results are promising. These studies have demonstrated the capability of marine bacteria to produce compounds not available from terrestrial sources. They also have led to an increase in knowledge of the many bioactive compounds produced by these microorganisms [11, 12].

Another major advance in the study of marine compounds has been the change in the nature of the studies performed with the isolated products. Nowadays, the compounds are systematically tested for relevant biomedical properties including antiproliferative effects. The major screening system is carried out by the National Cancer Institute of the USA. This system looks for selective activity in a panel of 60 human tumor cell lines [4]. Alternative strategies employ a more mechanistic-based approach, with systems designed to screen for substances with inhibitory properties towards specific enzymatic reactions. This type of assay offers specificity and can focus on a number of discrete drug targets. The potentially confounding effects of toxic components are also avoided, permitting the screening of crude extracts from marine organisms [13]. This type of screening can also be adapted to high-throughput screening, which offers the potential to readily screen hundreds of thousands of extracts in parallel against numerous therapeutic targets. Taken together these data show that the study of marine anticancer compounds is yielding not only the discovery and development of new drugs, but also the identification of new molecular targets for therapeutic intervention.

Marine-derived compounds in clinical development

Bryostatin-1

Bryostatin-1 is a macrocyclic lactone isolated from the marine invertebrate *Bugula neritine* [14]. Bryostatin-1 is a potent activator of protein kinase C (PKC), and has antagonistic effects on

tumor-promoting phorbol esters. Bryostatin-1 also has immunomodulatory functions, induces the differentiation of myeloid and lymphoid cell lines, platelet aggregation and promotes hematopoiesis [15]. Furthermore, bryostatin-1 inhibits the production of components of the matrix metalloproteinases family, down-regulates multidrug-resistance 1 (*MDR1*) gene expression, modulates *bcl-2* and *p53* gene expression and induces apoptosis [16, 17]. It has demonstrated significant antitumor activity in preclinical models against a wide spectrum of cell lines [18] and, in addition, has been shown to enhance the antitumor effects of various chemotherapeutic agents, such as cytosine arabinoside, gemcitabine, vincristine, cisplatin and paclitaxel [19].

Based on its novel mechanism of action and its potency, bryostatin-1 entered phase I trials using different infusion schedules [20–25]. None of these studies has thus far characterized the human pharmacokinetics of the agent due to the lack of a sensitive and reliable assay to determine plasma levels of bryostatin-1. Data regarding PKC modulation, a potential biological surrogate, have not been consistent. Thus, the optimal dose and schedule of administration has not yet been determined. The dose-limiting toxicity (DLT) of bryostatin-1 has consistently been severe myalgias, which were dose-related, cumulative and independent of the schedule of administration [20–25]. The pathogenesis of this phenomenon is uncertain and there are not consistent data to support an inflammatory or myolitic origin [21, 24]. The agent also produced hematological toxicity. Patients treated at doses over the maximum tolerated dose (MTD) had significant decrement in platelets, leukocytes and, particularly, hemoglobin in the immediate post-treatment period. They typically recovered to baseline shortly after treatment, with the exception of hemoglobin—a decrement which persisted 1–2 weeks after dosing [22, 23]. Biological studies did not observe any effects of the agent on bone marrow progenitors. This observation suggests that the hematological effects were due to peripheral blood cells pooling, rather than a decrease in production.

Objective responses were observed in phase I studies of bryostatin-1 in some solid tumors [24]. Based on these results, a large number of phase II studies of bryostatin-1 using various infusion regimens were conducted in both solid and hematological malignancies [20, 26–28]. To date, phase II studies on bryostatin-1 have failed to demonstrate any clinical meaningful activity. The toxicity profile was quite favorable with myalgias being the most common side-effect reported. These data, together with the synergy observed between bryostatin-1 and traditional cytotoxic drugs *in vitro*, suggested a potential role of bryostatin-1 as a modifier to traditional chemotherapy. Several phase I studies of bryostatin-1 in combination with cytotoxic agents have been conducted [29–35]. Myalgias have been the most frequent side-effect in combination regimens. Other reported toxicities appeared to be related to the cytotoxic drugs rather than bryostatin-1. The initial reports of phase II studies of bryostatin-1 in combination regimens are disappointing. One study reported some activity suggesting synergy [36], while others have failed to find activity [37–39].

In summary, despite the promising results in preclinical and early clinical studies with bryostatin-1, phase II studies have failed to show a significant benefit as a single agent. The reasons for this lack of efficacy are unclear, but might be related to pharmacological factors. Additional studies to improve the understanding of bryostatin-1 pharmacokinetics and pharmacodynamic effects in tumor tissues will aid in the further development of this agent.

Didemnins

The didemnins are a family of cyclic depsipeptides obtained from the Caribbean tunicate *Tridemnun solidum* [40]. Didemnin B (DB) was the most potent didemnin in the antitumor screening system that was selected for clinical development. DB inhibits the synthesis of RNA, DNA and proteins [40]. The agent demonstrated antitumoral activity against a variety of tumor models. The substantial evidence of activity in preclinical models with dose-dependent and tolerable toxicity profiles provided the impetus for phase I clinical trials, making DB the first marine natural product to be evaluated in clinical trials. The initial phase I trials of DB evaluated different schedules of administration [41–44]. The toxicity profile of DB was quite similar across the trials, with dose-dependent nausea and vomiting being the most commonly reported side-effects. Phase II trials using DB at the recommended doses were associated with poor efficacy, while trials using more aggressive regimens resulted in higher levels of toxicity, including cardiotoxicity [45–47]. These findings brought about the cessation of DB's clinical development.

Aplidine (dehydrodidemnin B) is a second-generation didemnin that was isolated from the Mediterranean tunicate, *Aplidium albicans* [48]. Aplidine interferes with the synthesis of DNA and proteins and induces G_1–G_2 cell cycle arrest [49]. Furthermore, aplidine possesses a unique and differential mechanism of cytotoxicity which involves the inhibition of ornithine descarboxylase, an enzyme that is critical in the process of tumor formation and growth [49]. Recent data also indicate that aplidine inhibits the expression of the vascular endothelial growth factor gene, having antiangiogenic effects [50].

In preclinical studies, aplidine was more active than DB and displayed substantial activity against a variety of solid tumor models, including tumors noted to be resistant to DB [51]. On the basis of its preclinical activity, aplidine entered phase I clinical trials, in patients with solid tumors and lymphomas, utilizing different schedules of administration (Table 2) [52–56]. Treatment with aplidine has generally been well tolerated, with the most common adverse events being asthenia, nausea, vomiting and transient transaminitis. Hypersensitivity reactions have also been reported. The agent does not induce hematological toxicity, mucositis or alopecia. The occurrence of neuromuscular toxicity with the elevation of creatine kinase levels has been dose-limiting in three of these studies [54–56]. Selected biopsies of affected muscles revealed muscular atrophy and loss of thick myosin filaments. Interestingly, the use of L-carnitine appears to prevent and ameliorate muscular toxicity [55]. Aplidine has shown some antitumor activity in phase I trials [54, 55], and is currently under active phase II development in solid tumors. Recent translational studies also indicate *in vitro* activity in acute lymphoid leukemia (ALL) and acute myeloid leukemia (AML) [57].

Dolastatins

Dolastatins are peptides isolated from *Dolabella auricularia*, a mollusk from the Indian Ocean. Within this family, the linear peptide dolastatin 10 and the desipeptide dolastatin 15 exhibit the most promising antiproliferative actions and, as a result, were chosen for development [58, 59]. The dolastatins inhibit cell proliferation and induce apoptosis in numerous malignant cell lines. These actions are mediated through interactions with tubulin, resulting in the alteration of microtubule function [59, 60]. Recent data indicate that the dolastatins also induce apoptosis in cancer cell lines [61]. Dolastatins exerted profound cytotoxic effects in animals bearing intraperitoneal tumors; in addition, they exhibited synergistic antitumor activity with vinca alkaloids and bryostatin-1 [17, 62]. Dolastatin 10 has been evaluated in various phase I clinical trials [63–66]. DLTs were myelosuppression and phlebitis. Preliminary data indicated that 40% of patients developed moderate peripheral neuropathy and patients with underlying neuropathy are at increased risk for this side-effect. Subsequent phase II studies failed to demonstrate activity in solid tumors. Despite this lack of significant clinical activity, the mechanism of action and favorable toxicity profile of dolastatin 10, coupled with ease of administration, make it an attractive agent for use in combination regimens.

The complexity and low yield of chemical synthesis of dolastatins, together with their poor water solubility, have been significant obstacles to broad clinical evaluation. These facts also have motivated the development of analog compounds. LU103793 is a stable and water soluble analog of dolastatin 15 that has shown prominent activity against a broad range of tumors [67]. LU103793 has been evaluated in five phase I clinical trials with different schedules of administration (Table 3) [68–72]. The side-effects of LU103793 depend on the schedule of administration. Cardiac toxicity consisting of hypertension and acute myocardial infarction in the pretreatment period was the DLT in studies in which the drug was given either as a rapid intravenous infusion every 3 weeks or on a weekly administration schedule. However, myelosuppression, particularly neutropenia, was the most frequent dose-limiting effect on schedules that involved 24-h infusion weekly and short infusion daily schemes.

In contrast to other tubulin interactive agents, LU103793 did not produce discernible peripheral neuropathy, although most subjects exposed to this drug have received a limited number of cycles, impairing the evaluation of cumulative effects. Phase II trials of LU103793 have failed to show activity in patients with melanoma [73] and breast cancer [74]. Phase II clinical trials of LU103793 are ongoing in breast, lung, ovarian, prostate and colon

Table 2. Phase I studies of aplidine

Author	No. pts	Schedule	Recommended phase II dose	DLT
Ciruelos et al. [52]	25	3-h i.v. infusion q2w	5 mg/m^2	Liver G3, renal G4, muscular G3
Maroun et al. [53]	27	1-h i.v. infusion × 5d q3w	1200 μg/m^2	Skin G3, diarrhea G3
Bowman et al. [54]	32	1-h i.v. infusion × 3w q4w	3200–3750 μg/m^2	Muscular G4
Armand et al. [55]	43	24-h i.v. infusion q2w	5 mg/m^2	Muscular G3
Anthoney et al. [56]	25	24-h i.v. infusion × 3w q4w	3750 μg/m^2	Muscular G4, liver G3

DLT, dose-limiting toxicity; G, grade; i.v., intravenous; pts, patients.

Table 3. Phase I trials of LU103973

Author	Schedule	Recommended phase II dose	DLT	Pharmacokinetics	Comments
Mross et al. [71]	5-min i.v. bolus q3w	10 mg/m^2	Cardiotoxicity	$t_{1/2}$, 9.6 h; V_d, 15 l, Cl, 18.3 ml/min, C_{max}, 3800–7640 nmol/l[a]	
Wolff et al. [69]	5-min i.v. bolus weekly × 4 q5w	5 mg/m^2	Cardiotoxicity		
Mross et al. [72]	24-h i.v. infusions weekly × 4 q5w	10 mg/m^2	Neutropenia		
Villalona-Calero et al. [68]	5-min i.v. bolus daily × 5 q3w	2.5 mg/m^2	Neutropenia, edema, hepatotoxicity	$t_{1/2}$, 12.3 ± 3.8 h; V_{ss}, 7.6 ± 2 l/m^2; Cl, 0.49 ± 0.18 l/h/m^2; C_{max}, 0.15–1.3 μmol/l	C_{max} correlated with neutropenia. Day 1 and 5 PKs were similar
Allen et al. [70]	5-min i.v. every other day × 3 q3 weeks	4.8 mg/m^2	Neutropenia		

[a]Parameters at 20 mg/m^2/dose level.

Cl, clearance; C_{max}, maximum concentration; DLT, dose-limiting toxicity; PK, pharmacokinetics; $t_{1/2}$, terminal half-life; V_d, volume of distribution; V_{ss}, volume at steady state.

cancer patients. In addition, the development of other dolastatin-type agents in phase I clinical studies continues [75, 76].

Ectenaisdin 743 (ET-743)

Ectenaisdins (Ets) are tetrahydroisoquinolone alkaloids isolated from *Ectenaiscidia turbinata*, a tunicate that grows on mangrove roots throughout the Caribbean sea. ET-743 was selected for clinical development because of its cytotoxic activity and its relative abundance within the tunicate compared with others Ets [77]. ET-743 alters the interaction of DNA with transcription factors and other proteins [78]. It also produces a delay in cell progression from G_1 to G_2 phase, inhibition of DNA synthesis and cell cycle arrest in G_2 phase, that eventually results in p53-independent apoptosis [79]. ET-743 inhibits translational activation of the *MDR1* gene. ET-743 also interacts with other molecular targets such as the microtubule network [79]. In the NCI human cell line screen, ET-743 was particularly active against cancer cell lines derived from melanoma and carcinomas of the colon, breast, lung, brain and ovary [80]. Cures were observed in a wide variety of solid tumor models [81].

Based on its novel mechanism of action, high potency and positive therapeutic index in preclinical studies, ET-743 entered phase I clinical trials (Table 4) [82–84]. In these trials, ET-743 was generally well tolerated with non-cumulative hematological and hepatic toxicities being the most commonly reported adverse events. The DLTs in all the phase I studies, except in the 72-h infusion schedule, were hematological toxicity and fatigue. Dose-related asymptomatic and reversible transaminitis was prevalent in phase I trials, but it was not a treatment-limiting toxicity. Grade 1–2 elevation of alkaline phosphatase (ALP) level has been reported in almost 50% of patients receiving treatment. Nausea, vomiting and asthenia were also reported but seldom severe. The most severe adverse events associated with ET-743 at the recommended phase II dose included long-lasting pancytopenia, renal and hepatic failure, and rhabdomyolysis. Across the different phase I trials evidence of antitumor activity was noted in patients with advanced and heavily pretreated ovarian, breast, and mesenchimal tumors. In some cases, the responses were long lasting in heavily pretreated patients.

At the beginning of the phase II program with the protracted infusion schedule, a number of patients with drug-induced life-threatening toxicities were reported. These toxicities included the sequence of severe reversible transaminitis, pancytopenia, reversible hiperbilirubinemia, rhabdomiolisis and renal failure. A pharmacokinetic/pharmacodynamic analysis was conducted to identify baseline and intercycle events that might characterize the risk. The result of this analysis identified the baseline biliary function as a reference parameter to identify patients eligible to receive full doses of ET-743. Moreover, an intercycle peak of alkaline phosphatase and/or bilirubin was also a clear indicator of high risk in successive cycles if full doses of ET-743 were given. These findings established an important rule in the clinical use of the drug. A recent analysis pooling the safety data of patients entered into phase II studies before and after this amendment has demonstrated a significant impact in the safety of ET-743 in adult patients with advanced disease; a similar safety profile in an

Table 4. Phase I studies of ET 743

Author	Schedule	Recommended phase II dose	Dose limiting toxicity (DLT)	Pharmacokinetics	Comments
Bowman et al., van Kesteren et al.	1-h infusion i.v. q3w	1000 μg/m^2	Thrombocytopenia, fatigue	–	Clinical improvement in one patient; stable disease in one patient with melanoma
Twelves et al.	3-h infusion i.v. weekly × 3 q4w	1650 μg/m^2	Hematogical, fatigue	–	–
Forouzesh et al.	3-h infusion i.v. weekly × 3 q4w	Ongoing	Ongoing	[a]Biexponential PK: initial $t_{1/2}$, 0.18–0.34 h, terminal $t_{1/2}$, 34–47 h, AUC 4.8–8.5 ng·h/ml, V_{ss} 1005–2052 l/m^2	One minor response in one patient with liposarcoma
Taama et al.	24-h infusion q3w	1500 μg/m^2	Neutropenia, thrombopenia	Cl, 30–60 l/h/m^2; V_{ss}, 1000–2000 l/m^2; terminal $t_{1/2}$, 20–40 h	Three partial responses (breast cancer, osteosarcoma and liposarcoma). Four patients (all with soft tissue sarcomas) stable disease lasting >3 months
Villalona-Calero et al.	1-h infusion daily × 5 q3w	325 μg/m^2	Neutropenia, thrombocytopenia	Data suggest that PK of ET-743 is dose independent	Three patients antitumor activity. Stable disease in one patient with renal carcinoma.
Ryan et al.	72-h infusion q3w	1050 μg/m^2	Transaminitis, rhabdomyolisis	Disposition biexponential Linear PK	Toxicity was schedule-dependent; the 72 h infusion resulted in decreased myelosuppression and comparable hepatotoxicity; antitumor activity was seen

[a]Data at dose level 1 (300 μg/m^2 weekly).

$t_{1/2}$, terminal half-life; AUC, area under the curve; Cl, clearance; DLT, dose-limiting toxicity; PK, pharmacokinetics; V_{ss}, volume at steady state; i.v., intravenous.

extensive compassionate cohort has been reported [85]. Phase II clinical trials have been performed to assess the activity of ET-743 in different tumors. Although the drug has displayed activity in several refractory tumor types, the most prominent data have been observed in patients with refractory soft tissue sarcoma (STS), ovarian and breast cancer. The overall response in phase II studies of ET-743 in patients with STS ranges from 10% in patients with prior treatment to 18% in first-line treatment. In addition, a relatively high number of patients had durable stable disease [86–90]. The true value of ET-743 in STS, however, remains to be established. Randomized, comparative trials providing survival and quality-of-life data are needed. Activity of ET-743 as second-line treatment in ovary [91] and breast carcinomas [92] has been reported. Phase III studies comparing conventional therapy versus ET-743 are necessary to fully elucidate the therapeutic potential of this agent.

Selected new natural marine products with promising applications in oncology

Over the last few years, a large number of novel compounds have been identified and characterized. Several of these agents are in the process of entering clinical trials (Table 5). Some of the newest and more interesting agents derived from marine sources have a common mechanism of action, which involves the disruption of microtubular function. The most relevant examples within this category include the halichondrins, spongistatin, curacin, laulimalide and discodermolide [93]. The majority of these compounds bind the vica alkaloids (halicondrins, spongistatin) or the colchicine-binding domain (curacin) inhibiting the polymerization of microtubules. Halichondrin B, a macrocyclic polyether isolated from the sponge *Halichondria okadai*, was selected for preclinical development by the NCI. Analogs derived from the total synthesis of halichondrin B have shown activity superior to the natural product. One of these, NSC707389, is now being tested in the clinical setting [94].

Other microtubule interacting agents, such as laulimalide, isolaulimalide, discodermolide and eleutherobin, interact with tubulin in a similar fashion to the taxanes, resulting in the assembly of tubulin and stabilization of the microtubules. In addition, laulimalide and isolaulimalide retained antitumor activity against the P-glycoprotein overexpressing multidrug resistant cell line, suggesting that they are poor substrates for transport by P-glycoprotein [93].

Other compounds of marine origin with promising activity include thiocoraline and kahalalide F. Thiocoraline is a novel bioactive depsipeptide isolated from *Micromonospora marine*, a marine microorganism located in the Mozambique Strait that inhibits RNA synthesis. Thiocoraline demonstrated potent and selective cytotoxic effects against lung and colon cancer cell lines as well as melanoma. Interestingly, this drug exerted preferential antiproliferative effects in colon cancer cell lines with defective p53 systems [95].

Thiocoraline represents a model of an anticancer agent acquired from marine microorganism and illustrates how the problems of drug supply can be overcome by artificial culture. Kahalalide F (KF) is a dessipeptide isolated from the mollusk *Elysia rubefescens* from Hawaii. KF induces cytotoxicity and blocks the cell cycle in G_1 phase in a p53-independent manner. *In vitro*, KF displayed activity against solid tumors with an interesting pattern of selectivity in prostate cancer cell lines. In addition, extensive *in vivo* work demonstrated that the agent had activity in breast and colon cancer. The intracellular target of kahalalide seems to be the lysosomas where the agent interferes with the organization of the organella. These results suggest that cells containing high lysosomal activity, such as prostate cancer cells, would be a suitable tumor type to explore the activity of this agent [96]. In phase I clinical trials of KF evaluating a continuous weekly infusion schedule in patients with advanced solid tumors, the DLT has been early-onset transaminitis. Other reported toxicities have been fatigue, headache, vomiting and pruritus limited to the hands [97, 98]. Hematological toxicities have not been observed. Additional studies of this agent are planned.

Conclusions

The marine ecosystem is an enormously rich source of natural products with potential therapeutic usefulness in oncology. Over the past few years, >2000 new compounds from various marine sources have been described and characterized. The significant expansion of this field is due to improvements in the technologies involved in sample collection, the closer collaboration among

Table 5. Selected marine-derived agents in preclinical development

Compound	Organism	Group	Location
Halichondrins B	*Halichondria okadai*	Porifera	Okinawa
Isohomohalichondrin B	*Lissodendoryx* sp.	Porifera	New Zealand
Laulimalide and isolaulimalide	*Cacospongia mycofijinsis*	Porifera	Marshall Islands
Discodermolide	*Discodermia dissoluta*	Porifera	–
Spongitatin	*Hyrtios erecta*	Porifera	–
Curacin	*Lyngbya majuscula*	Cyanobacteria	Curacao
Thiocoraline	*Micromonospora marina*	Actinobacteria	Mozambique Strait
Mycaperoxide B	*Mycala* sp.	Porifera	Thailand
Crambescidin-816	*Crambe crambe*	Porifera	Mediterranean

scientists from a variety of disciplines worldwide and the support of governmental institutions as well as pharmaceutical companies. Some of these compounds have been tested in clinical trials and are in advanced stages of development, while others are still in preclinical stages. These agents are characterized as having unique mechanisms of action and pharmacological properties. They represent potential candidates for the treatment of malignant disease, either to be used as single agents, or as part of a combination regimen. More recently, the focus of attention has shifted towards microscopic organisms. Microorganisms are abundant in the marine ecosystem and biologically rich. In fact, it is believed that a number of metabolites obtained from some macroorganisms may be produced by their associated microorganisms. In addition, microorganisms can be adapted to artificial culture conditions thus avoiding problems of collection and supply.

In summary, the marine world has become an important source of anticancer agents with novel mechanisms of action. The continuation of preclinical and clinical studies is required in order to assess the exact role of this new class of compound in the treatment of patients with cancer. It is anticipated that marine-derived anticancer drugs will represent valuable tools in the oncological armamentarium.

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