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Invited review

Bioactive peroxides as potential therapeutic agents

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Abstract

Present review describes research on more than 280 natural anticancer agents isolated from terrestrial and marine sources and synthetic biologically active peroxides. Intensive searches for new classes of pharmacologically potent agents produced by terrestrial and marine organisms have resulted in the discovery of dozens of compounds possessing high cytotoxic, antibacterial, antimalarial, and other activities as an important source of leads for drug discovery.

Keywords: Anticancer; Cytotoxic; Peroxides; Fatty acids; Terpenoids; Steroids; Alkaloids; Peptides

1. Introduction

More than 600 peroxides have been isolated and structurally characterized from natural sources, mainly as constituents of family Compositae and occur randomly in about 10 other plant families; they also were found in marine invertebrates, particularly in sponge species, and other organisms [1–6]. Among peroxides studied, fatty acid derivative, sesquiterpene endoperoxide, quinghaosu, has already been clinically applied as a new antimalarial agent [7–9].

Some synthesized peroxides as well as natural compounds are shown to possess many biological activities [10–13]. The preparation of chiral compounds in non-racemic form is a goal of great interest in organic synthesis, due to the large application these compounds have in several fields, such as in medicinal chemistry [14]. Interest in this field has been directed toward the use of biocatalysis for regioselective and stereoselective discriminations of alcohol functions, so as to achieve polyhydroxylated compounds in enantiopure form [15,16]. The enantioselective direct introduction of oxygen onto olefins with biocatalysis by haloperoxidases, in oxygenase-type reactions, is very useful and effective for this purpose [17,18]. Among naturally occurring peroxides fatty acid derivatives represented a large group compounds which are shown to possess cytotoxic, antibacterial, antimalarial, and other activities.

Ascaridole, the first natural peroxide was isolated from leaves of *Chenopodium ambrosioides* in 1895 by a German pharmacist living in Brazil and it has been attributed with most of the vermifuge (worm-expelling) actions of the plant. In the early 1900s it was one of the major anthelmintics used to treat ascarids and hookworms in humans, cats, dogs, horses, and pigs [19–28].

The first of marine natural peroxides to be reported was antitumor metabolite chondrillin, a six membered ring cycloperoxide found in 1976 by Wells [29] in the marine sponge *Chondrilla* sp.

In the past several decades, natural peroxides have been isolated from a wide variety of plants, and marine organisms. Extensive pharmacological screening performed on aquatic and/or terrestrial species resulted in discovery of novel antitumor, antibacterial, and antimalarial agents. The purpose of this review is to summarize antitumor, and also antibacterial, antimalarial and cytotoxic properties of more than 280 natural and synthesized peroxides, belonging to diverse structural classes, including fatty acids, terpenes, steroids, simple and complex lipids, and other metabolites.

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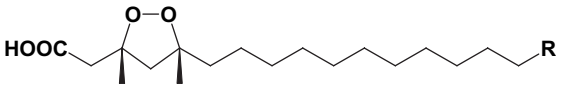
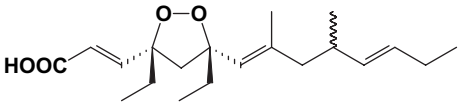
2. Anticancer fatty acid peroxides

Natural fatty acid peroxides that have potent biological activities with novel diverse structures are reviewed with classification as secondary metabolites such as 1,2-dioxolane carboxylates (five-membered ring cyclic peroxides), 1,2-dioxane carboxylates (six membered ring cyclic peroxides), cyclic peroxides with ring sizes greater than six, and their analogs and derivatives.

Saturated fatty acids (**1**–**5**), which contain 1,2-dioxolane ring were isolated from *Halichondriidae* marine sponges [30], are antitumor, antibacterial and antifungal agents. Dioxolane-3-acetic acids (**4**) and (**5**) at 0.5, and 10 μg/mL inhibited the growth of P388 mouse leukemia cells and HCT-8 human colon cells by 90–100% of control growth, respectively. The EtOAc extract of sponge *Plakortis halichondrioides* yielded a novel class of activators of cardiac SR-Ca^{2+}-pumping ATPase, plakortide E (**6**) [31].



1 R = Me; **2** R = Et; **3** R = Pr; **4** R = Bu; **5** R = Pen



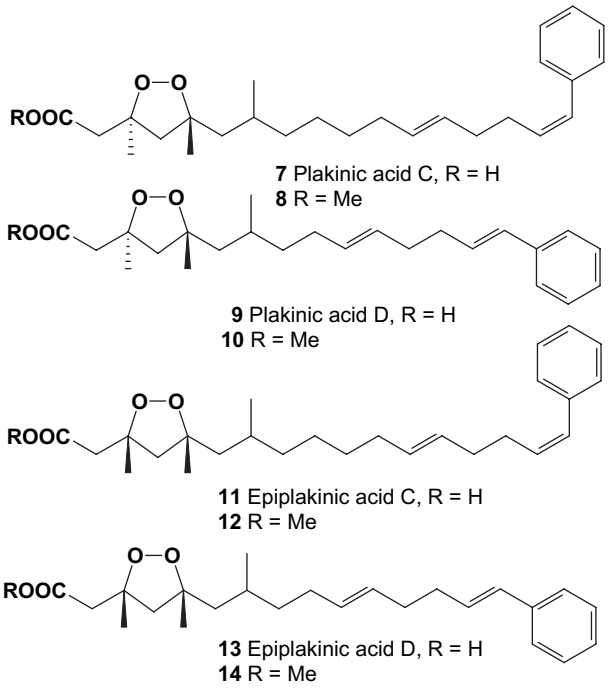
6 Plakortide E

Four metabolites containing five-membered peroxide rings, plakinic acids C **7**, D **9**, and epiplakinic acids C **11**, and D **13**, and their methyl esters **8**, **10**, **12**, **14**, respectively, have been isolated from a *Plakortis* sponge collected near the Fiji Islands [32]. These substances and their methyl esters exhibited cytotoxicity toward human epidermoid carcinoma (KB) cells, human colorectal adenocarcinoma (LoVo) cells, and L-1210 murine leukemia (Table 1). A shorter saturated chain analog, methyl ester of epiplakinic acid E **15**, was isolated from *Plakinastrella onkodes*, and it showed cytotoxic activity against human cancer cell lines A549 (IC_{50} 2.5 μg/mL), and P388 (IC_{50} 2.0 μg/mL) [33].

Table 1
Cytotoxic activity of peroxides plakinic acids and their methyl esters against some tumor cell lines (Ref. [32])

Compound	L-1210[a]	KB[b]	LoVo[b]
7	0.017	0.01	0.1
8	0.013	1.0	1.0
9	0.026	0.001	0.001
10	0.0043	1.0	0.1
11	0.052	0.01	1.0
12	0.29	0.1	0.1
13	0.017	0.1	1.0
14	0.003	0.1	0.1

[a] IC_{50} (μg/mL).
[b] MIC (μg/mL).



7 Plakinic acid C, R = H
8 R = Me

9 Plakinic acid D, R = H
10 R = Me

11 Epiplakinic acid C, R = H
12 R = Me

13 Epiplakinic acid D, R = H
14 R = Me

Two cyclic peroxides, epiplakinic acids G **16** and H **17**, were isolated from the deep-water sponge *Plakortis nigra* from Palau [34]. Epiplakinic acids G (**16**, IC_{50} 0.16 μM) and H (**17**, IC_{50} 0.39 μM) show cytotoxic activity against the HCT-116 human colon tumor cell line.

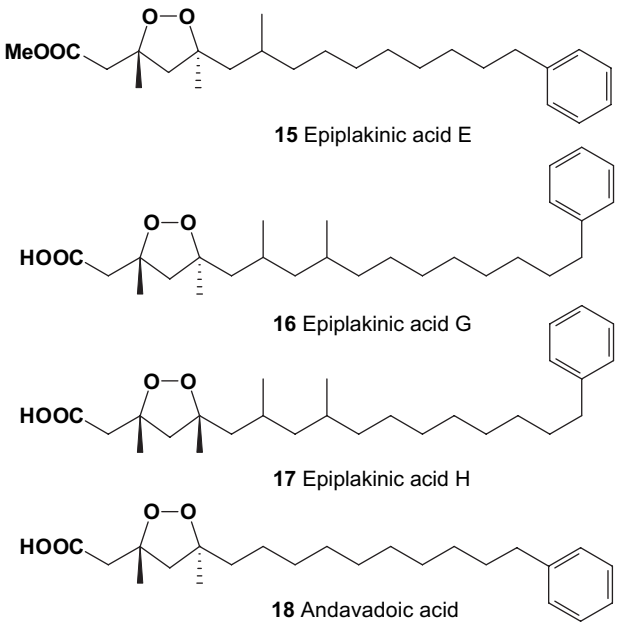
New 1,2-dioxolane, designated andavadoic acid **18**, isolated from the sponge *Plakortis* aff. *simplex*, collected from Madagascar, was found to be cytotoxic to a series of human tumor cells [35].

Andavadoic acid **18** has been found to be responsible for the cytotoxicity of the crude extract of the sponge. It demonstrated good activity (GI_{50} in the submicromolar range) against 13 human tumor cell lines (GI_{50} values in the submicromolar range): leukemia, K562 (ATCC-CCL-243); lung carcinoma A549 (ATCC-CCL-185); melanoma, SK-MEL-28 (ATCC-HTB-72); colon carcinoma, HT-29 (ATCC-HTB-38), LoVo (ATCC-CCL-229) and LoVo-Dox, MDR cell line; prostate carcinoma, DU145 (ATCC-HTB-81) and LNCaP (ATCC-CRL-1740); breast carcinoma SKBR3 (ATCC-HTB-30); ovary carcinoma SK-OV-3 (ATCC-HTB-77) JGROV and IGROV-ET resistant to ET-743; and pancreas carcinoma, PANC1 (ATCC-CRL-1469).

Table 2
Antitumor activity of compounds **19** and **20** (IC_{50} μg/mL, Ref. [36])

Tumor cell line	**19**	**20**
GXF 251L	>10	8.3
LXF 529L	>10	7.7
MAXF 401NL	>10	14.9[a]
MEXF 642NL	>10	7.2
RXF 486L	>10	14.4[a]
UXF 1138L	>10	14.1[a]

[a] Value extrapolated.

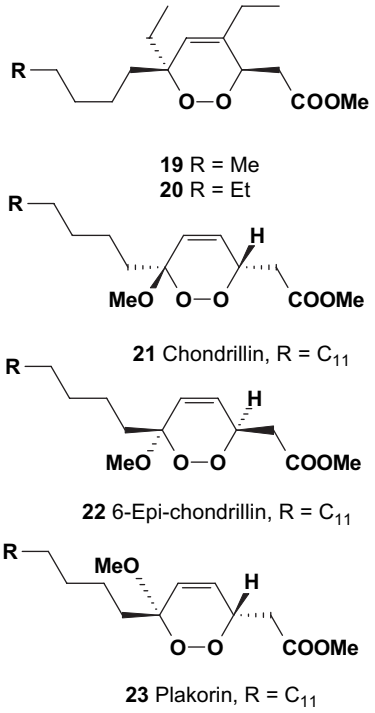


Two new cyclic peroxides, **19** and **20**, were isolated from the Norwegian sponge *Plakortis simplex* [36]. Peroxide **20** exhibited moderate cytotoxicity toward tumor cells, whereas peroxide **19** did not inhibit proliferation in any of the tumor cell lines tested up to a concentration of 10 μg/mL (Table 2). Peroxide **20** selectively inhibited proliferation in gastric cancer (GXF 251L), non-small cell lung cancer (LXFL 529L), and melanoma (MEXF 462NL) cell lines. Mammary (MAXF 401NL, $IC_{50} = 14.9$ μg/mL), renal (RXF 486L, $IC_{50} = 14.4$ μg/mL), and uterin (UXF 1138L, $IC_{50} = 14.1$ μg/mL) cancer cell lines were slightly less sensitive.

Chondrillin **21** was isolated from marine sponge belonging to the genus *Chondrilla* by Wells [29], and more recently, **21** and **22** were found in the extract of Taiwanese marine sponge *P. simplex* [37]. The antibiotic plakorin **23**, a six membered ring cycloperoxide was found in 1978 by Higgs and Faulkner [38] in the marine sponge *P. halicondrioides*. Chondrillin demonstrated antitumor activity against KB-16 cell line (IC_{50} μg/mL): 0.74 (Ref. [38]), A549 (0.3), HT-29 (1.1), P388 (2.4), and EL-4 (0.4) [39]. Cyclic peroxides: Chondrillin **21** and compounds **23**–**26** were prepared from marine sponge *Plakortis lita*, and are antitumor agents. Their antitumor activities were detected using the P388 mouse leukemia cell cytotoxicity assay. All the five metabolites **21**, **23**–**26** were cytotoxic, and had the greatest activity, inhibiting 50% of cell growth at 0.05 μg/mL, compared to the least active chondrillin with $IC_{50} = 5$ μg/mL [40].

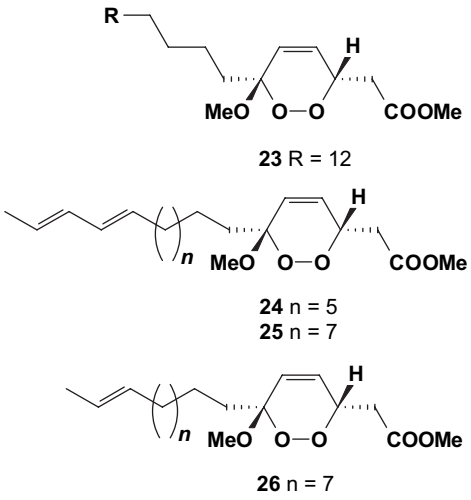
Compounds **22**, and **27**–**30** were identified from *P. lita* [41]. Both chondrillin **21** and **29** demonstrated cytotoxic activity against P388 cells, $ED_{50} > 10$ μg/mL. The epimer of chondrillin **21**, plakorin **23**, was identified from *Plakortis* sp. [42].

Plakorin is a potent activator of sarcoplasmic reticulum Ca^{2+}-ATPase, and also exhibited activity in vitro against murine lymphoma L-1210 cells (IC_{50} 0.85 μg/mL), and human epidermoid carcinoma KB cells ($IC_{50} = 1.8$ μg/mL) [43,44].

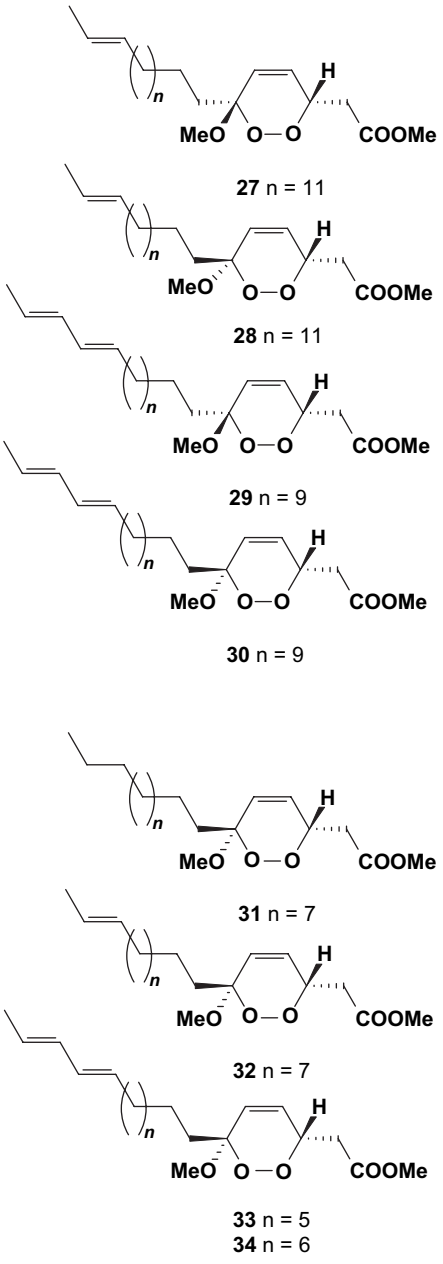


Antitumor compounds chondrillin **21**, and **31**–**34** also were identified from sponge *P. lita*, and these novel peroxy metabolites have shorter fatty chain-sides than plakorin [45].

Xestin A **35** and B **36** were produced by sponge *Xestospongia* sp. [46], and both peroxides showed cytotoxic activity against P388 cells, IC_{50} 0.3 and 3.0 μg/mL, respectively.

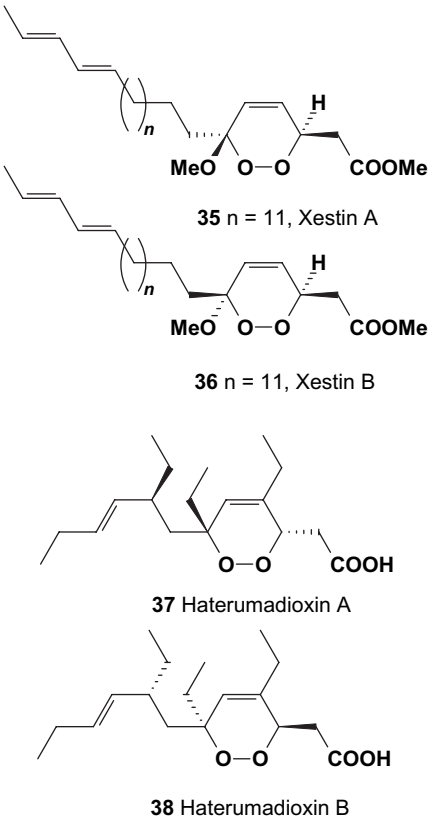


Other simpler types of peroxides were isolated from the Okinawan sponge *P. lita* [47]. Haterumadioxin A **37** and B **38** were evaluated against a human cancer cell line panel to show significant cytotoxicity against P833 cells, with IC_{50} values of 11.0 and 5.5 ng/mL, respectively. It was showed that haterumadioxin A inhibited growth of human cell lines: HBC-4, HBC-5, DMS273, DMS114, LOXIM-VI, OVCAR-4, ACHN, and MKN1 [47].

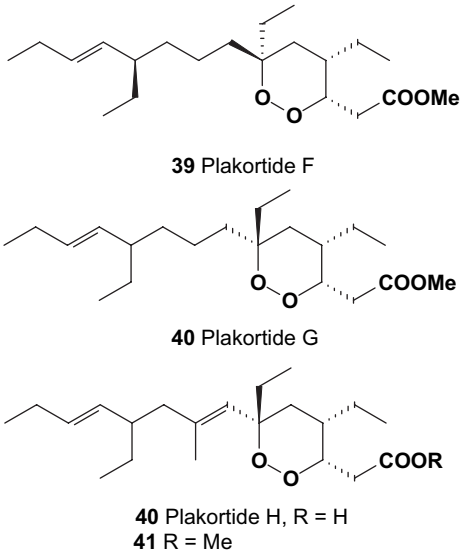


The peroxide-containing metabolite plakortide F (**39**), isolated from a Jamaican sponge, *Plakortis* sp., is a more typical of the genus *Plakortis* metabolite and was shown to exhibit significant activity against *Plasmodium falciparum* in vitro [48]. Agents of Ca^{2+} pumping activity of cardiac SR (sarcoplasmic reticulum), plakortides F (**39**), G (**40**), and H (**41**) have been isolated from the EtOAc extract of the Jamaican sponge *P. halichondrioides*. All three of the cyclic peroxides, plakortides F, G and H were found to significantly enhance Ca^{2+} uptake by SR [49].

Plakortide H (**40**), the novel cycloperoxide polyketides plakortide I (**42**) and J (**44**), and their methyl esters (**43**, **45**) have been isolated from the Caribbean marine sponge *P. simplex*. The isolated compounds exhibited good cytotoxic activity against WEHI 164, a murine fibrosarcoma cell line. The results are expressed as IC_{50} (μg/mL, the concentration that

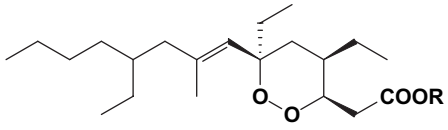


inhibited the cell growth by 50%): 7.1 for (**41**); 9.5 for (**43**); 8.2 for (**45**); and 2.5 for the mixture (**41**, **43**, **45**). It should be noted that plakortide H had been reported in 1996 by Patil et al. [49] as activator of cardiac SR–Ca^{2+}-ATPase [50].

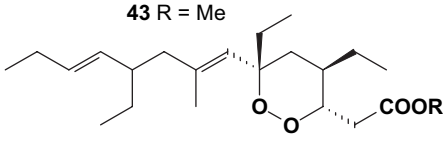


Four polyketide-derived cyclic peroxides, plakortides K (**46**), L (**47**), M (**48**), and N (**49**) were isolated from an undescribed sponge of the genus *Plakortis* collected at Discovery Bay, Jamaica [51]. Compounds (**46**, **47**) exhibited significant antimalarial activity against the W2 Clone of *P. falciparum* with an IC_{50} of 570 ng/mL. Two new polyketide

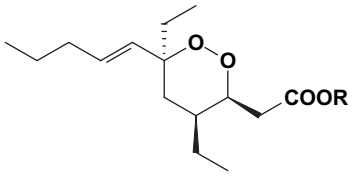
endoperoxides, namely, plakortide M (**48**) and plakortide N (**49**) were isolated from the marine sponge *P. halichondrioides* collected from Puerto Rico. Plakortides M and N exhibited potent cytotoxicity in the NCI human cancer screening program, whereas plakortide M Me ester displayed strong antimalarial activity against *P. falciparum* [52].



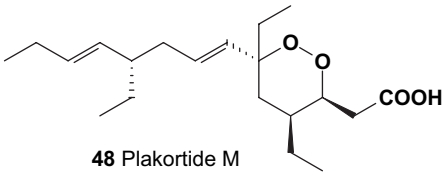
42 Plakortide I, R = H
43 R = Me



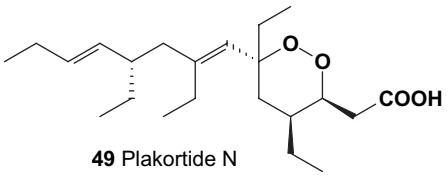
44 Plakortide J, R = H
45 R = Me



46 Plakortide K, R = Me
47 Plakortide L, R = H



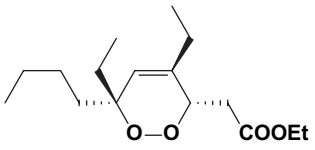
48 Plakortide M



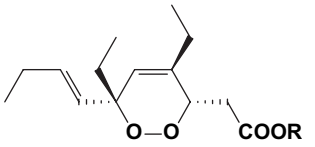
49 Plakortide N

Three cyclic peroxides, ethyl plakortide Z (**50**), ethyl ester didehydroplakortide Z (**51**), and methyl ester didehydroplakortide Z (**52**) were identified from the sponge *P. lita* from (Papua New Guinea). Compound **50** was equally active in vitro against solid tumor and L-1210 leukemia cell lines. Alternatively, **51** was observed in vitro to be moderately solid tumor selective but did not exhibit in vivo activity against solid tumors in mice [53].

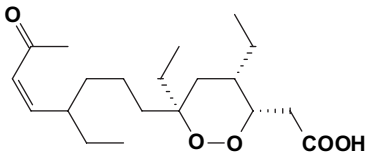
Two new cyclic peroxides, **53** and **54**, were isolated from sponge *Plakortis* sp. collected at Discovery Bay, Jamaica. Both **53** and **54** exhibited significant antimicrobial activity against pathogenic bacteria and fungi with IC_{50} values of 0.9–5.0 μg/mL and 0.7–8.0 μg/mL, respectively [54].



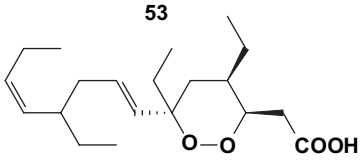
50 Plakortide Z



51 Didehydroplakortide Z, R = Et
52 R = Me

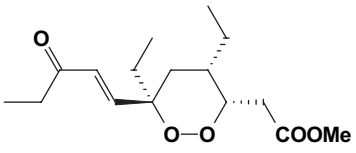


53

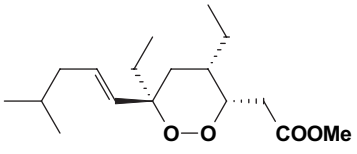


54

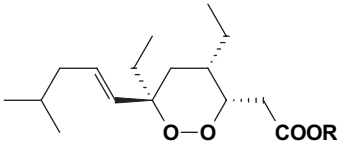
From the sponge *Plakortis* sp. collected at Jamaica, four cyclic peroxides, plakortolide I(M) **42**, J(N) **43**, K **44**, and L **45** were isolated [51]. Plakortolide I(M) is the first reported polyketide-derived peroxide with an α,β-unsaturated ketone moiety in the side chain and exhibits significant antimalarial activity against the W2 clone of *P. falciparum* with an IC_{50} value of 0.57 μg/mL [55].



55 Plakortolide I(M)



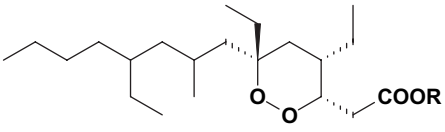
56 Plakortolide J



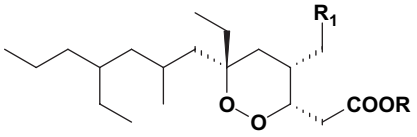
57 Plakortolide K, R = Me
58 Plakortolide L, R = H

The new plakortide Q has been isolated from the little studied marine sponge *Plakortis zyggompha*, together with the six new cyclic peroxide analogs in their Me ester forms: plakortide Q (**59**, ME **60**), 14-nor-plakortide Q (**61**, **62**), 14,16-dinor-plakortide Q (**63**, **64**), 11,12-didehydroplakortide Q (**65**, **66**), 11,12-didehydro-14-nor-plakortide Q (**67**, **68**), 11,12-didehydro-16-nor-plakortide Q (**70**, **71**), and 14,18-dinor-plakortide Q (**72**, **73**). The non-esterified peroxides exhibited more cytotoxic activity against human tumor cell lines than their corresponding Me esters (Table 3).

The acid carboxylic forms **59** and **61/62** were more cytotoxic (IC_{50} 2–9 μM) than the corresponding methyl esters **60** and **62** (Table 3). However, the bioactivity of **62** was comparable to that of **59**, and probably to that of **61** (supposed from **62**). As the mixture **61/65** (1:1 ratio) was twice more active than **59**, we assumed that **65** would have an activity close to 1 mM. This family of plakortides was probably responsible for the bioactivity of the crude extract of *P. zyggompha*. The plakortide Q (**59**) was also interestingly cytotoxic against the two human tumor cell lines A549 and HT-29 (IC_{50} 3.6 and 3.9 mM, respectively) [56].

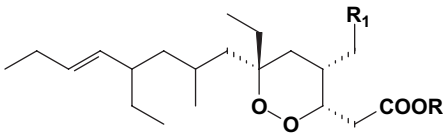


59 Plakortide Q, R = H
60 Plakortide Q ME, R = Me

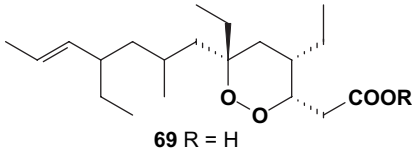


61 R = H, R_1 = Me
62 R,R_1 = Me
63 R,R_1 = H
64 R = Me, R_1 = H

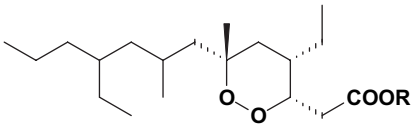
The cytotoxic cyclic peroxides, methyl capucinoate A **73**, **74** and **75** were identified from the Dominican marine sponges *Plakinastrella onkodes* by cytotoxicity-guided fractionation [57]. The cyclic peroxides **73** (B16F1, IC_{50}, 12 ng/mL), **74** (B16F1, IC_{50}, 12 ng/mL) and **75** (P388, IC_{50}, 55 ng/mL), showed in vitro cytotoxicity, but none of them showed in vivo activity against murine leukemia P388.



65 R = H, R_1 = Me
66 R,R_1 = Me
67 R,R1 = H
68 R = Me, R1 = H

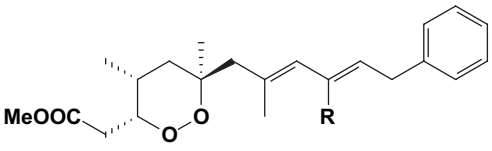


69 R = H
70 R = Me

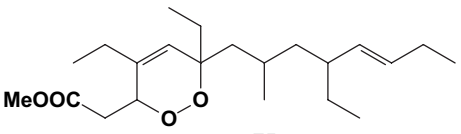


71 R = H
72 R = Me

Three new peroxylactones, plakortolides B **76**, D **77**, and C **78**, and a new peroxy ester, epiplakinic acid E Me ester, were isolated and characterized from a previously unstudied marine sponge, *P. onkodes*. A mixture of steroidal peroxides was also found in this organism. Plakortolides B and D, and epiplakinic acid E Me ester, were evaluated for biological activity and found to show cytotoxicity against the A549 human lung carcinoma and P388 murine leukemia cell lines, and to effect adhesion in an assay employing the EL-4.IL-2 cell line, which correlates with signal transduction activity (Table 4) [33,58].



73 Capucinoate A, R = H
74 Capucinoate B, R = Me



75

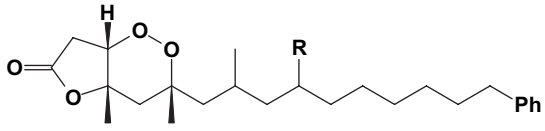
Table 3
Cytotoxic activity of plantide Q derivatives (IC_{50} μM, Ref. [56])

Compound	MDA-MB231	A549	HT-29
59	9.0	3.6	3.9
60	18.4	7.8	13.0
61/65	4.6	4.0	1.9
62	24.4	14.0	12.3
66	4.1	6.8	12.8
68/63	28.9	23.6	27.2

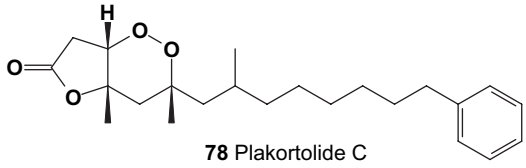
Table 4
Antitumor activities for some peroxy metabolites (IC_{50} μg/mL, Refs. [33,58])

Compound	A549	P388	EL-4
11	2.0	2.5	4.6
21	0.3	2.4	0.4
76	1.3	0.4	4.4
77	3.8	0.8	15.8

Plakortolide B **76** induced cell adhesion in the EL-4.E-2 cell line, which corresponded to very modest agonistic activity against a suite of protein kinase C isoenzymes [59,60] (activity at 50 pg/mL: α − 19%, β-I − 13%, β-II− 27%, δ − 9%, ε − 38%, and γ − 9%). In contrast, chondrillin induced cell adhesion in the EL-4.E-2 cell line but expressed modest antagonistic activity against the PKC isoenzymes (IC_{50} values (μg/mL): α − +36, β-I − +49, β-II − +49, δ − +23, ε − +30, γ − >150, and ζ − +43).

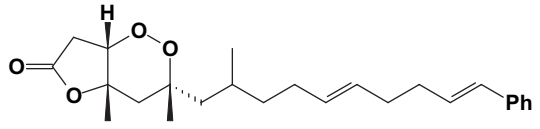


76 Plakortolide B, R = H
77 Plakortolide D, R = Me

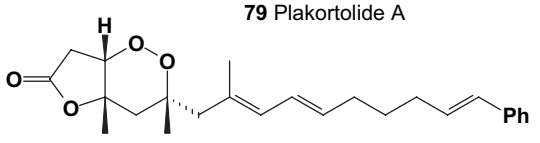


78 Plakortolide C

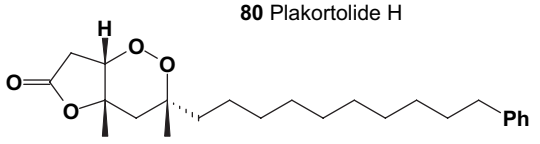
A cytotoxic cyclic peroxide-containing metabolite plakortolide A **79** has been identified from a *Plakortis* sponge [61]. Bioactive compounds from the MeOH–EtOAc extract of the sponge *P*. aff. *simplex*, collected in Madagascar, was found to be cytotoxic to a series of human tumor cells. From this sponge, three compounds, 1,2-dioxane peroxylactones named plakortolides H (**80**) and I (**81**), and 1,2-dioxolane, designated andavadoic acid **18**, have been isolated and their structures elucidated [62]. Andavadoic acid (**18**) showed significant activity against 13 tumor cells with GI_{50} values in the submicromolar range.



79 Plakortolide A

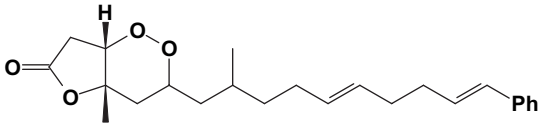


80 Plakortolide H

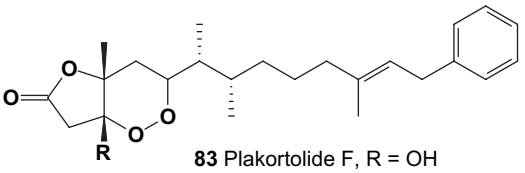


81 Plakortolide I

Marine sponge *P*. *onkodes* collected in Jamaica yielded three cyclic peroxides, including the known plakortolide and two new analogs of (**82**) plakortolides F (**83**) and G (**84**). Compounds (**82**) and (**84**) exhibited potent inhibitory activity against the protozoan *Toxoplasma gondii* in HFF cells and represent the first marine natural products reported with *T*. *gondii* inhibitory activity [63].

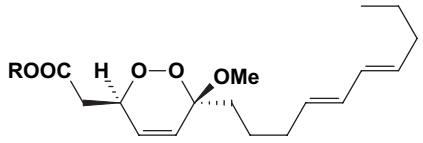


82 Plakortolide

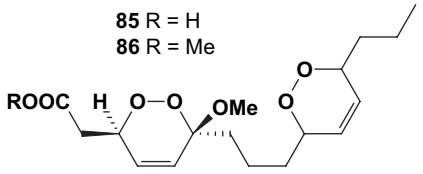


83 Plakortolide F, R = OH
84 Plakortolide G, R = H

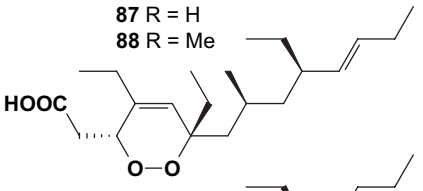
Cyclic peroxide-containing polyketide C16 acids and their Me esters **85**–**88** have been isolated from the Indo-Pacific marine sponge *P*. aff. *simplex* [64]. Compounds **84**, **86** are proposed to contain a single 1,2-dioxene ring, while **87**, **88** incorporate two 1,2-dioxene rings. The methyl esters were found to be active against cultured P388 murine leukemia cells. Two antifungal and cytotoxic cyclic peroxide-containing acids, **89** and **90**, were isolated from a marine sponge, *Plakortis angulospiculatus*, which was collected by scuba off the coast of Venezuela. The acids are potent cytotoxic and antifungal agents although their esters are only cytotoxic [65].



85 R = H
86 R = Me



87 R = H
88 R = Me



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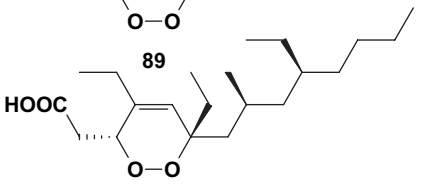


90

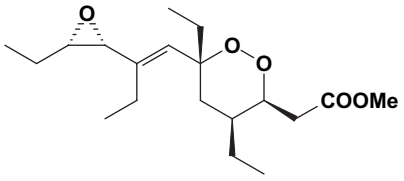
Two new cancer cell growth inhibiting cyclic peroxides, plakorstatins 1 (**91**) and 2 (**91**, **92** isomer at epoxide ring), were isolated from the Indonesian marine sponge *P*. *nigra*. The structures of plakorstatins (**91**) and (**92**) including relative configuration were elucidated on the basis of mass and 2D NMR spectroscopic interpretations, and their biological activity against some human cancer cell lines are shown in Table 5. These are the first plakortides with an epoxy group in the side chain. Plakorstatin 2 was found to differ from plakorstatin (**91**)

Table 5
Cancer cell growth inhibitory activity (ED_{50} values for P388 and GI_{50} values for other cancer cell lines[a]) for plakorstatins **91** and **92** (Ref. [66])

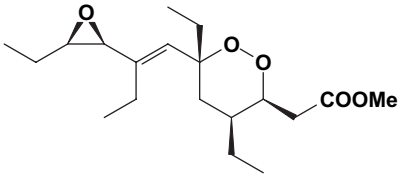
Compound	A	B	C	D	E	F	G
91	1.1	>10	>10	1.8	>10	>10	>10
92	0.91	6.4	3.8	1.6	>10	6.7	1.7

[a] Cancer cell type: P388 (**A**, lymphocytic leukemia); BXPC3 (**B**, pancreas adenocalcinoma); MCF7 (**C**, breast adenocarcinoma); SF-268 (**D**, CNS glioblastoma); NCI-H460 (**E**, lung large cell); KM20L2 (**F**, colon adenocarcinoma); DU145 (**G**, prostate carcinoma).

only in the configuration of the epoxide group. Both exhibited moderate cancer cell growth inhibition against the murine P388 lymphocytic leukemia cell line with ED_{50} values of 1.1 and 0.91 μg/mL, for peroxides (**91**) and (**92**), respectively [66].



91 Plakorstatin 1

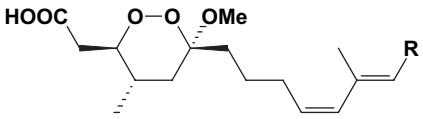


92 Plakorstatin 2

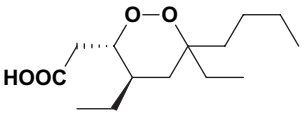
Manadic acid A **93** and B **94** were recovered from extracts of Indonizean *Plakortis* sp. sponge [67]. Both peroxides were moderately active against various antitumor cell lines. Two short branched-chain acids, **95** and **96**, were isolated from a sponge collected in New Guinea, a *Callyspongia* sp. [68]. These metabolites inhibited leukemia cell growth with ED_{50} values of 5.5 and 2.6 μg/mL for **95** and **96**, respectively.

Two 3,6-epidioxy-7,10-tetrahydrofurano C26 unsaturated fatty acids, stolonic acids A **97** and B **98**, were isolated from a previously undescribed ascidian species, *Stolonica* sp. collected off the Maldive Islands in the Indian Ocean. Both compounds exhibited antiproliferative activity against selected human melanoma and ovarian tumor cell lines, with IC_{50} values of approximately 0.05–0.1 μg/mL [69].

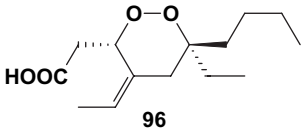
Stolonoxide A **99** (as Na salt **100**, and methyl ester **101**), a novel peroxide possessing an unprecedented molecular arrangement, has been isolated as its Me ester from the marine Mediterranean tunicate *Stolonica socialis* [70]. A series of stolonoxide A related compounds (**102–108**) were independently isolated by bioassay guided screening from researchers of the University of Cadiz (Spain) [50]. A strong cytotoxicity (IC_{50} 0.1 μg/mL) against several mammalian tumor cell lines for purified and for a crude mixture of (**99–108**) has been found and showed in Table 6 [71–73].



93 Manadic acid A, R = Me
94 Manadic acid B, R = Et



95



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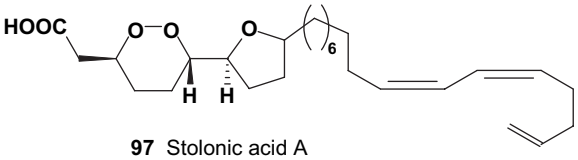
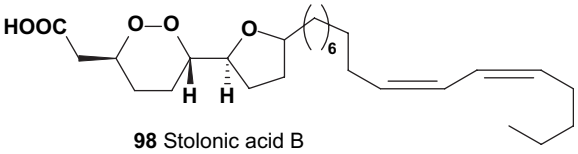
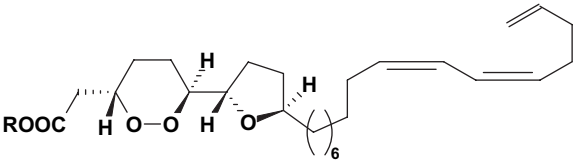


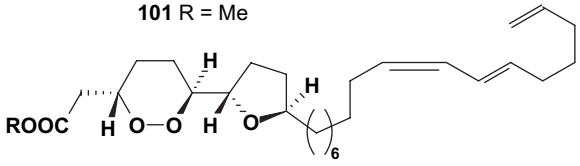
97 Stolonic acid A



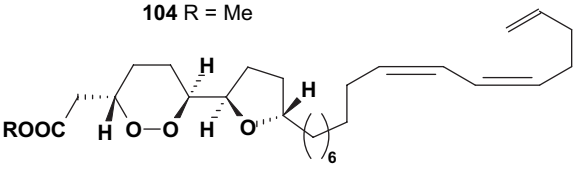
98 Stolonic acid B



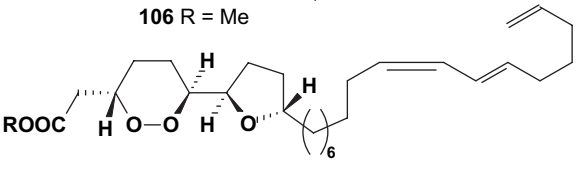
99 Stolonoxide A, R = H
100 R = Na,
101 R = Me



102 Stolonoxide B, R = H
103 R = Na,
104 R = Me



105 Stolonoxide C, R = Na
106 R = Me

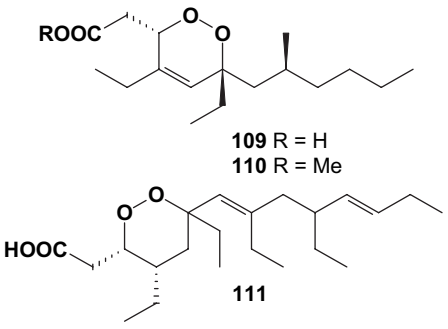


107 Stolonoxide D, R = Na
108 R = Me

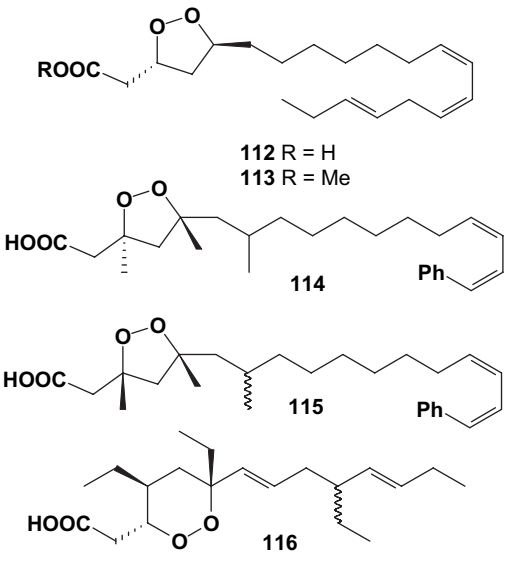
Table 6
Cytotoxic activity of stolonoxides against tumor cell lines (IC_{50} μg/mL, Ref. [71])

Compound	P388	A549	HT-29	MEL28	DU145
99	0.01	0.10	0.10	0.10	0.10
100	0.01	0.10	0.10	0.10	0.10
101	0.05	0.10	0.10	0.10	0.10
102	0.05	0.10	0.10	0.10	0.10
103	0.10	0.10	0.10	–	0.10
105	0.50	0.10	0.10	0.50	0.10
106	0.50	0.10	0.05	0.50	0.10
107	0.50	0.10	0.05	0.10	0.10
108	0.01	0.01	0.05	0.10	0.10
Doxorubicin	0.02	0.002	0.05	0.02	–

Three cyclic peroxides (**109**–**111**) have shown strong in vitro antiproliferative effects on promastigotes of *Leishmania mexicana*, a flagellate protozoan that causes leishmaniasis; they have been isolated from the Palauan sponge *Plakortis* aff. *angulospiculatus* [74].

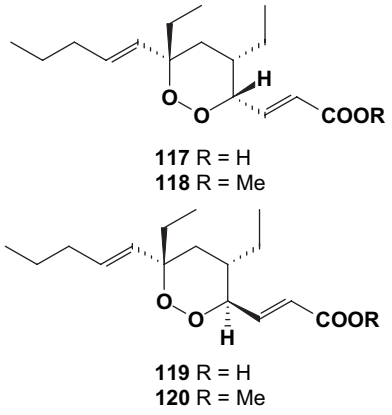


Three cyclic peroxides (**81**, **112**, **113**) were isolated from the hexane extract of a *Plakinastrella* sp. from the Philippines. The major compounds in the sponge showed activity against *Candida albicans* [75].

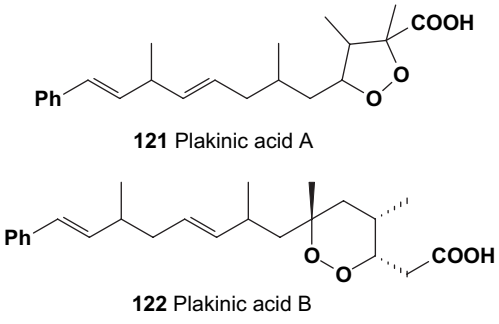


A new 1,2-dioxolane peroxide acids, (**114**, **115**) have been isolated from the sponge *P. halichondrioides*.and 1,2-dioxane ring peroxide acid (**116**) has been isolated from *P. onkodes*. The three new compounds exhibit moderate activity against the fungal pathogen *C. albicans* with MICs of 1.6, 1.6, and 5.0 μg/mL, respectively, for **114**, **115**, and **116**. Compound **116** also showed in vitro inhibition of the fungal pathogen *A. fumigatus* with an IC_{90} value of 5.6 μg/mL [76].

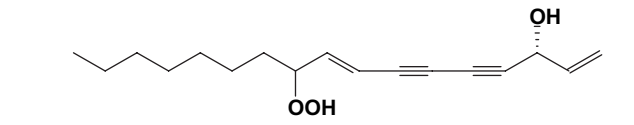
The Caribbean sponge *Chondrosia collectrix* contained four antimicrobial metabolites (**117**–**120**) [77]. Plakinic acids A (**121**) and B (**122**), two peroxy acids isolated from a Caribbean sponge are shown to have antifungal activity [78]. Although the isolated acids are potent antimicrobial compounds, the methyl esters are essentially inactive.



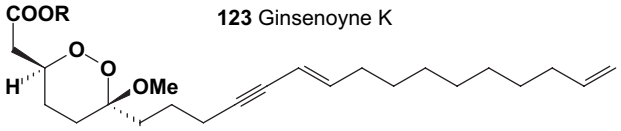
Three acetylenic cytotoxic compounds have been isolated from plant and marine sponges. Ginsenoyne K (**123**) was isolated from a hexane extract of the roots of *Panax ginseng* [79]. This metabolite showed activity against cancer cell lines IC_{50} of 1.8 μg/mL against L-1210 mouse leukemic cells and 10.5 μg/mL against HeLa cells [80].



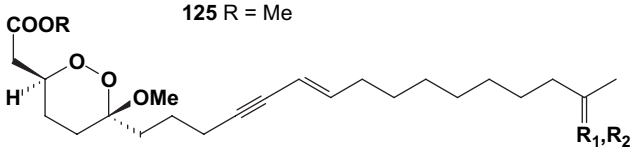
Two new acetogenin peroxides and their methyl esters, named peroxyacarnoic acid A (**124**, **125**), peroxyacarnoic acids C (**126**, **127**) and D (**128**, **129**) were isolated from the sponge *Acarnus bicladotylota* [81,82]. Isolated methyl esters of fatty acids from marine sponge and mixture showed strong antifungal activity [83].



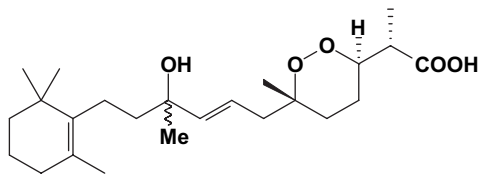
123 Ginsenoyne K



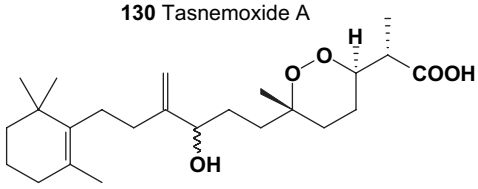
124 Peroxyacarnoic acid A, R = H
125 R = Me



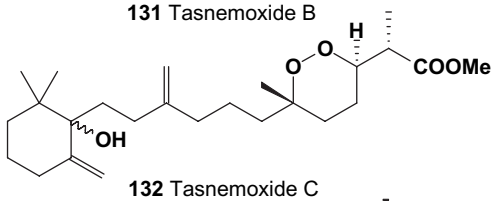
126 Peroxyacarnoic acid C, R = H, R_1,R_2 = O
127 R = Me, R_1,R_2 = O
128 Peroxyacarnoic acid D, R,R_1,R_2 = H
129 R = Me, R_1,R_2 = H



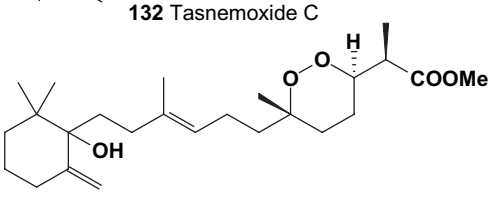
130 Tasnemoxide A



131 Tasnemoxide B



132 Tasnemoxide C



133 Hurghaperoxide

3. Bioactive terpenoid peroxides

Some identified terpenoids from marine and terrestrial showed cytotoxic activities against tumor cell lines. Tasnemoxides A–C (**130–132**), three new cytotoxic cyclic norsesterterpene peroxides were isolated from the Red Sea sponge *Diacarnus erythraenus*, together with the known compound sigmosceptrellin B. Isolated metabolites show cytotoxicity ($IC_{50} > 1$ μg/mL) against the three types of cells, including murine leukemia (P388; ATCC: CCL 46), human lung carcinoma (A549; ATCC: CCL 8), and human colon carcinoma (HT-29; ATCC: HTB 38) [84]. Norsesterterpene cyclic peroxide, named hurghaperoxide (**133**) with a similar structure, was isolated from an undescribed Red Sea sponge [85].

Two new norsesterterpene 1,2-dioxanes, mycaperoxides A (**134**) and B (**135**), were isolated from a Thai sponge of the genus *Mycale*. Mycaperoxides A and B showed significant cytotoxicity (IC_{50} 0.5–1.0 μg/mL) against the cell lines of P388, A549, and HT-29 and displayed antiviral activity (IC_{50} 0.25–1.0 μg/mL) against vesicular stomatitis virus and herpes simplex virus type-1. Both compounds also inhibited the growth of the Gram-positive bacteria *Bacillus subtilis* and *Staphylococcus aureus* [86,87], and a norsesterterpene mycaperoxide G Me ester (**136**) was obtained from southern Australian marine sponges, *Mycale* sp. [88].

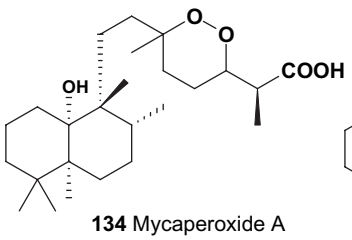
Mycaperoxide H (**137**), a new cyclic norsesterterpene peroxide, was isolated from a Thai marine sponge *Mycale* sp. Mycaperoxide H was cytotoxic against HeLa cells with an IC_{50} value of 0.8 μg/mL [89]. A cytotoxic norsesterterpenoid, mycaleperoxide **138**, was isolated from sponge *Mycale izuensis* collected in Thailand [90].

The lipophilic extract of the Red Sea sponge *Diacarnus erythraenus* contain cyclic peroxide, aikupikoxide A **139**, B **140**, C **141**, and D **142**, as well as the known norterpene peroxides muqubilin (also known as prianicin A) **143**, and nuapapuin A Me ester **144**. Isolated metabolites **139–142** show cytotoxicity (IC_{50} >1 μg/mL) against the three types of cells, including murine leukemia (P388; ATCC: CCL 46), human lung carcinoma (A549; ATCC: CCL 8), and human colon carcinoma (HT-29; ATCC: HTB 38) [91]. Norsesterterpenoid peroxide, prianicin A (**143**) from Red sponge *Prianos* sp. showed antibacterial activity against *Streptococcus* sp. [92]. It was fourfold more effective than tetracycline against β-hemolytic *Streptococcus*.

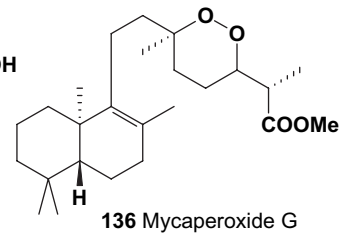
The marine sponge *Diacarnus* cf. *spinopoculum* has provided a series of norterpenes, including eight peroxides **145–152** [93]. Eight of these compounds represent additional examples of the muqubilin/sigmosceptrellin classes (norsesterterpene peroxides) or the nuapapuin class (norditerpene peroxides). Also isolated were dinorditerpenones which are biosynthetically related to the muqubilin/sigmosceptrellin structure classes. All eleven compounds were evaluated for their cytotoxic properties using a soft agar assay system and the NCI's 60 cell line screen (Table 7). On overall, the norsesterterpene peroxides were less selective as cytotoxins than norditerpene peroxide analogs. Two compounds, nuapapuin A Me ester and nuapapuin B, which were somewhat selective in their cytotoxic behavior, were selected for further in vivo evaluation.

The isomeric norsesterterpenes sigmasceptrellin A (**153**), C (**156**), and B (**155**) were isolated as main compounds from the ichthyotoxic fraction of the *Sigmosceptrella laevis* [94]. Sigmosceptrellin A methyl ester (**154**), a novel ichtyotoxic norsesterterpenoid also was isolated from the sponge *S. laevis* [95].

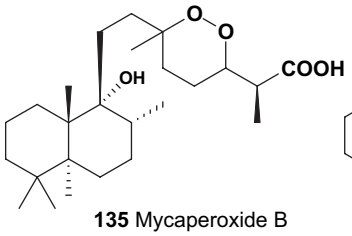
Ascaridole (also known as ascaridol; ascarisin; 1,4-epidioxy-*p*-menth-2-ene, **157**) is a bicyclic monoterpene that has an unusual bridging peroxide functional group. Ascaridole has been documented with sedative and pain-relieving properties as well as antifungal effects [96]. Ascaridole was found to



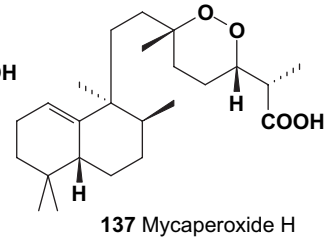
134 Mycaperoxide A



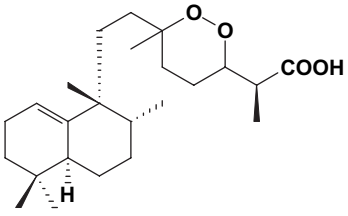
136 Mycaperoxide G



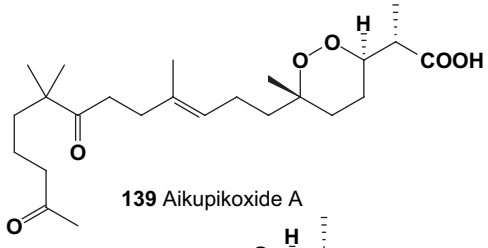
135 Mycaperoxide B



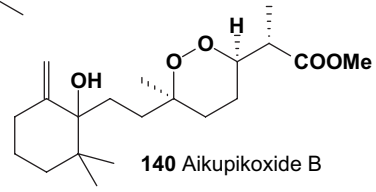
137 Mycaperoxide H



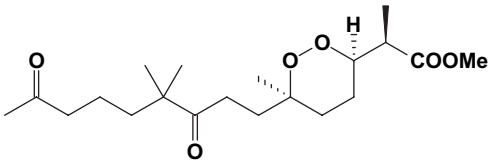
138 Mycaleperoxide



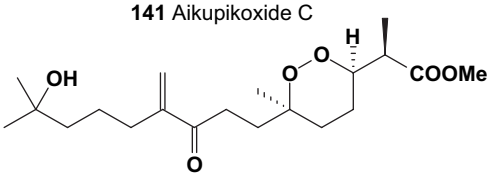
139 Aikupikoxide A



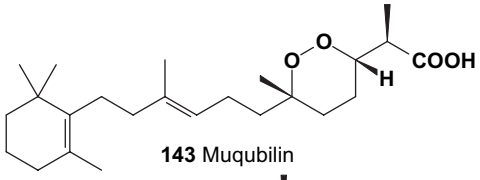
140 Aikupikoxide B



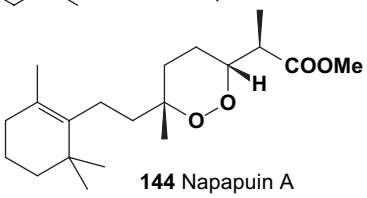
141 Aikupikoxide C



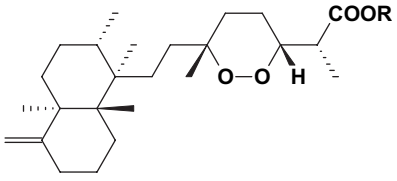
142 Aikupikoxide D



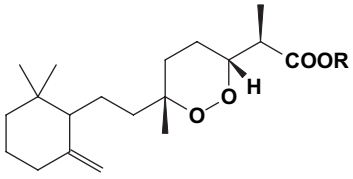
143 Muqubilin



144 Napapuin A



145 Sigmosceptrellin A, R = Me
146 Sigmosceptrellin B, R = H



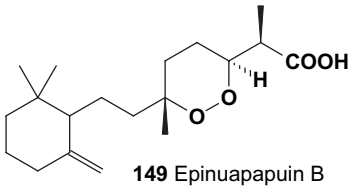
147 Napapuin B, R = H
148 Napapuin B Me ester, R = Me

be a potent inhibitor in vitro development of *P. falciparum* [97], *Trypanosoma cruzi* [98], and *Leishmania amazonensis* [99]. Ascaridole also showed antitumor activity against different tumor cell lines in vitro (CCRF-CEM, HL60, MDA-MB-231). The findings are the first hint that ascaridole may be an interesting novel candidate drug for cancer treatment [100].

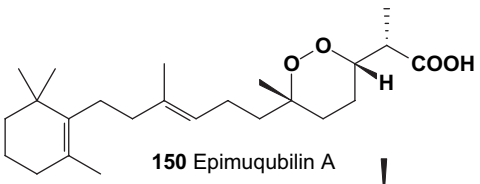
Table 7
In vivo growth inhibition from NCT's 60 cell lines[a] (GI_{50} μM, Ref. [92])

No.	HL60	MOLT-4	A549	KM12	LOX	IGRO	7860	BT549
145	0.14	0.98	1.45	0.94		0.12	0.61	0.96
146	0.14	0.84	0.94	0.95	0.16	0.10	0.50	0.96
147	1.63	2.16	3.05	4.82	0.25	0.63	0.94	1.05
148	1.60	>5.0	0.64	0.40	0.47	0.50	0.27	4.95
149	2.06	>5.0	>5.0	>5.0		1.73	>5.0	>5.0
150	>5.0	>5.0	>5.0	>5.0	>5.0	>5.0	>5.0	>5.0
151	>5.0	>5.0	>5.0	>5.0		>5.0	>5.0	>5.0
152	>5.0	>5.0	>5.0	>5.0	>5.0	2.42	>5.0	

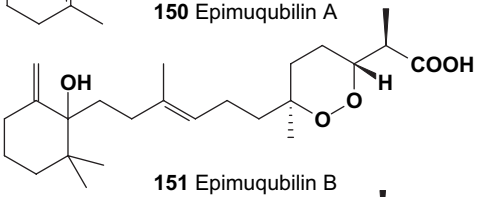
[a] Key: HL-60 (TB)/MOLT-4, leukemia; A549/ATCC, non-small cell lung cancer; KM12, colon cancer; LOX IMVI, melanoma; IGROV1, ovarian cancer; 786-0, renal cancer; BT549, breast cancer.



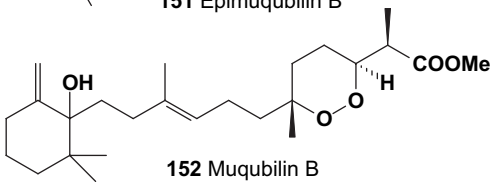
149 Epinuapapuin B



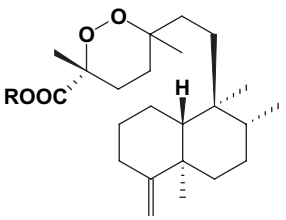
150 Epimuqubilin A



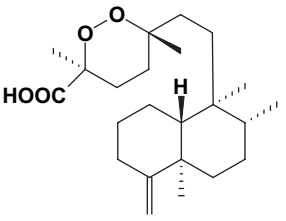
151 Epimuqubilin B



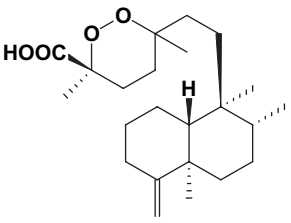
152 Muqubilin B



153 Sigmosceptrellin A, R = H
154 R = Me



155 Sigmosceptrellin B



156 Sigmosceptrellin C

Ascaridole was isolated from leaves and/or seeds of the genus *Chenopodium*: *Chenopodium murale*, *Chenopodium foliosum*, *Chenopodium rubrum*, *Chenopodium missouriense*, *Chenopodium opulifolium*, *Chenopodium polyspermum*, *Chenopodium vulvaria*, *Chenopodium album*, *Chenopodium ambrosioides*, *Chenopodium ficifolium*, *Chenopodium quinoa*, *Chenopodium botrys*, and *Chenopodium urbicum* [101]. The genus *Chenopodium* includes a variety of weedy herbs (more than 200 species) native to much of Europe, Asia, India, China and both North and South America [102]. Goosefoot is common name for the genus *Chenopodium*, as well as for the goosefoot family Chenopodiaceae. The *C. ambrosioides*, which has been successfully used in chorea, and the *C. botrys*, which has been used with advantage in catarrh and humoral asthma, as an expectorant, are both indigenous, and though less powerful, possess somewhat similar properties.

The genus *Chenopodium* has been utilized as an important grain and vegetable crop being rich in protein, carotenoid, ascorbic acid and a wide range of minerals. However, at present, the medicinal uses of *Chenopodium* are not widely known. Various plant parts of different species have been traditionally used in the treatment of several disorders [27]. *C. ambrosioides* (also known as American wormseed oil, chenopodium oil, or Baltimore oil) is rich of monoterpenes [98]. The seed and fruit contain a large amount of essential oil which has a main active chemical in it called ascaridole. Herb *C. ambrosioides* are plants widely known in popular medicine as antihelminthic, vermifuge, emmenagogue and abortifacient [103,104]. They are used for the treatment of digestive, respiratory, uro-genital, vascular and nervous disorders, for metabolic disturbances such as diabetes and hypercholesterolemia, and as sedative, antipyretic and antirrheumatic [105–107]. Ascaridole has shown inhititory effect against several mouse skin cancer cell lines [108]. Biosynthesis of ascaridole has recently been reported [101,109] (Fig. 1).

Antibacterial agent, 1-methyl-5-(1-methylethyl)-3,6,7-trioxatricyclo[3.2.2.02,4]nonane (**158**) has been isolated from leaf oil of aromatic plant *Curcuma longa* (Bangladesh) [110]. This compound has shown antibacterial different activity against: *Alternaria alternate*, *Bacillus cereus*, *Bacillus megaterium*, *B. subtilis*, *Botryosphaeria rhodina*, *Colletotrichum corchori*, *Curvularia lunata*, *Escherichia coli*, *Fusarium equiseti*, *Macrophomina phaseolina*, *Pseudomonas aeruginosa*,

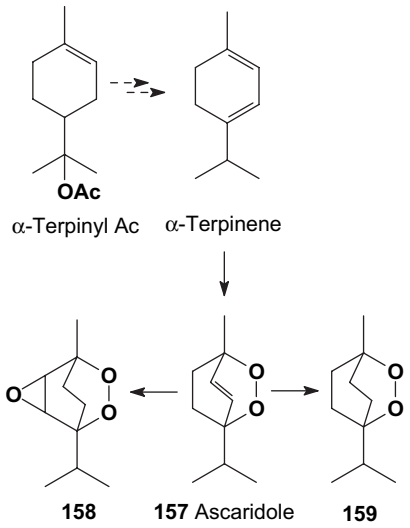


Fig. 1. Proposed biosynthesis of ascaridole and two of it's peroxy derivatives.

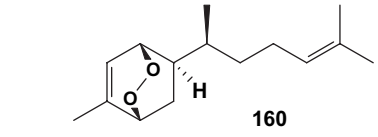
Salmonella typhi, *Shigella dysenteriae*, *Shigella sonnei*, *Staphylococcus aureus*, and *Vibrio cholerae*.

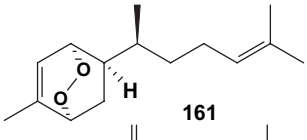
Dihydroascaridole (**159**) for the first time was isolated from *Chenopodium* oil by Paget, in 1938 [111], and also was found in some species of the genus *Chenopodium* [101]. Dihydroascaridole showed the antimalarial activity in vitro against *P. falciparum* and in mice against *P. berghei* [112,113].

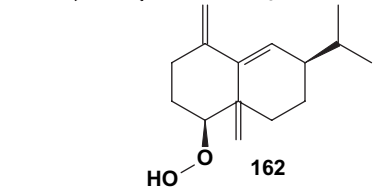
Artemisia species (family Asteraceae), widespread in nature, are frequently utilized for the treatment of diseases such as malaria, hepatitis, cancer, inflammation, and infections by fungi, bacteria, and viruses. Furthermore, some *Artemisia* constituents exhibited antitumor, antibacterial and other activities [114]. Artemisia (*Artemisia annua*), known in the United States as sweet Annie or annual wormwood, is an annual herb native to China, and Vietnam, where it is known as *qinghao*. An aquatic extract of *Artemisia capillaris* (0.5–5 μg/mL) inhibited the secretion of EtOH-induced interleukin-1α (IL-1α) and tumor necrosis factor-α (TNF-α). *A. capillaris* also inhibited the EtOH-, IL-1α-, and TNF-α-induced cytotoxicity. Furthermore, it was found that *A. capillaris* inhibited the EtOH-induced apoptosis of Hep G2 cells [115]. The water and methanol extracts of *Artemisia argyi* showed significant cytotoxicity against J774A.1 cells but not so much against normal leukocytes [116]. Hepatocarcinoma cells SMMC-7721 were treated by the essential oil of *Artemisia annul*, and it was demonstrated that the essential oil of *A. annul* could induce apoptosis of cultured SMMC-7721 [117]. Several anticancer, and other bioactive peroxy metabolites have been isolated from the genus *Artemisia*.

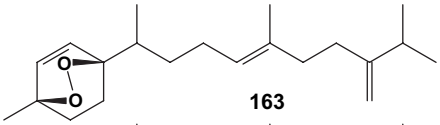
Two sesquiterpene endoperoxides, (1*S*,4*R*,6*R*)-1,4-endoperoxybisabola-2,10-diene (**160**), (1*R*,4*S*,6*R*)-1,4-endoperoxybisabola-2,10-diene (**161**), and a sesquiterpene hydroperoxide, 1β-hydroperoxygermacra-4(15),5,10(14)-triene (**162**) were isolated from the aerial parts of *Artemisia stolonifera* (Compositae). Compounds (**160**) and (**161**) exhibited cytotoxicity against five human tumor cell lines with their ED_{50} values ranging from 0.20 to 5.43 μg/mL and from <0.1 to 0.87 μg/mL, respectively [118]. Two new diastereomeric homoditerpene peroxides (**163**, **164**) were isolated from the aerial parts of wormwood (*Artemisia absinthium*). Both compounds showed antimalarial activity in vitro with an EC_{50} of 1 μg/mL [119].

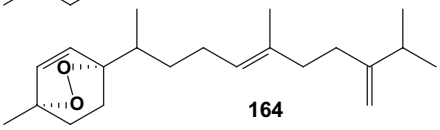
Artemisinin (**165**, also has other names as Qinghaosu, Huanghuahaosu, Qing Hau Sau, Qing Hau Su, Arteannuin, and Artemef), sesquiterpene lactone endoperoxide was originally developed in 1971 in China from the plant *A. annua* (sweet wormwood) [120]. Artemisinin is the active ingredient in qinghao, a Chinese herbal tea that have been used for 150 years to treat malaria and haemorrhoids. Now artemisinin and these three derivatives are being used around the world as effective new antimalarial drugs in the fight against falciparum malaria, including multi-drug-resistant *P. falciparum*. At the present time new artemisinin analogues and/or derivatives are being developed and studies of their structure–activity relationships, their antimalarial mechanisms, their interaction with ferrous ions and the DNA damage associated with these processes are being actively pursued. In addition, recent studies also indicate



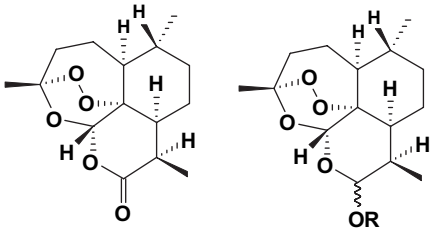






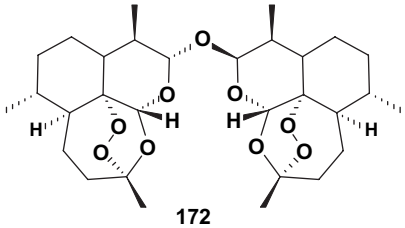


that some artemisinin derivatives have other bioactivities, including antiparasitic (against *Schistosoma japonicum*, *T. gondii* and so on) and anticancer activities [120]. Artemisinin has been shown to selectively kill cancer cells in vitro and retard the growth of implanted fibrosarcoma tumors in rats [121]. A series of artemisinin-related synthesized endoperoxides were tested for cytotoxicity to Ehrlich ascites tumor cells using the microculture tetrazolium assay. Artemisinin had an IC_{50} value of 29.8 μM, and their IC_{50} values ranged from 12.2 to 19.9 μM, and shown in Table 8 [122].



165 Artemisinin (Qinghaosu)

166 R = H (α+β)
167 R = β-Me
168 R = β-Et
169 R = α-$COCH_2CH_2CO_2Na$
170 R = β-$CH_2C_6H_4CO_2H$
171 R = β-$CH_2CC_6H_4CO_2Na$



172

Table 8
The in vitro cytotoxicity of artemisinin-related endoperoxides to Erlich Ascites tumor cells (Ref. [122])

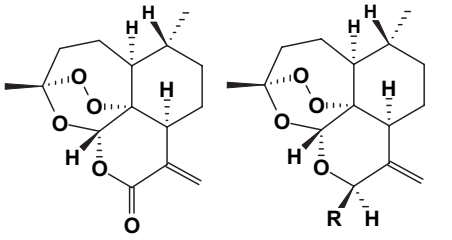
Compound	IC_{50} (μM)	IC_{80} (μM)
165 Artemisinin	29.8	346
166 Dihydroartemisinin	83.4	233
167 Artemether	14.3	>125
168 Arteether	13.1	212
169 Sodium artesunate	19.9	53.6
170 Artelinic acid	16.1	103
171 Sodium artelinate	12.2	47.9
172 Ether dimer of **165**	1.4	2.4
173 Artemisitene	6.8	16.0
174 Hydroperoxyartemisitene	37.8	59.2
175 Ethylperoxyartemisitene	11.5	38.3
176 11-Hydroxyartemisinin	48.7	125
177 11-Hydroxy-11-*epi*-artemisinin	>125	>125
178 Anhydrodihydroartemisinin	29.2	207
179 Formate ester	91.0	156
180 Ketone	>175	>175

On investigation of eight species of *Artemisia* collected in various regions of India and screened by TLC, IR, and HPLC methods, only *A. annua* showed the presence of artemisinin [123].

The discovery of antimalarial activity of naturally occurring peroxides such as artemisinin (**165**) has stimulated recent interest in the development of methods for synthesis of new cyclic peroxide systems. In this respect, unusual the spiro-peroxide, 1,2,6,7-tetraoxaspiro[7.11]nonadecane (**181**) has been synthesized, which was found to exhibit antimalarial activities in vitro and in vivo comparable with those reported for artemisinin [124]. Although a number of natural products containing cyclic peroxide substructural units of various types have been reported to date, there are relatively few examples of molecules containing the peroxylactone moiety; noteworthy examples include the two diterpenoids (**182**, **183**) isolated from the marine alga *Taonia atomaria* (Dictyotaceae family) [125], and arteannuin H (**184**) isolated from *A. annua* [126].

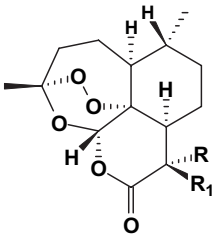
The in vitro cytotoxic activity of the artemisinin (**165**) and some chemically prepared derivatives, which have been found to display cytotoxicity to cloned murine Ehrlich ascites tumor (EAT) cells and human HeLa cells and against murine bone marrow using a clonogenic assay for committed progenitor cells of the granulocyte–monocyte lineage (CFU-GM assay) was reported [127]. Comparing artemisinin to deoxyartemisinin, the endoperoxide group appeared to play a role in cytotoxicity to CFU-GM cells. Dimers of dihydroartemisinin and dihydrodeoxyartemisinin revealed that the stereochemistry of the ether linkage of the dimers was a more important determinant for this cytotoxic activity. The nonsymmetrical dimer of dihydroartemisinin and the corresponding endoperoxide-lacking dimer of dihydrodeoxyartemisinin were equally cytotoxic to CFU-GM cells.

Despite the differences between both systems, it may be stated that most compounds displayed higher cytotoxicity to CFU-GM cells than to EAT cells. Dimers of dihydroartemisinin (**185**, **186**) were selected as potential antitumor compounds and subjected to the National Cancer Institute drug-screening program consisting of about sixty human cancer cell lines derived from nine different tissues (Table 9). Both compounds displayed the same specific cytotoxicity pattern. Throughout the screen dimer **185** was more active than **186** (Tables 10 and 11).

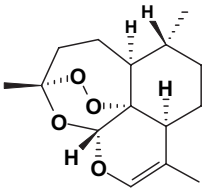


173

174 R = OOH
175 R = COOEt



176 R = Me, R_1 = OH
177 R = OH, R_1 = Me



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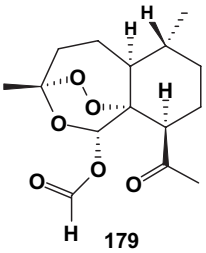

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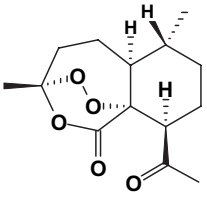

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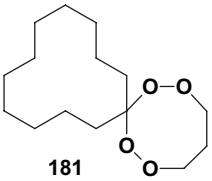
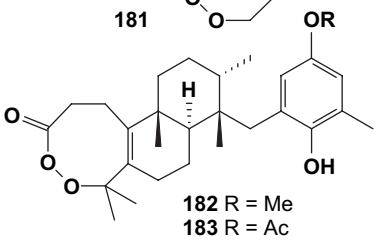
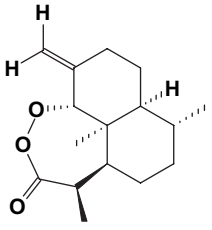


181



182 R = Me
183 R = Ac



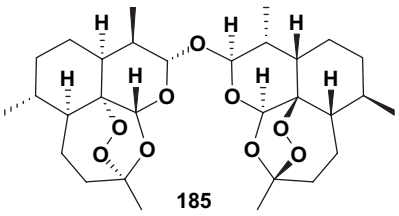
184 Arteannuin H

Table 9
The in vitro cytotoxicity of artemisinin derivatives to tumor cells (IC_{50} μM, Ref. [127])

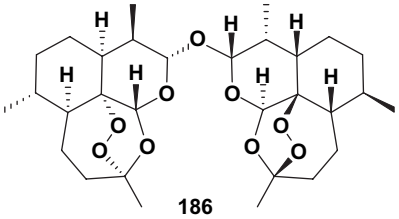
Compound	CFU-GM	EAT[a]	EAT[b]
165	385	>1000	723
173	1.7	10.0	11.3
185	3.6	55.6	3.6
186	52.5	–	31.2

[a] Clonogenic assay.
[b] MMT assay.

The genus *Achillea* (family Asteraceae) comprises about 100 species that commonly occur in temperate regions throughout the Old World, especially on higher mountains of the Mediterranean. Due to numerous medicinal properties, aerial parts of the members of the genus are used widely in traditional medicine. The extracts of aerial parts of *Achillea clavennae*, *A. holosericea*, *A. lingulata* and *A. millefolium* show the strongest antimicrobial activity against five bacteria (*S. aureus*, *E. coli*, *Klebsiella pneumoniae*, *P. aeruginosa* and *Salmonella enteritidis*) and two fungi (*Aspergillus niger* and *C. albicans*) [128].



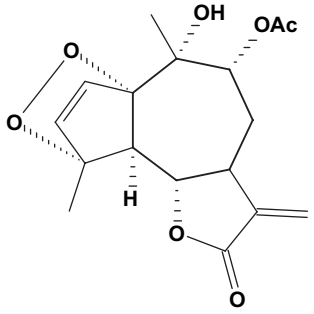
185



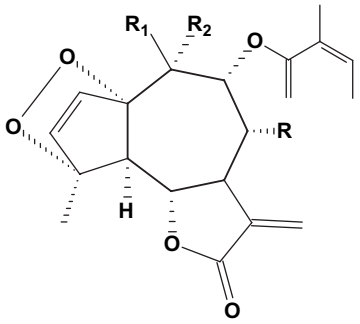
186

The antimicrobial activity of the essential oil from *Achillea fragrantissima* against *C. albicans* [129], extract of *A. clavennae* against Gram-positive (*B. subtilis*, *B. cereus*, *S. aureus*, *Streptococcus faecalis*), Gram-negative (*E. coli*, *K. pneumoniae*, *P. aeruginosa*, *Proteus mirabilis*) and fungal organisms (*A. niger*, *A. fumigatus*, *C. albicans*) was reported [130]. The antimicrobial activities of the essential oils of *Achillea setacea* and *Achillea teretifolia* against 14 microorganisms were also reported [131].

Essential oils exhibited inhibitory effects on *Clostridium perfringens*, *Acinetobacter lwoffii* and *C. albicans* with a range of minimum inhibitory concentration values extending from 0.28 to 2.25 mg/mL. The cytotoxicity and antioxidant properties of herb extracts of *Achillea alexandri-regis* were also studied [132]. Combined chloroform and ethylacetate extracts exhibited a pronounced cytotoxic effect against HeLa cancer cells ($IC_{50} = 25.92$ μg/mL), and lower cytotoxicity against K562 leukemia cells ($IC_{50} = 48.59$ μg/mL). The methanol extract was found to be a moderately cytotoxic in vitro agent



187 Apressin



188 R = H, R_1 = Me, R_2 = OH
189 R = H, R_1 = OH, R_2 = Me
190 R,R_2 = OH, R_1 = Me

against HeLa and K562 cells. Anticancer agent apressin (**187**) was isolated from flowers of *Achillea depressa* [133], and the aerial parts of *Achillea clavennae* [134]. It has shown cytotoxic activity against cancer cell lines (IC_{50} μg/mL): HeLa

Table 10
GI_{50} values (μM) of artemisinin dimeric compounds (Ref. [127])

Tumor cell lines	**185**	**186**
Leukemia		
CCRF-GEM	0.045	7.51
HB-60(TB)	0.028	0.61
K-652	0.068	1.04
MOLT-4	0.057	2.37
RPMI-8226	0.013	0.66
Non-small lung cancer		
A549/ATCC	0.098	18.7
EKVX	0.076	2.16
HOP-62	10.2	>25
HOP-92	0.121	3.66
NCI-H23	0.046	1.15
NCI-H322M	10.8	>25
NCI-H460	0.068	>25
Colon cancer		
COLO 205	0.044	1.17
HCC-2998	0.45	>25
HCT-116	0.019	0.71
HCT15	0.024	0.74
HT-29	0.05	1.32
KM12	0.033	0.88
SW-620	0.043	1.16
CNS cancer		
SF-295	0.59	–
SNB-19	1.71	>25
SNB-75	0.22	–
U251	0.065	>25

Table 11
GI_{50} values (μM) of artemisinin dimeric compounds (Ref. [127])

Tumor cell lines	**185**	**186**
Melanoma		
LOX IMVI	0.03	1.0
SK-MEL-2	0.50	16.9
SK-MEL-28	12.3	>25
SK-MEL-5	0.053	3.6
UACC-257	0.093	>25
UACC-62	0.081	2.0
Ovarian cancer		
IGROV1	1.05	>25
OVCAR-3	0.46	8.2
OVCAR-4	0.29	8.6
OVCAR-5	0.39	>25
OVCAR-8	0.09	1.2
SK-OV-3	0.48	>25
Renal cancer		
786-0	0.37	22.0
A498	11.6	>25
ACHN	0.10	3.5
CAKI-1	0.28	>25
RXF 393	0.47	–
SN12C	0.28	16.8
TK-10	0.24	–
UO-31	0.063	1.7
Prostate cancer		
PC-3	0.015	0.19
DU145	5.1	>25
Breast cancer		
MCF7	0.11	1.51
MCF7/ADR-RES	0.22	1.45
RES MDA-MB-231/ATCC	0.40	>25
MDA-MB-435	0.061	–
MDA-N	0.59	3.68
BT-549	0.058	1.3
T-47D	0.03	1.2

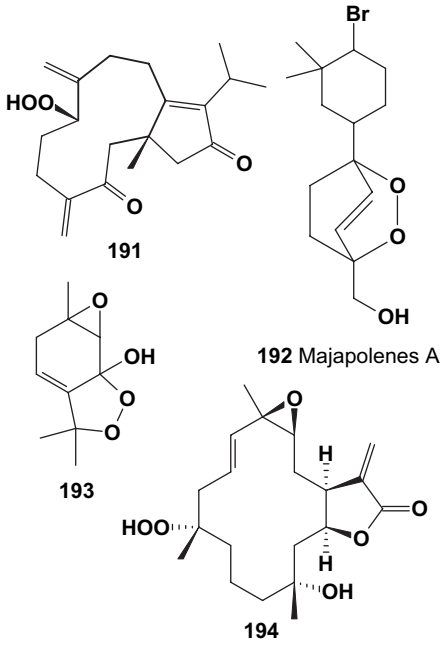
(7.36), Fem-X (5.27), K562 (4.44), PBMC (15.89), and PBMC+PHA (8.78). Bohlmann and Knoll reported the isolation of three bioactive guaianolides similar to apressin (**188**–**190**) from two South African *Anthanasia* species [135].

A new cytotoxic dolabellane diterpene, (1*R*,7*R*)-7-hydroperoxydolabella-4(16),8(17),11(12)-triene-3,13-dione (**191**) has been isolated from the Formosan soft coral *Clavularia inflata*. Compound (**191**), which contains a secondary hydroperoxy group, exhibited cytotoxicity against A549, HT-29, and P388 cell lines with ED_{50} values of 0.56, 0.31, and 0.052 μg/mL, respectively [136].

Sesquiterpene, majapolenes A (**192**, a dioxabicyclo[2.2.2]-alkene) was isolated from a Philippine collection of *Laurencia majuscula*. This metabolite was found as a major component of a Philippine collection of *Laurencia caraibica*. Compound **192** also represents the major and only active component of the *L. mjuscula* extract. It displayed modest mean response parameter values for all NCI 60 cell lines of 0.4 μM for GI_{50}, 0.9 μM for TGI and 2.8 μM for LC_{50} (50% net cell death) [137].

Extracts of *Adenosma caeruleum*, which is used in Vietnamese folk medicine, are showed to have moderate inhibitory activity on the tube-like formation induced by human umbilical venous endothelial cells in the in vitro angiogenesis assay [138]. A monoterpenoid peroxide (**193**) was isolated from the aerial parts of *Adenosma indiana*, and showed activity against the viral hepatitis [139].

A novel hydroperoxide cembrane-type diterpenoid (**194**) was isolated from the soft coral *Sarcophyton crassocaule* collected from the Xisha Islands in the South China Sea. It exhibited strong cytotoxicity against the P388 cell line with IC_{50} value of 0.1 μg/mL [140].



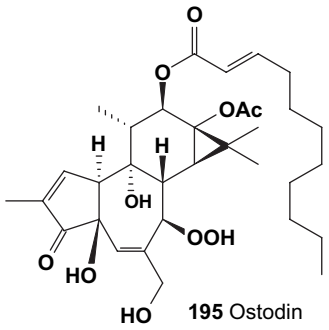
Cytotoxic compound, ostodin (**195**), the Δ5,6-7β-hydroperoxide was isolated from a chloroform extract of the stems and fruits of *Ostodes paniculata* (Euphorbiaceae). It showed anticancer activity against P388 and KB in vitro cell lines at $ED_{50} \leq 4$ mg/mL) [141].

Cytotoxic cembranoid, planaxool (**196**), was isolated from the marine mollusk *Planaxis sulcatus*. Planaxool showed cytotoxicity (IC_{100}) against L-1210 (mouse murine leukemia) cell line at a level of 2.4 μg/mL [142]. Mulin-11,3-dien-20-oic acid (**197**), showed trypanocidal activity against all stages of *T. cruzi*, including intracellular amastigotes. At 10 μM, this compound displayed a strong lytic activity [143].

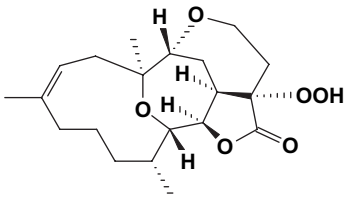
Cytotoxic hydroperoxy sesquiterpene lactone peroxyferolide (**198**) was isolated from *Liriodendron tulipifera*. It possesses antitumor activity against KB cells with an ED_{50}, 0.29 μg/mL [144].

New hydroperoxy lepidozenes (**199**–**201**) were recovered from extracts of marine anemone *Anthopleura pacifica* [145]. Isolated sesquiterpenoids exhibit the following cytotoxicity against murine melanoma cells (IC_{50} μg/mL): **199**, 0.7; **200**, 2.3 and **201**, 0.9.

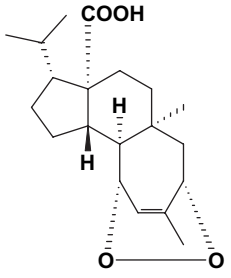
Nardosinone (**202**) for the first time was identified from *Nardostachys chinensis* (Valerianaceae), in 1965 [146], and structure was confirmed a decade later [147]. More recently, it has been detected in the same plant [148,149]. It showed



195 Ostodin

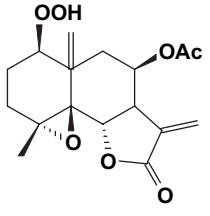


196 Planaxool

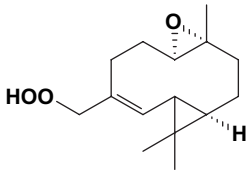


197 Mulinic acid

cytotoxic activity against P388 cells [150]. Nardosinone-induced enhancement of the NGF-action was completely blocked by PD98059, a representative mitogen activated protein kinase, and it is also a kinase inhibitor [151,152].



198 Peroxyferolide



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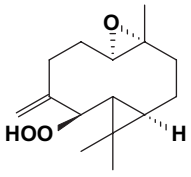
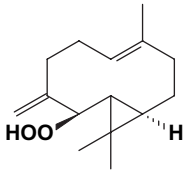


200



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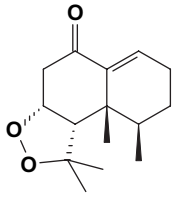


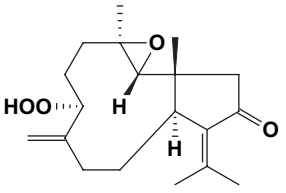
202 Nardosinone

New diterpenoid, (**203**) having a dolabellane skeleton was isolated from the Okinawan soft coral of the genus *Clavularia*. This diterpenoid showed cytotoxic activity against several tumor cell lines [153].

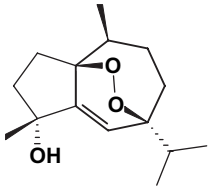
Oxygenated sesquiterpenoid, peroxygibberol (**204**), was isolated from a Formosan soft coral, *Sinularia gibberosa*. The sesquiterpene **204** exhibited moderate cytotoxicity against the growth of the Hepa59T/VGH cell line at ED_{50} 8.2 μg/mL [154].

Two major carotane sesquiterpenes, rugosal A (**205**) and rugosic acid A (**206**) were isolated from *Rosa rugosa* leaves [155]. The occurrence of rugosal A in the exudate suggests a possible defensive role of the glandular trichome against pest organisms, as it has antifeedant activity against tobacco cutworm larvae. Yingzhaosu A (**207**) was isolated from Chinese medicinal herb *Artabotrys uncinatus*, and it showed strong antimalarial activity against *P. falciparum* [156].

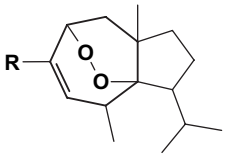
Herb *Juniperus phoenicea* (Phoenician Juniper or Arâr) is a juniper found throughout the Mediterranean region, from Morocco and Portugal east to Turkey and Egypt, and also on Madeira and the Canary Islands, and on the mountains of western Saudi Arabia near the Red Sea [157]. The essential oils of a juniper have shown antimicrobial activity against *C. albicans*, *S. aureus*, *E. coli*, and *P. aeruginosa* [158]. New oxygenated diterpenic acids isolated as their Me ester were (**208**, **209**) isolated from Moroccan *J. phoenicea* and *Juniperus thurifera* var. *africana* (Cupressaceae) [159]. The cytotoxicity of the abietane diterpenoids was reported against five cell lines (Table 12).



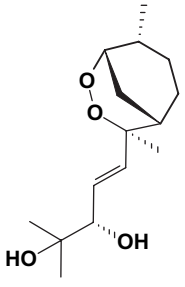
203



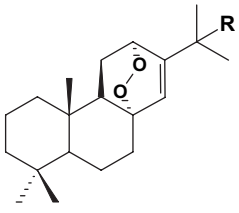
204 Peroxygibberol



205 Rugosal A,
R = CHO
206 Rugosic acid A,
R = COOH



207 Yingzhaosu A



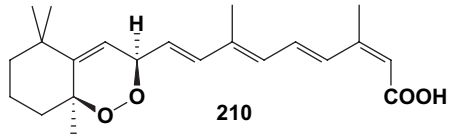
208 R = OOH
209 R = OH

Table 12
Cytotoxic activity oxygenated diterpenic acids Me (IC_{50} μg/mL, Ref. [157])

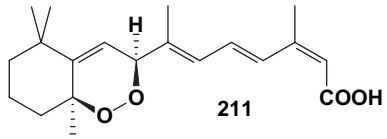
Cell line	A549	H-116	PSN1	T98G	SKBR3
208	5.0	2.5	5.0	>5.0	>5.0
209	>5.0	>5.0	>5.0	>5.0	>5.0
Cyclohex[a]	0.1	0.1	0.01	2.5	0.05

[a] Cycloheximide.

Synthesis of the β-carotene oxidation products, 5*S*,8*S*-epidioxy-5,8-dihydroretinoic acid (**210**) and 13-*cis*-5*S*,8*S*-epidioxy-5,8-dihydroretinoic acid (**211**) was achieved in six steps starting from β-ionone [160]. Evaluation of the photooxygenation products (**210**, **211**) on a panel of five cancer cell lines showed a significant cytotoxic activity (Table 13).

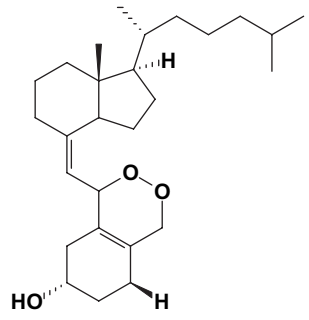


210



211

Epidioxyvitamin D3 derivatives were prepared by photochemical oxygenation of vitamin D3 derivative and possessed antileukemia activity with minute effects on Ca^{2+} metabolism [161]. Photochemical oxygenation of 24*R*-hydroxyvitamin D3 in C_6H_6 containing Rose Bengal gave the two C6 isomers of epidioxysecocholestadienediol (**212**, **213**). Anticancer activity at 25 μg/mL (**212**) and 250 μg/mL (**213**) against human myeloid leukemia cells was reported.



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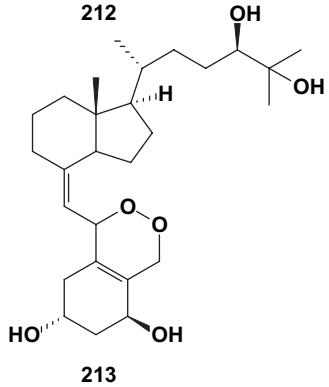

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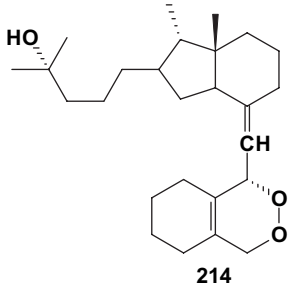
Table 13
Cytotoxicities (IC_{50} μg/mL, Ref. [160]) of RA oxidation products **210** and **211** against a panel of five cancer cell lines

Cell line[a]	NCI-H460	MCF-7	SF-268	MIA-PaCa2	HeLa
210	21.0	21.9	NA	13.9	19.8
211	2.1	2.7	3.5	1.7	2.0
Dox	0.01	0.07	0.04	0.05	ND

NA, not active at 30 μM; ND, not determined.
[a] Key: NCI-H460, human non-small cell lung cancer; MCF-7, human breast cancer; SF-268, human CNS cancer (glioma); MIA Pa Ca-2, metastatic human pancreatic cancer; HeLa, human cervical carcinoma. Doxorubicin (Dox) was used as the positive control.

Two anticancer compounds (**214**, **215**) were prepared by oxidation of 25-hydroxyvitamin D3 [162]. Both isolated products inhibited the growth of myelogenous leukemia HL60 cells.



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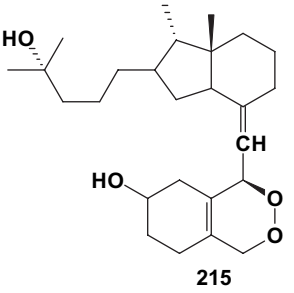


215

4. Steroidal peroxy metabolites

Many steroids containing (-OOH and/or –O-O-) group(s) have been isolated from aquatic and terrestrial organisms [2,4]. 24-Hydroperoxy-24-vinylcholesterol (**216**), and 29-hydroperoxystigmasta-5,24(28)-dien-3β-ol (**217**), highly cytotoxic compounds, were recovered from the brown alga *Turbinaria conoids* [163], and more recently from the methanolic extract of the Indonesian red alga *Ceratodictyon spongiosum* and its symbiotic sponge *Sigmadocia symbiotica* [164]. Reported activities are shown in Table 14.

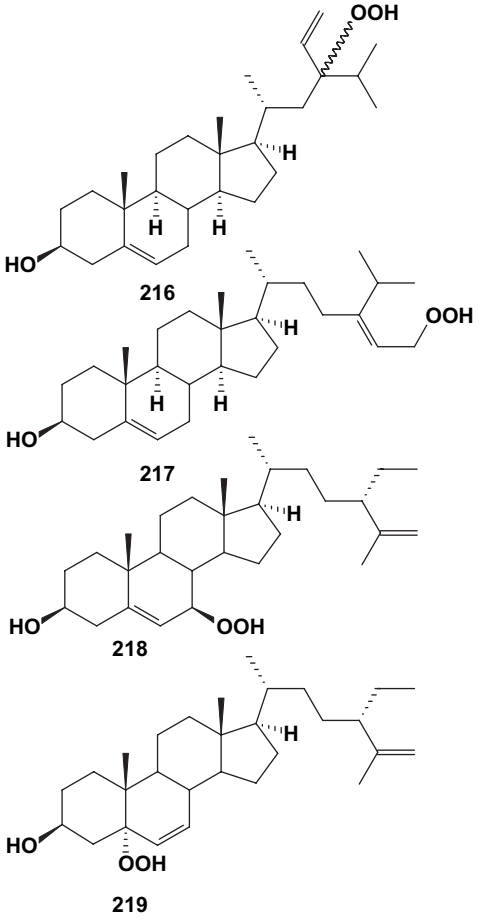
The cytotoxic steroid (**217**) obtained from *Ciona intestinalis* was prepared by photooxidation of fucosterol in the presence of eosin; small amounts of (**217**). Compound (**217**) and intermediates have showed cytotoxicity against L-1210 leukemia cells [165].

Table 14
Cytotoxicity peroxy sterols from marine algae (ED_{50} μg/mL, Refs. [163,166])

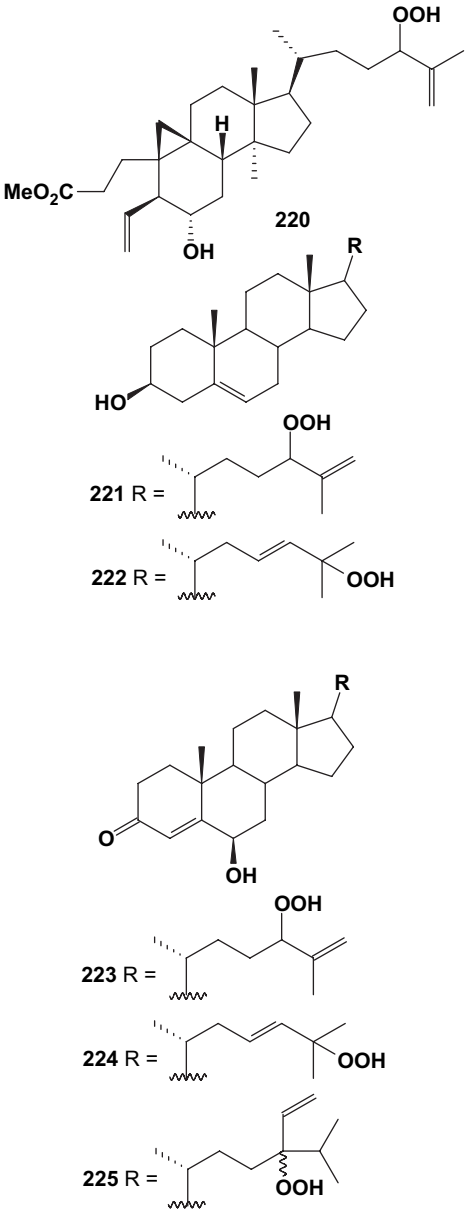
Sterol	P388	KB	A549	HY-29
216	0.3	1.8	7.1	5.9
217	0.5	2.2	2.2	5.0
218	0.4	1.0	0.5	1.0
219	0.5	1.0	1.1	0.9

The two hydroperoxy clerosterols, (24*S*)-24-ethyl-7-oxocholesta-5,25-dien-3β-ol (**218**), and (24*S*)-24-ethyl-cholesta-5,25-dien-3β,7α-diol (**219**), were isolated from the marine green alga *Codium arabicum*. Isolated peroxides have showed significant cytotoxicity toward various cancer cell lines (Table 14) [166].

The (6*S*)-hydroxy-29-nor-3,4-seco-cycloart-4(30),24-dien-3-oic acid (**220**) was isolated from the aerial parts of *Antirhea acutata* [167]. Sterol (**220**) has shown moderate inhibitory activities in cyclooxygenase-1 and -2 assays (IC_{50} 43.7 and 4.7 μM, respectively).



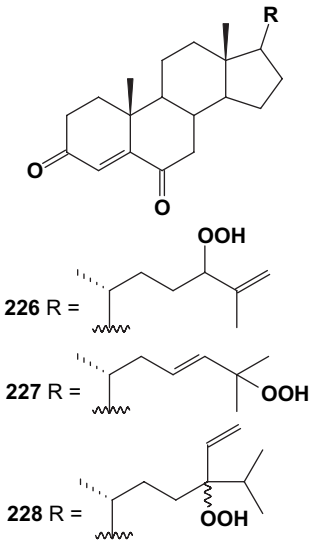
Several cytotoxic oxygenated desmosterols (**221**–**224**, **226**, **227**) were obtained from extract of the red alga *Galaxaura marginata*, which was collected near Taiwan [168,169]. Isolated sterols exhibited significant cytotoxicity toward several cancer cell lines (Table 15).

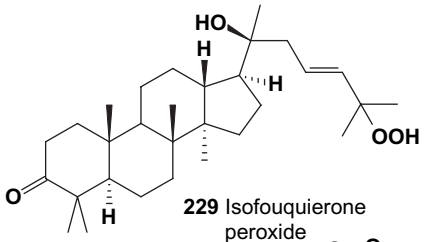


Two additional cytotoxic sterols, e.g. 24-hydroperoxy-24-ethylcholesta-4,28(29)-dien-3,6-dione (**225**), and 24-hydroperoxy-6β-hydroxy-24-ethylcholesta-4,28(29)-dien-3-one (**228**) were isolated from the marine brown alga *Turbinaria conoides* [170], and their activities are presented in Table 15.

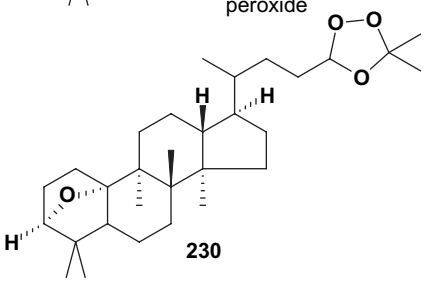
Table 15
Cytotoxic activity of oxygenated desmosterols (ED_{50} μg/mL, Refs. [168–170])

Sterol	P388	KB	A549	HT-29
221	0.26	0.33	0.64	0.43
222	0.22	1.41	1.68	1.27
223	0.22	0.79	0.58	0.47
224	0.28	0.40	1.00	0.63
225	0.40	1.80	1.80	1.70
226/227	0.53	1.15	0.88	1.48
228	0.80	4.00	2.50	1.40





229 Isofouquierone peroxide



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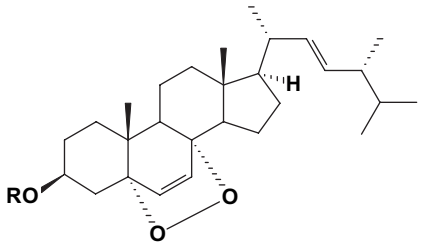


231 Ergosterol peroxide, R = H
232 R = 3β-O-Glc

The genus of *Rhus* contains approximately 250 species of woody shrubs and small trees in the family Anacardiaceae. *Rhus javanica* is a tall, broad leaved tree that is distributed in Korea, Japan and China. Its barks and leaves are used as traditional remedies of dysentery and diarrhea in Korea [171]. Recently, the antineoplastic effect of *R. javanica* has been reported [172]. The antioxidant, cytotoxic, and antitumorigenic activities of a fractionated, ethanol extract derived from *Rhus verniciflua*, a plant indigenous to Korea, China, and Japan were also reported [173]. Cytotoxic dammarane triterpene isofouquierone peroxide (**229**) was isolated from the stem bark of *R. javanica* [174].

From the dried and fresh aerial parts of *Senecio selloi* (Asteraceae), the new triterpene ozonide (**230**) was isolated. Natural sterol has shown antiplasmodial activity but synthesized analog is inactive [175].

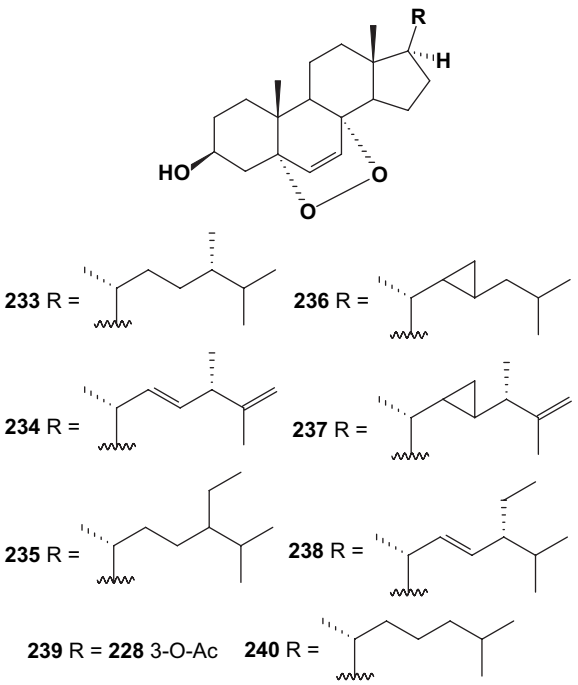
The ubiquitous ergosterol peroxide (**231**) was isolated from number of sources [2,4], marine as well as terrestrial. In addition, a glucosylated derivative of ergosterol peroxide (**232**) has been obtained from extract of *Hericum erinacens* [176]. Ergosterol peroxide (**232**) showed potent cytotoxicity against mouse lymphemia L-1210/v/c in vitro, but exhibited no significant antitumor activity against either leukemia P388 in CDF1 mouse or sarcoma 180 in mouse [177]. It displayed potent antitumor activity against Walker 256 carcinosarcoma and MCF7 breast cancer cell lines [178]. Ergosterol peroxide was found to be a greater inhibitor to the proliferation of K562, Jurkat, WM-1341, HL60 and RPM1-8226 tumor cell lines by 10–40% at 10 μg/mL [179].

Ergosterol peroxide from the marine sponge *Spirastrella abata* showed cytotoxicity against five human solid tumor cell lines including A549, SK-OV-3, SK-MEL-2, XF498, and HCT15 [180], and also against human gastric tumor cell line (SNU-1), human hepatoma cell line (SNU-354), human colorectal tumor cell line (SNU-C4) and murine sarcoma 180 and were of 18.7, 158.2, 84.6 and 74.1 μM (IC_{50}), respectively [181].

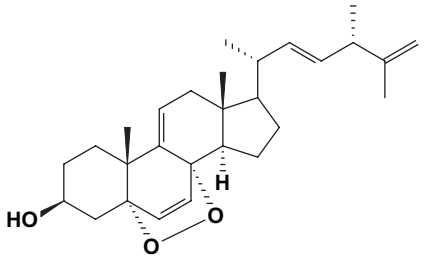
Ergosterol peroxide was isolated from the fruiting bodies of *Ganoderma applanatum*, and it was found to exhibit potent rat lens aldose reductase inhibition, with IC_{50} value being 15.4 μg/mL [182]. Ergosterol peroxide was isolated for the first time in *Oryza sativa*. This is the first report of potential allelopathic activity of steroids on weeds based on their phytotoxicity on barnyardgrass (Echinochloa crus-galli) as target species [183]. Among the lipophilic extracts of seven traditional edible mushrooms, the acetone extract of *Sarcodon aspratus* markedly inhibited the growth of HL60 human leukemia cells and induced apoptosis after 24 h incubation. The major active component was identified as ergosterol peroxide [184]. It completely inhibited growth and induced apoptosis of HL60 cells at a concentration of 25 μM. The glycosylated form of ergosterol peroxide (**232**) from the mycelia of *Cordyceps sinensis* was found to be a greater inhibitor to the proliferation of K562, Jurkat, WM-1341, HL60 and RPM1-8226 tumor cell lines by 10–40% at 10 μg/mL than it's previously identified aglycon, 5α,8α-epidioxy-24(*R*)-methylcholesta-6,22-dien-3β-ol [185].

A number of other steroidal endoperoxides (**233**–**240**) have been reported which differ in the nature of the side chain. These peroxides have been identified from marine and terrestrial sources [2,4]. Among isolated compounds **238** was found to be an inhibitor of induced inflammation and tumor promotion in mouse studies [177]. A series of the cytotoxic components were isolated from the marine sponge *Spirastrella abata*,

5α,8α-epidioxy Δ6 sterols and 5α,8α-epidioxy Δ6,9(11) sterols (**233**–**235**, **238**, and **230**). These compounds showed cytotoxicity against five human solid tumor cell lines including A549, SK-OV-3, SK-MEL-2, XF498, and HCT15 [180].



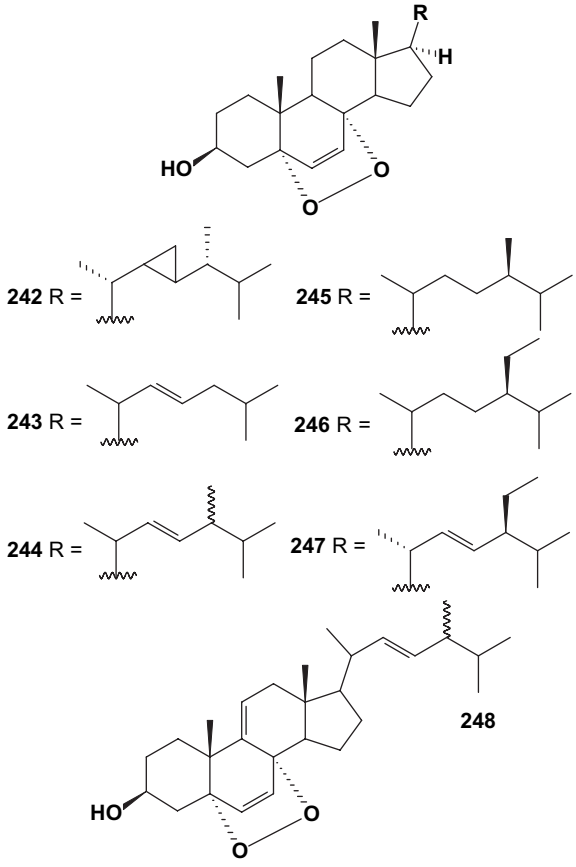
A new epidioxyergostane-type steroid, 9(11)-dehydroaxinysterol (**241**), as well as axinysterol (**234**) were isolated from the Okinawan sponge of the genus *Axinyssa* [186]. Epidioxysterol (**241**) was found to show significant growth inhibitory effects against human cancer cell lines (Table 16). Compound **234** has shown the lethal bioassay for brine shrimp.



241 9(11)-Dehydroaxinysterol

A new sterol, (22*R*,23*R*,24*R*)-5α,8α-epidioxy-22,23-methylene-24-methylcholest-6-en-3β-ol (**242**) has been isolated from a soft coral *Sinularia* sp. [187]. Cytotoxicity of sterol toward various cancer cell lines was at 0.4 μg/mL for P388, 2.1 for KB, 2.7 for A549, and 1.4 for HT-29. Clastogenic and cytotoxic activities of the fractions of 5α,8α-epidioxysterols of the marine sponge *Ircinia campana* (class Demospongia) collected in the Colombian Caribbean, was reported [188]. The results showed that at the highest concentrations, 422.5 μg/mL and 42.25 μg/mL they were cytotoxic, while at the concentration of 4.22 μg/mL they did not show genotoxic effect on the lymphocytes. From this cytotoxic fraction sterol (**242**) was isolated.

Several of unusual steroids belonging to the 5α,8α-epidioxy sterol family (**233**, **235**, **238**, **240**, **243**–**248**) have been isolated from the lipid extract of the marine sponge *Luffariella* cf. *variabilis* from Mayotte (Indian Ocean) [189]. The mixture of the 10 steroids showed inhibitive activity against the human T cell leukemia/lymphotropic virus type I (HTLV-I) and also displayed cytotoxic activity against the human breast cancer cell line (MCF7WT).



Sterol with antitumor activity (**233**) was isolated from the edible mushroom *S. aspratus* (BERK S. ITO) [190]. Purified sterol was a more effective inhibitor of HL60 leukemia cell growth and stronger apoptosis-inducer than ergosterol peroxide (**231**). It selectively suppressed the growth of HT-29 human colon adenocarcinoma cells but did not suppress WI38 normal human fibroblasts. In addition, sterol (**248**) also was isolated. Cytotoxic sterol (**244**) was also isolated from fruiting bodies of formosan *Ganoderma lucidum* [191], it exhibited potent inhibition of KB cells and human PLC/PRF/5 cells in vitro, at ED_{50} 9.97 and 10.99 μg/mL, respectively.

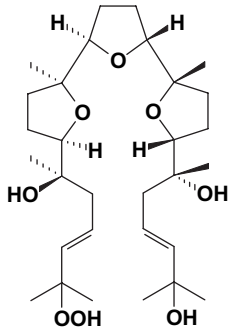
5. Miscellaneous anticancer peroxides

Eurycoma longifolia (Tongkat Ali or Pasak Bumi) is a flowering plant in the family *Simaroubaceae*, native to Indonesia and Malaysia. Historically, it has been used as medicine by

Table 16
Growth inhibitory activity of **241** against human cancer cell lines (Ref. [186])

Origin of cancer	Cell line	IC_{50} (μg/mL)
Breast	HBC-4	0.85
	BSY-1	0.60
	HBC-5	0.96
	MCF-7	0.36
	MDA-MB231	1.26
Lung	NCI-H23	0.54
	NCI-H226	0.63
	NCI-H522	0.57
	NCI-H460	0.61
	A549	0.96
	DMS273	0.54
	DMS114	0.48
Stomach	ST-4	0.69
	MKN1	0.42
	MKN7	0.48
	MKN28	0.84
	MKN45	0.54
	MKN74	0.54
Kidney	RXF-63IL	0.72
	ACHN	0.51
Colon	HCC-2998	0.57
	KM-12	0.60
	HT-29	0.57
	HCT-15	0.75
	HCT-116	0.48
Ovary	OVCAR-3	0.19
	OVCAR-4	0.60
	OVCAR-5	0.54
	OVCAR-8	0.22
	SK-OV-3	0.81
CNS	U251	0.63
	SF-268	1.02
	SF-295	0.75
	SF-539	0.84
	SNB-75	2.16
	SNB-78	1.17
Prostate	DU145	0.54
	PC-3	0.57
Melanoma	LOX-IMVI	0.60

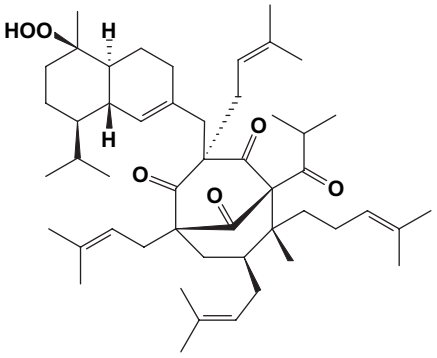
the folk in it's countries of origin as a libido enhancer and to treat various sexual dysfunctions. It recently was reported that *E. longifolia* caused increased muscle strength and size when compared to a placebo [192]. Longilene peroxide (**249**), from *E. longifolia*, represents a new qualene-type triterpene. The compound exhibits cytotoxic activity against KB cells, IC_{50} 5.3 μg/mL [193,194].



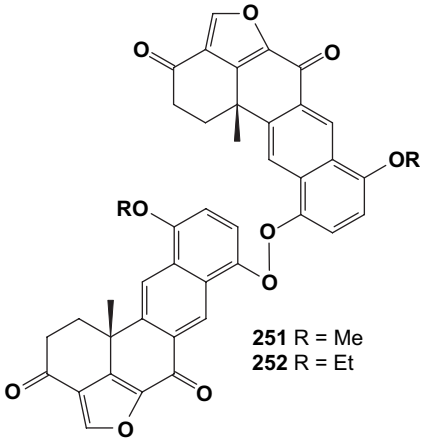
249

A new C50-hydroperoxide with a hitherto unknown constitution, containing cadinane hydroperoxide and hyperfoin as partial structures, was isolated from the stems and leaves of St. John's wort (*Hypericum perforatum*, Hypericaceae). This unusual compound entitled hydroperoxycadiforin (**250**) contains a hydroperoxo group in its molecule [195]. Hyperforin itself was shown to inhibit or modulate several neurotransmitter systems in vitro. It is a potent uptake inhibitor of serotonin, dopamine, noradrenaline and GABA, with IC_{50} values of about 0.05–0.1 μg/mL, and of L-glutamate at 0.5 μg/mL in synaptosomal preparations [196]. Hyperforin has been demonstrated as a modulator of several neuronal ion channels, and inhibits smooth muscle contraction induced by various neurotransmitters. Hyperforin (0.3–10 μg/mL) caused a concentration-dependent elevation of Ca^{2+} and extracellular acidification rate, and both of these effects are independent of extracellular Ca^{2+} [197].

Two novel pentacyclic polyketide dimers, dihalenaquinolides A (**251**) and B (**252**), have been isolated from the marine sponge *Petrosia elastica* collected in Nan-wan, Taiwan [198]. The cytotoxic activity of the isolated pentacyclic hydroquinones and quinones were tested in vitro against human prostate (PC3) and hepatoma (Hep3B) tumor cell lines, and results are shown in Table 17.



250 Hydroperoxycadiforin



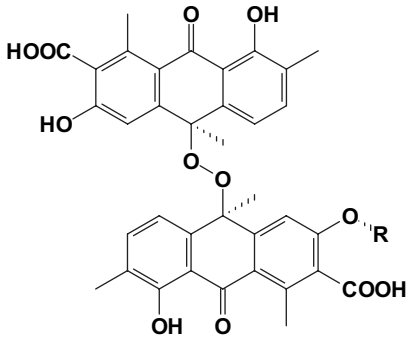
251 R = Me
252 R = Et

Table 17
Cytotoxicity of polycyclic hydroquinones (Ref. [198])

Compound	PC-3	HERP3B
251	56	5
252	35	5
Taxol (0.1 μg/mL)	80	85

PC-3, human prostate cancer cells; Herp3b, human hapatoma cancer cells.

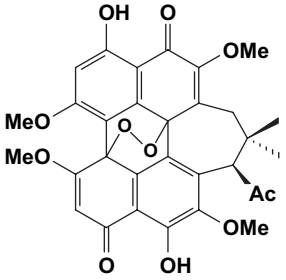
The novel antibiotic oxanthromicin (**253**) was isolated from *Actinomadura* SCC 1646 [199,200]. Compound (**254**) showed good activity in vitro against dermatophytic fungi and moderate activity against *C. albicans* and *S. aureus*. Adxanthromycins A (**254**) and B (**255**) are new inhibitors of ICAM-1/LFA-1 mediated cell adhesion molecule isolated from the fermentation broth of *Streptomyces* sp. NA-148. Adxanthromycins A and B inhibited the formation of the JY cell aggregates from 1.5 μg/mL, respectively, in a dose-dependent manner. Components A and B also inhibited a human T cell leukemia cell line SKW-3 adhesion to soluble ICAM-1 in a dose-dependent manner with an IC_{50} of 18.8 μg/mL and 25.0 μg/mL, respectively [201,202].



253 Oxanthromicin, R = H
254 Adxanthromycin A, R = Gal
255 Adxanthromycin B, R = Gal(4-1)Gal

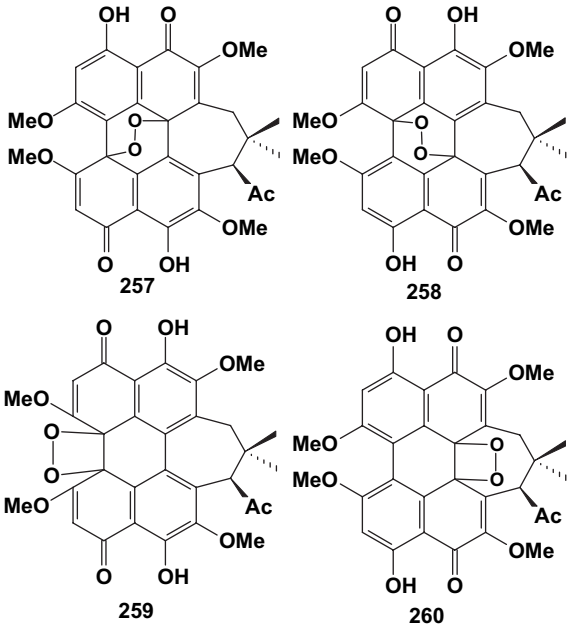
Hypocrellins are dark red pigments having the perylenequinone structure, with photodynamic activity toward microorganisms. These pigments produced by the fungus *Hypocrella bambusae* [203,204], and *Shiraia bambusicoia* [205–207]. All isolated metabolites have shown anticancer activities [205,208,209], and antiviral activity against the human immunodeficiency virus (HIV-1) [210]. Thus, hypocrellin D significantly inhibited the growth of tumor cell lines Bel-7721, A549 and Anip-973 with IC_{50} values of 1.8, 8.8, 38.4 μg/mL, respectively [205]. Natural cytotoxic peroxyhypocrellin (**256**) was isolated from *S. bambusicoia* [206]. Several cytotoxic peroxyhypocrellins (**257–260**) were obtained by the photooxidation of hypocrellin A [203].

The mechanism of photooxidation of perylenequinonoid pigment, hypocrellin A (HA), was reported [211]. It was found that singlet oxygen plays a key role in the photooxididation of HA. The following processes were detected to be involved in the photooxidation of HA using ^{18}O atom labeling. After the HA was photoexcited, it tautomerizes and dissociates to produce the anion, which reacts with singlet oxygen generated



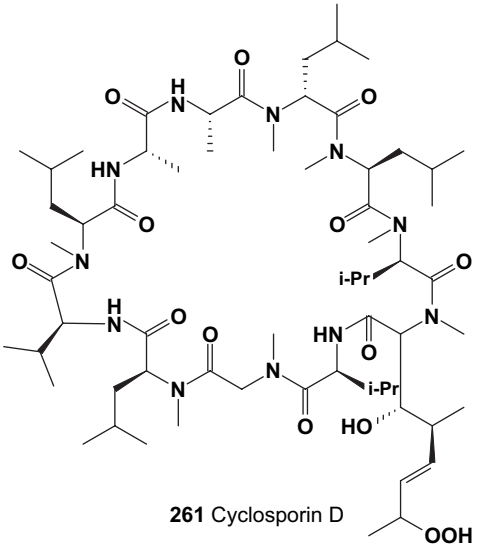
256 Peroxyhypocrellin

during irradiation, resulting in the formation of an endoperoxide. The endoperoxide then rearranges to form a dioxetane which undergoes cycloreversion to produce the final product, a di-α-naphthoquinone. Of these factors, the extremely strong effect of pH on the photooxidation of hypocrellin A may be used as a basis for the selective localization of hypocrellin A in tumor tissue. On the basis of the proposed mechanism for the photooxidation of HA, it may be possible to design and synthesize an optimal tumor selective PQP photosensitizer for the photodynamic therapy of human tumors.

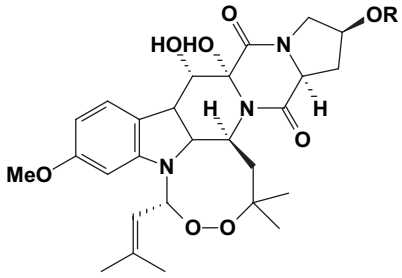


Cyclosporins are produced by certain species of the filamentous fungi, belonging to the genus *Tolypocladium* [212,213]. Some cyclic peptides and depsipeptides are synthesized in microorganisms by large multienzymes called nonribosomal peptide synthetases. The structures of peptide products originating in this way are complex and diverse and are microorganism-specific. Proven cytotoxic, anti-inflammatory, anticancer, and immunosuppressive activities of some cyclic peptides indicate that these molecules may contribute to the synergistic array of fungal virulence factors and support microbial invasion during fungal infection. The interesting structure of Cyclosporin D (**261**) with –OOH group, was assigned to a new metabolite of the fungus *Tolypocladium*

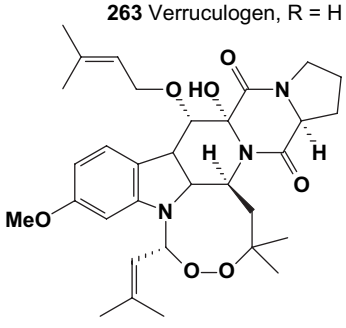
terricola [214]. Rare alkaloids, acetoxyverruculogen (**262**), and mycotoxin, verruculogen (**263**, also known as verruculogen TR 1), a tremorgenic metabolites were isolated from *Penicillium verruculosum*. The biosynthetic pathway for **262** was examined with [^{13}C]acetate [215]. Fumitremorgin A (**264**) known as neurotropic metabolite [216] was produced by fungus *Aspergillus fumigatus* [217]. Verruculogen showed cytotoxicity against A549 (lung cell line) and Hep G2 (liver cells) [218].



261 Cyclosporin D

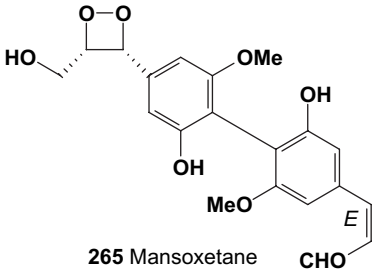


262 R = Ac
263 Verruculogen, R = H

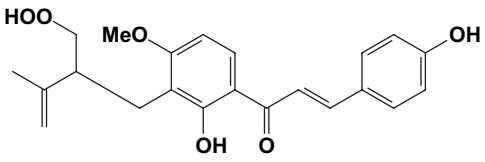


264 Fumitremorgin A

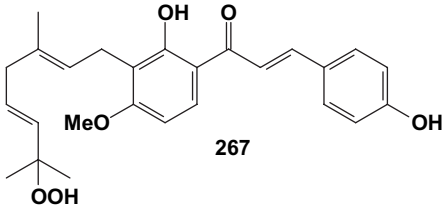
The unique neolignan mansoxetane (**265**), isolated from the heartwood of *Mansonia gagei*, is the first example of a biphenylneolignan with a dioxetane ring discovered in nature [219], although many synthesized compounds with the same dioxetane ring have showed anticancer activity [220,221]. Xanthoangelol E (**266**), which is isolated from the root of *Angelica keiskei* (Umbelliferae), shows the effects of xanthoangelol, on NF-κB activation and ET-1 gene expression in cultured porcine aortic endothelial cells [222]. It has the potential to modulate arachidonic acid metabolism in platelets [223], and inhibited the phenylephrine-induced vasoconstriction through endothelium-dependent endothelium-derived relaxing factor production and/or nitric oxide production [224]. Xanthoangelol E, also effectively inhibited the production of TXB2 and HHT from exogenous AA in platelets [225]. Therapeutic agent **267** isolated from roots of *A. keiskei*, is also used for insulin-related disease [226].



265 Mansoxetane

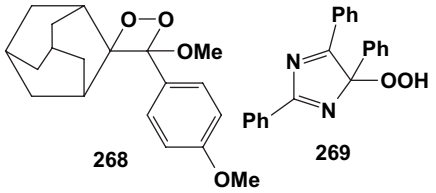


266 Xanthoangelol E



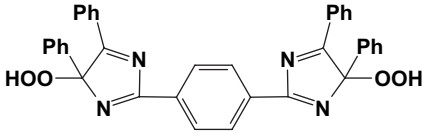
267

Antitumor agents (**268–270**) for heat generation or singlet oxygen generation, destroy cancer cells by the action of heat and/or singlet oxygen are effective as a new cancer remedy alleviating the burden to be imposed on the patients, have been synthesized, tested for anticancer activity, and obtained a wholly satisfactory results [227].



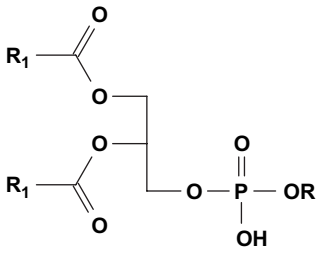
268

269

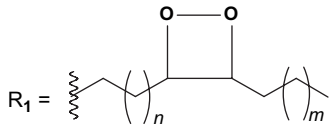


270

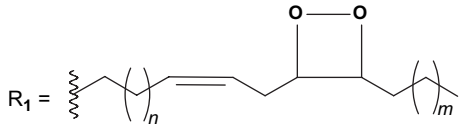
The unique dioxetanilated phosphatidic acids (**271**–**279**) as novel anticancer agents were synthesized [228]. All prepared phospholipids with dioxetane containing ring(s) fatty acids have shown anticancer activity against L-1210 tumor cells.



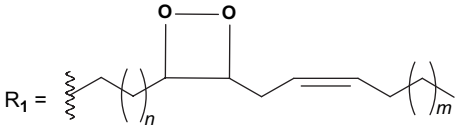
271 R = Serine
272 R = Inositol
273 R = Ethanolamine



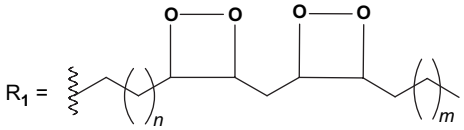
274 n = m = 6



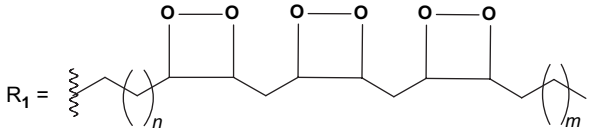
275 n = 6, m = 3



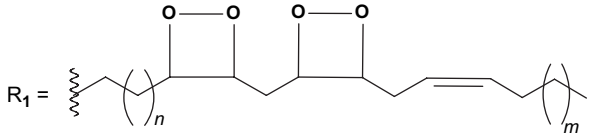
276 n = 6, m = 3



277 n = 6; m = 3



278 n = 6; m = 3



279 n = 6; m = 3

Linoleic acid and trilinolenoylglycerol dioxetanes were prepared by ozonation of linoleic acid Me at −80 °C in Me_2CO [229]. Obtained trilinolenoylglycerols with dioxetane group(s), and linoleic acid Me dioxetanes, also were cytotoxic against L-1210 leukemia cells [229].

Lipoxygenases (linoleate: oxygen oxidoreductase, LOX) are a family of monomeric non-heme, non-sulfur iron dioxygenases, which catalyze the conversion of polyunsaturated fatty acids into conjugated hydroperoxides [230]. The unsaturated fatty acids, which are essential in humans, are absent in most bacteria and thus LOXs are also absent in typical prokaryotes. LOXs are widely expressed in animal and plant cells, sometimes at high level, and their activity may initiate the synthesis of a signaling molecule or may induce structural or metabolic changes in the cell [231]. LOX products have been shown to induce programmed cell death in human T cells [232], neutrophils [233], PC12h cells [234] and Jurkat cells [235].

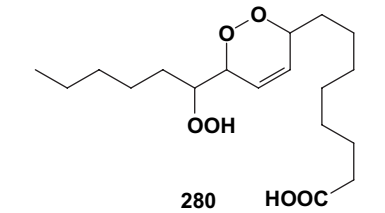
Fatty acid hydroperoxides can be cleaved off from the phospholipids by phospholipase A2 [236], e.g. by its mitochondrial Ca^{2+}-dependent isoform induced by superoxide [237] or by the Ca^{2+}-independent isoform. Fatty acid hydroperoxides are transient, non-radical but reactive species, which are eventually degraded by glutathione peroxidase or phospholipid hydroperoxide glutathione peroxidase to the corresponding hydroxyl fatty acids. Fatty acid hydroperoxides may also decompose to toxic epoxy acids and α,β,γ,δ-unsaturated aldehydes [238]. Hydroperoxides and/or epoxides of unsaturated fatty acids were detected, isolated, and identified from different sources [239–241]. Free radical-initiated autoxidation of polyunsaturated fatty acids has been implicated in numerous human diseases, including atherosclerosis and cancer [242].

Ultraweak chemiluminescence arising from lipoperoxidation has been attributed by several authors to the radiative deactivation of singlet oxygen and triplet carbonyl products [243–247]. The latter emitters have been suggested to come from annihilation of RO and ROO radicals as well as from the thermolysis of dioxetane intermediates formed by (2 + 2) cycloaddition of one O_2 to polyunsaturated fatty acids. Interesting experimental data have been obtained by Brazil researchers [248]. Authors studied the formation of free radicals which formed dioxetane derivatives of some polyunsaturated fatty acids, as intermediates. Reexamining the chemiluminescence properties of dioxygenated tetramethylethylene and linoleic acid and comparing them with those of tetraethyldioxetane, a hindered dioxetane, and corroborated with the literature information, authors conclude that only steric hindrance leads to dioxetane formation upon singlet oxygen addition to electron-poor olefins, albeit in very low yields. Two linoleic acid derivatives containing the dioxetane group (**275**, **276**), and other three peroxides (**280**–**282**) were identified [248]. 1,2-Dioxetane derivative of oleic acid Me (**274**) was obtained by oxidation of methyl oleate, and then reduction with $SnCl_2$ [249].

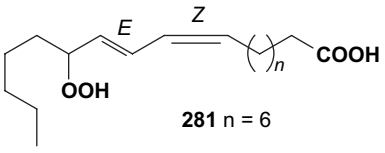
Dioxetane-induced oxidations of guanine base in DNA and 2′-deoxyguanosine, genotoxicity and mutagenicity of dioxetanes in human cells, inhibitory effect of fatty acid ester hydroperoxides in oxidative DNA damage induced by photosensitizers, intercalating hydroperoxides and *N*-hydroxypyridinethiones as photochemical hydroxyl radical sources for oxidative DNA damage, and genotoxicity of furocoumarin hydroperoxides in cells has recently been observed [250].

6. Concluding remarks

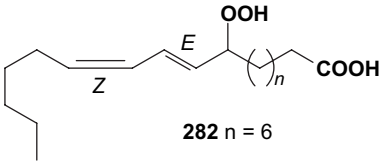
Natural and synthesized peroxy compounds have attracted the attention of biologists and chemists throughout the world for the past 10 decades. As a result of the potential for new drug discovery, terrestrial and aquatic natural products have attracted scientists from different disciplines, such as organic chemistry, bioorganic chemistry, pharmacology, ecology and



280



281 n = 6



282 n = 6

biology. This interest has led to the discovery of almost 600 natural peroxy products to date and many of the compounds have shown very promising biological activity. Natural as well as synthesized peroxides are highly toxic and can damage cellular macromolecules, including lipids, proteins and DNA. Furthermore, these compounds participate in free radical reactions that generate more reactive organic radicals which thereby increase their toxicity, and many from them are shown to have cytotoxic, antibacterial, and/or antimicrobial activities. Organic peroxides are well established synthetic agents in the manufacture of many pharmaceutical intermediates, herbicides, insecticides and various other fine chemicals. Organic peroxides offer opportunities to reduce the number of reaction steps in synthetic routes applying classical synthetic procedures.

References

[1] V.M. Dembitsky, T.A. Gloriozova, V.V. Poroikov, Mini Rev. Med. Chem. 7 (2007) 571–589.
[2] D.A. Casteel, Nat. Prod. Rep. 16 (1999) 55–73.
[3] M. Jung, H. Kim, K. Lee, M. Park, Mini Rev. Med. Chem. 3 (2003) 159–165.
[4] D.A. Casteel, Nat. Prod. Rep. 9 (1992) 289–312.
[5] K.R. Lee, Saengyak Hakhoechi (in Korean) 22 (1991) 145–155.
[6] S. Matsugo, N. Kayamori, T. Konishi, Niigata Yakka Daigaku Kenkyu Hokoku (in Japanese) 6 (1986) 23–37.
[7] A. Omari, P. Garner, Clin. Evid. 9 (2003) 850–860.
[8] A. Omari, P. Garner, Clin. Evid. 11 (2004) 1047–1057.
[9] C.W. Jefford, Curr. Opin. Investig. Drugs 5 (2004) 866–872.
[10] Y. Tang, Y. Dong, J.L. Vennerstrom, Med. Res. Rev. 24 (2004) 425–428.
[11] K.J. McCullough, M. Nojima, Curr. Org. Chem. 5 (2001) 601–636.
[12] W. Ando (Ed.), Organic Peroxides, J. Wiley & Sons, New York, NY, 1992.
[13] M. Ghezai, W. Ming, S. Bala, H. Daryle, Coord. Chem. Rev. 219–221 (2001) 789–820.
[14] A.N. Collins, G.N. Sheldrake, J. Crosby, Chirality in Industry, J. Wiley & Sons, New York, 1998.
[15] C. Sanfilippo, A. Patti, M. Piattelli, G. Nicolosi, Tetrahedron: Asymmetry 9 (1998) 2809–2817.
[16] C. Sanfilippo, A. Patti, G. Nicolosi, Tetrahedron: Asymmetry 10 (1999) 3273–3276.
[17] V.M. Dembitsky, Tetrahedron 59 (2003) 4701–4720.
[18] M.C.R. Franssen, Catalysis Today 22 (1994) 441–457.
[19] A. Sekera, J. Rahm, Cesk. Farm. 1 (1952) 471–477.
[20] H. Ziemann, Rundschau Deut. Tech. 18 (1938) 9.
[21] Y.R. Naves, Parfums de France 13 (1935) 4–13.
[22] G. Duviau, J. Med. Bord. 130 (1953) 44–51.
[23] A. Wautie, Parasitica, 2 (1946) 44–67.
[24] F. Guimaraes, C. R. Seances Soc. Biol. Fil. 96 (1927) 1251–1253.
[25] E.C. Faust, JAMA, J. Am. Med. Assoc. 108 (1937) 386–392.
[26] D.L.J. Opdyke, Food Cosmet. Toxicol. 14 (Suppl.) (1976) 713–715.
[27] A. Bhargava, S. Shukla, D. Ohri, J. Med. Arom. Plant Sci., 27 (2005) 309–319.
[28] A. Harder, Parasitol. Res., 88 (2002) 477–480.
[29] R.J. Wells, Tetrahedron Lett., 17 (1976) 2637–2638.
[30] A.D. Patil, PCT Int. Appl. US Patent 86–825920 19860204, 1987, 29 pp.
[31] A.D. Patil, A.J. Freyer, M.F. Bean, B.K. Carte, J.W. Westley, R.K. Johnson, Tetrahedron, 52 (1996) 377–394.
[32] B.S. Davidson, J. Org. Chem. 56 (1991) 6722–6724.
[33] P.A. Horton, R.E. Longley, M. Kelly-Borges, O.J. McConnell, L.M. Ballas, J. Nat. Prod. 57 (1994) 1374–1381.
[34] J. Sandler, P.L. Colin, J.N.A. Hooper, D.J. Faulkner, J. Nat. Prod. 65 (2002) 1258–1261.
[35] A. Rudi, R. Afanii, L.G. Gravalos, M. Aknin, E. Gaydou, J. Vacelet, Y. Kashman, J. Nat. Prod. 66 (2003) 682–685.
[36] M. Holzwarth, J.-M. Trendel, P. Albrecht, A. Maier, W. Michaelis, J. Nat. Prod. 68 (2005) 759–761.
[37] Y.C. Shen, C.V. Prakash, Y.H. Kuo, J. Nat. Prod. 64 (2001) 324–327.
[38] M.D. Higgs, D.J. Faulkner, J. Org. Chem. 43 (1978) 3454–3457.
[39] R.E. Longley, O.J. McConnell, E. Essich, D. Harmody, J. Nat. Prod. 56 (1993) 915–920.
[40] T. Higa, S. Sakemi Jr., C. Christophersen, PCT Int. Appl. WO 8704709 A1 19870813 Application WO87-US135 19870121, 1987, 31 pp.
[41] F.S. De Guzman, F.J. Schmitz, J. Nat. Prod. 53 (1990) 926–931.
[42] T. Murayama, Y. Ohizumi, H. Nakamura, T. Sasaki, J. Kobayashi, Experientia 45 (1989) 898–899.
[43] J. Kobayashi, T. Murayama, Y. Oizumi, Jpn. Kokai Tokkyo Koho, Japanese Patent JP 02229185, 1990.
[44] P.H. Dussault, C.T. Eary, K.R. Woller, J. Org. Chem. 64 (1999) 1789–1797.
[45] S. Sakemi, T. Higa, U. Anthoni, C. Christophersen, Tetrahedron 43 (1987) 263–268.
[46] E. Quinoa, E. Kho, L.V. Manes, P. Crews, G.J. Bakus, J. Org. Chem. 51 (1986) 4260–4264.
[47] N. Takada, M. Watanabe, A. Yamada, K. Suenaga, K. Yamada, K. Ueda, D. Uemura, J. Nat. Prod. 64 (2001) 356–359.
[48] D.J. Gochfeld, M.T. Hamann, J. Nat. Prod. 64 (2001) 1477–1479.
[49] A.D. Patil, A.J. Freyer, B. Carte, R.K. Johnson, P. Lahouratate, J. Nat. Prod. 59 (1996) 219–223.
[50] E. Fattorusso, O. Taglialatela-Scafati, M. Di Rosa, A. Ianaro, Tetrahedron 56 (2000) 7959–7967.
[51] J.-F. Hu, H.-F. Gao, M. Kelly, M.T. Hamann, Tetrahedron 57 (2001) 9379–9383.
[52] M. Del Sol Jimenez, S.P. Garzon, A.D. Rodriguez, J. Nat. Prod. 66 (2003) 655–661.
[53] B. Harrison, P. Crews, J. Nat. Prod. 61 (1998) 1033–1037.
[54] C.W. Lim, Y.J. Cha, J.S. Kim, J. Fisheries Sci. Technol. 8 (2005) 6–9.

[55] E. Fattorusso, S. Parapini, C. Campagnuolo, N. Basilico, O. Taglialatela-Scafati, D. Taramelli, J. Antimicrob. Chemother. 50 (2002) 883–888.
[56] F. Berrue, O.P. Thomas, C. Funel-Le Bon, F. Reyes, P. Amade, Tetrahedron 61 (2005) 11843–11849.
[57] D.E. Williams, T.M. Allen, R. Van Soest, H.W. Behrisch, R.J. Andersen, J. Nat. Prod. 64 (2001) 281–285.
[58] Y. Chen, K.B. Killday, P.J. McCarthy, R. Schimoler, K. Chilson, C. Selitrennikoff, S.A. Pomponi, A.E. Wright, J. Nat. Prod. 64 (2001) 262–264.
[59] D.J. Bums, J. Bloomenthal, M. Lee, R.M. Bell, J. Biol. Chem. 265 (1990) 12044–12051.
[60] P. Basta, M.B. Strickland, W. Holmes, C.R. Loomis, L.M. Ballas, D.J. Burns, Biochem. Biophys. Acta 1132 (1992) 154–160.
[61] B.S. Davidson, Tetrahedron Lett. 32 (1991) 7167–7170.
[62] See Ref. [35].
[63] T.L. Perry, A. Dickerson, A.A. Khan, R.K. Kondru, D.N. Beratan, P. Wipf, M. Kelly, M.T. Hamann, Tetrahedron 57 (2001) 1483–1487.
[64] A. Rudi, R. Talpir, Y. Kashman, Y. Benayahu, M. Schleyer, J. Nat. Prod. 56 (1993) 2178–2182.
[65] S.P. Gunasekera, M. Gunasekera, G.P. Gunawardana, P. McCarthy, N. Burres, J. Nat. Prod. 53 (1990) 669–674.
[66] G.R. Pettit, T. Nogawa, J.C. Knight, D.L. Doubek, J.N.A. Hooper, J. Nat. Prod. 67 (2004) 1611–1613.
[67] T. Ichiba, P.J. Scheuer, M. Kelly-Borges, Tetrahedron 51 (1995) 12195–12202.
[68] S.I. Toth, F.J. Schmitz, J. Nat. Prod. 57 (1994) 123–127.
[69] M.T. Davies-Coleman, C.L. Cantrell, K.R. Gustafson, J.A. Beutler, L.K. Pannell, M.R. Boyd, J. Nat. Prod. 63 (2000) 1411–1413.
[70] A. Fontana, M.C. Gonzalez, M. Gavagnin, J. Templado, G. Cimino, Tetrahedron Lett. 41 (2000) 429–432.
[71] R. Duran, E. Zubı'a, M.J. Ortega, S. Naranjo, J. Salva, Tetrahedron 56 (2000) 6031–6037.
[72] R. Duran, E. Zubia, M.J. Ortega, J. Salva, J.L. Fernandez Puentes, D. Garcia Gravalos, L. Naranjo, PCT Int. Appl., Great Britain Patent WO 2001083477 A1 20011108, 2001, 24 pp.
[73] A. Fontana, G. Cimino, M. Gavagnin, M.C. Gonzalez, E. Estornell, J. Med. Chem. 44 (2001) 2362–2365.
[74] R.S. Compagnone, I.C. Pina, H.R. Rangel, F. Dagger, A.I. Suarez, M.V.R. Reddy, D.J. Faulkner, Tetrahedron 54 (1998) 3057–3068.
[75] A. Qureshi, J. Salva, M.K. Harper, D.J. Faulkner, J. Nat. Prod. 61 (1998) 1539–1542.
[76] Y. Chen, P.J. McCarthy, D.K. Harmody, R. Schimoler-O'Rourke, K. Chilson, C. Selitrennikoff, S.A. Pomponi, A.E. Wright, J. Nat. Prod. 65 (2002) 1509–1512.
[77] D.B. Stierle, D.J. Faulkner, J. Org. Chem. 44 (1979) 964–968.
[78] D.W. Phillipson, K.L. Rinehart Jr., J. Am. Chem. Soc. 105 (1983) 7735–7736.
[79] K. Hirakura, M. Morita, K. Nakajima, Y. Ikeya, H. Mitsuhashi, Phytochemistry 31 (1992) 899–903.
[80] K. Hirakura, H. Mihashi, T. Fujihashi, T. Okuma, Jpn. Kokai Tokkyo Koho, Japanese Patent JP 04352742 A2 19921207 Heisei, 1992, 10 pp.
[81] T. Yosief, A. Rudi, Y. Wolde-ab, Y. Kashman, J. Nat. Prod. 61 (1998) 491–493.
[82] A. Fontana, G. D'Ippoloto, L. D'Souza, E. Mollo, P.S. Parameswaram, G. Cimino, J. Nat. Prod. 64 (2001) 131–133.
[83] P. Dussault, C. Xu, J.M. Raible, Abstracts of Papers, 229th ACS National Meeting, San Diego, CA, United States, March, 2005, pp. 13–17.
[84] D.T. Youssef, J. Nat. Prod. 67 (2004) 112–114.
[85] Y. Guo, M. Gavagnin, E. Mollo, E. Trivellone, G. Cimino, N.A. Hamdy, I. Fakhr, M. Pansini, Nat. Prod. Lett. 9 (1996) 105–112.
[86] J. Tanaka, T. Higa, K. Suwanborirux, U. Kokpol, G. Bernardinelli, C.W. Jefford, J. Org. Chem. 58 (1993) 2999–3002.
[87] J.-I. Tanaka, T. Higa, K. Suwanoborirux, C.W. Jefford, G. Bernardinelli, D.G. Gravalos, Eur. Pat. Appl. European Patent EP 610076 A1 19940810, 1994, 8 pp.
[88] R.J. Capon, S.J. Rochfort, S.P.B. Ovenden, R.P. Metzger, J. Nat. Prod. 61 (1998) 525–528.
[89] P. Phuwapraisirisan, S. Matsunaga, N. Fusetani, N. Chaitanawisuti, S. Kritsanapuntu, P. Menasveta, J. Nat. Prod. 66 (2003) 289–291.
[90] P. Phuwapraisirisan, S. Matsunaga, N. Fusetani, Org. Lett. 26 (2005) 2233–2236.
[91] D.T. Youssef, W.Y. Yoshida, M. Kelly, P.J. Scheuer, J. Nat. Prod. 64 (2001) 1332–1335.
[92] S. Sokoloff, S. Halevy, V. Usieli, A. Colorni, S. Sarel, Experientia 38 (1982) 337–338.
[93] S. Sperry, F.A. Valeriote, T.H. Corbett, P. Crews, J. Nat. Prod. 61 (1998) 241–247.
[94] M. Albericci, J.C. Braekman, D. Daloze, B. Tursch, Tetrahedron 38 (1982) 1881–1890.
[95] M. Albericci, M. Collart-Lempereur, J.C. Braekman, D. Daloze, B. Tursch, J.P. Declercq, G. Germain, M. Van Meerssche, Tetrahedron Lett. 29 (1979) 2687–2690.
[96] P.W. Pare, J. Zajicek, V.L. Ferracini, I.S. Melo, Biochem. Syst. Ecol. 21 (1993) 649–653.
[97] Y. Pollack, R. Segal, J. Golenser, Parasitol. Res. 76 (1990) 570–572.
[98] F. Kiuchi, Y. Itano, N. Uchiyama, G. Honda, A. Tsubouchi, J. Nakajima-Shimada, T. Aoki, J. Nat. Prod. 65 (2002) 509–512.
[99] L. Monzote, A.M. Montalvo, S. Almanonni, R. Scull, M. Miranda, J. Abreu, Chemotherapy 52 (2006) 130–136.
[100] T. Efferth, A. Olbrich, A. Sauerbrey, D.D. Ross, E. Gebhart, M. Neugebauer, Anticancer Res. 22C (2002) 4221–4224.
[101] V.M. Dembitsky, S. Abu-Lafi, L.O. Hanus, Biores. Technol. 98 (2007) in press.
[102] B.D. Smith, Proc. Natl. Acad. Sci. USA. 103 (2006) 12223–12228.
[103] A.H. Beckett, M. Donbrow, G.O. Jolliffe, J. Pharm. Pharmacol. 7 (1955) 55–65.
[104] A.H. Beckett, G.O. Jolliffe, J. Pharm. Pharmacol. 7 (1955) 606–607.
[105] M.M. Kliks, Soc. Sci. Med. 21 (1985) 879–886.
[106] D. MacDonald, K. VanCrey, P. Harrison, P.K. Rangachari, J. Rosenfeld, C. Warren, G. Sorger, J. Ethnopharmacol. 92 (2004) 215–221.
[107] E. Okuyama, K. Umeyama, Y. Saito, M. Yamazaki, M. Satake, Chem. Pharm. Bull. 41 (1993) 1309–1311.
[108] B.L. Van Duuren, L. Orris, N. Nelson, J. Natl. Cancer Inst. 35 (1965) 707–717.
[109] J.-F. Cavalli, F. Tomi, A.-F. Bernardini, J. Casanova, Phytochem. Anal. 15 (2004) 275–279.
[110] J.U. Chowdhury, M. Yusuf, J. Begum, A.J. Taniya, M.E. Hossain, M.A. Hossain, Indian Perfumer. 49 (2005) 61–65.
[111] H. Paget, J. Chem. Soc. (1938) 829–833.
[112] L. Carballeira, R.A. Mosquera, M.A. Rios, J. Comput. Chem. 10 (1989) 911–920.
[113] S. Rebello, S.F.G. da Costa, J.T. Rico, C.R. Seances Soc. Biol. Fil. 98 (1928) 995–997.
[114] R.X. Tan, W.F. Zheng, H.Q. Tang, Planta Med. 64 (1998) 295–302.
[115] H.-N. Koo, S.-H. Hong, H.-J. Jeong, E.-H. Lee, N.-G. Kim, S.-D. Choi, K.-W. Ra, K.-S. Kim, B.-K. Kang, J.-J. Kim, J.G. Oh, H.-M. Kim, Immunopharm. Immunotoxicol. 24 (2002) 441–453.
[116] T.E. Lee, S.W. Park, T.J. Min, J. Biochem. Mol. Biol. 32 (1999) 585–593.
[117] Y. Li, M. Li, L. Wang, Z. Jiang, W. Li, H. Li, Sichuan Da Xue Xue Bao Yi Xue Ban 35 (2004) 337–339.
[118] H.C. Kwon, S.U. Choi, K.R. Lee, Archiv. Pharmacal Res. 23 (2000) 151–154.
[119] G. Ruecker, D. Manns, S. Wilbert, Phytochemistry 31 (1992) 340–342.
[120] Y. Li, Y.L. Wu, Med. Trop. (Mars) 58 (Suppl. 3) (1998) 9–12.
[121] N.P. Singh, C. Lai Henry, Anticancer Res. 24 (2004) 2277–2780.
[122] H.J. Woerdenbag, T.A. Moskal, N. Pras, T.M. Malingre, F.S. El-Feraly, H.H. Kampinga, A.W. Konings, J. Nat. Prod. 56 (1993) 849–856.
[123] S. Balachandran, R.A. Vishwakarma, S.P. Popli, Indian J. Pharm. Sci. 49 (1987) 152–154.
[124] H.-S. Kim, Y. Nagai, K. Ono, K. Begum, Y. Wataya, Y. Hamada, K. Tsuchiya, A. Masuyama, M. Nojima, K.J. McCullough, J. Med. Chem. 44 (2001) 2357–2361.
[125] A.G. Gonzalez, J.D. Martin, C. Perez, J. Rovirosa, B. Tagle, J. Clardy, Chem. Lett. 10 (1984) 1649–1652.

[126] L.-K. Sy, G.D. Brown, Tetrahedron 54 (1998) 4345–4356.
[127] A.C. Beekman, P.K. Wierenga, H.J. Woerdenbag, W. Van Uden, N. Pras, A.W. Konings, F.S. El-Feraly, A.M. Galal, H.V. Wikstrom, Planta Med. 64 (1998) 615–619.
[128] G. Stojanovic, N. Radulovic, T. Hashimoto, R. Palic, J. Ethnopharmacol. 101 (2005) 185–190.
[129] S. Barel, R. Segal, J. Yashphe, J. Ethnopharmacol. 33 (1991) 187–191.
[130] N. Bezic, M. Skocibusic, V. Dunkic, A. Radonic, Phytother. Res. 17 (2003) 1037–1040.
[131] M. Unlu, D. Daferera, E. Donmez, M. Polissiou, B. Tepe, A. Sokmen, J. Ethnopharmacol. 83 (2002) 117–121.
[132] T. Kundakovic, T. Stanojkovic, Z. Juranic, N. Kovacevic, Pharmazie 60 (2005) 319–320.
[133] E. Tsankova, U. Jacobsson Kempe, T. Norin, I. Ognyanov, Phytochemistry 20 (1981) 1436–1438.
[134] S. Trifunovic, V. Vajs, Z. Juranic, Z. Zizak, V. Tesevic, S. Macura, S. Milosavljevic, Phytochemistry 67 (2006) 887–893.
[135] F. Bohlmann, K.H. Knoll, Phytochemistry 18 (1979) 995–1001.
[136] C.-Y. Duh, M.-C. Chia, S.-K. Wang, H.-J. Chen, A.A.H. El-Gamal, C.F. Dai, J. Nat. Prod. 64 (2001) 1028–1031.
[137] K.L. Erickson, J.A. Beutler, G.N. Gray, J.H. Cardellina II, M.R. Boyd, J. Nat. Prod. 58 (1995) 1848–1860.
[138] N.-H. Nam, H.-M. Kim, K.-H. Bae, B.-Z. Ahn, Phytotherapy Res. 17 (2003) 107–111.
[139] T.C. Le, Q.H. Nguyen, M.P. Le, V.H. Le, T.T. Le, D.M. Pham, T.D. Nguyen, T.L.H. Nguyen, V.B. Nguyen, Tap Chi Duoc Hoc (Vietnam) 2–3 (1987) 12–17.
[140] X.-H. Xu, C.-H. Kong, C.-J. Lin, X. Wang, Y.-D. Zhu, H.-S. Yang, Chin. J. Chem. 21 (2003) 1506–1509.
[141] S.S. Handa, A.D. Kinghorn, G.A. Cordell, N.R. Farnsworth, J. Nat. Prod. 46 (1983) 123–126.
[142] M. Alam, G.E. Martin, A.S. Zektzer, A.J. Weinheimer, R. Sanduja, M.A. Ghuman, J. Nat. Prod. 56 (1993) 774–779.
[143] J.E. Araya, I. Neira, S. da Silva, R.A. Mortara, P. Manque, E. Cordero, H. Sagua, A. Loyola, J. Borquez, G. Morales, J. Gonzalez, Memorias do Instituto Oswaldo Cruz (Sao Paulo, Brazil) 98 (2003) 413–418.
[144] R.W. Doskotch, F.S. El-Feraly, E.H. Fairchild, C.T. Huang, J. Chem. Soc. Chem. Commun. (1976) 402.
[145] G.C. Zheng, A. Ichikawa, M.O. Ishitsuka, T. Kusumi, H. Yamamoto, H. Kakisawa, J. Org. Chem. 55 (1990) 3677–3679.
[146] K.E. Schulte, G. Glauch, G. Rucker, Nardosinon, ein neuer inhaltsstoff von nardostachys chinensis batalin, Tetrahedron Lett. 35 (1965) 3083–3084.
[147] G. Rucker, K.H. Kahrs, H.W. Hembeck, Arch. Pharm. (Weinheim) 308 (1975) 858–862.
[148] H. Itokawa, K. Masuyama, H. Morita, K. Takeya, Chem. Pharm. Bull. (Tokyo) 41 (1993) 1183–1184.
[149] Y. Zhang, Y. Lu, L. Zhang, Q.T. Zheng, L.Z. Xu, S.L. Yang, J. Nat. Prod. 68 (2005) 1131–1133.
[150] G. Ruecker, L. Shide, A. Olbrich, Arch. Pharm. (Weinheim) 323 (1990) 171–175.
[151] P. Li, K. Matsunaga, K. Yamamoto, R. Yoshikawa, K. Kawashima, Y. Ohizumi, Neurosci. Lett. 273 (1999) 53–56.
[152] P. Li, K. Matsunaga, T. Yamakuni, Y. Ohizumi, Brain Res. Dev. Brain Res. 145 (2003) 177–183.
[153] Y. Yamada, Yakugaku Zasshi 122 (2002) 727–743.
[154] A.F. Ahmed, Y.-H. Kuo, C.-F. Dai, J.-H. Sheu, J. Nat. Prod. 68 (2005) 1208–1212.
[155] Y. Hashidoko, S. Tahara, J. Mizutani, Phytochemistry 31 (1992) 779–782.
[156] X. Liang, in: H.M. Chang (Ed.), Advances in Chinese Medical Materials Research, International Symposium, World Sci., Singapore, Singapore, 1985, pp. 427–432.
[157] R.P. Adams, Junipers of the World: The Genus Juniperus, Trafford Publ., Vancouver, B.C., Canada, 2004, p. 282.
[158] A. Angioni, A. Barra, M.T. Russo, V. Coroneo, S. Dessi, P. Cabras, J. Agric. Food Chem. 51 (2003) 3073–3078.
[159] A.F. Barrero, J.F. Quilez del Moral, M.M. Herrador, M. Akssira, A. Bennamara, S. Akkad, M. Aitigri, Phytochemistry 65 (2004) 2507–2515.
[160] E.M.K. Wijeratne, M.X. Liu, N.B. Kantipudi, C.B. Brochini, A.A.L. Gunatilaka, L.M. Canfield, Bioorg. Med. Chem. 14 (2006) 7875–7879.
[161] H. Takayama, S. Yamada, K. Nakayama, T. Suda, Vitamin D3 derivatives, European Patent EP 81793 A1 19830622, Application EP 82–111302 19821207, 1983, 11 pp.
[162] H. Takayama, Jpn. Kokai Tokkyo Koho, Japanese Patent JP 58032823 A2 19830225 Showa, 1983, 4 pp.
[163] J.H. Sheu, G.H. Wang, P.J. Sung, Y.H. Chiu, C.Y. Duh, Planta Med. 63 (1997) 571–572.
[164] J.M. Lo, W.L. Wang, Y.M. Chiang, C.M. Chen, J. Chin. Chem. Soc. 48 (2001) 821–826.
[165] M. Guyot, E. Morel, C. Belaud, J. Chem. Res. Synop. 8 (1983) 188–189.
[166] J.H. Sheu, C.-C. Liaw, C.-Y. Duh, J. Nat. Prod. 58 (1995) 1521–1526.
[167] D. Lee, M. Cuendet, F. Axelrod, P.I. Cahvez, H.H.S. Fong, J.M. Pezzuto, A.D. Kinghorn, Tetrahedron 57 (2001) 7107–7112.
[168] J.-H. Sheu, S.-Y. Huang, C.-Y. Duh, Cytotoxic oxygenated desmosterols of the red alga *Galaxaura marginata*, J. Nat. Prod. 59 (1996) 23–26.
[169] J.-H. Sheu, S.-Y. Huang, G.-H. Wang, C.-Y. Duh, J. Nat. Prod. 60 (1997) 900–903.
[170] J.H. Sheu, G.H. Wang, P.J. Sung, C.-Y. Duh, J. Nat. Prod., 62 (1999) 224–247.
[171] B.S. Jung, M.K. Shin, Hyang-Yak (Saeng-Yak) Dae-Sa-Jeon, Young Lim Publications, Seoul, 1990, pp. 380–381.
[172] J.G. Park, J.W. Hyun, K.H. Lim, J.E. Shin, Y.J. Won, Y.D. Yi, K.H. Shin, I.M. Chang, Korean J. Pharmacog. 24 (1993) 223–230.
[173] D.D. Kitts, K.-T. Lim, J. Toxicol. Environ. Health 64A (2001) 357–371.
[174] I.S. Lee, S.R. Oh, K.S. Ahn, H.K. Lee, Chem. Pharm. Bull. 49 (2001) 1024–1026.
[175] G. Ruecker, D. Manns, E.P. Schenkel, R. Hartmann, B.M. Heinzmann, Arch. Pharm. (Weinheim) 336 (2003) 205–207.
[176] Y. Takaishi, M. Uda, T. Ohashi, K. Nakano, K. Murakami, T. Tommiatsu, Phytochemistry 30 (1991) 4117–4120.
[177] S. Matsueda, M. Shimoyama, T. Imaizumi, Y. Tsushima, Yakugaku Zasshi 102 (1982) 347–351.
[178] K. Kahlos, L. Kangas, R. Hiltunen, Planta Med. 55 (1989) 389–390.
[179] Y.C. Kuo, S.C. Wenig, C.J. Chou, T.T. Chang, W.J. Tsai, Br. J. Pharmacol. 140 (2003) 895–906.
[180] K.S. Im, K.I. Nam, C.J. Sim, J.H. Jung, Saengyak Hakhoechi 31 (2000) 401–406.
[181] K.S. Nam, Y.S. Jo, Y.H. Kim, J.W. Hyun, H.W. Kim, Life Sci. 69 (2001) 229–237.
[182] S.H. Lee, S.H. Shim, J.S. Kim, S.S. Kang, Arch. Pharm. Res. 29 (2006) 479–483.
[183] F.A. Macias, N. Chinchilla, R.M. Varela, J.M. Molinillo, Steroids 71 (2006) 603–608.
[184] T. Takei, M. Yoshida, M. Ohnishi-Kameyama, M. Kobori, Biosci. Biotechnol. Biochem. 69 (2005) 212–215.
[185] J.W. Bok, L. Lermer, J. Chilton, H.G. Klingeman, G.H.N. Towers, Phytochemistry 51 (1999) 891–898.
[186] M. Iwashima, I. Terada, K. Iguchi, T. Yamori, Chem. Pharm. Bull. 50 (2002) 1286–1289.
[187] J.-H. Sheu, K.-C. Chang, C.-Y. Duh, J. Nat. Prod. 63 (2000) 149–151.
[188] C. Gonzalez, A. Pareja, M.E. Marquez, D.M. Marquez, A. Martinez, E. Higuita, Acta Farmaceutica Bonaerense (Colombia) 24 (2005) 75–79.
[189] A. Gauvin, J. Smadja, M. Aknin, R. Faure, E.-M. Gaydou, Canadian J. Chem. 78 (2000) 986–992.
[190] M. Kobori, M. Yoshida, M. Ohnishi-Kameyama, T. Takei, H. Shinmoto, Biol. Pharm. Bull. 29 (2006) 755–759.
[191] C.N. Lin, W.P. Tome, S.J. Won, J. Nat. Prod. 54 (1991) 998–1002.
[192] S. Hamzah, A. Yusof, Br. J. Sports Med. 37 (2003) 464–470.
[193] H. Itokawa, E. Kishi, H. Morita, K. Takeya, Y. Iitaka, Chem. Lett. (1991) 2221–2222.

[194] H. Morita, E. Kishi, K. Takeya, H. Itokawa, Y. Iitaka, Phytochemistry 34 (1993) 765–771.
[195] G. Ruecker, D. Manns, R. Hartmann, U. Bonsels, Arch. Pharm. (Weinheim) 328 (1995) 725–730.
[196] S.S. Chatterjee, M. Noldner, E. Koch, C. Erdelmeier, Pharmacopsychiatry 31 (1998) 7–15.
[197] E. Koch, S.S. Chatterjee, Pharmacopsychiatry 34 (Suppl. 1) (2001) S70–S73.
[198] Y.C. Shen, C.V.S. Prakash, J.-H. Guh, Tetrahedron Lett. 45 (2004) 2463–2465.
[199] M. Patel, A.C. Horan, V.P. Gullo, D. Loebenberg, J.A. Marquez, G.H. Miller, J.A. Waitz, J. Antibiot. 37 (1984) 413–415.
[200] J.J.K. Wright, Y. Merill, M.S. Puar, A.T. McPhail, J. Chem. Soc. Chem. Commun. 7 (1984) 473–474.
[201] S. Takahashi, T. Nakano, T. Koiwa, T. Noshita, S. Funayama, H. Koshino, A. Nakagawa, J. Antibiot. (Tokyo) 53 (2000) 163–170.
[202] T. Nakano, T. Koiwa, S. Takahashi, A. Nakagawa, J. Antibiot. (Tokyo) 53 (2000) 12–18.
[203] W.S. Chen, Y.T. Chen, X.Y. Wan, E. Friedrichs, H. Puff, E. Breitmaier, Liebigs Ann. Chem. 10 (1981) 1880–1885.
[204] D. Zhenjun, J.W. Lown, Photochem. Photobiol. 52 (1990) 609–616.
[205] L.Z. Fang, C. Qing, H.J. Shao, Y.D. Yang, Z.J. Dong, F. Wang, W. Zhao, W.Q. Yang, J.K. Liu, J. Antibiot. (Tokyo) 59 (2006) 351–354.
[206] Y. Shen, X. Rong, Z. Gao, Zhongguo 27 (2002) 674–676.
[207] T. Kishi, S. Tahara, N. Taniguchi, M. Tsuda, C. Tanaka, S. Takahashi, Planta Med. 57 (1991) 376–379.
[208] Z. Diwu, Photochem. Photobiol. 61 (1995) 529–539.
[209] Z. Diwu, C. Zhang, J.W. Lown, Anticancer Drug 8 (1993) 129–143.
[210] J.B. Hudson, J. Zhou, J. Chen, L. Harris, L. Yip, G.H.N. Towers, Photochem. Photobiol. 60 (1994) 253–255.
[211] Z. Diwu, J.W. Lown, J. Photochem. Photobiol. 18B (1993) 145–154.
[212] A. Jegorov, V. Matha, P. Sedmera, V. Havlicek, J. Stuchlik, P. Seidel, P. Simek, Phytochemistry 38 (1995) 403–407.
[213] V. Matha, A. Jegorov, J. Weiser, J.S. Pillai, Cytobios 69 (1992) 163–170.
[214] P. Sedmera, V. Havlicek, A. Jegorov, A.L. Segre, Tetrahedron Lett. 36 (1995) 6953–6956.
[215] M. Uramoto, M. Tanabe, K. Hirotsu, J. Clardy, Heterocycles 17 (1982) 349–354.
[216] M. Yamazaki, Seikagaku 61 (1989) 207–210.
[217] N. Eickman, J. Clardy, R.J. Cole, J.W. Kirksey, Tetrahedron Lett. 12 (1975) 1051–1054.
[218] J. Bunger, G. Westphal, A. Monnich, B. Hinnendahl, E. Hallier, M. Muller, Toxicology 202 (2004) 199–211.
[219] W. Chavasiri, Tetrahedron Lett. 44 (2003) 6759–6761.
[220] L. Nassi, B. Epe, D. Schiffmann, W. Adam, A. Beinhauer, A. Griesbeck, Carcinogenesis 8 (1987) 947–953.
[221] E. Emmanouilidou, P.C. Ioannou, T.K. Christopoulos, K. Polizois, Anal. Biochem. 313 (2003) 97–105.
[222] M. Sugii, M. Ohkita, M. Taniguchi, K. Baba, Y. Kawai, C. Tahara, M. Takaoka, Y. Matsumura, Biol. Pharm. Bull. 28 (2005) 607–610.
[223] T. Fujita, S. Sakuma, T. Sumiya, H. Nishida, Y. Fujimoto, K. Baba, M. Kozawa, Res. Commun. Chem. Pathol. Pharmacol. 77 (1992) 227–240.
[224] M. Matsuura, Y. Kimura, K. Nakata, K. Baba, H. Okuda, Planta Med. 67 (2001) 230–235.
[225] K. Baba, M. Taniguchi, K. Nakata, Foods and Food Ingredients (Japan) 178 (1998) 52–60.
[226] T. Enoki, E. Kobayashi, K. Ogawa, Y. Kudo, M. Tanabe, K. Sugiyama, H. Ohnogi, H. Sagawa, I. Kato, PCT Int. Appl. Japanese Patent WO 2004096198 A1 20041111, 2004, 289 pp.
[227] M. Kimura, H. Iwagaki, M. Tsunenaga, and S. Inoue, PCT Int. Appl. Japanese Patent JP 6017 20050330, 2005, 40 pp.
[228] K. Hirasawa, Mi. Murakami, Y. Sato, Jpn. Kokai Tokkyo Koho, Japanese Patent JP 63014788 A2 19880121 Showa, 1988, 8 pp.
[229] K. Hirasawa, M. Murakami, Y. Sato, Jpn. Kokai Tokkyo Koho, Japanese Patent JP 63014720 A2 19880121 Showa, 1988, 3 pp.
[230] G. Spiteller, Free Radic. Biol. Med. 41 (2006) 362–387.
[231] M. Maccarrone, G. Melino, A. Finazzi-Agro, Cell Death Differ. 8 (2001) 776–784.
[232] P.A. Sandstrom, D. Pardi, P.W. Tebbey, R.W. Dudek, D.M. Terrian, T.M. Folks, T.M. Buttke, FEBS Lett. 365 (1995) 66–70.
[233] M.-J. Hébert, T. Takano, H. Holthöfer, H.R. Brady, J. Immunol. 157 (1996) 3105–3115.
[234] H. Aoshima, T. Satoh, N. Sakai, M. Yamada, Y. Enokido, T. Ikeuchi, H. Hatanaka, Biochim. Biophys. Acta 1345 (1997) 35–42.
[235] S.D. Liberles, S.L. Schreiber, Chem. Biol. 7 (2000) 365–372.
[236] S. Miyamoto, C. Dupas, K. Murota, J. Terao, Lipids 38 (2003) 641–649.
[237] A. Guidarelli, O. Cantoni, Biochem. J. 366 (2002) 307–314.
[238] G. Spiteller, Ann. N. Y. Acad. Sci. 959 (2002) 30–44.
[239] D. Lapenna, G. Ciofani, Cristiana Bruno, Sante D. Pierdomenico, F. Cuccurullo, Br. J. Pharm. 133 (2001) 739–745.
[240] R. Castoria, C. De Luca, A.A. Fabbri, S. Passi, C. Fanelli, J. Toxicol. Toxin Rev. 8 (1989) 349–360.
[241] P. Spiteller, W. Kern, J. Reiner, G. Spiteller, Biochim. Biophys. Acta 1531 (2001) 188–208.
[242] H. Yin, N.A. Porter, Antioxid. Redox Signal. 7 (2005) 170–184.
[243] R. Medina-Navarro, G. Duran-Reyes, J.J. Hicks, Endocr. Res. 25 (1999) 263–280.
[244] J.F. Griveau, E. Dumont, P. Renard, J.P. Callegari, D. Le Lannou, J. Reprod. Fertil. 103 (1995) 17–26.
[245] W. Torinuku, T. Miura, Singlet oxygen and ultraweak chemiluminescence in rat skin, Tohoku J. Exp. Med. 135 (1981) 387–393.
[246] L. Cavallini, A. Bindoli, F. Macri, Chem. Biol. Interact. 28 (1979) 139–146.
[247] G.C. Gisler, J. Diaz, N. Duran, Physiol. Chem. Phys. 14 (1982) 335–342.
[248] P. Di Mascio, L.H. Catalani, E.J.H. Bechara, Free Radic. Biol. Med. 12 (1992) 471–478.
[249] A. Hautfenne, M. Loncin, C. Paquot, Oleagineux (Paris) 21 (1966) 303–308.
[250] W. Adam, S. Andler, D. Ballmaier, S. Emmert, B. Epe, G. Grimm, K. Mielke, M. Moller, T.M. Runger, C.R. Saha-Moller, A. Schonberger, H. Stopper, Recent Results Cancer Res. 143 (1997) 21–34.