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THE HETEROCYCLIC NATURAL PRODUCTS OF GORGONIAN CORALS OF GENUS *BRIAREUM* EXCLUSIVE OF BRIARANE-TYPE DITERPENOIDS

Ping-Jyun Sung* and Ming-Chyuan Chen

National Museum of Marine Biology and Aquarium, 2 Houwan Road, Checheng, Pingtung 944, Taiwan R.O.C. E-mail: pjsung@nmmba.gov.tw

Abstract – An overview on the heterocyclic natural products of gorgonian octocorals of genus *Briareum* is presented. Based on previous studies, most of the heterocyclic natural products from *Briareum* are diterpenoid metabolites, including eunicellin, asbestinane, cembrane, and briarane-type derivatives. The structures, names, biological activities, and references of these natural products exclusive of briaranes are described in this review.

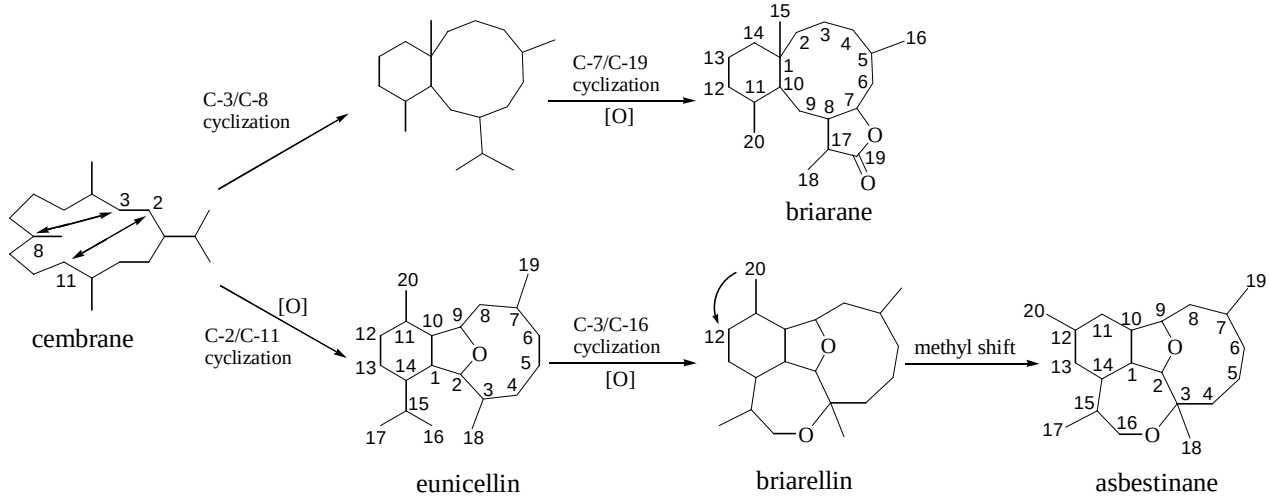
1. INTRODUCTION

Gorgonian corals of genus *Briareum* (phylum Cnidaria, class Anthozoa, subclass Octocorallia, order Gorgonacea, suborder Scleraxonia, family Briareidae), which can be found on Caribbean waters and West Pacific Ocean, have been investigated extensively for many years and yielded a number of novel secondary metabolites. Studies on the chemical constituents of gorgonian corals of genus *Briareum* were began in the early 1960's with the discovery of crystalline terpenoid lactones and the occurrence of a nitrogen-containing metabolite, taurobetaine, in the gorgonian coral *Briareum asbestinum*, collected at Bahamian waters.[1–3]

Two West Indian gorgonians, *B. asbestinum* and *B. polyanthes*, and two Indo-Pacific gorgonians, *B. excavatum* and *B. stechei*, along with several gorgonians identified as *Briareum* spp., have been studied for their chemical constituents. In addition, *Briareum* was considered to be synonymous with the Indo-Pacific gorgonian corals of genus *Solenopodium*.[4] Based on previous studies, gorgonians of genus *Briareum* are particularly rich in diterpenoids such as eunicellin, asbestinane, and briarane-type compounds, and some of these metabolites possess potential biological activities. Due to the briarane diterpenoids have been reviewed previously,[5] therefore, this review was consisted of the heterocyclic natural products isolated from gorgonians of genus *Briareum* exclusive of briarane-type metabolites.

2. *Briareum asbestinum*

The gorgonian *B. asbestinum* is only found in Caribbean waters, and this species is commonly distributed from Southern Florida through most of the West Indies. *B. asbestinum* played an important role in the studies of gorgonian corals, the first briarellin-type eunicellin (briarellin A),[6] asbestinane (asbestinin-1),[7] and briarane (briarein A)[8] metabolites all were isolated from this organism. Moreover, the phylogenetic relationships studies indicated that the taxonomic position of *B. asbestinum* is out of ordinary in subclass Octocorallia.[9,10] These results seem to explain the diversity of chemical constituents in *B. asbestinum*.

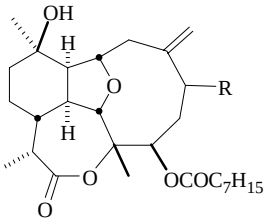
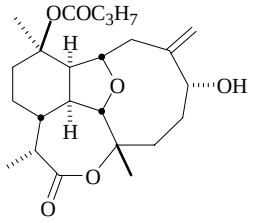
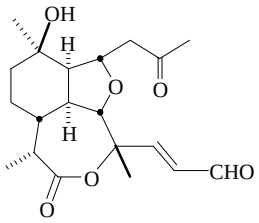
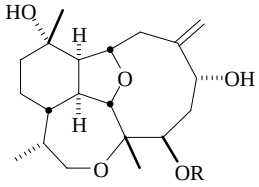
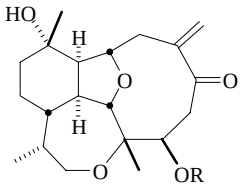
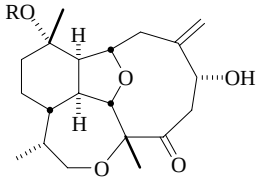
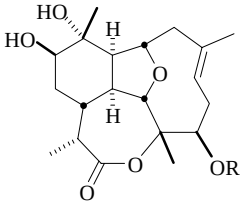
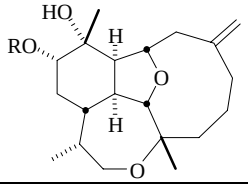


Scheme 1. The proposed biosynthetic pathway among the cembrane, eunicellin, briarellin, asbestinane, and briarane carbon skeletons. The number system shown in briarane, eunicellin, and asbestinane are those presently in use.

2.1. Eunicellin-Type Metabolites

Eunicellin-type metabolites are produced by the C-2/C-11 cyclization of cembrane derivatives (Scheme 1), and all the metabolites of this type possessed a bicyclo[8.4.0] system in structures. Five new metabolites, briarellins A–D (**1**–**4**) and *seco*-briarellin (**5**), featuring the eunicellin carbon skeleton, were isolated from the organic extracts of *B. asbestinum*, collected at Mona Island, Puerto Rico (Table 1).[6] In a later study, the other five new compounds, briarellins E–I (**6**–**10**) were also obtained from this organism (Table 1).[11] The structures, including the relative stereochemistry of metabolites (**1**–**10**) were elucidated by the interpretations of extensive spectral analysis (IR, MS, 1D, and 2D NMR),[6,11,12] and the structure of briarellin C (**3**) was further confirmed by the butyrylation of briarellin B (**2**).[6] It is noted that the oxygen-bearing functional groups link across the C-3/C-16 in briarellins (**1**–**10**) are rarely found in other eunicellin-type metabolites.[13] The *seco*-briarellin (**5**), which contained only three rings and thus comprised a novel type of briarellin diterpenoid. Except for *seco*-briarellin (**5**), the compound of this type is only found in an Indo-Pacific soft coral, *Pachyclavularia violacea*.[14] Briarellins A (**1**) and E (**6**) exhibited moderate cytotoxicity toward HeLa tumor cells.[6,11]

Table 1. The Briarellin-Type Metabolites from *B. asbestinum*

Structure	No.	Name	Biological activity	Ref.
	**1**	briarellin A (R = β-OH)	IC_{50} (HeLa) = 20.0 μg/mL	6,12
	2	briarellin B (R = α-OH)		6,12
	3	briarellin C (R = α-$OCOC_3H_7$)		6,12
	**4**	briarellin D		6,12
	**5**	*seco*-briarellin		6,12
	**6**	briarellin E (R = COC_7H_{15})	IC_{50} (HeLa) = 20.0 μg/mL	11,12
	**7**	briarellin F (R = COC_7H_{15})		11,12
	**8**	briarellin G (R = COC_7H_{15})		11,12
	**9**	briarellin H (R = COC_7H_{15})		11,12
	**10**	briarellin I (R = COC_7H_{15})		11,12

2.2. Asbestinane-Type Metabolites

Asbestinane-type metabolites are a group of irregular diterpenoids that are related to the briarellin-based compounds by migration of the methyl group (CH_3-20) attaching from C-11 to C-12 in the six-membered

ring (Scheme 1). The first asbestinane-type compounds, asbestinin-1 to -5 (**11**–**15**) (Table 2), were isolated from the hexane extract of *B. asbestinum* collected at Lighthouse Reef and at Carrie Bow Cay, Belize.[7] The structure and relative stereochemistry of asbestinin-1 (**11**) were established by a single-crystal X-Ray diffraction analysis on its corresponding diol derivative, and the structures of asbestinin-2 to -5 (**12**–**15**) were determined by the combination of spectral analysis (IR, MS, ^{1}H, and ^{13}C NMR). Furthermore, the relationships between these five metabolites (asbestinin-1 to -5) were established by the preparation of chemical derivatives of these metabolites.[7] Two new asbestinanes, asbestinin epoxide (**16**) and asbestinin-5 acetate (**17**), were obtained from the toxic extracts of *B. asbestinum*, collected at the Bay Islands of Honduras (Table 2). The structures of metabolites (**16** and **17**) were established by spectral data analysis including detailed NMR chemical shift calculations. Moreover, the structures of asbestinins (**16** and **17**) were further confirmed by the epoxidation of asbestinin-2 (**12**) and by the acetylation of asbestinin-5 (**15**), respectively. The NMR spectral data for asbestinin-1 (**11**), -2 (**12**), -4 (**14**), and -5 (**15**) were reassigned in this study.[15]

Four new 4-deoxyasbestinanes, 4-deoxyasbestinin A (**18**), 11-acetoxy-4-deoxyasbestinin B (**19**), 4-deoxy-asbestinin C (**20**), and 11-acetoxy-4-deoxyasbestinin D (**21**) were isolated from the hexane extract of *B. asbestinum*, collected from Palomino Key, Puerto Rico (Table 2). The gross structures of asbestinins (**18**–**21**) were established by extensive 2D NMR studies, including INADEQUATE experiments, and the relative stereochemistry of these metabolites was determined by NOESY spectrum. Based on the combination of detailed NMR chemical shifts analysis and coupling constant calculations, the C-6/C-7 double bonds in metabolites (**18** and **19**) were proved to exist in *E* configuration, and the geometry of C-6/C-7 double bonds in metabolites (**20** and **21**) were assigned as *Z* configuration.[16,17] In the biological activity testing, the 4-deoxyasbestinins (**18**–**21**) exhibited cytotoxicity toward CHO-K1 tumor cells and showed antimicrobial activity.[16]

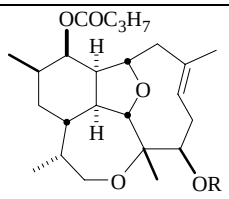
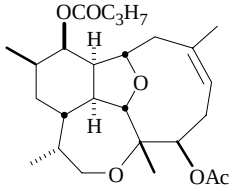
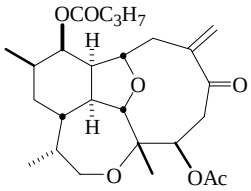
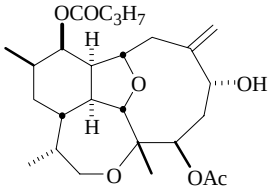
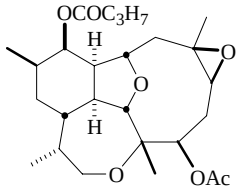
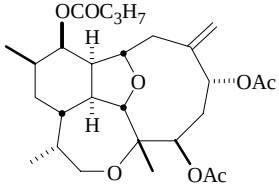
The specimen of *B. asbestinum*, collected off the Mona Island, Puerto Rico, yielded five new asbestinane-type metabolites, asbestinin-6 to -10 (**22**–**26**). The structures, including the relative configuration of asbestinins (**22**–**26**) were established by spectroscopic methods (IR, MS, 1D, and 2D NMR), and the structures of metabolites (**22** and **24**) were further established by the reduction of these two metabolites converting into the known semi-synthetic product, the diol derivative of asbestinin-1.[7,18] In the cytotoxicity testing, some of these metabolites were found to exhibit cytotoxicity toward a limited panel of human tumor cell lines (Table 2).[18]

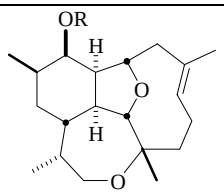
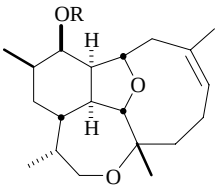
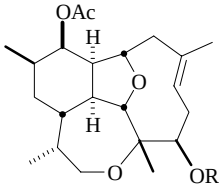
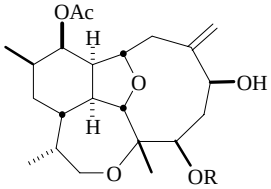
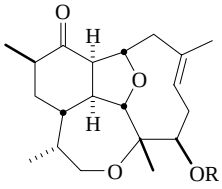
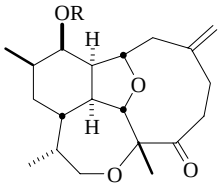
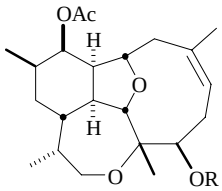
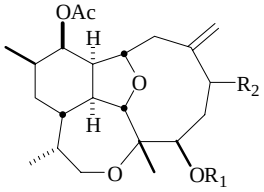
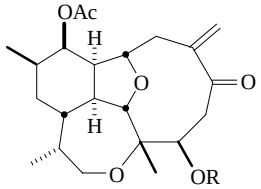
Seventeen new asbestinanes, including asbestinin-11 to -23 (**27**–**39**), 11-acetoxy-4-deoxyasbestinin E (**40**), 11-acetoxy-4-deoxyasbestinin F (**41**), 4-deoxyasbestinin G (**42**), and *seco*-asbestinin (**43**) were isolated from the hexane extract of *B. asbestinum*, collected at Puerto Rico (Table 2). The structures and relative stereochemistry of metabolites (**27**–**43**) were elucidated by extensive spectral analysis (IR, UV, MS, 1D,

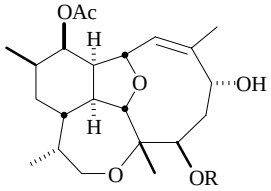
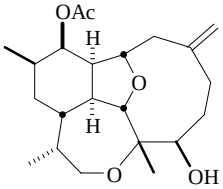
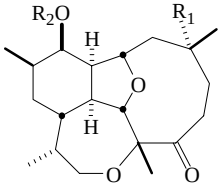
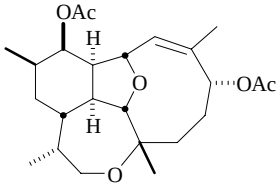
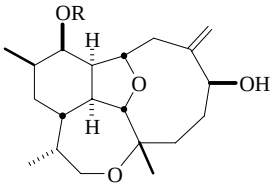
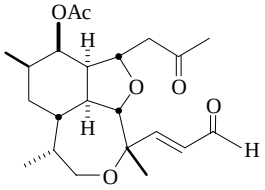
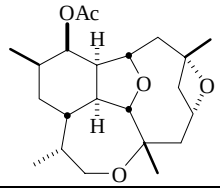
and 2D NMR) and by comparison the related spectral and physical data with those reported previously.[19,20] The structures of asbestinin-13 (**29**), -16 (**32**), -20 (**36**), and -23 (**39**) were further confirmed by chemical evidences. Oxidation of asbestinin-13 (**29**) and -20 (**36**) were found to give the asbestinin-16 (**32**) and -10 (**26**), respectively. Besides, hydrogenation of asbestinin-10 (**26**) gave a product identical with asbestinin-23 (**39**).[19]

An unnamed novel asbestinane (**44**) was obtained from *B. asbestinum*, collected off South-West Tobago. The structure, including the relative stereochemistry of metabolite (**44**) was elucidated by spectral analysis and by comparison the ^{1}H NMR spectral data with those of 11-acetoxy-4-deoxyasbestinin B (**19**).[16,21] It is noted that asbestinane (**44**) is the only asbestinane-type metabolite that possesses an oxetane ring in structure (Table 2).[21]

Table 2. The Asbestinane-Type Metabolites from *B. asbestinum*

Structure	No.	Name	Biological activity	Ref.
	**11**	asbestinin-1 (R = Ac)	showed the ability to antagonize the effects of acetylcholine on guinea pig ileum preparation at a level of 13% at a concentration of 16 μg/mL.	7,15
	13	asbestinin-3 (R = H)		7
	**12**	asbestinin-2		7,15
	**14**	asbestinin-4		7,15
	**15**	asbestinin-5	showed the ability to antagonize the effects of acetylcholine on guinea pig ileum preparation at a level of 38%, and exhibited histamine antagonism at a level of 40%, respectively (16 μg/mL).	7,15
	**16**	asbestinin epoxide		15
	**17**	asbestinin-5 acetate		15

Structure	No.	Name	Biological activity	Ref.
	**18**	4-deoxyasbestinin A (R = COC_3H_7)	ED_{50} (CHO-K1) = 3.35 μg/mL	16
	19	11-acetoxy-4-deoxyasbestinin B (R = Ac)	ED_{50} (CHO-K1) = 2.50 μg/mL	16
	**20**	4-deoxyasbestinin C (R = COC_3H_7)	ED_{50} (CHO-K1) = 3.55 μg/mL	16
	21	11-acetoxy-4-deoxyasbestinin D (R = Ac)	ED_{50} (CHO-K1) = 4.82 μg/mL Compounds (**18**–**21**) exhibited anti-microbial activity against *Klebsiella pneumoniae*.	16,17
	**22**	asbestinin-6 (R = COC_7H_{15})	IC_{50} (MCF-7, CCRF-CEM, HCT-116) = 1.5, 0.5, 5 μg/mL	18,19
	**23**	asbestinin-7 (R = COC_7H_{15})	IC_{50} (MCF-7, CCRF-CEM, HCT-116) = 9, 0.15, 5 μg/mL	18,19
	**24**	asbestinin-8 (R = COC_7H_{15})	IC_{50} (MCF-7, CCRF-CEM, HCT-116) = > 50, 2.5, 10 μg/mL	18
	**25**	asbestinin-9 (R = COC_3H_7)	IC_{50} (SK5-MEL, A-498, HCT-116) = > 50, > 50, 20 μg/mL	18
	26	asbestinin-10 (R = Ac)	IC_{50} (SK5-MEL, A-498, HCT-116) = > 50, 15, > 50 μg/mL	18
	**27**	asbestinin-11 (R = COC_7H_{15})		19
	28	asbestinin-12 (R = Ac)		19
	**29**	asbestinin-13 (R_1 = COC_7H_{15}, R_2 = α-OH)		19
	30	asbestinin-14 (R_1 = COC_5H_{11}, R_2 = α-OH)		19
	31	asbestinin-15 (R_1 = Ac, R_2 = β-OH)		19
	33	asbestinin-17 (R_1 = Ac, R_2 = α-OH)		19
	**32**	asbestinin-16 (R = COC_7H_{15})		19
	35	asbestinin-19 (R = Ac)		19

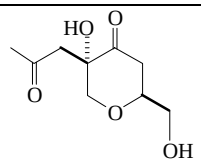
Structure	No.	Name	Biological activity	Ref.
	**34**	asbestinin-18 (R = COC_7H_{15})		19
	**36**	asbestinin-20		19
	**37**	asbestinin-21 (R_1 = OH, R_2 = Ac)		19
	38	asbestinin-22 (R_1 = OH, R_2 = COC_3H_7)		19
	39	asbestinin-23 (R_1 = H, R_2 = Ac)		19
	**40**	11-acetoxy-4-deoxyasbestinin E		19
	**41**	11-acetoxy-4-deoxyasbestinin F (R = Ac)		19
	42	4-deoxyasbestinin G (R = COC_3H_7)		19
	**43**	*seco*-asbestinin		19,20
	**44**			21

Each species of octocoral seems to have its own specific set of secondary metabolites.[22] *B. asbestinum* was proven to possess a diverse array of natural products and particularly rich in diterpenoids, such as briarellins and asbestinanes, and these metabolites are rarely found in other marine organisms. Terpenoid compounds are often present in large amounts in marine invertebrates, and as a major class, represent the largest percentage of natural products isolated from marine organisms,[23] and marine invertebrates were proven to possess the ability to synthesize the terpenoid metabolites.[24] It is important to note that the asbestinane-type metabolites are only found in *B. asbestinum*, and suggesting that this type of metabolites could be a decisive indicator in the chemotaxonomy for *B. asbestinum*. Asbestinane-type metabolites have been studied for their potential functions in defensive chemistry of colonies of *B. asbestinum*.[25,26]

3. *Briareum polyanthes*

A novel antimicrobial pyranone metabolite, bissetone (**45**), was isolated from this organism collected at Bermuda (Table 3). The structure, including the relative stereochemistry of metabolite (**45**) was determined on the basis of X-Ray analysis of the *p*-bromobenzoate derivative of compound (**45**).[27]

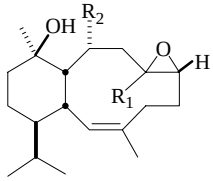
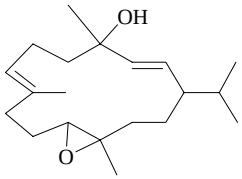
Table 3. The Pyranone Secondary Metabolite from *B. polyanthes*

Structure	No.	Name	Biological activity	Ref.
	**45**	bissetone	inhibition of the Gram-negative bacteria *Pseudomonas aeruginosa* and *Xanthomonas campestris* at 0.5 and 0.25 mg/disk, respectively, (2 mm zone in the impregnated disk assay).	27

4. *Briareum stechei*

Three new eunicellin-type metabolites, solenopodins A–C (**46**–**48**), along with an unnamed cembrane-type diterpenoid (**49**) were isolated from *B. stechei*, collected at Australian Great Barrier Reef (Table 4).[28] The gross structures and relative configuration of metabolites (**46**–**48**) were determined with spectroscopic methods (IR, MS, 1D, and 2D NMR). However, the stereochemistry of cembranoid (**49**) was not elucidated completely.[28]

Table 4. The Eunicellin and Cembrane-Type Metabolites from *B. stechei*

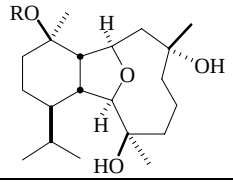
Structure	No.	Name	Ref.
	**46**	solenopodin A (R_1 = α-CH_3, R_2 = OAc)	28
	47	solenopodin B (R_1 = β-CH_3, R_2 = OAc)	28
	48	solenopodin C (R_1 = α-CH_3, R_2 = H)	28
	**49**		28

The chemical constituents of *B. stechei* indicated that a cembranoid metabolite co-occurs with eunicellin and briarane-type metabolites. This is consistent with a biosynthetic pathway wherein a cembrane intermediate serves as a precursor to the briarane and eunicellin skeletons.[28]

5. *Briareum* spp.

A new eunicellin-type compound (**50**) and its 4-acetoxy derivative (**51**) were isolated from the gorgonian *Briareum* sp., collected at Rib Reef, Australia. The structures, including the relative stereochemistry of diterpenoids (**50** and **51**) were elucidated by extensive spectroscopic methods (IR, MS, 1D, and 2D NMR) (Table 5).[29]

Table 5. The Eunicellin Metabolites from *Briareum* spp.

Structure	No.	Name	Ref.
	**50**	(1*R**,4*R**,5*S**,6*R**,8*R**,12*R**,13*R**,14*R**)-cladiellane-4,8,12-triol (R = H)	29
	51	(1*R**,4*R**,5*S**,6*R**,8*R**,12*R**,13*R**,14*R**)-4-acetoxycladiellane-8,12-diol (R = Ac)	29

Furthermore, a famous marine toxin, palytoxin (PTX),[30,31] was also found in a gorgonian identified as *Briareum* sp., collected at Lizard Island, Australia.[32] This toxin was supposed to accumulate in the organs of coral reef animals by the transport and sequestration in food chains.[33]

CONCLUSION

Based on previous studies, exclusive of briarane-type diterpenoids,[5] a number of heterocyclic natural products were obtained from the gorgonian octocorals of genus *Briareum*. A chemical class distribution of the heterocyclic natural products (**1**–**51**) compiled in this review, indicate that the diterpenoid metabolites are the major components of the heterocyclic metabolites isolated from *Briareum*. According to the chemical class distribution of these diterpenoids, they are 34 asbestinanes; 15 eunicellins; and a cembrane.

The marine organisms have been and will continue to be a rich source of novel bioactive substances for drug discovery. In above natural products isolated from the gorgonian corals of genus *Briareum*, palytoxin exhibited high toxicity in mice (LD_{50} = 0.15 mg/kg);[30] 11 metabolites (compounds **1**, **6**, and **18**–**26**) were cytotoxic toward the testing tumor cells;[6,11,16,18] five metabolites (compounds **18**–**21** and **45**) possessed antimicrobial activity;[16,27] and two metabolites (compounds **11** and **15**) showed antagonistic effect.[15] These results indicate that the heterocyclic natural products from gorgonians of genus *Briareum* are potential for pharmaceutical researches in the future.

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