Structures of Agelasines, Diterpenes Having a 9-Methyladeninium Chromophore Isolated from the Okinawan Marine Sponge *Agelas nakamurai* Hoshino[1)]

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Novel diterpenes, agelasine-A, -B, -C, -D, -E, and -F were isolated from the Okinawan marine sponge *Agelas nakamurai* Hoshino and their structures were established to be monocyclic and bicyclic diterpenes having a 9-methyladeninium unit by spectral analyses and chemical transformations.

Since Cullen and Devlin reported the isolation of agelasine, a quaternary 9-methyladenine derivative of an unidentified diterpene as a constituent of the marine sponge *Agelas dispar* in 1975,[3)] a variety of novel compounds having a polar functionality attached to a terpenoid moiety, the latter varying from acyclic sesqui to a mono or bicyclic diterpene skeleton have been isolated from marine sponges of the genus *Agelas* as bioactive metabolites. Included among these were terpenoid derivatives of hypotaurocyamine, agelasidine-A, -B and -C (**1**, **2**, and **3**)[4,5)] and seven diterpene derivatives of quaternary 9-methyladenine, agelasine-A, -B, -C, -D, and -E (**4**, **5**, **6**, **7**, **8**), agelasine-F (ageline-A, **9**), and ageline-B (**10**) isolated from the Okinawan marine sponge *Agelas nakamurai* by us[6,7)] and from a Pacific marine sponge *Agelas* sp. by Capon and Faulkner[8)] (Fig. 1). These compounds exhibited interesting bioactivities: Inhibitory effects on growth of microorganisms, contractive responses of smooth muscles and enzymic reactions of Na,K-ATPase. We

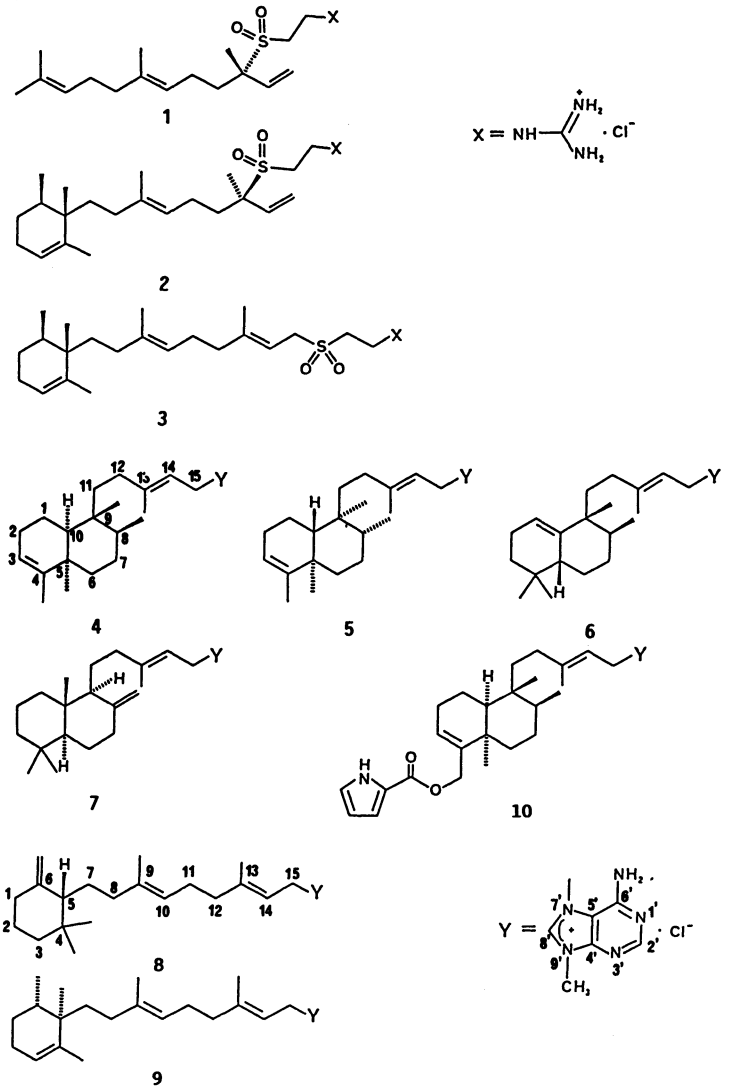


Fig. 1.

Table 1. ^{13}C NMR Data for Agelasine-A (**4**), -B (**5**), -C (**6**), -D (**7**), -E (**8**), -F (**9**), 7,9-Dimethyladeninium Perchlorate (**16**), and Methyl Kolavenate (**24**)[a)]

Carbon	4	5	6	7	8	9	16	24
1	17.6t	18.3t	121.0d	39.4t	38.0t	25.2t		18.4t
2	24.0t	27.5t	23.9t	20.3t	23.5t	26.8t		27.5t
3	123.1d	120.3d	38.2t	43.2t	36.1t	122.4d		120.5d
4	139.7s	144.3s	32.1s	34.4s	34.6s	139.3s		144.4s
5	40.2s	38.7s	44.5d	56.8d	53.5d	40.1s		38.8s
6	36.1t	36.3t	24.5t	25.5t	149.0s	33.0d		36.4t
7	28.8t	26.9t	29.9t	40.2t	24.6t	34.0t		26.9t
8	37.4d	36.3d	40.2d	149.0s	32.3t	35.0t		36.4d
9	37.7s	38.2s	43.8s	57.4d	136.2s	136.5s		38.3t
10	44.7d	46.4d	142.5s	40.6s	122.7d	123.7d		46.6d
11	33.0t	33.1t	34.2t	39.4t	26.0t	26.1t		34.7t
12	36.9t	36.8t	35.4t	22.5t	39.4t	39.4t		36.9t
13	147.9s	147.5s	149.3s	149.5s	146.8s	146.8s		161.6s
14	115.7d	115.7d	115.4d	115.9d	115.2d	115.2d		114.9d
15	48.7t	48.7t	48.8t	48.8t	48.4t	48.4t		162.3s
4-CH_3	19.7q	18.3q	22.6q	22.1q	26.0q	18.9q		19.2q
			28.6q	34.0q	28.2q			
5-CH_3	33.0q	19.9q				15.6q		20.0q
6-CH_3						20.8q		
6-=CH_2					108.6t			
8-CH_3	16.0q	16.0q	16.0q					16.0q
8-=CH_2				106.9t				
9-CH_3	17.8q	17.9q	26.3q		15.9q	16.0q		18.1q
10-CH_3				15.0q				
13-CH_3	17.3q	17.5q	17.3q	17.1q	17.2q	17.2q		18.4q
2′	155.8d	156.0d	156.7d	156.9d	155.2d	155.7d	155.4d	
4′	149.4s	149.5s	150.5s	150.7s	149.2s	149.2s	148.6s	
5′	109.7s	109.7s	110.8s	111.0s	109.5s	109.5s	109.7s	
6′	152.4s	152.5s	153.8s	153.9s	152.2s	152.2s	152.4s	
8′	141.7d	141.7d	142.0d	142.6d	141.2d	141.2d	141.9d	
9′-NCH_3	32.0q	32.0q	32.1q	32.0q	31.7q	31.7q	31.3q	
7′-NCH_3							36.2q	

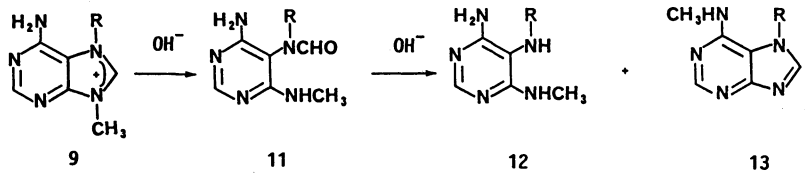
a) δ in ppm, **4**—**9** and **24** in $CDCl_3$ and **16** in DMSO-d_6.

have recently reported the isolation and structures of agelasine-A—F briefly. In this paper, we describe the structural elucidation of the compounds in detail.

Specimens of *Agelas nakamurai* were collected at Zanpa Cape, Okinawa using SCUBA (−10—−20m), frozen shortly for transportation and stored at −20° C until workup. The chloroform-soluble material of methanolic extract of the sponge was chromatographed on a silica-gel column with 3:12:2:2 chloroform–1-butanol–acetic acid–water as eluent to give two fractions. The less polar fraction was repeatedly fractionated by HPLC on a C_{18} column using a methanol–water solvent system containing 0.2 M† sodium chloride to give agelasine-A, agelasine-E and agelasine-F (ageline-A), and a mixture of agelasine-B, -C, and -D. The mixture was further repeatedly chromatographed by HPLC using a C_8 column with methanol–water containing 0.2 M sodium chloride as a mobile phase to yield agelasine-B, -C, and -D. Agelasine-A—F are ammonium salts and isolated as chloride forms. The isolation yields were 0.037, 0.1, 0.01, 0.0056, 0.031, and 0.08% for agelasine-A—F respectively, from the wet weight of the sponge.

Agelasine-A—F (**4**—**9**) showed a number of spectral features in common: Their field desorption mass spectra (FDMS) contained identical principal ions at *m/z* 422 (M^+—Cl) and their high-resolution EI mass spectra (HREIMS) exhibited the principal ions at *m/z* 421.3216, 421.3204, 421.3177, 421.3182, 421.3202, and 421.3201 (M^+—HCl), respectively, revealing the same molecular formula, $C_{26}H_{40}N_5Cl$ (Calcd for $C_{26}H_{39}N_5$: M—HCl 421.3202). Their UV spectra showed alike absorption maxima at 272 nm with very similar ε values, indicating the presence of a same UV absorbing chromophore in these components. The presence of a common main fragment ion at *m/z* 149 in their electron impact mass spectra suggested that the chromophore was composed of $C_6H_7N_5$ (HREIMS *m/z* 149.0722, Calcd for $C_6H_7N_5$: 149.0709). This was supported by the facts that their ^1H NMR spectra contained four common signals corresponding seven protons assignable to NH_2 (δ=6.76—6.98), N-CH_3 (δ=4.10) and two aromatic protons (δ=8.48—8.50, 2′-H and δ=10.91—10.94, 8′-H) and their ^{13}C NMR spectra (Table 1) possessed six common signals due to a heteroaromatic ring. The common chromophore was assigned as a

† 1 M=1 mol dm^{-3}.



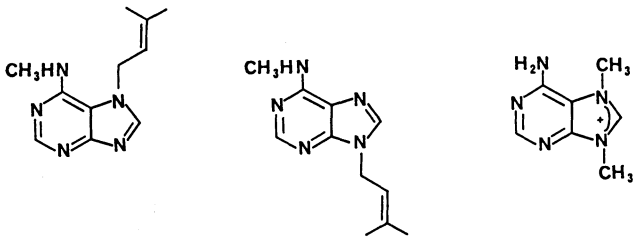
9 11 12 13

Fig. 2.

quaternary 9-methyladenine moiety on the basis of above spectral data and their following chemical behavior.

Agelasines were quite stable under acidic conditions, whereas the compounds were very unstable under basic conditions, even on a alumina column these compounds decomposed to yield less polar materials (Fig. 2). Agelasine-F (**9**), upon treatment with a base, was converted to the corresponding formamide **11**, whose ^{1}H NMR spectrum for the nitrogen-containing portion showed signals at δ 2.97 (d, 3H, J=5 Hz) for N-CH_3, 4.80 (brs, 2H) due to NH_2, and 7.96, and 8.14 (s, each 1H) assignable to the formamide group and 2′-H, respectively. Its electron impact mass spectrum (EIMS) exhibited a molecular ion at m/z 439 corresponding to a molecular formula $C_{26}H_{41}N_5O$. These data were in good agreement with literature values.[3] Furthermore, **9** was heated with 40% KOH at 110° C in a sealed tube to give two products. The less polar product was a white powder; UV absorption maximum at 272 nm (ε 8350); its ^{1}H NMR spectrum contained signals at δ 2.99 (d, 3H, J=5 Hz) for N-CH_3, 3.40 (d, 2H, J=7.5 Hz) due to N-methylene and 7.99 (s, 1H) arising from 2′-H. Lack of the signal for the formamide and the molecular ion at m/z 411 encouraged us to formulate the nitrogenous portion of the molecule as **12**. The polar product was identified as 6-N-methyl-7-alkyladenine derivative (**13**) which had been obtained by Cullen and Devlin from the cyclization of a relevant formamide with sodium hydride in N,N-dimethylacetamide. The structure was confirmed by comparing the spectral data of **13** with those of 6-N-methyl-7-prenyladenine (**14**) and 6-N-methyl-9-prenyladenine (**15**). During these chemical conversions no notable change could be found for the structure of its alkyl moiety.

The other components demonstrated identical behavior on treating with a base. The structure of nitrogenous portion of agelasines was resultantly confirmed by the comparison of the spectral data with those of 7,9-dimethyladeninium perchlorate (**16**).[9a] Above results established that agelasines contained a common polar group, quaternary 9-methyladeninium, and differed only in hydrocarbon portion which was composed of $C_{20}H_{33}$, a diterpene moiety. Further examination of the ^{1}H and ^{13}C NMR spectra of agelasines made it apparent that agelasines contained a common terminal grouping -C(CH_3)=CH-CH_2-Y, whereby the hydrocarbon portion of the molecule connected to 7-N atom of the 9-methyladenine. The E geometry of the double bond was established by their



14 15 16

Fig. 3.

high field ^{13}C NMR signals for vinyl methyls (δ=17.1—17.5). The elucidation of the structures of remaining parts for each component was performed as follows.

Agelasine-A and -B (**4**, **5**), both exhibited one vinyl proton signal, two singlet, one doublet and one vinyl methyl signals at δ 5.28 (brs), 0.79 (s), 1.02 (s), 0.73 (d, J=5.8 Hz) and 1.67 (brs) for **4** and δ 5.17 (brs), 0.70 (s), 0.97 (s), 0.76 (d, J=5.2 Hz) and 1.57 (brs) for **5**, respectively, in their ^{1}H NMR spectra and the ^{13}C NMR signals for a trisubstituted double bond at δ 123.1 (d) and 139.7 (s) for **4** and 120.3 (d), 144.3 (s) for **5**, respectively. This spectral feature is consistent with a clerodane skeleton, a bicyclic ring system. For clerodane type compounds, the ^{13}C NMR chemical shift of 5-methyl usually provides particularly valuable piece of structural evidence for the mode of ring fusion: That of the trans-isomer occurs ca. 11—12 ppm upfield from that of the cis-isomer.[10] Indeed, on inspection of their ^{13}C NMR data, it was found that **4** exhibited a diagnostic low field signal (δ=33.4) due to the angular methyl while **5** showed a high field signal for the angular methyl (δ=19.9). Therefore cis and trans fused clerodane skeleton were assigned to **4** and **5**, respectively.

Since there are four asymmetric centers for clerodane ring system, for the gross structure there are sixteen stereochemical variations, wherein only six have been reported. However **4** on treating with a mixture of hydrochloric acid and acetic acid, converted to **17a** (Fig. 4), leaving only two asymmetric centers at 8 and 9 positions. **17a** can be correlated to **18a**, **18b**, and **18c**. For **18a**[11] having *trans*-8,9-dimethyl, the three singlet methyl signals occurred at δ 0.98, while for **18b**[11] and **18c**[12] having *cis*-8,9-dimethyl, two singlet methyl signals appeared at low field (δ=0.98 for **18b**, and 0.97 and 0.98 for **18c**, respectively), the other was at somewhat high field (δ=0.78 for **18b** and 0.84 for **18c**). The similarity of chemical shifts for three methyl signals in **17a** (δ=0.79, 0.94, and 0.96) clearly favored

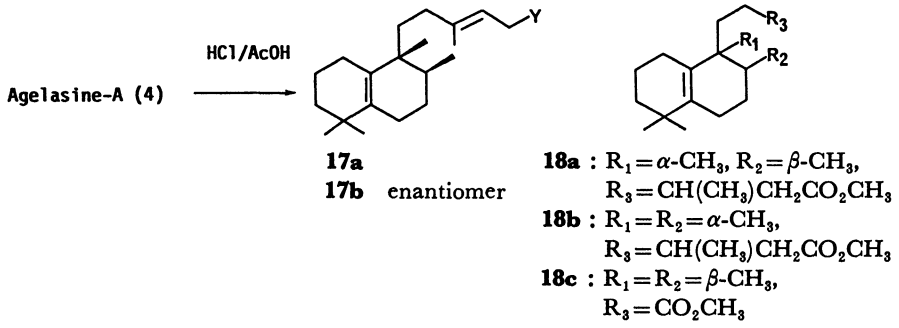


Fig. 4.

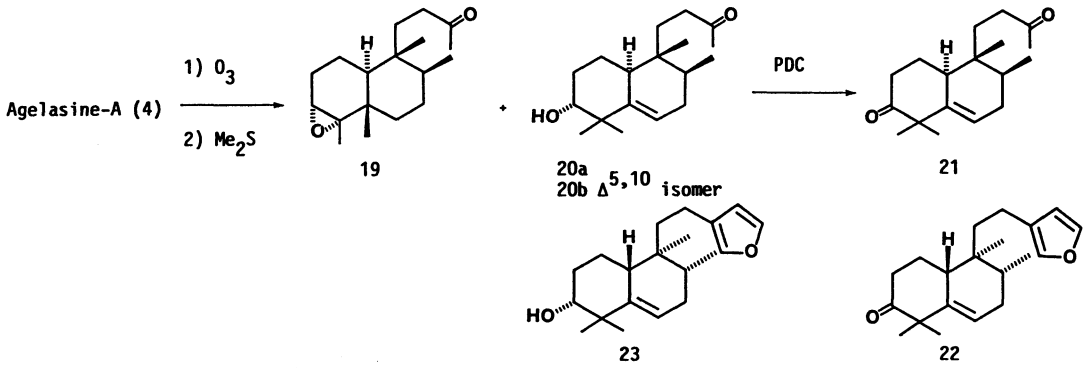
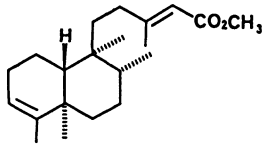


Fig. 5.

the relative stereochemistry of **18b** and **18c** over that of **18a**. Since the specific rotation of **17a** ($[\alpha]_D$ −14.7°) was same in sign to that of **18c** ($[\alpha]_D$ −52.9°), the absolute configuration of **17a** is probably identical to that of **18c**. It was confirmed by comparing **17a** with the enantiomeric **17b** which was obtained from agelasine-B (**5**). For agelasine-A, the remaining problem involved the deduction of the configuration of C-5 or C-10. Ozonolysis of **4** followed by reduction with dimethyl sulfide furnished an epoxide **19** and two olefinic alcohols **20a** and **20b** (Fig. 5), whose structures were assigned on the basis of their spectral data: **20a** showed an olefinic proton signal at δ 5.55 (m, 1H) and two ^{13}C NMR signals at δ 144.8 (s) and 118.4 (d) for a trisubstituted vinyl group while **20b** showed no olefinic proton signal and two ^{13}C NMR signals at δ 131.3 (s) and 135.1 (s) for a tetrasubstituted double bond. **20a** was oxidized with pyridinium dichromate (PDC) to yield a diketone **21**, which can be correlated to **22** obtained from a solidago diterpene via **23** by McCrindle et al.[13] The comparison of the ^1H NMR spectral data between **21** and **22** revealed close similarity. But **21** showed negative Cotton effect ($[\theta]_{293}$ −3480), which was opposit to that of **22** ($[\theta]_{291}$ +3320) suggesting that the absolute configuration of C-10 in **21** is reversed to that of **22**. Since **4** had a cis-fused decalin skeleton, absolute configurations for all asymmetric centers in **4** thus were well established. On the other hand, there were some differences in chemical shifts for methyls between **20a** and **23**; δ=0.65 (s), 0.81 (d, J=6.5 Hz), 0.94 (s), and 1.13 (s) for **20a** and 0.72 (s), 0.87 (d, J=6 Hz), 1.06 (s), and 1.17 (s) for **23** respectively. This may be explained by different orientation of the hydroxyl group with respect to 10-H between them. In **23**, the hydroxyl group was situated anti and oriented axially, but in **20a** the hydroxyl group must be situated syn and adopted equatorial orientation, which was indicated by the coupling constants of 3-H proton signal at δ 3.23 (dd, J=4 and 11 Hz). This revealed that the epoxy group in **19** and the hydroxyl group in **20b** were also α-oriented.



24

Fig. 6.

Treatment of agelasine-B with a mixture of hydrochloric acid and acetic acid as well as agelasine-A furnished a tetrasubstituted olefinic compound. As **17b** was identical to **17a** in all respects except the sign of optical rotation ($[\alpha]_D$ +17.1°), it must be an antipode with α-dimethyl groups at 8 and 9 positions. Since agelasine-B has a trans-fused clerodane skeleton, there were only two stereochemical alternatives: 5β-methyl and 10α-proton or 5α-methyl and 10β-proton. The latter was present in methyl kolavenate (**24**)[14] (Fig. 6). The chemical shifts of four methyl signals in **5**, δ=0.70 (s), 0.76 (d, J=5.2 Hz), 0.97 (s) and 1.57 (s), were closely parallel to those of **24**, δ=0.76 (s), 0.85 (d),1.02 (s) and

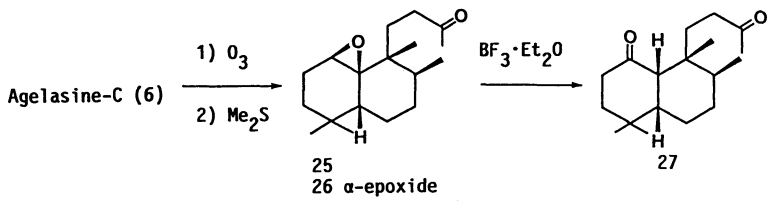


Fig. 7.

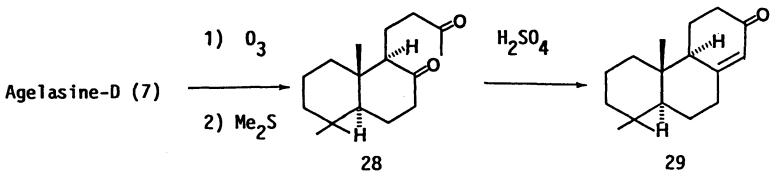


Fig. 8.

1.60 (s), and comparison of ^{13}CNMR spectrum of **5** with that of **24** revealed an excellent correlation for the relevant signals (Table 1). Since the optical rotation of **5** ($[\alpha]_D$ −21.5°) is same in sign to that of **24** ($[\delta]_D$ −58°), the absolute configuration of **5** must be identical to that of **24** at all asymmetric centers. Recently, the structure of agelasine-B (**5**) was confirmed by the synthesis of **5** from **24** by Iio et al.[15]

The ^{1}HNMR spectrum of agelasine-C (**6**) showed a vinyl proton signal at δ 5.31, three singlet methyl signals at δ 0.82, 0.84, and 0.87 and one doublet methyl signal at δ 0.79 (J=6.8 Hz). Thus a rearranged labdane skeleton with 1, 10- or 5,6-double bond was assumed for its gross structure. However, lack of a high field methyl signal, which had been observed for 9-methyl in **20a** (δ=0.65), favored the double bond arrangement at 1,10-position. An acid catalyzed rearrangement of **6** as well as **4** yielded **17a**, indicating the stereochemistry of C-8 and C-9 as illustrated in **6**. Furthermore, reductive ozonolysis workup of **6** (ozone and dimethyl sulfide) gave two epoxides **25** and **26** (Fig. 7). Both epoxides were very hindered and the attempt to reduce them with lithium triethylhydroborate was failed. Then **26** was treated with boron trifluoride etherate to yield a diketone **27** whose ^{1}HNMR spectrum contained signals for α-protons to the carbonyl group at 1-position, δ=2.12 (2-H, ddd, 1H, J=2.3, 4.4, and 13.5 Hz), 2.42 (2-H, dt, 1H, J=6.3 and 13.5 Hz) and 2.48 (10-H, d, 1H, J=5.0 Hz) besides four methyl signals at δ=0.97 (s), 1.03 (d, J=6.9 Hz), 1.17 (s) and 1.23 (s). On irradiation at δ=1.44 (8-H), the doublet methyl signal for C-8 methyl was collapsed to a singlet, whereas the α-proton signals were not affected. This result companied with the small J-value of 10-H signal (5.0 Hz) provided valid evidence for that **27** has a cis-fused decalin skeleton with a carbonyl group at **1** position. In addition, the observation of nuclear Overhauser effects between both the two singlet methyl signals at C-4 and C-9 (δ=0.97 and 1.23) and the 10-H signal (δ=2.48) required a non-steroid-like conformation for **27** which exhibited negative

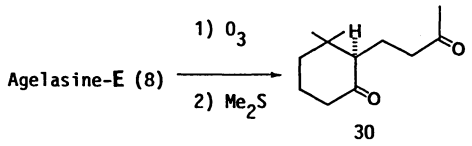


Fig. 9.

Cotton effect ($[\theta]_{298}$ −2370). On the basis of the octant rule, the absolute configuration of **27** was determined as illustrated. Since **27** might be formed from the α-epoxide **26** via a trans antiparallel hydride shift from C-1 to an incipient cationic center at 10-position and the configuration of C-5 was uneffected by the rearrangement conditions employed, the epoxy groups in **25** and **26** might be β- and α-oriented and the stereochemistry of **6** should be as illustrated.

Agelasine-D (**7**) showed three singlet methyl signals at δ 0.65, 0.79, and 0.87 and two vinyl proton signals at δ 4.43 (brs) and 4.81 (brs) for an exomethylene group which was further suggested by the ^{13}CNMR signals at δ 106.9 (t) and 149.0 (s). These data are compatible with a labdane ring system, a well studied skeleton. The stereochemistry of **7** was established by following chemical conversion (Fig. 8). Ozonolysis of **7** followed by reduction with dimethyl sulfide afforded a diketone **28**, which was treated with sulfuric acid to yield an enone **29**. Its spectral data and optical properties were identical with those of an authentic enone.[16,17]

The ^{13}CNMR spectrum of agelasine-E (**8**) showed six olefinic carbon signals at δ 108.3 (t), 115.2 (d), 122.7 (d), 136.2 (s), 146.8 (s), and 149.0 (s) due to its hydrocarbon portion, indicating the presence of three double bonds and a ring. In addition, its ^{1}HNMR spectrum contained signals at δ 5.00 (brs, 1H) and 1.57 (s, 3H) for additional group $-C(CH_3)=CH-CH_2-$, in which the geometry of the double bond was assigned to be *E* on the basis of a high field resonace of an olefinic methyl carbon (δ=15.9), leaving a terminal monocyclic moiety. The remaining ^{1}HNMR data showed two singlet methyl signals at δ 0.82 and 0.90 and an exomethylene proton signals at δ 4.53 (brd,

J=1.5 Hz) and 4.76 (brd, J=1.5 Hz) due to the terminal cyclic part. A 1-alkyl-2-methylene-6,6-dimethylcyclohexane ring system was assigned for the terminal residue. The structure of the diterpene portion in **8** was confirmed by ozonolysis results (Fig. 9). **8** was treated with ozone and worked up with dimethyl sulfide to give a diketone **30** and an unidentified compound **31** arising from the linear polyisoprenyl chain. The CD spectrum of **30** showed a negative Cotton effect ($\Delta\varepsilon_{292}$ −1.57) which was opposite to that of a known diketone having a 5*S* configuration ($\Delta\varepsilon_{296}$ +1.57),[18] indicating the absolute configuration of **8** to be 5*S*. The reduction of **31** with sodium borohydride followed by acetylation with acetic anhydride and pyridine to give 1,4-diacetoxypentane.

The spectral data of agelasine-F (**9**) was in good agreement with those of ageline-A and an identical structure was proposed for it.[8] However, the absolute configuration of **9** was independently established by a successive chemical conversions outlined as following (Fig. 10). The ozonolysis of **9** followed by reductive workup (ozone at −78° C and sodium borohydride) furnished an epoxide **32** and 1,4-pentanediol. **32** was further reduced with lithium triethylhydroborate to give a diol **33**, which was acetylated and then was submitted to dehydration with thionyl chloride/pyridine to yield a 2:1 mixture of an exocyclic olefin **35** and its endo-isomer. The mixture without separation was deacetylated with sodium methoxide to give olefinic alcohol **36**, which then was oxidized with pyridinium dichromate to yield a mixture containing a compound **37**. The mixture was treated with ozone, reduced with dimethyl sulfide and cyclized in methanolic KOH under an argon atmosphere to yield a cis-octalone **38**, which was identical in all respects with an authentic sample.[19] The CD spectrum of **38** exhibited a positive Cotton effect at 236 nm ([θ] +5800) and a negative one at 335 nm ([θ] −390) in hexane, indicating the absolute configuration of the ring junction methyl group as illustrated. On the other hand, ozonolysis of **9** (reductive workup with dimethyl sulfide) furnished an enol ether **39** and a ketone **40**. **39** may be formed via a trans-opening of an epoxide and was characterized by the carbon signals at δ 92.8 (d) and 146.9 (s). It then was treated with *p*-bromobenzoyl chloride and triethylamine to yield **41**. The structure of **41** was elucidated by the interpretation of its ^{1}HNMR spectrum and extensive homodecoupling experiments run at 400 MHz. The axial orientation of *p*-bromobenzoyloxy group and equatorial orientation of 1-methyl were indicated by the coupling constants of the proton signals at 1 and 4 position (J_{1-2a}=13 Hz and J_{3a-4}=J_{3b-4}=3 Hz). Therefore the downfield shift of the signal for 10-methyl (δ=0.93) by the benzoylation ($\Delta\delta$ 0.12) revealed that the *p*-bromobenzoyloxy group i.e. the epoxy group and 10-methyl group in **32** were situated syn. The CD spectrum of compound **40** showed a positive Cotton effect ([θ]$_{295}$ +131). Hence it could in turn be used as a standard to deduce the stereochemistry of agelasidine-B and -C.[5]

Experimental

General. All mp's were obtained on a Yanagimoto micro melting point apparatus and uncorrected. Optical rotations were measured on a Union automatic polarimeter PM-201. IR spectra were recorded on a Hitachi 260-50 spectrophotometer. ^{1}HNMR spectra were taken on JEOL FX-90Q (90 MHz), Bruker WH-270 (270 MHz), or Bruker AM-400 (400 MHz) instruments. ^{13}CNMR spectra were recorded with a JEOL FX-90Q (22.5 MHz) spectrometer. FD mass spectra and HREI mass spectra were measured on a Hitachi M-80A mass spectrometer. CD spectra were obtained on a JASCO J-40A spectrophotometer. UV spectra were taken on a Varian Cary-17 instrument.

Collection, Extraction, and Separation. The marine sponge *Agelas nakamurai* Hoshino (5 Kg, wet weight) was collected at Zampa Cape, Okinawa in 1981 using SCUBA

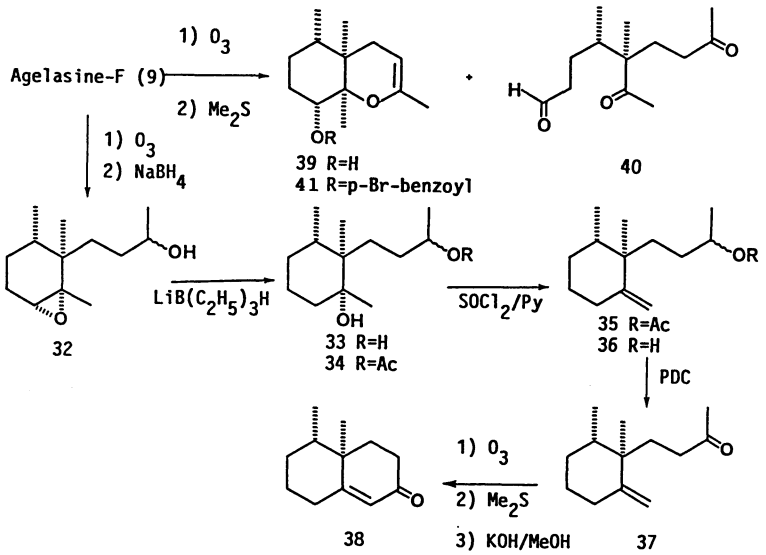


Fig. 10.

(−10—−20 m), shortly frozen and shipped via air to Tokyo, then stored at −20° C until workup. The sponge was cut into small pieces and extracted with methanol (3×40 l). The solvent was evaporated under reduced pressure to give a crude extract (195 g), which was dissolved in methanol. The methanol-soluble material (125 g) was partitioned between chloroform and water. Each 12 g of chloroform-soluble material (60 g) was chromatographed on a silica-gel column (Wako gel C-300, Wako Chemical, 50×600 mm) with 3:12:2:2 chloroform-1-butanol-acetic acid-water as eluent, monitered by TLC. The less polar fraction contained agelasidines (positive coloration with Sakaguchi reagent) and the polar fractions were consisted of agelasines (slightly positive coloration with Sakaguchi reagent and strong UV absorption). The polar fraction was purified by preparative HPLC on a C_{18} column (Nomura Chemical Co. Ltd. Develosil ODS 15/30, 21×250 mm×3) with 8:2 methanol-water containing 0.2 M sodium chloride (flow rate 28 ml min^{-1}) to give a mixture of agelasines (22 g). A part (2 g) of the mixture was separated on a C_{18} column (Develosil ODS-5, 10×250 mm) with 8:2 methanol-water containing 0.2 M sodium chloride (flow rate 4 ml min^{-1}) to yield agelasine-A (**4**, t_R=17.3 min, 170 mg), a mixture of agelasine-B, -C, and -D (t_R=18 min, 822 mg), and agelasine-E (**8**, t_R=19.8 min, 255 mg) and agelasine-F (**9**, t_R=21 min, 368 mg). The mixture was chromatographed on a C_8 column (Develosil C_8-5, 10×250 mm) using methanol-water (73:27) containing 0.2 M sodium chloride as eluent to give a mixture of agelasine-B and -D (t_R=32.3 min, 643 mg), and agelasine-C (**6**, t_R=39 min, 143 mg). The former was further separated by HPLC on a C_8 column (Develosil C_8-5, 10×250 mm) with 7:3 methanol-water containing 0.2 M sodium chloride to furnish agelasine-B (**5**, t_R=53.5 min, 220 mg) and agelasine-D (**7**, t_R=56.3 min, 24 mg).

Agelasine-A (4). Amorphous solid; mp 173—174° C; $[\alpha]_D$ −31.3° (*c* 0.59, CH_3OH); UV (CH_3OH) 272 nm (ε 8910); IR ($CHCl_3$) 3320, 3150, 2950, 1640, 1610, 1455, 1380, 1300, 1230, 1190, 1090 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.73 (d, 3H, *J*=5.8 Hz), 0.79 (s, 3H), 1.02 (s, 3H), 1.67 (brs, 3H), 1.88 (brs, 3H), 1.0—2.1 (m, 14H), 4.10 (s, 3H), 5.26 (brs, 1H), 5.41 (brt, 1H, *J*=6.5 Hz), 5.71 (brd, 2H, *J*=6.5 Hz), 6.98 (brs, 2H, exch), 8.48 (s, 1H), 10.81 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 150, 81, 41, 69, 55, 216, 217, 163, 298, 406; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3216. Calcd for $C_{26}H_{39}N_5$: M−HCl 421.3202.

Agelasine-B (5). Amorphous solid; mp 167—170° C; $[\alpha]_D$ −21.5° (*c* 1.0, CH_3OH); UV (CH_3OH) 272 nm (ε 8240); IR ($CHCl_3$) 3370, 3160, 2960, 1640, 1610, 1590, 1460, 1385, 1300, 1240, 1195, 1090 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.70 (s, 3H), 0.76 (d, 3H, *J*=5.2 Hz), 0.97 (s, 3H), 1.57 (s, 3H), 1.86 (brs, 3H), 0.7—2.2 (m, 14H), 4.10 (s, 3H), 5.17 (brs, 1H), 5.41 (brt, 1H, *J*=6.5 Hz), 5.71 (brd, 2H, *J*=6.5 Hz), 6.84 (brs, 2H, exch), 8.50 (s, 1H), 10.89 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 95, 121, 107, 189, 163; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3204. Calcd for $C_{26}H_{39}N_5$: M−HCl 421.3202.

Agelasine-C (6). Amorphous solid; mp 176—179° C; $[\alpha]_D$ −55.1° (*c* 2.04, CH_3OH); UV (CH_3OH) 272 nm (ε 8340); IR (KBr) 3300, 3140, 2965, 1645, 1615, 1470, 1380, 1360, 1230, 1180 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.79 (d, 3H, *J*=6.8 Hz), 0.82 (s, 3H), 0.84 (s, 3H), 0.87 (s, 3H), 0.7—2.2 (m, 14H), 4.10 (s, 3H), 5.31 (brt, 1H, *J*=6.2 Hz), 5.41 (brt, 1H, *J*=6.5 Hz), 5.70 (brd, 2H, *J*=6.5 Hz), 6.84 (brs, 2H, exch), 8.50 (s, 1H), 10.88 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 191, 177, 135, 122, 95, 107; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3177. Calcd for $C_{26}H_{39}N_5$: M−HCl 421.3202.

Agelasine-D (7). Amorphous solid; mp 175—176° C; $[\alpha]_D$ +10.4° (*c* 1.05, CH_3OH); UV (CH_3OH) 272 nm (ε 9180); IR ($CHCl_3$) 3300, 3150, 2950, 1640, 1610, 1590, 1455, 1390, 1300, 1090, 895 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.65 (s, 3H), 0.79 (s, 3H), 0.87 (s, 3H), 0.6—2.4 (m, 16H), 1.86 (s, 3H), 4.10 (s, 3H), 4.43 (brs, 1H), 4.81 (brs, 1H), 5.41 (brt, 1H, *J*=6.5 Hz), 5.72 (brd, 2H, *J*=6.5 Hz), 6.75 (brs, 2H, exch), 8.50 (s, 1H), 10.94 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 81, 95, 69, 137, 109, 121, 217, 293; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3182. Calcd for $C_{26}H_{39}N_5$: M−HCl 421.3202.

Agelasine-E (8). Amorphous solid; mp 180—182° C; $[\alpha]_D$ −17.1° (*c* 1.88, CH_3OH); UV (CH_3OH) 272 nm (ε 9720); IR ($CHCl_3$) 3330, 3150, 2950, 1640, 1610, 1590, 1455, 1405, 1380, 1365, 1300, 1235, 1190, 1090 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.82 (s, 3H), 0.90 (s, 3H), 1.56 (s, 3H), 1.86 (s, 3H), 0.7—2.2 (m, 15H), 4.10 (s, 3H), 4.53 (brd, 1H, *J*=1.5 Hz), 4.76 (brd, 1H, *J*=1.5 Hz), 5.00 (brs, 1H), 5.46 (brt, 1H, *J*=6.5 Hz), 5.72 (brd, 2H, *J*=6.5 Hz), 6.87 (brs, 2H, exch), 8.50 (s, 1H), 10.83 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 216, 81, 95, 69, 109; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3202. Calcd for $C_{26}H_{39}N_5$: M−HCl 421.3202.

Agelasine-F (9). Amorphous solid; mp 178—180° C; $[\alpha]_D$ −5.5° (*c* 2.55, CH_3OH); UV (CH_3OH) 272 nm (ε 7700); IR ($CHCl_3$) 3300, 3150, 2970, 1640, 1590, 1455, 1380, 1300, 1235, 1090 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.84 (s, 3H), 0.85 (d, 3H, *J*=7 Hz), 1.57 (s, 3H), 1.59 (s, 3H), 1.86 (s, 3H), 1.0—2.2 (m, 13H), 4.10 (s, 3H), 5.02 (brs, 1H), 5.42 (brs, 1H), 5.47 (brt, 1H, *J*=7 Hz), 5.72 (brd, 2H, *J*=7 Hz), 6.87 (brs, 2H, exch), 8.50 (s, 1H), 10.83 (s, 1H); EIMS *m/z* 421 (M^+−HCl), 149, 150, 216, 217, 298, 123, 121; FDMS *m/z* 422 (M^+−Cl); Found: *m/z* 421.3201, 149.0722. Calcd for $C_{26}H_{39}N_5$ and $C_6H_7N_5$: M−HCl 421.3202 and 149.0709.

Acid Catalyzed Rearrangement of Agelasine-A (4). **4** (10 mg, 0.022 mmol) was treated with a solution of acetic acid (0.4 ml) and 12 M HCl (0.1 ml) at room temperature using HPLC to follow the reaction. After 30 min the starting meterial was converted completely to give single product. The reagents were removed in vacuo and 1 M NaCl solution (7 ml) was added. The solution was extracted with ethyl acetate (2×15 ml). The extract was concentrated to yield **17a** (10 mg, 100% theoretical). **17a**: Colorless crystals; ^{1}H NMR ($CDCl_3$, 270 MHz) δ=0.79 (s, 3H), 0.80 (d, 3H, *J*=6 Hz), 0.94 (s, 3H), 0.96 (s, 3H), 1.86 (brs, 3H), 1.0—2.0 (m, 15H), 4.08 (brs, 3H), 5.40 (brt, 1H, *J*=6 Hz), 5.67 (brd, 2H, *J*=6 Hz), 7.04 (brs, 2H exch), 8.46 (brs, 1H), 10.70 (s, 1H); ^{13}C NMR ($CDCl_3$, 67.5 MHz) δ=16.3 (q), 17.6 (q), 20.0 (t), 21.2 (q), 25.2 (t), 25.8 (t), 27.2 (t), 27.8 (q), 29.3 (q), 32.0 (q), 33.7 (d), 34.4 (t), 34.6 (s), 40.0 (t), 40.7 (s), 48.8 (t), 109.9 (s), 115.5 (d), 132.0 (s), 137.5 (s), 141.9 (d), 147.8 (s), 149.6 (s), 152.5 (s), 156.0 (d); $[\alpha]_D$ −14.7° (*c* 0.42, CH_3OH).

Ozonolysis of Agelasine-A (4). A solution of **4** (30 mg, 0.066 mmol) in methanol (3 ml) was saturated with ozone for 30 min at −78° C. The excess ozone was removed by a N_2 stream and dimethyl sulfide (0.055 ml) was added. The solution was stirred at 0° C for 30 min and room temperature overnight. The solvent was removed under reduced pressure and the residue was separated by HPLC on a C_{18} column (Develosil ODS-5, 10×250 mm) with methanol-water (8:2) to give the olefinic alcohol **20a** (4 mg, t_R=13.38 min, 22% theoretical), the epoxide **19** (1.5 mg, t_R=20.63 min, 8.3% theoretical) and the olefinic alcohol **20b** (3 mg, t_R=23.85 min, 16.6% theoretical). **20a**: A colorless syrup; ^{1}H NMR ($CDCl_3$,

90 MHz) δ=0.65 (s, 3H), 0.81 (d, 3H, J=6.5 Hz), 0.94 (s, 3H), 1.13 (s, 3H), 2.16 (s, 3H), 3.23 (dd, 1H, J=4, 11 Hz), 5.55 (m, 1H); EIMS m/z 278 (M^+), 189, 187, 119, 207, 227, 161, 224, 260. **19**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.81 (d, 3H, J=6 Hz), 0.87 (s, 3H), 1.09 (s, 3H), 1.32 (s, 3H), 2.15 (s, 3H), 2.98 (brd, 1H, J=3.5 Hz); EIMS m/z 278 (M^+), 207, 189, 149, 121, 123, 119, 109, 107, 95, 249, 221. **20b**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.83 (d, 3H, J=4.8 Hz), 0.85 (s, 3H), 1.01 (s, 3H), 1.02 (s, 3H), 2.12 (s, 3H), 3.52 (m, 1H); EIMS m/z 278 (M^+), 189, 207, 119, 135, 187, 105, 227, 260, 244, 242.

Oxidation of Olefinic Alcohol (20a). To a solution of **20a** (2.4 mg, 0.0086 mmol) in CH_2Cl_2 (1 ml), pyridinium dichromate (13 mg) was added. The reaction mixture was stirred at room temperature overnight. Diethyl ether (5 ml) was added, and then the reaction mixture was filtrated to remove the solid and concentrated under reduced pressure. The residue was purified by a short silica-gel column (hexane–ethyl acetate 17:3) to yield the olefinic diketone **21** (2 mg, 80% theoretical). **21**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.69 (s, 3H), 0.83 (d, 3H, J=6 Hz), 1.24 (s, 6H), 2.18 (s, 3H), 5.26 (m, 1H); EIMS m/z 276 (M^+), 205, 206, 163, 187, 93, 119, 121, 203, 191, 107; CD $[\theta]_{293}$ −3480.

Acid Catalyzed Rearrangement of Agelasine-B (5). **5** (10 mg, 0.022 mmol) was treated with a solution of acetic acid (0.4 ml) and 12 M HCl (0.1 ml) at room temperature for 3 h. The reaction mixture was dried in vacuo and the residue was separated by HPLC on a C_8 column (Develosil C_8-5 packed column, 10×250 mm) using methanol–water (8:2) containing 0.2 M sodium chloride as a mobile phase. The main fraction was concentrated under reduced pressure and the resultant water layer was extracted with ethyl acetate. Concentration of the extract gave **17b** (7 mg, 70% theoretical). **17b**, amorphous solid, was identical with **17a** in all respects except $[\alpha]_D^{25}$ +17.4° (c 0.35, CH_3OH).

Acid Catalyzed Rearrangement of Agelasine-C (6). **6** (12 mg, 0.026 mmol) was treated with a solution of acetic acid (0.48 ml) and 12 M HCl (0.32 ml) at room temperature for 3 h and worked up as above to yield **17a** (8.8 mg, 73% theoretical), which was identical with that obtained from agelasine-A, and showed $[\alpha]_D$ −15.9° (c 0.44, CH_3OH).

Ozonolysis of Agelasine-C (6). Ozone was bubbled in the solution of **6** (70 mg, 0.15 mmol) in methanol (50 ml) at −78° C for 1 h. The excess ozone was removed by N_2 flow. Dimethyl sulfide (1 ml) was added, then the solution was allowed to warm up to 0° C and then room temperature (in 30 min each). The reaction mixture was dried under reduced pressure and the residue was submitted to silica-gel column chromatography (hexane–ethyl acetate 95:5) to furnish two epoxides **25** and **26** (9 mg, 21% and 13 mg, 31% theoretical, respectively). **25**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.68 (s, 3H), 0.79 (s, 3H), 0.93 (d, 3H, J=7.2 Hz), 1.06 (s, 3H), 2.15 (s, 3H), 3.04 (brd, 1H, J=4 Hz), 1.0—2.7 (m, 14H); EIMS m/z 278 (M^+), 207, 263, 189, 137, 119; $[\alpha]_D^{25}$ −36.7° (c 0.60, CH_3OH). **26**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.64 (s, 3H), 0.86 (s, 6H), 1.03 (d, 3H, J=7.2 Hz), 2.16 (s, 3H), 2.91 (brd, 1H, J=4 Hz), 1.0—2.6 (m, 14H); EIMS m/z 278 (M^+), 207, 163, 137, 83, 55, 67, 95, 105, 119, 121; $[\alpha]_D^{25}$ +6.5° (c 1.85, CH_3OH).

Reaction of 26 with Boron Trifluoride Etherate. A solution of **26** (3.5 mg, 0.013 mmol) in anhydrous benzene (1.5 ml) was treated with boron trifluoride etherate (0.1 ml) for 10 min at room temperature. The solution was diluted with ethyl acetate (5 ml) and extracted with aqueous sodium hydrogencarbonate (10%, 10 ml), then washed with brine (10 ml), and dried under anhydrous sodium sulfate. Evaporation left a residue, which was submitted to silica-gel column chromatography (hexane–ethyl acetate 85:15) to yield the diketone **27** (1.5 mg, 43% theoretical). **27**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.97 (s, 3H), 1.03 (d, 3H, J=6.9 Hz), 1.13 (s, 3H), 1.23 (s, 3H), 2.12 (ddd, 1H, J=2.3, 4.4, 13.5 Hz), 2.17 (s, 3H), 2.26 (m, 2H), 2.42 (dt, 1H, J=6.3, 13.5 Hz), 2.48 (d, 1H, J=5 Hz); EIMS m/z 278 (M^+), 207, 125, 151, 208, 95, 109, 152, 119, 161, 189; CD $[\theta]_{298}$ −2370.

Ozonolysis of Agelasine-D (7). A solution of **7** (5 mg, 0.011 mmol) in methanol (0.5 ml) was treated with ozone at −78° C for 30 min. After removing excess ozone, dimethyl sulfide (0.03 ml) was added, then the solution was kept at 0° C for 30 min and at room temperature for 3 h. Evaporation of the solvent left a residue, which was submitted to silica-gel column chromatography (hexane–ethyl acetate 85:15) to afford the diketone **28** (1.5 mg, 52% theoretical). An additional amount (ca. 18 mg) was obtained from the ozonolysis of the mixture of agelasines. **28**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.72 (s, 3H), 0.84 (s, 3H), 0.95 (s, 3H), 2.08 (s, 3H), 1.0—2.8 (m, 16H); ^{13}C NMR ($CDCl_3$, 22.5 MHz) δ=14.6 (q), 16.3 (q), 19.0 (t), 21.7 (q), 24.0 (t), 29.9 (q), 33.6 (q), 33.7 (t), 39.2 (t), 42.0 (t), 42.6 (t), 42.7 (s), 42.8 (t), 54.2 (d), 63.2 (d), 208.9 (s), 211.9 (s); IR ($CHCl_3$) 1710 cm^{-1}; EIMS m/z 264 (M^+), 249, 81, 95, 67, 55, 121, 139, 231; $[\alpha]_D^{25}$ −28.5° (c 1.5, CH_3OH).

Conversion of 28 to the Enone (29). Concentrated sulfuric acid (0.13 ml) was added dropwise to the solution of **28** (16 mg, 0.061 mmol) in methanol (2.4 ml). The solution was heated to boiling, then cooled, diluted with water (15 ml) and extracted with diethyl ether (2×15 ml). Evaporation of the solvent left a residue, which was submitted to silica-gel column chromatography (hexane–ethyl acetate 9:1) to give the enone **29** (12.2 mg, 76.2% theoretical): Colorless crystals; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.80 (s, 3H), 0.87 (s, 3H), 0.92 (s, 3H), 5.88 (brs, 1H); ^{13}C NMR ($CDCl_3$, 22.5 MHz) δ=15.1 (q), 18.6 (t), 20.4 (t), 21.9 (q), 21.9 (t), 33.3 (s), 33.5 (q), 36.7 (t), 38.9 (s), 39.2 (t), 41.7 (t), 51.5 (d), 53.9 (d), 125.7 (d), 165.7 (s), 199.4 (s); IR ($CHCl_3$) 1680 cm^{-1}; EIMS m/z 246 (M^+), 110, 137, 123, 231; $[\alpha]_D^{25}$ +36.0° (c 0.5, CH_3OH); UV (CH_3OH) 242 (ε 20050), 310 nm (360); CD $[\theta]_{242}$ +38460, $[\theta]_{317}$ −8720.

Ozonolysis of Agelasine-E (8). A solution of **8** (26 mg, 0.057 mmol) in methanol (3 ml) was saturated with ozone at −78 °C for 30 min. After removing the excess ozone dimethyl sulfide (0.075 ml) was added, then the solution was allowed to warm up to 0° C (30 min) and room temperature overnight. The reaction mixture was dried under reduced pressure and the residue was submitted to silica-gel column chromatography (hexane–ethyl acetate 85:15) to yield the diketone **30** (10 mg, 90% theoretical) and the compound **31** (5.1 mg). **30**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 270 MHz) δ=0.79 (s, 3H), 1.09 (s, 3H), 2.12 (s, 3H), 1.5—2.6 (m, 11H); ^{13}C NMR ($CDCl_3$, 22.5 MHz) δ=18.3 (t), 21.8 (q), 23.1 (t), 29.5 (q), 39.5 (t), 39.7 (s), 41.4 (t), 42.6 (t), 60.1 (d), 208.6 (s), 213.0 (s); IR ($CHCl_3$) 1710 cm^{-1}: EIMS m/z 196 (M^+), 181, 111, 163, 139, 121, 95; CD $[\theta]_{292}$ −5174, $\Delta\varepsilon_{292}$ −1.57.

Reduction and Acetylation of 31. **31** (5 mg) was reduced with sodium borohydride (10 mg) in methanol (1.5 ml), workup in usual way to give a crude product, which was purified by silica-gel column chromatography (chloroform–methanol 9:1) to yield 1,4-pentanediol (4.5 mg), which was acetylated with acetic anhydride (0.1 ml) and pyridine

(0.1 ml) to give 1,4-diacetoxypentane.

Ozonolysis of Agelasine-F (9) with Sodium Borohydride Reduction. A solution of **9** (50 mg, 0.11 mmol) in methanol (3 ml) was treated with ozone at −78° C for 15 min. After removing the excess ozone, sodium borohydride (105 mg, in three portions) was added. The reaction mixture was stirred and allowed to warm up to 0° C and room temperature (in 30 min each), then was neutralized with acetic acid. Removing solvent left a residue, which was submitted to silica-gel column chromatography (chloroform–methanol 95:5) to furnish the epoxide **32** (13.8 mg, 54% theoretical) and 1,4-pentanediol (3 mg). **32**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.77 (d, 3H, J=6 Hz), 0.88 (s, 3H), 1.20 (d, 3H, J=5 Hz), 1.24 (s, 3H), 2.98 (brs, 1H), 3.76 (brq, 1H, J=7 Hz); ^{13}C NMR ($CDCl_3$, 22.5 MHz) δ=15.9 (q), 18.1 (q), 20.8 (q), 23.5 (q), 24.2 (t), 25.9 (t), 33.3 (d), 33.6 (t), 34.4 (t), 38.4 (s), 62.5 (d), 62.8 (s), 68.8 (d); EIMS m/z 212 (M^+), 197, 126, 109, 153, 167, 179.

Reduction of the Epoxide 32. A solution of **32** (13 mg, 0.061 mmol) in anhydrous THF (1 ml) was treated with lithium triethylhydroborate (1 M, 0.5 ml) under nitrogen atmosphere and the reaction mixture was stirred at room temperature overnight. The solvent was evaporated and the residue was dissolved in a solution of chloroform and methanol (15 ml, 95:5). The solution was passed through a short silica-gel column to give the pure diol **33** (11 mg, 84% theoretical): Amorphous solid; mp 121.5—123 °C; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.88 (d, 3H, J=6 Hz), 0.90 (s, 3H), 1.22 (d, 3H, J=6.5 Hz), 1.26 (s, 3H), 3.68 (m, 1H); EIMS m/z 214 (M^+), 196, 181, 178, 170, 163, 138, 96, 149.

Acetylation of 33. A mixture of **33** (8 mg, 0.037 mmol), acetic anhydride (0.1 ml), triethylamine (0.1 ml) and 4-dimethylaminopyridine (1 mg) was stirred at room temperature for 1 h. The product was purified by silica-gel column chromatography (ethyl acetate–hexane 15:85) to yield the monoacetate **34** (8.5 mg, 88% theoretical): A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.85 (d, 3H, J=6 Hz), 0.88 (s, 3H), 1.21 (d, 3H, J=6.5 Hz), 1.25 (s, 3H), 2.02 (s, 3H), 4.78 (hexalet, 1H, J=6 Hz); EIMS m/z 256 (M^+), 239, 196, 181, 178, 163, 154, 149, 136, 138.

Dehydration of 34. A solution of **34** (8 mg, 0.031 mmol) in pyridine (1 ml) was treated with thionyl chloride (0.1 ml) at −5° C for 15 min. The reaction mixture was diluted with diethyl ether (5 ml), quenched with water (5 ml) and then separated. The aqueous layer was extracted with diethyl ether (5 ml). The combined ether layer was dried over anhydrous sodium sulfate and concentrated to give a residue, which was chromatographed on a silica-gel column (hexane–ethyl acetate 85:15) to give a mixture of the *exo*-olefin 35 and its *endo*-isomer with a ratio 2:1 (total 7.5 mg): A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.84 (d, 3H, J=6 Hz), 0.85 (s, 3H×1/3), 0.91 (s, 3H×2/3), 1.20 (d, 3H, J=6.5 Hz), 2.01 (s, 3H), 4.54 (d, 1H×2/3, J=2 Hz), 4.78 (brd, 1H×2/3, J=2 Hz), 4.84 (hexalet, 1H, J=6 Hz), 5.40 (brs, 1H×1/3); EIMS m/z 239 (M^++1), 123, 178, 149, 136.

Deacetylation of 35. A solution of **35** and its isomer (7 mg, 0.029 mmol) in methanol (1 ml) was treated with 1 M sodium methoxide (0.5 ml in three portion) under nitrogen atmosphere, then silica-gel column chromatography of the product gave a mixture of **36** and its *endo*-olefin isomer (5 mg): A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=3.73 (hexalet, 1H, J=6 Hz), 4.54 (d, 1H×2/3, J=2 Hz), 4.78 (brs, 1H×2/3), 5.40 (brs, 1H×1/3); EIMS m/z 196 (M^+), 123, 178, 163, 121.

Oxidation of 36. To a solution of **36** and its *endo*-olefin isomer (5 mg, 0.026 mmol) in anhydrous dichloromethane (1 ml) pyridinium dichromate (10 mg) was added and the reaction mixture was stirred under nitrogen atmosphere for 6 h, then it was diluted with diethyl ether (5 ml) and filtrated to remove the solid. The filtrate was passed through a short silica-gel column and concentrated to yield the olefinic ketone **37** and its *endo*-olefin isomer (3 mg): A syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.84 (d, 3H, J=6 Hz), 0.88 (s, 3H×1/3), 0.90 (s, 3H×2/3), 2.12 (s, 3H), 4.54 (d, 1H×2/3, J=2 Hz), 4.79 (brs, 1H×2/3), 5.40 (brs, 1H×1/3); EIMS m/z 194 (M^+), 123, 176, 161.

Conversion of 37 to the Octalone 38. A solution of **37** and its isomer (3 mg, 0.015 mmol) in methanol (1 ml) was saturated with ozone at −78° C for 5 min. After removing the excess ozone, dimethyl sulfide (0.007 ml) was added, then the reaction mixture was allowed to warm up to 0° C and room temperature (in 30 min each). After removing the solvent and excess reagent under reduced pressure, 5% KOH aqueous solution (0.1 ml) and methanol (1 ml) were added. The solution was refluxed for 3 h under argon atmosphere, then cooled to room temperature. Evaporation left a residue, which was chromatographed on a silica-gel column (hexane–ethyl acetate 85:15) to yield the octalone **38** (1 mg), which was further purified by TLC (hexane–ethyl acetate 85:15). ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.91 (brs, 3H, J=6Hz), 1.11 (s, 3H), 5.74 (brs, 1H); EIMS m/z 178 (M^+), 136, 163, 121, 107; CD $[\theta]_{335}$ −390, $[\theta]_{236}$ +5800.

Ozonolysis of Agelasine-F (9) with Dimethyl Sulfide Reduction. Ozone was bubbled in a solution of **9** (59 mg, 0.11 mmol) in methanol (3 ml) at −78° C for 30 min. After removing the excess ozone, dimethyl sulfide (0.5 ml) was added, then the solution was allowed to warm up to 0° C and room temperature (in 30 min each) and stirred overnight. Usual workup and HPLC separation (Develosil 60-3, 10× 250 mm, benzene–acetone 90:10) furnished the enol ether **39** (7.6 mg) and the aldehyde **40**. **39**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.80 (d, 3H, J=6.5 Hz), 0.93 (s, 3H), 1.24 (s, 3H), 1.67 (d, 3H, J=1.5 Hz), 3.68 (brs, 1H), 4.32 (m, 1H); ^{13}C NMR ($CDCl_3$, 22.5 MHz) δ=16.2 (q), 18.0 (q), 18.8 (q), 20.0 (q), 25.2 (t), 29.0 (t), 30.3 (t), 32.1 (d), 35.7 (s), 74.8 (d), 79.6 (s), 92.8 (d), 146.9 (s); EIMS m/z 210 (M^+), 149, 125, 123, 109. **40**: A colorless syrup; ^{1}H NMR ($CDCl_3$, 90 MHz) δ=0.85 (d, 3H, J=7 Hz), 1.03 (s, 3H), 2.11 (s, 6H), 9.75 (t, 1H, J=2 Hz); EIMS m/z 226 (M^+), 107, 123, 121, 142, 147, 165, 183; CD $[\theta]_{296}$ +130.

***p*-Bromobenzoylation of 39.** A mixutre of **39** (5 mg, 0.024 mmol), triethylamine, *p*-bromobenzoylchloride (20 mg) and anhydrous benzene (0.5 ml) was stirred at room temperature overnight. Usual workup and silica-gel column chromatography (hexane–ethyl acetate 96:4) yield the ester **41** (2.5 mg), a colorless syrup; ^{1}H NMR ($CDCl_3$, 400 MHz) δ=0.83 (d, 3H, J=6.6 Hz), 1.05 (s, 3H), 1.18 (s, 3H), 1.35 (dm, 1H, J= 13 Hz), 1.53 (dq, 1H, J=13, 4 Hz), 1.70 (s, 3H), 1.73 (dm, 1H, J=13 Hz), 1.78 (dm, 1H, J=17 Hz), 1.82 (m, 1H), 1.96 (dd, 1H, J=17, 5 Hz), 2.13 (tt, 1H, J=13 and 3 Hz), 4.19 (brd, 1H, J=5 Hz), 5.06 (brt, 1H, J=3 Hz).

Treatment of 9 with Sodium Carbonate. A solution of **9** (5 mg, 0.0011 mmol) in water (2 ml) was treated with 2 M sodium carbonate aqueous solution (0.01 ml) for 2 h, and then it was extracted with ethyl acetate. Concentration of the extracts gave the formamide **11** (5 mg, 100% yield): Amor-

phous solid; UV (CH_3OH) 260 (ε 4360), 270 (pH 2, 9160), 260 nm (pH 11, 8930); ^1HNMR ($CDCl_3$, 90 MHz) δ=2.97 (d, J=5 Hz), 4.14 (d, 3H, J=7 Hz), 4.80 (brs, 2H), 7.96 (s, 1H), 8.14 (s, 1H); EIMS m/z 439 (M^+), 396.

Hydrolysis of 9 with KOH. A mixture of **9** (10 mg, 0.0022 mmol), methanol (2 ml) and 40% KOH aqueous solution (1 ml) was heated in a sealed tube at 115° C for 3 h. The reaction mixture was dried under reduced pressure to give a residue, which was submitted to TLC separation (chloroform–methanol 95:5) to yield the less polar product **12** (0.5 mg, 6% yield) and the polar product **13** (4 mg). **12**: Amorphous solid; UV (CH_3OH) 272 nm (ε 8350); ^1HNMR ($CDCl_3$, 90 MHz) δ=2.99 (d, 3H, J=5 Hz), 3.40 (d, 2H, J=7.5 Hz), 7.99 (s, 1H); EIMS m/z 411 (M^+), 396, 139. **13**: UV (CH_3OH) 272 nm (ε 8520), 277 (8360), 282 (pH 2); ^1HNMR ($CDCl_3$, 90 MHz) δ=3.10 (d, 3H, J=5 Hz), 4.87 (d, 2H, J=7 Hz), 7.85 (s, 1H), 8.55 (s, 1H); EIMS m/z 421 (M^+), 406, 298, 149.

Preparation of 6-*N*-Methyl-7-prenyladenine (14), 6-*N*-Methyl-9-prenyladenine (15), and 7,9-Dimethyladeninium Perchlorate (16). Compounds **14**, **15**, and **16** were prepared by the methods described in literatures.[9] **14**: Amorphous solid; mp 157—158° C; UV (CH_3OH) 272 (sh, ε 12300), 277 nm (12600); IR (KBr) 1670, 1615, 1580, 1480, 1460, 1390, 1350 cm^{-1}; ^1HNMR ($CDCl_3$) δ=1.84 (s, 3H), 1.85 (s, 3H), 3.08 (d, 3H, J=4.8 Hz, NMe), 4.85 (d, 2H, J=6.2 Hz, NCH_2), 5.30 (brs, 1H, NH), 5.40 (t, 1H, J=6.2 Hz, =CH), 7.80 (s, 1H, 8-H), 8.48 (s, 1H, 2-H); ^{13}CNMR ($CDCl_3$) δ=18.2 (q), 25.6 (q), 28.1 (q, NMe), 45.8 (t, NCH_2), 112.2 (s, C-5), 119.0 (d, =CH), 139.6 (s, =C), 143.6 (d, C-8), 151.6 (s, C-6), 153.0 (d, C-2), 159.6 (s, C-4); EIMS m/z 217 (M^+). **15**: Crystals; mp 110—111° C; UV (CH_3OH) 267 nm (ε 16300); IR (KBr) 3280, 2920, 1615, 1580, 1400, 1330, 1305, 1225 cm^{-1}; ^1HNMR ($CDCl_3$) δ=1.80 (s, 6H), 3.19 (d, 3H, J=4.9 Hz, NMe), 4.73 (d, 2H, J=6.0 Hz, NCH_2), 5.42 (t, 1H, J=6.0 Hz, =CH), 6.12 (brs, 1H, NH), 7.69 (s, 8-H), 8.42 (s, 2-H); ^{13}CNMR ($CDCl_3$) δ=17.7 (q), 25.3 (q), 27.4 (q, NMe), 40.2 (t, NCH_2), 117.8 (d, =CH), 119.6 (s, C-5), 138.3 (s, =C), 138.7 (d, C-8), 148.7 (s, C-6), 152.8 (d, C-2), 155.3 (s, C-4); EIMS m/z 217 (M^+). **16**: Rods; mp 293—294° C; UV (CH_3OH) 212 (ε 12900), 272 nm (8080); IR (KBr) 3410, 3100, 1675, 1620, 1140, 1110, 1080, 620 cm^{-1}; ^1HNMR (DMSO-d_6) δ=3.86 (s, 3-H, 9-NMe), 4.17 (s, 3H, 7-NMe), 7.93 (brs, 2H, NH_2), 8.40 (s, 1H, 2-H), 9.59 (s, 1H, 8-H); EIMS m/z 163 (M^+-HClO_4).

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