New Lanostanoids, Elfvingic Acids A−H, from the Fruit Body of *Elfvingia applanata*

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Eight new lanostanoids, 12α,15*β*-dihydroxy-3,7,11,23-tetraoxolanost-8,(20*Z*)(22)-dien-26-oic acid (**1**), 7*β*,8*β*-epoxy-15*β*,20*S*-dihydroxy-3,12,23-trioxolanost-9(11),16-dien-26-oic acid (**2**), 7*β*,8*β*-epoxy-3*β*,15*β*,20*S*-trihydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid (**3**), 7*β*,8*β*-epoxy-2α,3*β*,15*β*,20*S*-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid (**4**), 7*β*,8*β*-epoxy-3*β*,15*β*,20*S*,28-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid (**5**), 7*β*,8*β*-epoxy-3*β*,15*β*,19,20*S*-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid (**6**), 8*β*,15*β*,20*S*-trihydroxy-3,7,12,23-tetraoxolanost-9(11),16-dien-26-oic acid (**7**), 7α,8α-epoxy-15*β*,23*ξ*-dihydroxy-12-oxo-3,4-secolanost-4(28),9(11),(20*Z*)(22)-trien-3,26-dioic acid (**8**), and the methyl ester of **8** (**8a**) were isolated from the fruit bodies of *Elfvingia applanata*. Their structures were established primarily by NMR experiments, and their biological activity against Kato III and Ehlrich cells was investigated.

In the course of our research program aimed at the discovery of biologically active compounds from fungi, we have initiated the chemical study of *Elfvingia applanata* (Pers.) Pat. (Ganodermataceae), which grows on the trees in broad-leaved forests and is distributed throughout Japan. *E. applanata* has been used in traditional medicine as a treatment for cancer.[1,2] An earlier chemical constituent study of this fungus resulted in the isolation of lanostane triterpenes.[3,4] Eight new lanostenoid triterpenes, elfvingic acids A (**1**), B (**2**), C (**3**), D (**4**), E (**5**), F (**6**), G (**7**), and H (**8**), and the methyl ester of **8** (**8a**) (Chart 1) were isolated from the 70% EtOH extract of the fruit bodies of *E. applanata*. We describe here the isolation and structure elucidation of **1**−**8** and **8a**, primarily by extensive NMR, and their biological activity against Kato III and Ehlrich cells.

Results and Discussion

Elfvingic acid A (**1**) gave a $[M + Na]^+$ peak at *m/z* 551.2634 (HRFABMS). This corresponds to a molecular formula $C_{30}H_{40}O_8$, requiring 11 unsaturation equivalents. The IR spectrum of **1** showed absorption at 3400 (OH), 1705, 1685, and 1670 cm^{-1} (C=O). Absorption at 249.5 nm in the UV spectrum suggested the presence of a conjugated carbonyl system in **1**. The ^1H NMR spectrum of **1** exhibited six singlet methyls at *δ* 1.08, 1.10, 1.29, 1.32, 1.89, and 2.55, one doublet methyl at *δ* 1.34, two oxymethines at *δ* 4.22 and 4.90, and one olefinic proton at *δ* 6.65. The 30 carbon signals observed in the ^{13}C NMR spectrum were sorted, by DEPT experiment, into seven methyl carbons; five methylene carbons; five methine carbons, two of which had oxygen substituents (*δ* 79.8 and 76.8); four sp^3 quaternary carbons; four sp^2 carbons [*δ* 157.5 (s), 152.0 (s), 149.8 (s), and 125.3 (d)]; and five carbonyl carbons (*δ* 215.2, 203.8, 202.7, 198.9, and 178.5) (Table 1). These data suggest that **1** is a tetracyclic triterpene. The structure of **1** was deduced from detailed analysis of ^1H and ^{13}C NMR data aided with 2D NMR including COSY, HMQC, HMBC, and ROESY experiments. The COSY spectrum revealed connectivities of C-1 to C-2, C-5 to C-6, C-15 to C-17, and C-24 to C-27. HMBC correlations from the protons of the seven methyl groups completed the definition of all of the functional groups in the lanostane framework. That is, long-range correlations of Me-18 (*δ* 1.29) to C-12, C-13, C-14, and C-17; of Me-19 (*δ* 1.32) to C-1, C-5, C-9, and C-10; of Me-21 (*δ* 2.55) to C-17, C-20, and C-22; of Me-27 (*δ* 1.34) to C-24, C-25, and C-26; of Me-28 (*δ* 1.08) and Me-29 (*δ* 1.10) to C-3, C-4, and C-5; and of Me-30 (*δ* 1.89) to C-8, C-13, C-14, and C-15 revealed two double bonds at the C-8 and C-20 positions, in which two secondary hydroxy groups were attached to C-12 and C-15, and two of five carbonyl groups were attached at C-3 and C-26, respectively. The remaining three carbonyl functions were determined to be at C-7, C-11, and C-23 by HMBC correlations of H_2-6 (*δ* 2.74, and 2.53) to C-7; of H-12 (*δ* 4.22) to C-9 and C-11; and of H-22 (*δ* 6.65) and H_2-24 (*δ* 3.17, and 2.55) to C-23. The relative stereochemistry of **1** was confirmed by ROESY analysis described as follows, except for C-25. Namely, significant ROESY correlations of H-12 to Me-18 and of Me-30 to H-15 [4.90 (br d, J = 6.0 Hz)] and to H-17 [*δ* 3.70 (dd, J = 9.6. 9.3 Hz)] indicated α-orientations of 12-OH and H-17 and *β*-orientation of 15-OH. The ROE between Me-21 and H-22 indicated the *Z* configuration of the double bond at C-20 and C-22 (Table 2). Therefore, **1** was formulated as 12α,15*β*-dihydroxy-3,7,11,23-tetraoxolanosta-8,(20*Z*)(22)-dien-26-oic acid.

Elfvingic acid B (**2**) gave a $[M + Na]^+$ peak at *m/z* 551.2625 in its HRFABMS, corresponding to a molecular formula $C_{30}H_{40}O_8$ and requiring 11 unsaturation equivalents. The IR spectrum of **2** indicated hydroxy (3400 cm^{-1}) and carbonyl (1710 and 1665 cm^{-1}) functions. The ^{13}C NMR spectrum of **2** showed three carbonyls (*δ* 213.2, 208.2, and 204.0), one acid carboxyl (*δ* 178.5), two trisubstituted double bonds (*δ* 163.5, 158.8, 127.3, and 126.2), and two hydroxy carbons [*δ* 79.6 (d) and 72.3 (s)]. The presence of a trisubstituted epoxy group was recognized by the characteristic signals at *δ* 63.2 (s) and δ_C 57.6 (d); δ_H 3.97 (d, J = 5.2 Hz) in the ^{13}C NMR and the HMQC spectra. The

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Chart 1

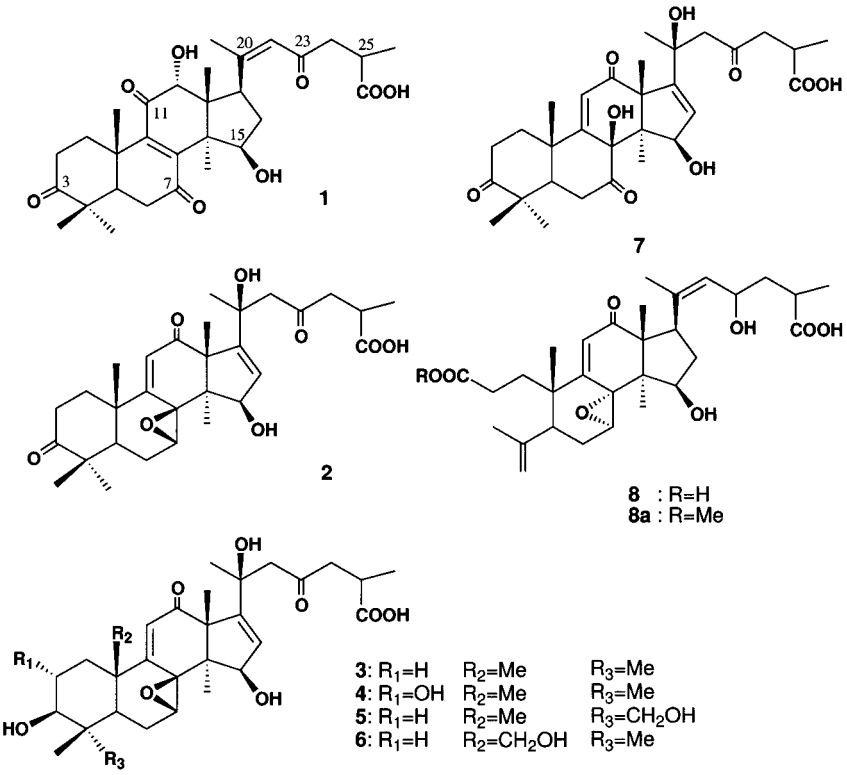

Table 1. ^{13}C NMR Spectral Data (δ) for **1**−**8** and **8a** (in pyridine-d_5, 125 MHz)

	1	2	3	4	5	6	7	8	8a
1	35.6	37.3	37.3	46.1	37.2	31.7	37.1	37.9	37.4
2	34.6	34.2	28.2	68.1	27.9	28.2	34.7	30.1	30.1
3	215.2	213.2	77.4	82.8	71.3	77.5	212.8	176.1	173.9
4	47.2	47.8	40.3	40.4	44.3	40.3	48.0	145.6	145.5
5	50.5	49.8	49.2	49.2	41.4	49.8	48.1	44.3	44.2
6	37.9	21.9	21.8	21.9	21.5	21.5	36.2	27.9	27.8
7	202.7	57.6	58.5	58.3	58.5	58.6	205.5	63.7	63.9
8	152.0	63.2	63.4	63.4	63.5	64.1	81.6	67.0	67.0
9	149.8	163.5	165.4	164.7	165.6	160.4	164.0	164.1	163.6
10	39.7	38.2	38.9	40.1	38.7	45.4	40.4	44.4	44.2
11	203.8	126.2	125.9	125.9	125.8	130.1	124.9	130.3	130.4
12	79.8	204.0	204.2	204.1	204.3	204.0	205.0	203.6	203.0
13	54.1	63.6	63.9	63.9	63.9	64.0	66.9	60.3	60.3
14	51.0	47.8	48.1	48.1	48.0	48.1	49.0	53.9	53.8
15	76.8	79.6	79.9	79.8	79.8	80.0	81.7	76.9	76.5
16	36.5	127.3	127.3	127.2	127.2	127.0	126.3	40.3	40.3
17	48.1	158.8	159.0	158.9	158.9	159.0	159.6	47.1	47.1
18	18.8	27.8	28.3	28.3	28.3	28.2	30.9	19.8	19.8
19	18.8	20.6	21.8	23.2	22.6	60.0	20.0	24.1	24.1
20	157.5	72.3	72.5	72.5	72.4	72.4	72.7	141.8	141.7
21	21.2	29.7	30.0	29.9	29.9	29.9	30.0	19.8	19.8
22	125.3	54.8	55.1	55.1	55.0	55.1	55.2	127.0	127.0
23	198.9	208.2	208.1	208.9	208.1	208.0	207.9	75.9	76.0
24	48.7	48.9	49.1	49.1	49.1	49.1	49.2	37.4	37.2
25	36.2	35.6	35.9	35.9	35.9	35.8	36.0	35.0	35.0
26	178.5	178.5	178.5	178.5	178.4	178.4	178.5	179.9	179.9
27	18.2	17.7	17.7	18.1	18.1	18.0	18.1	16.1	16.1
28	27.6	24.7	28.6	29.1	65.3	28.7	25.0	23.8	23.6
29	20.6	21.9	16.2	17.3	13.0	17.0	22.0	115.5	115.6
30	27.8	25.3	25.6	25.6	25.5	25.7	26.1	21.4	21.4
OMe									51.7

features of the ^{13}C NMR spectrum for **2** were similar to those obtained for applanoxidic acid G;[4] however, compound **2** showed significant differences for the chemical shifts of carbons at C-5−C-10, C-13, and C-14, suggesting the presence of the 7β,8β-epoxy ring in **2** (see Experimental Section). The stereochemistry of **2** was confirmed on the basis of the ROESY experiment, except for C-25. First, the configuration of H-15 [δ 4.49 (d, J = 3.0 Hz)] was assigned as α on the basis of the ROESY correlation between H-15 and Me-30 (δ 1.09). Second, significant ROEs were observed connecting Me-30 to H-7 (δ 3.97) and H-7 to H-15, supporting the assignments of 7β,8β-epoxy function. From the analysis of all of these data and from biogenetic considerations, the structure of **2** was formulated as 7β,8β-epoxy-15β,20*S*-dihydroxy-3,12,23-trioxolanost-9(11),16-dien-26-oic acid.

Table 2. Key ROESY Correlations for Compounds **1**−**8** (in pyridine-d_5, 600 MHz)

H	1	2	3	4	5	6	7	8
1		H-11	H-11	H-11	H-11	H-11	H-11	H-5
2β		H_3-19, H_3-29		H_3-19, H_3-29			H_3-19, H_3-29	
3			H-5, H_3-28	H-5, H_3-28	H-5, H_2-28	H-5, H_3-28		
5		H_3-28	H-3, H_3-28	H-3, H_3-28	H-3, H_2-28	H-3, H_3-28	H_3-28	H_2-1
7		H-15, H_3-30	H-15, H_3-30	H-15, H_3-30	H-15, H_3-30	H-15, H_3-30		H-15, H_3-18
8							H_3-18	
11		H_2-1	H_2-1	H_2-1	H_2-1	H_2-1	H_2-1	H_3-19
12	H_3-18, H_3-19, H_3-21							
15	H_2-16, H_3-30	H-7, H_3-30	H-7, H_3-30	H-7, H_3-30	H-7, H_3-30	H-7, H_3-30	H_3-30	H-7, H_3-30
16	H-15	H_3-21, H_2-22	H_3-21, H_2-22	H_3-21, H_2-22	H_3-21, H_2-22	H_3-21, H_2-22	H_3-21, H_2-22	
17	H-12, H_3-21, H_3-30							H_3-30
18	H-12						H-8	H-7, H_3-21
19	H-12, H_3-29	H-2β, H_3-29	H_3-29	H-2β, H_3-29	H_3-29	H_3-29	H-2β, H_3-29	H-11, H_3-29
21	H-12, H-22	H_2-16	H_2-16	H_2-16	H_2-16	H_2-16	H_2-16, H_3-30	H_3-18, H-22
22	H_3-21	H_2-16	H_2-16	H_2-16	H_2-16	H_2-16	H_2-16	H_3-21
28		H-5	H-3, H-5	H-3, H-5	H-3, H-5	H-3, H-5	H-5	
29	H_3-19	H-2β, H_3-19	H_3-19	H-2β, H_3-19	H_3-19	H_2-19	H-2β, H_3-19	H_3-19
30	H-15, H-17	H-7, H-15	H-7, H-15	H-7, H-15	H-7, H-15	H-7, H-15	H-15, H_3-21	H-15, H-17

Elfvingic acid C (**3**) has the molecular formula $C_{30}H_{42}O_8$ by the ^{13}C NMR data of **3** and HRFABMS, which differs from **2** by containing two additional hydrogen atoms. Comparison of NMR data of **3** with those of **2** revealed that **3** has a hydroxy at C-3 [δ 77.4 (d)] instead of a carbonyl function in **2**. Indeed, H-3 was observed at δ 3.41 as the double doublet shape (J = 12.3, 4.2 Hz), suggesting β-orientation of 3-OH. The connectivity of the HMBC and ROESY experiments and biogenetic considerations supported the assumed structure of **3** as 7β,8β-epoxy-3β,15β,20*S*-trihydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid.

Elfvingic acid D (**4**) has the molecular formula $C_{30}H_{42}O_9$ by the ^{13}C NMR data of **4** and HRFABMS, which differs from **3** by containing an additional oxygen atom. Comparison of NMR data of **4** with those of **3** revealed that **4** has a hydroxy at C-2 [δ 68.1 (d)] instead of a methylene function in **3**. Indeed, H-3 was observed at δ 3.36 as the doublet shape (J = 9.6 Hz), suggesting α-orientation of 2-OH and β-orientation of 3-OH, respectively. The connectivities of the HMBC and ROESY experiments and biogenetic considerations supported the assumed structure of **4** as 7β,8β-epoxy-2α,3β,15β,20*S*-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid.

The molecular formula $C_{30}H_{42}O_9$ of elfvingic acid E (**5**) was deduced from HRFABMS. The ^1H NMR spectrum of **5** exhibited the presences of five singlet methyls at δ 1.01, 1.04, 1.32, 1.72, and 2.13, one doublet methyl at δ 1.38, and the lack of one singlet methyl as compared with **5** in the same lanostane framework. Indeed, in the NMR and HMQC spectra, the signals due to a hydroxymethyl group appeared at δ_H 3.61 and δ_H 4.20 (each 1H, d, J = 11.2 Hz)]; δ_C 65.3 (t), in addition to the six methyl carbons. By comparison of NMR data of **5** with those of **3**, only the C-3 and C-5 carbons around the C-4 position in **5** were shifted, suggesting the CH_2OH to be at the C-28 or C-29 position. The key ROEs between H-3 [δ 4.18 (1H, dd, J = 11.0, 4.5 Hz)] and these protons (δ 4.20 and 3.61) confirmed a CH_2OH group at the C-28 position. The connectivities of the HMBC and ROESY experiments and biogenetic considerations supported the assumed structure of **5** as 7β,8β-epoxy-3β,15β,20*S*,28-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid.

The ^1H NMR spectrum of elfvingic acid F (**6**) exhibited the same features as **5**, indicating the presence of a CH_2-OH group. Nevertheless, the chemical shifts due to the CH_2OH function were slightly different, with C-28 at δ 60.0 (t) and protons at δ 4.40 and 4.19 (each d, J = 10.2 Hz), along with six methyls in the ^1H NMR spectrum. On comparison of ^{13}C NMR data with those of **3**, the C-1, C-9, and C-10 positions were shifted, suggesting the CH_2OH to be at the C-19 position. The NOE between H_2-19 and Me-29 (δ 1.07) confirmed this position in a similar manner as **5**. The connectivities of the HMBC and ROESY experiments and biogenetic considerations supported the assumed structure of **6** as 7β,8β-epoxy-3β,15β,19,20*S*-tetrahydroxy-12,23-dioxolanost-9(11),16-dien-26-oic acid.

Elfvingic acid G (**7**) gave a $[M + Na]^+$ peak at *m*/*z* 567.2571 in its HRFABMS ($C_{30}H_{40}O_9$). In the ^1H NMR spectrum of **7**, the characteristic epoxy proton due to H-7 [δ 3.97 (d)] observed in **2** was absent. On the other hand, one carbonyl (δ 205.5) and one *tert*-carbinol (δ 81.6) signal appeared in the ^{13}C NMR and DEPT spectra, suggesting the opening of the epoxy function at C-7 and C-8. Indeed, HMBC experiments confirmed these assignments; that is, long-range connections of Me-30 (δ 1.04) to C-8 (δ 81.6), C-13, C-14, and C-15; of HO-8 (δ 7.96) to C-8, C-9, and C-14; and of H_2-6 (δ 2.48 and 3.40) to C-7 (δ 205.5) were observed, respectively. In the ROESY experiment, significant ROEs were observed between 8-OH (δ 7.96) and Me-18 (δ 2.27) and between H-15 [δ 5.42 (d, J = 3.0 Hz)] and Me-30 (δ 1.04), indicating a β-orientation of 8-OH. The connectivity of the HMBC and ROESY experiments and biogenetic considerations supported the assumed structure of **7** as 8β,15β,20*S*-trihydroxy-3,7,12,23-tetraoxolanost-9(11),16-dien-26-oic acid.

Elfvingic acid H (**8**) gave a $[M + Na]^+$ peak at *m*/*z* 553.2780 in its HRFABMS ($C_{30}H_{42}O_8$). The ^1H NMR spectrum of **8** exhibited six singlet methyls at δ 1.08, 1.21, 1.22, 1.60, 1.70, and 2.15, one doublet methyl at δ 1.21, three oxymethines at δ 4.26, 5.34, and 5.48, characteristic exomethylene signals at δ 4.92 and 4.82, and two olefinic proton signals at δ 5.95 and 6.39. The structure of **8** was determined by a combination of COSY, DEPT, HMQC, HMBC, and ROESY experiments. The above data implied the presence of double bonds at the C-4(28), C-9, and C-20 positions, of the epoxy group at C-7 and C-8, of two secondary hydroxy groups at C-15 and C-23, and of two carboxylic acids at C-3 and C-26, respectively. The relative stereochemistry of **8** was confirmed by ROESY analysis except for C-23 and C-25. Namely, ROESY correlations of Me-30 (δ 1.22) between H-15 (δ 4.26) and H-17 [δ 3.30 (dd, J = 11.4, 7.5 Hz)] indicated β-orientation of 15-OH and α-orientation of H-17. A significant ROE between Me-18 (δ 1.60) and H-7 (δ 5.34) revealed the β-orientation of H-7. Also, the high-field shift of Me-18 (δ 1.60) was brought by the disappearance of the anisotropic effect with the 7β,8β-epoxy ring observed in **2**−**5**, implying that the epoxy ring

in **8** was α-oriented. The ROE between Me-21 (δ 2.15) and H-22 (δ 5.95) indicated *Z* configuration of the double bond at C-20. The connectivity of the HMBC and ROESY experiments supported the assumed structure of **8** as 7α,8α-epoxy-15β,23ξ-dihydroxy-12-oxo-3,4-secolanosta-4(28),9,(20*Z*)(22)-trien-3,26-dioic acid.

Compound **8a** gave a $[M + Na]^+$ peak at *m/z* 567.2936 in its HRFABMS ($C_{31}H_{44}O_8$), which differs from **8** by containing CH_2. The NMR spectrum of **8a** exhibited the same features as **8**, indicating the presence of a COOMe group. Compound **8a** was formulated as the C-3 methyl ester of **8** by an HMBC experiment.

Compounds **1**−**8** and **8a** were tested for cytotoxicities against Kato III and Ehlrich cells.[5,6] Compound **8a** showed potent cytotoxicity in vitro, with IC_{50} values of 1.1 μg/mL for both cells (positive control, hinokitiol 0.6 μg/mL).

Experimental Section

General Experimental Procedures. Melting points were measured with a Yanagimoto micromelting point apparatus and were uncorrected. Optical rotations were taken on a JASCO DIP-360 polarimeter. IR spectra were recorded on a JASCO FT/IR-5300, UV spectra were recorded on a HITACHI U-3000, CD spectra were recorded on a JASCO J-500C, and NMR spectra were obtained on a Varian UNITY 600 spectrometer in C_5D_5N and $CDCl_3$ using TMS as internal standard. NMR experiments included COSY, DEPT, HMQC, HMBC, and ROESY. Coupling constants (*J* values) are given in Hz. The FABMS (Xe gun, 10 kV, triethylene glycol as the matrix) was measured on a JEOL JMS-PX303 mass spectrometer.

Plant Material. The fruit bodies of *Elfvingia applanata* were collected in Nagano, Japan, in autumn 1999. A voucher specimen (TB 2065) is deposited in the Herbarium of the Faculty of Pharmaceutical Sciences, Tokushima Bunri University, Tokushima, Japan.

Extraction and Isolation. The fresh fruit bodies (850 g) of *E. applanata* were extracted with 70% EtOH at room temparature for 6 weeks. The ethanolic extract was partitioned between EtOAc and H_2O. The EtOAc-soluble portion (12.2 g) was subjected to Si gel column chromatography with CH_2Cl_2−MeOH−H_2O (25:1:0−25:10:0.1) to afford seven fractions (fractions 1−7). Fraction 2 was passed through Sephadex LH-20 (MeOH) and successively purified by preparative HPLC (ODS, 72% MeOH) to afford elfvingic acid H methyl ester (**8a**, 5 mg). Similarly, purification by Sephadex LH-20 (MeOH) and preparative HPLC (ODS, 50−55% MeOH) afforded elfvingic acids A (**1**, 23 mg), G (**7**, 15 mg), and H (**8**, 6 mg) from fraction 3, elfvingic acids B (**2**, 50 mg) and C (**3**, 60 mg) from fraction 4, elfvingic acids B (**2**, 14 mg), C (**3**, 18 mg), and D (**4**, 25 mg) from fraction 5, and elfvingic acids C (**3**, 9 mg), D (**4**, 12 mg), E (**5**, 6 mg), and F (**6**, 15 mg) from fraction 6.

Elfvingic acid A (1): amorphous powder; $[\alpha]^{25}_D$ +52.4° (*c* 1.1, MeOH); UV (MeOH) λ_{max} (log ε) 261 (sh), 255 (4.22), 249.5 (4.26), 244.5 (4.21); CD (MeOH) Δε +3.83 (275, peak), −0.90 (247, trough), +3.94 (221.5, peak); FT-IR (film) ν_{max} 3400, 1705, 1685, 1670 cm^{-1}; ^1H NMR (C_5D_5N) δ 1.08 (3H, s, Me-28), 1.10 (3H, s, Me-29), 1.29 (3H, s, Me-18), 1.32 (3H, s, Me-19), 1.34 (3H, d, *J* = 7.4 Hz, Me-27), 1.89 (3H, s, Me-30), 2.53 (1H, m, H_2-6), 2.55 (3H, s, Me-21), 2.55 (1H, dd, *J* = 17.5, 5.9 Hz, H_2-24), 2.74 (1H, m, H_2-6), 3.17 (1H, dd, *J* = 17.5, 8.2 Hz, H_2-24), 3.70 (1H, dd, *J* = 9.6, 9.3 Hz, H-17), 4.22 (1H, s, H-12), 4.90 (1H, br d, *J* = 6.0 Hz, H-15), 6.65 (1H, s, H-22); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* $[M - H]^-$ 527; HRFABMS *m/z* $[M + Na]^+$ 551.2634 (calcd for $C_{30}H_{40}O_8$ + Na, 551.2621).

Elfvingic acid B (2): amorphous powder; $[\alpha]^{25}_D$ −36.0° (*c* 1.6, MeOH); UV (MeOH) λ_{max} (log ε) 246 (3.91); CD (MeOH) Δε −1.55 (292, trough), +5.80 (221, peak); FT-IR (film) ν_{max} 3400, 1710, 1665 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.00 (3H, s, Me-30), 1.13 (3H, s, Me-28), 1.15 (3H, s, Me-29), 1.20 (3H, d, *J* = 7.1 Hz, Me-27), 1.43 (3H, s, Me-19), 1.46 (3H, s, Me-21), 1.62 (1H, dd, *J* = 12.5, 5.6 Hz, H-5), 1.67 (1H, dd, *J* = 12.4, 5.7 Hz, H-5), 1.84 (3H, s, Me-18), 2.65 (1H, dd, *J* = 18.5, 5.4 Hz, H_2-24), 2.81, 3.02 (each 1H, d, *J* = 14.4 Hz, H_2-22), 3.10 (1H, dd, *J* = 18.5, 8.0 Hz, H_2-24), 2.94 (1H, m, H-25), 3.85 (1H, d, *J* = 5.5 Hz, H-7), 4.23 (1H, d, *J* = 3.0 Hz, H-15), 5.66 (1H, d, *J* = 3.0 Hz, H-16), 6.05 (1H, s, H-11); ^{13}C NMR ($CDCl_3$) δ 16.9 (Me-27), 20.5 (Me-21), 21.6 (C-6), 21.9 (Me-29), 24.5 (Me-28), 25.0 (Me-30), 27.4 (Me-19), 29.3 (Me-18), 33.7 (C-2), 34.5 (C-25), 37.1 (C-1), 37.9 (C-10), 46.8 (C-14), 47.7 (C-4), 47.8 (C-24), 49.5 (C-5), 53.7 (C-22), 57.5 (C-7), 63.1 (C-8), 63.2 (C-13), 71.6 (C-20), 79.2 (C-15), 125.4 (C-15), 126.1 (C-11), 158.6 (C-17), 161.9 (C-9), 179.6 (C-26), 203.3 (C-12), 207.8 (C-23), 213.5 (C-3); ^1H NMR (C_5D_5N) δ 1.02 (3H, s, Me-28), 1.09 (3H, s, Me-30), 1.15 (3H, s, Me-29), 1.34 (3H, d, *J* = 7.1 Hz, Me-27), 1.41 (3H, s, Me-19), 1.67 (1H, dd, *J* = 12.4, 5.7 Hz, H-5), 1.74 (3H, s, Me-21), 2.14 (3H, s, Me-18), 2.97 (1H, dd, *J* = 18.4, 5.8 Hz, H_2-24), 3.17, 3.30 (each 1H, d, *J* = 13.6 Hz, H_2-22), 3.55 (1H, dd, *J* = 18.4, 7.4 Hz, H_2-24), 3.97 (1H, d, *J* = 5.2 Hz, H-7), 4.49 (1H, d, *J* = 3.0 Hz, H-15), 6.04 (1H, d, *J* = 3.0 Hz, H-16), 6.14 (1H, s, H-11); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* $[M - H]^-$ 527; HRFABMS *m/z* $[M + Na]^+$ 551.2625 (calcd for $C_{30}H_{40}O_8$ + Na, 551.2621).

Elfvingic acid C (3): amorphous powder; $[\alpha]^{25}_D$ −28.2° (*c* 0.3, MeOH); UV (MeOH) λ_{max} (log ε) 261.5 (sh), 255.5 (4.09), 249.5 (4.10), 244.5 (sh); CD (MeOH) nm Δε +6.62 (221, peak); FT-IR ν_{max} (film) 3400, 1705, 1685, 1665 cm^{-1}; ^1H NMR (C_5D_5N) δ 1.07 (3H, s, Me-29), 1.12 (3H, s, Me-28), 1.20 (3H, s, Me-30), 1.25 (3H, s, Me-19), 1.39 (3H, d, *J* = 7.1 Hz, Me-27), 1.75 (3H, s, Me-21), 2.12 (3H, s, Me-18), 2.97 (1H, dd, *J* = 18.4, 5.8 Hz, H_2-24), 3.17, 3.30 (each1H, d, *J* = 13.6 Hz, H_2-22), 3.41 (1H, dd, *J* = 12.3, 4.2 Hz, ME), 3.55 (1H, dd, *J* = 18.4, 7.4 Hz, H_2-24), 3.94 (1H, d, *J* = 5.2 Hz, H-7), 4.49 (1H, d, *J* = 3.0 Hz, H-15), 6.03 (1H, d, *J* = 3.0 Hz, H-16), 6.15 (1H, s, H-11); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* $[M - H]^-$ 529; HRFABMS *m/z* $[M + Na]^+$ 553.2787 (calcd for $C_{30}H_{42}O_8$ + Na, 553.2777).

Elfvingic acid D (4): amorphous powder; $[\alpha]^{25}_D$ −17.7° (*c* 0.2, MeOH); UV (MeOH) λ_{max} (log ε) 261 (3.88), 255.5 (3.98), 250.5 (3.98), 244.5 (3.92); CD (MeOH) Δε −0.65 (290, trough), +1.09 (247, peak), +19.4 (220, peak); FT-IR (film) ν_{max} 3400, 1705, 1650 cm^{-1}; ^1H NMR (C_5D_5N) δ 1.10 (6H, s, Me-28 and Me-29), 1.24 (3H, s, Me-30), 1.35 (3H, s, Me-19), 1.38 (3H, d, *J* = 6.3 Hz, Me-27), 1.74 (3H, s, Me-21), 2.13 (3H, s, Me-18), 2.97 (1H, dd, *J* = 18.4, 5.8 Hz, H_2-24), 3.16, 3.28 (each1H, d, *J* = 13.6 Hz, H_2-22), 3.36 (1H, d, *J* = 9.6 Hz, H-3), 3.56 (1H, dd, *J* = 18.4, 7.3 Hz, H_2-24), 3.95 (1H, d, *J* = 6.3 Hz, H-7), 4.18 (1H, dt, *J* = 6.3, 9.6 Hz, H-2), 4.50 (1H, d, *J* = 3.0 Hz, H-15), 6.03 (1H, d, *J* = 3.0 Hz, H-16), 6.32 (1H, s, H-11); ^{13}C NMR (C_5D_5N) see Table 1; FABMS *m/z* $[M - H]^-$ 545; HRFABMS *m/z* $[M + Na]^+$ 569.2754 (calcd for $C_{30}H_{42}O_9$ + Na, 569.2726).

Elfvingic acid E (5): amorphous powder; $[\alpha]^{25}_D$ −8.36° (*c* 0.3, MeOH); UV (MeOH) λ_{max} (log ε) 248.5 (3.33); CD (MeOH) Δε +0.97 (220.5, peak); FT-IR (film) ν_{max} 3400, 1700, 1665 cm^{-1}; ^1H NMR (C_5D_5N) δ 1.01 (3H, s, Me-30), (3H, s, Me-29), 1.32 (3H, s, Me-19), 1.38 (3H, d, *J* = 7.5 Hz, Me-27), 1.72 (3H, s, Me-21), 2.13 (3H, s, Me-18), 2.96 (1H, dd, *J* = 18.0, 5.8 Hz, H_2-24), 3.15, 3.30 (each1H, d, *J* = 13.6 Hz, H_2-22), 3.56 (1H, dd, *J* = 18.0, 7.5 Hz, H_2-24), 3.61 (1H, d, *J* = 11.2 Hz, H_2-28), 3.90 (1H, d, *J* = 6.0 Hz, H-7), 4.18 (1H, dd, *J* = 11.0, 4.5 Hz, H-3), 4.20 (1H, d, *J* = 11.2 Hz, H_2-28), 4.41 (1H, d, *J* = 3.0 Hz, H-15), 5.99 (1H, d, *J* = 3.0 Hz, H-16), 6.17 (1H, s, H-11); ^{13}C NMR (C_5D_5N) see Table 1; FABMS *m/z* $[M - H]^-$ 545; HRFABMS *m/z* $[M + Na]^+$ 569.2684 (calcd for $C_{30}H_{42}O_9$ + Na, 569.2726).

Elfvingic acid F (6): amorphous powder; $[\alpha]^{25}_D$ +26.1° (*c* 0.2, MeOH); UV (MeOH) λ_{max} (log ε) 250.5 (3.98); CD (MeOH) Δε +0.97 (220.5, peak); FT-IR (film) ν_{max} 3400, 1705, 1685 cm^{-1}; ^1H NMR (C_5D_5N) δ 1.07 (3H, s, Me-29), 1.19 (3H, s, Me-30), 1.24 (3H, s, Me-28), 1.35 (3H, d, *J* = 7.1 Hz, Me-27), 1.75 (3H, s, Me-21), 2.30 (3H, s, Me-18), 2.48 (1H, dd, *J* = 15.1, 4.4 Hz, H_2-6), 2.94 (1H, dd, *J* = 18.3, 5.9 Hz, H_2-24), 3.15, 3.28 (each 1H, d, *J* = 13.6 Hz, H_2-22), 3.40 (1H, t, *J* = 15.1 Hz, H_2-6), 3.50 (1H, dd, *J* = 11.4, 4.0 Hz, H-3), 3.54 (1H, dd, *J* = 18.3, 7.5 Hz, H_2-24), 4.00 (1H, d, *J* = 5.8 Hz, H-7), 4.19, 4.40 (each 1H, d, *J* = 10.2 Hz, H_2-19), 4.52 (1H, d, *J* = 3.0 Hz, H-15), 6.04 (1H, d, *J* = 3.0 Hz, H-16), 6.39 (1H, s, H-11); ^{13}C NMR

(C_5D_5N), see Table 1; FABMS *m/z* [M − H]$^-$ 545; HRFABMS *m/z* [M + Na]$^+$ 569.2754 (calcd for $C_{30}H_{42}O_9$ + Na, 569.2726).

Elfvingic acid G (7): amorphous powder; $[\alpha]^{25}_D$ −2.4° (*c* 1.3, MeOH); UV (MeOH) λ_{max} (log ϵ) 262 (sh), 255.5 (sh), 250 (sh), 240.5 (4.12); CD (MeOH) $\Delta\epsilon$ −1.62 (261, trough), +5.44 (230, peak); FT-IR (film) ν_{max} 3400, 1710, 1665 cm^{-1}; ^{1}H NMR (C_5D_5N) δ 1.04 (3H, s, Me-30), 1.08 (3H, s, Me-29), 1.14 (3H, s, Me-28), 1.38 (3H, d, *J* = 7.5 Hz, Me-27), 1.70 (6H, s, Me-19 and Me-21), 2.27 (3H, s, Me-18), 2.48 (1H, m, H_2-6), 2.98 (1H, dd, *J* = 18.1, 5.8 Hz, H_2-24), 3.16, 3.23 (each1H, d, *J* = 13.5 Hz, H_2-22), 3.30 (1H, dd, *J* = 11.4, 7.5 Hz, H-17), 3.40 (1H, m, H_2-6), 3.57 (1H, dd, *J* = 18.1, 7.5 Hz, H_2-24), 5.42 (1H, d, *J* = 3.0 Hz, H-15), 5.92 (1H, d, *J* = 3.0 Hz, H-16), 6.11 (1H, s, H-11), 7.96 (8-OH); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* [M − H]$^-$ 543; HRFABMS *m/z* [M + Na]$^+$ 567.2571 (calcd for $C_{30}H_{40}O_9$ + Na, 567.2570).

Elfvingic acid H (8): amorphous powder; $[\alpha]^{25}_D$ +73.3° (*c* 0.3, MeOH); UV (MeOH) λ_{max} (log ϵ) 255 (4.19), 250 (4.19), 254; CD (MeOH) $\Delta\epsilon$ +14.9 (253.5, peak); FT-IR (film) ν_{max} 3400, 1700, 1665 cm^{-1}; ^{1}H NMR (C_5D_5N) δ 1.08 (3H, s, Me-19), 1.21 (3H, d, *J* = 7.4 Hz, Me-27), 1.22 (3H, s, Me-30), 1.60 (3H, s, Me-18), 1.70 (3H, s, Me-29), 2.15 (3H, s, Me-21), 3.16 (1H, dd, *J* = 12.3, 4.2 Hz, H-5), 3.30 (1H, dd, *J* = 11.4, 7.5 Hz, H-17), 4.26 (1H, d, *J* = 6.3 Hz, H-15), 4.82, 4.92 (each1H, br s, H_2-28), 5.34 (1H, d, *J* = 3.8 Hz, H-7), 5.48 (1H, ddd, *J* = 8.5, 5.2, 5.2 Hz, H-23), 5.95 (1H, br d, *J* = 8.5 Hz, H-22), 6.39 (1H, s, H-11); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* [M − H]$^-$ 529; HRFABMS *m/z* [M + Na]$^+$ 553.2780 (calcd for $C_{30}H_{42}O_8$ + Na, 553.2777).

Compound 8a: amorphous powder; $[\alpha]^{25}_D$ +16.7° (*c* 0.3, MeOH); ^{1}H NMR (C_5D_5N) δ 0.89 (3H, s, Me-19), 1.19 (3H, s, Me-30), 1.21 (3H, d, *J* = 7.4 Hz, Me-27), 1.59 (3H, s, Me-18), 1.68 (3H, s, Me-29), 2.15 (3H, s, Me-21), 3.07 (1H, dd, *J* = 12.4, 4.2 Hz, H-5), 3.30 (1H, dd, *J* = 11.4, 7.5 Hz, H-17), 3.62 (3H, s, OMe), 4.26 (1H, d, *J* = 6.3 Hz, H-15), 4.78, 4.92 (each 1H, br s, H_2-28), 5.33 (1H, d, *J* = 3.4 Hz, H-7), 5.48 (1H, ddd, *J* = 8.5, 5.2, 5.2 Hz, H-23), 5.95 (1H, br d, *J* = 8.7 Hz, H-22), 6.35 (1H, s, H-11); ^{13}C NMR (C_5D_5N), see Table 1; FABMS *m/z* [M − H]$^-$ 543; HRFABMS *m/z* [M + Na]$^+$ 567.2936 (calcd for $C_{31}H_{44}O_8$ + Na, 567.2934).

Culture Medium. Complete RPMI-1640 medium (Nissui Pharmaceutical Co., Ltd. Japan) containing 100 μg/mL streptomycin and 0.0035 μL/mL 2-mercaptoethanol and Dulbecco's modified Eagle's medium (DMEM, Nissui Pharmaceutical Co., Ltd. Japan) containing 50 μg/mL kanamycin sulfate and 4 mM L-glutamin were used throughout the study.

Cytotoxic Assay. Human stomach cancer cells Kato-III were maintained in DMEM and RPMI-1640 (1:1) supplemented 10% heat-inactivated fetal bovine serum (FBS, Gibco BRL). Cells were cultured in the medium at 37 °C in a humidified atmosphere of 5% CO_2 and 95% air throughout the study. Kato III cells in the exponential growth phase were plated in 96-well flat-bottom microplates at a density of 3 × 10^3 cells per 100 μL in each well and grown for 24 h in the medium. After removal of medium, 100 μL of fresh medium with various concentrations of test compounds was added. After 72 h culturing, cell growth was measured by the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method. Test compounds **1**−**8** and **8a** were dissolved in dimethyl sulfoxide (DMSO).

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