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Three New Acyclic Diterpene Glycosides from the Aerial Parts of Paprika and Pimiento

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Paprika and pimiento are used as vegetables and spices. We have obtained three new acyclic diterpene glycosides, called capsianosides XVII (1), V methyl ester (2) and XIV (3) together with capsianosides I[1)] and II[2)] from the aerial parts of paprika and pimiento. The structures of these compounds have been established by ^{1}H- and ^{13}C-NMR spectra and two-dimensional NMR methods.

Key words paprika; pimiento; capsianoside; acyclic diterpene glycoside; Solanaceae; *Capsicum annuum*

The isolation and structure elucidation of acyclic diterpene glycosides designated as capsianosides II, IX—X, XIII, and XV—XVI from the fruits of paprika *Capsicum annuum* L. var. *grossum*[2)] and C—F,[3)] and I′—III and IV from the fruits of pimiento *Capsicum annuum* L. var. *grossum*[1)] have been reported. Now our study is focused on the water-soluble constituents of the aerial parts of paprika and pimiento. In this study, we have isolated three new acyclic diterpene glycosides named capsianosides XVII (**1**), V methyl ester (**2**) and XIV (**3**) along with capsianosides I[1)] and II.[2)] The distribution of these new compounds is summarized in Table 1.

Capsianoside XVII (**1**): An amorphous powder, $[\alpha]_D$ $-53.6°$ (MeOH), was found to have a molecular formula $C_{44}H_{74}O_{21}Na$ at *m/z* 961.4678 by HR-FAB-MS. The ^{13}C-NMR spectrum of **1** showed signals due to eight double bond carbons at δ 115.8, 144.5, 126.0, 135.9, 152.6, 110.5, 131.1, and 132.9; one methylene carbon at δ 67.9, one methine carbon at δ 75.3 and one quarternary carbon at δ 81.5 with an oxygen function; three methyl carbons at δ 16.3, 21.9, and 23.3; and six methylene carbons δ 42.7, 23.6, 36.9, 35.0, 32.5, and 27.3. Further, the ^{1}H-NMR spectrum disclosed the presence of terminal vinyl protons at δ 5.20 (1H, br d, $J=11.0$ Hz, H-1*b*), 5.23 (1H, br d, $J=17.7$ Hz, H-1*a*), and 5.93 (1H, dd, $J=11.0$, 17.7 Hz, H-2), two olefinic protons at δ 5.15 (1H, m, H-6) and 5.45 (1H, t, H-14); one secondary hydroxyl group at δ 3.99 (1H, m, H-10); seven methylene groups at δ 1.61×2 (each 2H, m, H_2-4, 9), 1.95—2.30 (8H, m, H_2-5, 8, 12, 13), 4.16 (1H, d, $J=11.6$ Hz, H-17*b*), and 4.34 (1H, d, $J=11.6$ Hz, H-17*a*); one olefinic methylene group at δ 4.89 (1H, s, H-18*b*) and 5.02 (1H, s, H-18*a*); and three methyl groups at δ 1.38 (3H, s, H_3-20), 1.61 (3H, s, H_3-19), and 1.78 (3H, s, H_3-16). The position of olefinic methylene was determined by HMBC.

The ^{1}H-NMR signals due to the sugar part of **1** showed signals ascribable to 2 mol of hexosyl and 2 mol of deoxyhexosyl moieties from the evidence of signals ascribable to four anomeric protons at δ 4.21 (1H, d, $J=7.9$ Hz), 4.35 (1H, d, $J=7.9$ Hz), 4.71 (1H, s), and 4.81 (1H, s). Moreover, the ^{13}C-NMR spectra also exhibited four anomeric carbon signals at δ 99.6, 102.5, 101.8, and 102.8. The following HMBC as shown in Fig. 1 correlations revealed the sugar connectivities: correlations between the signal at δ_H 4.35 (1H, d, $J=7.9$ Hz) and the signal at δ_C 81.5 (aglycone C-3) showing the Glc I H-1 to be linked to the C-3 of the aglycone; the signal δ_H 4.21 (1H, d, $J=7.9$ Hz) and the signal at δ_C 67.9 (aglycone C-17) exhibiting the Glc II H-1 to be

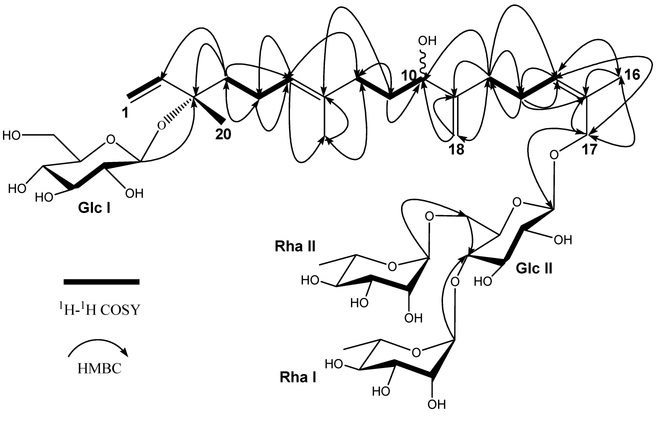

Fig. 1. Key HMBC and ^{1}H–^{1}H COSY of **1**

Table 1. Distribution of Capsianosides in Paprika and Pimiento

Capsianoside	Monomers													Dimers			
	I	I′	II	III	IV	V methy ester	IX	X	XIII	XIV	XV	XVI	XVII	C	D	E	F
C. annuum L. var. *grossum* (paprika, fruits[2)])			O	O			O	O	O		O	O					
C. annuum L. var. *grossum* (paprika, aerial part)			O			O							O				
C. annuum L. var. *grossum* (pimiento, fruits[1)])		O	O	O	O									O	O	O	O
C. annuum L. var. *grossum* (pimiento, aerial part)	O		O							O							

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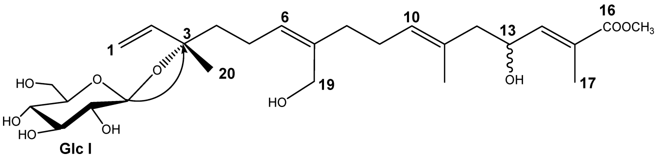

Fig. 2. Structure of **2**

linked to the C-17 of the aglycone; the signal at δ_H 4.81 (1H, s) and the signal at δ_C 79.5 (Glc II C-4) indicating the Rha I H-1 to bound to the Glc II C-4; the signal at δ_H 4.71 (1H, s) and the signal at δ_C 67.1 (Glc II C-6) showing the Rha II H-1 to be linked to the Glc II C-6. From this evidence, **1** was characterized as shown in Fig. 1.

Capsianoside V Methyl Ester (**2**): An amorphous powder, $[\alpha]_D$ $-4.7°$ (MeOH), was found to have a molecular formula $C_{27}H_{44}O_{10}Na$ at *m/z* 551.2766 by HR-FAB-MS. Analysis of the NMR data of compound **2** in comparison with those of capsianoside V$^{1)}$ showed a similar structure. Compound **2** displayed the signals from one hexosyl anomeric proton at δ 4.36 (1H, d, $J=7.3$ Hz) in the ^{1}H-NMR spectrum. Moreover, a singlet at δ 4.09 (2H, s) due to a hydroxymethyl group and four methyl groups at δ 1.39 (3H, s, H_3-20), 1.61 (3H, s, H_3-18), 1.77 (3H, s, H_3-17) and 3.74 (3H, s, $-OCH_3$) was observed in **2**. The ^{13}C-NMR signals could be assigned to one anomeric carbon signal (δ_C 99.6), one hydroxymethine group (δ_C 67.9), one carbonyl ester function (δ_C 170.1), and one –OMe group (δ_C 52.1). The HMBC correlations (Fig. 2) between the signal at δ_H 4.36 (1H, d, $J=7.3$ Hz) and the signal at δ_C 81.3 (aglycone C-3) clarified its sugar connectivities.

Capsianoside XIV (**3**): An amorphous powder, $[\alpha]_D$ $-20.4°$ (MeOH), was found to have a molecular formula $C_{38}H_{64}O_{16}Na$ at *m/z* 799.4161 by HR-FAB-MS. A comparative analysis of the ^{1}H- and ^{13}C-NMR data of **3** with those of capsianoside II$^{2)}$ suggested the presence of a 6*E*,10*E*,14*Z*,17-hydroxygeranyllinalool moiety along with an oligosaccharide chain and clearly indicated structural similarity between the two compounds. The major difference in compound **3** with respect to capsianoside II$^{2)}$ was a lack of signals due to the diglucosyl residue linked to the C-3 position and that compound **3** exhibited one additional hydroxymethyl group. Compound **3** displayed signals due to one hexosyl and two deoxyhexosyl anomeric protons at δ 4.21 (1H, d, $J=7.9$ Hz), 4.71 (1H, s), and 4.84 (1H, s) in the ^{1}H-NMR spectrum. Moreover, in **3**, a singlet at δ 4.09 (2H, s) due to a hydroxymethyl group and three methyl groups at δ 1.25 (3H, s, H_3-20), 1.61 (3H, s, H_3-18), and 1.77 (3H, s, H_3-16) were observed, one less than that in capsianoside II$^{2)}$ when compared with the ^{1}H-NMR spectra of capsianoside II.$^{2)}$ The location of the hydroxymethyl group at C-19 was derived from the HMBC spectrum, which showed correlations between H_2-19 (δ_H 4.09) and C-6 (δ_C 129.2), C-7 (δ_C 138.1), and C-8 (δ_C 36.0) and was also supported by hydroxylation shifts observed in the ^{13}C-NMR spectrum for C-6, C-7, and C-8 with respect to the same carbon signals in capsianoside II.$^{2)}$ The ^{13}C-NMR spectrum of **3** exhibited changes in the chemical shifts assignable to C-2, C-3, and C-20 in the aglycone part of $+2.0$, -8.2, and $+4.3$ ppm, respectively, by comparison with those of capsianoside II.$^{2)}$ Thus, this showed detachment of the diglucosyl residue connected at C-3 of aglycone. The location of the sugar units and interglycosidic linkages were determined by the aid of the HMBC spectrum. Cross-peaks between the anomeric proton (δ 4.21) of Glc II and C-17 of aglycone (δ 67.9), anomeric proton (δ 4.84) of Rha I and C-

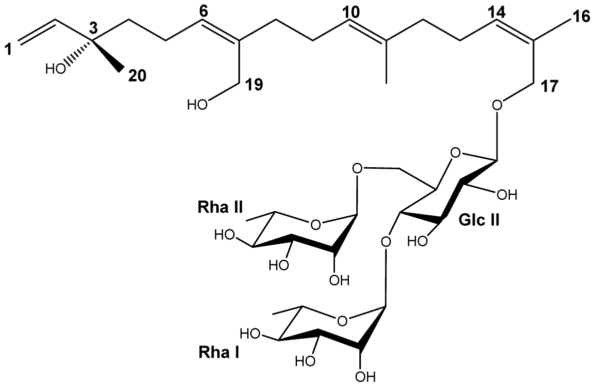

Fig. 3. Structure of **3**

Table 2. ^{13}C-NMR Data of Compounds **1**, **2** and **3** in CD_3OD (125 MHz)

Position	1	2	3
1	115.8	115.7	112.2
2	144.5	144.5	146.4
3	81.5	81.3	73.9
4	42.7	42.9	43.7
5	23.6	23.2	23.3
6	126.0	129.3	129.2
7	135.9	139.4	138.1
8	36.9	35.8	36.0
9	35.0	27.8	27.9
10	75.3	129.4	126.0
11	152.6	132.1	135.6
12	32.5	48.0	40.9
13	27.3	67.9	27.3
14	131.1	141.2	131.3
15	132.9	131.9	132.5
16	21.9	170.1	21.9
17	67.9	13.1	67.9
18	110.5	16.8	16.3
19	16.3	60.0	60.0
20	23.3	23.6	27.7
–OMe		52.1	
Glc I-1	99.6	99.6	
2	75.3	75.3	
3	78.3	78.4	
4	71.8	71.9	
5	77.7	77.7	
6	62.9	62.9	
Glc II-1	102.5		102.3
2	75.3		75.3
3	76.8		76.8
4	79.5		79.5
5	75.6		75.5
6	67.1		67.0
Rha I-1	102.8		102.8
2	72.4		72.4
3	72.6		72.6
4	74.1		74.1
5	69.9		69.9
6	17.9		17.9
Rha II-2	101.8		101.7
2	72.3		72.3
3	72.3		72.3
4	73.9		73.9
5	70.7		70.7
6	18.2		18.2

4 of Glc II (δ 79.5), and anomeric proton (δ 4.71) of Rha II and C-6 of Glc II (δ 67.0) clarified the location of the sugar chain at C-17 and the interglycosidic linkages, which were identical to those observed for capsianoside II.[2] Therefore, the structure of capsianoside XIV (**3**) was defined as 17-*O*-α-L-rhamnopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→6)]-β-D-glucopyranoside-6*E*,10*E*,14*Z*-(3*S*)-19-hydroxygeranyllinalool as shown in Fig. 3.

Discussion

A series of capsianosides obtained from *Capsicum* plants are chemically very interesting in respects to rare naturally occurring compound. In this time, from the aerial parts of paprika and pimiento, five acyclic diterpene glycosides have been isolated. Among which three ones were new compounds. Their chemical structures have been characterized, however, the respective configurations at C-10 and C-13 in compound **1** and **2** could not be decided owing to scarcely amount available. But we have now engaged with *Capsicum chinense*, so when we have much abundant amount of the relating compound, we will solve the above remaining problems. Since the configuration at C-3 was previously determined after isolation of the aglycone part by enzymatic hydrolysis, the configurations at C-3 of compounds **1**—**3** have been regarded as identical with those of previously isolated compounds.

As listed in Table 1, restricting to paprika and pimiento, four dimeric acyclic diterpene glycosides were obtained from the fruits of pimiento, meanwhile, from the aerial parts, they were not obtained.

Experimental

The optical rotations were measured with a JASCO DIP-1000 (l=0.5) automatic digital polarimeter. The ^{1}H- and ^{13}C-NMR spectra were measured with JEOL-α-500 NMR spectrometers, and the chemical shifts are given on a δ (ppm) scale with tetramethylsilane as the internal standard. The HR-FAB-MS spectra were measured with a JEOL JMS-DX303HF spectrometer and taken in a glycerol matrix containing NaI. TLC was performed on silica gel plates (Kieselgel 60 F_{254}, Merck) and RP C_{18} silica gel plates (Merck). The spots on TLC were visualized by UV light (254/366 nm) and sprayed with 10% H_2SO_4, followed by heating. Column chromatography was carried out on a Diaion HP-20 (Mitsubishi Chemical Ind.), ODS (Wako Pure Chemical Industries, Ltd., Fuji Silysia Chemical, Ltd., Japan), and silica gel 60 (spherical, 40—100 mm, and 230—400 mesh ASTM, Kanto Chemical Co., Inc.).

Plant Material The aerial parts of paprika *Capsicum annuum* L. var. *grossum* BAILEY and pimiento *Capsicum annuum* L. var. *grossum* were harvested at Nagao Farm, Kumamoto Prefecture, Japan.

Extraction and Isolation The aerial parts of paprika (1.55 kg) were extracted successively with 100% MeOH (three times). After evaporation of MeOH *in vacuo*, the residue (75.54 g) was partitioned between *n*-hexane and an aqueous fraction. The aqueous fraction was eluted from Diaion HP-20 using H_2O and MeOH (100%). The fraction (8.63 g) eluted with MeOH was divided into two parts: one part (3.08 g) was subjected to ODS (Wako) column chromatography (eluted with 50—70% MeOH), and the remainder (5.55 g) was subjected to MCI column chromatography (eluted with 40—100% MeOH). The fractions eluted with MeOH/H_2O were subsequently subjected to silica gel column chromatography ($CHCl_3$/MeOH/H_2O solvent system, 8 : 2 : 0.2—7 : 3 : 0.5), and ODS (Chromatorex) column chromatography (MeOH/H_2O solvent system, 50—60% MeOH). From the aerial parts of Paprika, capsianoside II[2] (65.6 mg), XVII (**1**) (2.3 mg), and V methyl ester (**2**) (9.3 mg) were obtained.

The aerial parts of pimiento (700 g) were extracted with 100% MeOH. After evaporation of MeOH *in vacuo*, the residue (46.67 g) was partitioned between *n*-hexane and an aqueous fraction. The aqueous fraction was eluted from Diaion HP-20 using H_2O and MeOH (100%). The fraction (4.95 g) eluted with MeOH was subjected to ODS (Wako) column chromatography (eluted with 50—70% MeOH). The fractions eluted with MeOH/H_2O were subsequently subjected to silica gel column chromatography ($CHCl_3$/MeOH/H_2O solvent system, 9 : 1 : 0.1—5.7 : 1 : 0.1), and ODS (Chromatorex) column chromatography (MeOH/H_2O solvent system, 60% MeOH). From the aerial parts of Pimiento, we obtained capsianoside I[1] (4.9 mg), II[2] (24.9 mg), and XIV (**3**) (1.4 mg).

Capsianoside XVII (**1**): An amorphous powder, $[\alpha]_D^{29}$ $-53.6°$ (c=0.23, MeOH), positive HR-FAB-MS $[M+Na]^+$ m/z 961.4678 (Calcd for $C_{44}H_{74}O_{21}Na$, 961.4620). ^{1}H-NMR (CD_3OD) δ: 1.26 (3H, d, J=6.1 Hz, Rha H_3-6), 1.27 (3H, d, J=6.1 Hz, Rha H_3-6), 1.38, 1.61, 1.78 (each 3H, s, H_3-20, H_3-19, H_3-16), 1.61×2 (each 2H, m, H_2-4, H_2-9), 1.95—2.30 (8H, m, H_2-5, H_2-8, H_2-12, H_2-13), 3.17—4.00 (m, sugar), 3.99 (1H, m, H-10), 4.16, 4.34 (each 1H, d, J=11.6 Hz, H_2-17), 4.21 (1H, d, J=7.9 Hz, Glc III H-1), 4.35 (1H, d, J=7.9 Hz, Glc I H-1), 4.71 (1H, s, Rha II H-1), 4.81 (1H, s, Rha I H-1), 4.89 (1H, s, H-18*b*), 5.02 (1H, s, H-18*a*), 5.15 (1H, m, H-6), 5.20 (1H, br d, J=11.0 Hz, H-1*b*), 5.23 (1H, br d, J=17.7 Hz, H-1*a*), 5.45 (1H, t, H-14), 5.93 (1H, dd, J=11.0, 17.7 Hz, H-2).

Capsianoside V Methyl Ester (**2**): An amorphous powder, $[\alpha]_D^{29}$ $-4.7°$ (c=0.93, MeOH), positive HR-FAB-MS $[M+Na]^+$ m/z 551.2766 (Calcd for $C_{27}H_{44}O_{10}Na$, 551.2832). ^{1}H-NMR (CD_3OD) δ: 1.39, 1.61, 1.77 (each 3H, s, H_3-20, H_3-18, H_3-17), 1.60 (2H, m, H_2-4), 1.95—2.40 (10H, m, H_2-5, H_2-8, H_2-9, H_2-12), 3.15—3.82 (m, sugar), 4.09 (2H, s, H_2-19), 4.36 (1H, d, J=7.3 Hz, Glc I H-1), 4.54 (1H, m, H-13), 5.18 (1H, m, H-6), 5.20 (1H, br d, J=11.0 Hz, H-1*b*), 5.24 (1H, br d, J=17.7 Hz, H-1*a*), 5.25 (1H, m, H-10), 5.95 (1H, dd, J=11.0, 17.7 Hz, H-2), 6.60 (1H, d, J=8.5 Hz, H-14).

Capsianoside XIV (**3**): An amorphous powder, $[\alpha]_D^{27}$ $-20.4°$ (c=0.14, MeOH), positive HR-FAB-MS $[M+Na]^+$ m/z 799.4161 (Calcd for $C_{38}H_{64}O_{16}Na$, 799.4092). ^{1}H-NMR (CD_3OD) δ: 1.25 (3H, s, H_3-20), 1.26 (3H, d, J=6.1 Hz, Rha H_3-6), 1.29 (3H, d, J=6.7 Hz, Rha H_3-6), 1.61, 1.77 (each 3H, s, H_3-18, H_3-16), 1.63 (2H, m, H_2-4), 1.99—2.20 (10H, m, H_2-5, H_2-8, H_2-9, H_2-12, H_2-13), 3.20—4.01 (m, sugar), 4.09 (2H, s, H_2-19), 4.14, 4.32 (each 1H, d, J=11.6 Hz, H_2-17), 4.21 (1H, d, J=7.9 Hz, Glc III H-1), 4.71 (1H, s, Rha II H-1), 4.84 (1H, s, Rha I H-1), 5.03 (1H, br d, J=11.0 Hz, H-1*b*), 5.15 (1H, m, H-10), 5.20 (1H, br d, J=17.4 Hz, H-1*a*), 5.29 (1H, t, J=7.0 Hz, H-6), 5.39 (1H, t, J=6.7 Hz, H-14), 5.91 (1H, dd, J=11.0, 17.4 Hz, H-2).

References

1) Izumitani Y., Yahara S., Nohara T., *Chem. Pharm. Bull.*, **38**, 1299—1307 (1990).
2) Lee J. H., Kiyota N., Ikeda T., Nohara T., *Chem. Pharm. Bull.*, **54**, 1365—1369 (2006).
3) Lee J. H., Kiyota N., Ikeda T., Nohara T., *Chem. Pharm. Bull.*, **55**, 1151—1156 (2007).