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Studies on the Constituents of *Momordica charantia* L. IV.[1] Characterization of the New Cucurbitacin Glycosides of the Immature Fruits. (2) Structures of the Bitter Glycosides, Momordicosides K and L[2]

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The structures of momordicosides K and L, bitter principles in the fruits of *Momordica charantia* L. (Cucurbitaceae), were elucidated as 7-*O*-β-D-glucopyranosides of 3β,7β-dihydroxy-25-methoxy-cucurbita-5,23-dien-19-al and 3β,7β,25-trihydroxy-cucurbita-5,23-dien-19-al, respectively, from spectral and chemical evidence.

Keywords——*Momordica charantia*; Cucurbitaceae; tetracyclic triterpene glycoside; cucurbitacin; momordicosides K and L; bitter glycoside; 3β,7β-dihydroxy-25-methoxy-cucurbita-5,23-dien-19-al; 3β,7β,25-trihydroxy-cucurbita-5,23-dien-19-al

From the many species of the Cucurbitaceae which are known to possess medicinal or toxic properties, various bitter principles have been isolated and their structures have been elucidated. They are characterized by the presence of a unique carbon skeleton and a more or less highly oxygenated side chain and nucleus, C-3 and C-11 being invariably oxygenated.

Momordicosides K and L (abbreviated as K and L, respectively), isolated as bitter principles from the immature fruits of *Momordica charantia* L., have been characterized as unique cucurbitacins having a formyl group and a 7-*O*-β-D-glucopyranosyl group, and this paper deals with their structures.

Table I. ^{13}C-NMR Chemical Shifts of Momordicosides K and L, and Their Derivatives [(a) Aglycone Moiety, (s) Sugar Moiety]

Momordicosides	Quat. C	Oxygen-bearing C	Olefinic C
K[a]	41.8	(a) 50.0 q, 71.5 d, 74.7 s, 74.7 d, 207.0 d	121.9 d, 128.0 d
	45.6, 47.9	(s) 62.7 t, 71.7 d, 75.4 d, 78.3 d, 78.5 d 106.8 d	137.3 d, 147.1 s
L[a]	41.8	(a) 69.6 s, 71.5 d, 74.7 d, 206.9 d	122.0 d, 123.8 d
	45.5, 47.9	(s) 62.8 t, 71.5 d, 75.4 d, 78.5 d (×2?) 101.4 d	141.3 d, 147.2 s
K-red[b]	38.9, 42.0	(a) 50.3 q, 65.5 t, 72.8 d, 74.9 d, 74.9 s	120.9 d, 128.5 d
	46.1, 47.6	(s) 62.8 t, 71.7 d, 76.0 d, 78.9 d, 79.0 d 101.2 d	137.5 d
Krr-1-1 (=F_1H-lag)	38.6, 46.2 48.5, 50.0	76.9 d, 78.6 t, 90.0 s	
Krr-2-2	39.6, 41.7 46.4, 49.4	65.5 t, 76.0 d	120.4 d, 142.9 s

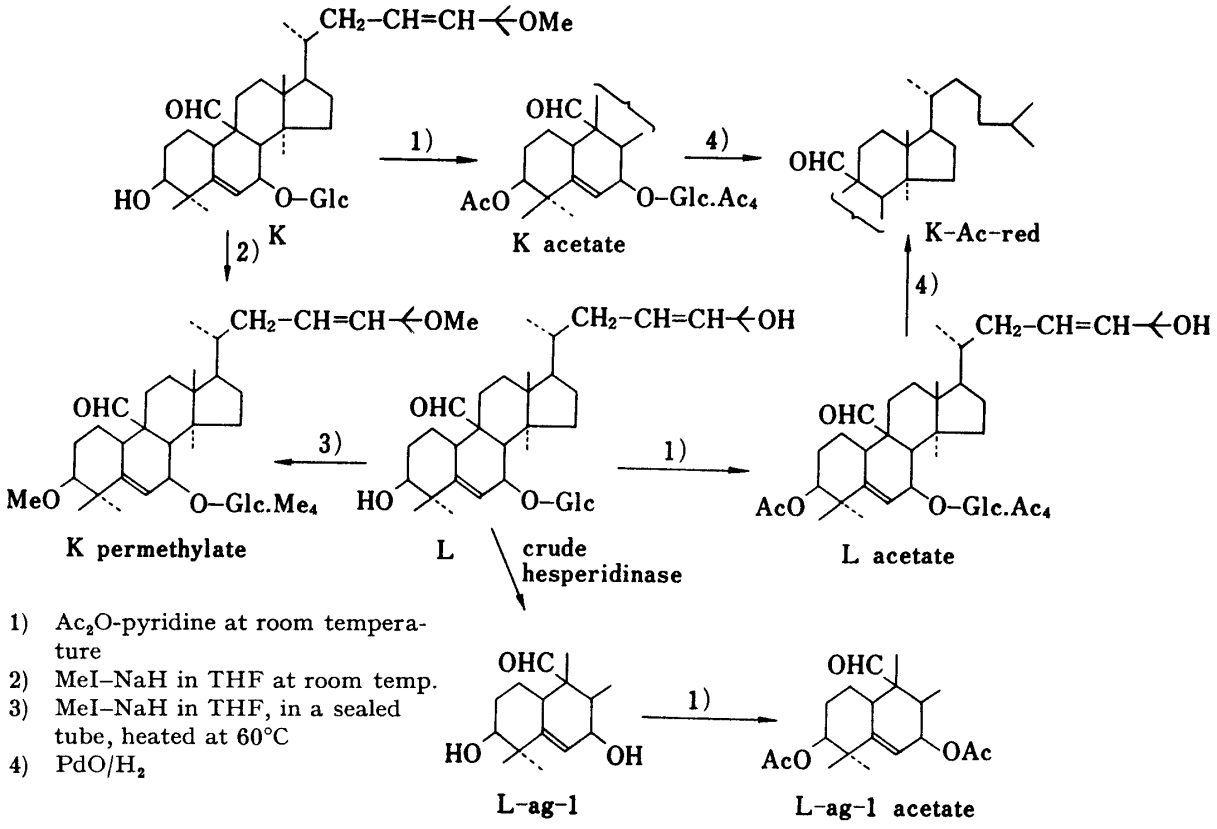
a) The 4th quaternary carbon signal may overlap on the signal at δ 41.8.
b) The 4th olefinic carbon signal may be masked by the signal of pyridine.

Momordicoside K, the less polar bitter glycoside, was found to be $C_{37}H_{60}O_9 \cdot 1/2H_2O$, and it gave a penta *O*-acetate, $C_{47}H_{70}O_{14} \cdot 1/2H_2O$. The ^{1}H nuclear magnetic resonance (^{1}H-NMR) spectrum suggested this compound to be a tetracyclic triterpene glycoside having a $-C(OMe)Me_2$

group (δ 1.31, 6H, s, and δ 3.21, 3H, s, OMe), at least one double bond (δ 6.17, br d, J=6 Hz) and a formyl group (δ 10.43, H, s) on a quaternary carbon atom. The ^{13}C nuclear magnetic resonance (^{13}C-NMR) spectrum (Table) disclosed the presence of a methoxyl group on a quaternary carbon, a glucopyranosyl group, two oxymethine groups, a formyl group, a trisubstituted double bond and a disubstituted double bond.

Momordicoside L, $C_{36}H_{58}O_9 \cdot 2H_2O$, was isolated as a penta-*O*-acetate, $C_{46}H_{68}O_{14}$. The acetate showed the infrared (IR) absorption of a hydroxyl group at 3400 cm^{-1} indicating the presence of a rather unreactive hydroxyl group. The ^1H-NMR spectra of L and its acetate were quite similar to those of K and its acetate, respectively, except that the former two showed no methoxyl signal and that they showed at δ 1.53 the singlet of two equivalent methyl groups on the oxygenated quaternary carbon atom. K and its acetate showed the corresponding signal at δ 1.31.

These spectral features suggested K and L to have the same skeleton and L to be the hydroxyl derivative of K, which has a methoxyl group.



When K acetate was hydrogenated over PdO, a compound (K-Ac-red) having no methoxyl group was obtained. The ^1H-NMR spectrum of K-Ac-red indicated that it retains acetylated glucosyl and secondary acetoxyl groups, a trisubstituted double bond and a formyl group, but the disubstituted double bond was saturated. L acetate gave the same product on catalytic hydrogenation.

Methylation of L gave a hexa-*O*-methylate, and this compound was identical with K permethylate. The ^1H-NMR spectrum of K permethylate showed the signal of the anomeric proton at δ 4.58 as a doublet (J=8 Hz) indicating the configuration of the glucopyranosyl groups of K and L to be β.

L gave, on enzymatic hydrolysis, an aglycone (L-ag-1), $C_{30}H_{48}O_4$, which provided a diacetate on acetylation with Ac_2O–pyridine at room temperature. The ^1H-NMR spectrum

of L-ag-1 showed the signals of two hydroxymethine protons (δ 3.80, H, br s; δ 4.35, H, br d, J=5 Hz), olefinic protons (δ 5.90, 2H, br s; δ 6.24, H, br d, J=5 Hz) and a formyl proton (δ 10.58, H, s). One of the hydroxymethine protons was coupled with an olefinic proton, and it follows that L-ag-1 has two secodary hydroxyl groups, one isolated, and the other a hydroxyl group on a carbon allylic to the trisubstituted double bond. L showed the ^{1}H-NMR signal of an isolated hydroxymethine proton at δ 3.81 (br s) and this signal was shifted downfield by acetylation. This fact apparently indicates that the sugar moieties of K and L are linked not to the isolated hydroxyl group, but to the allylic one.

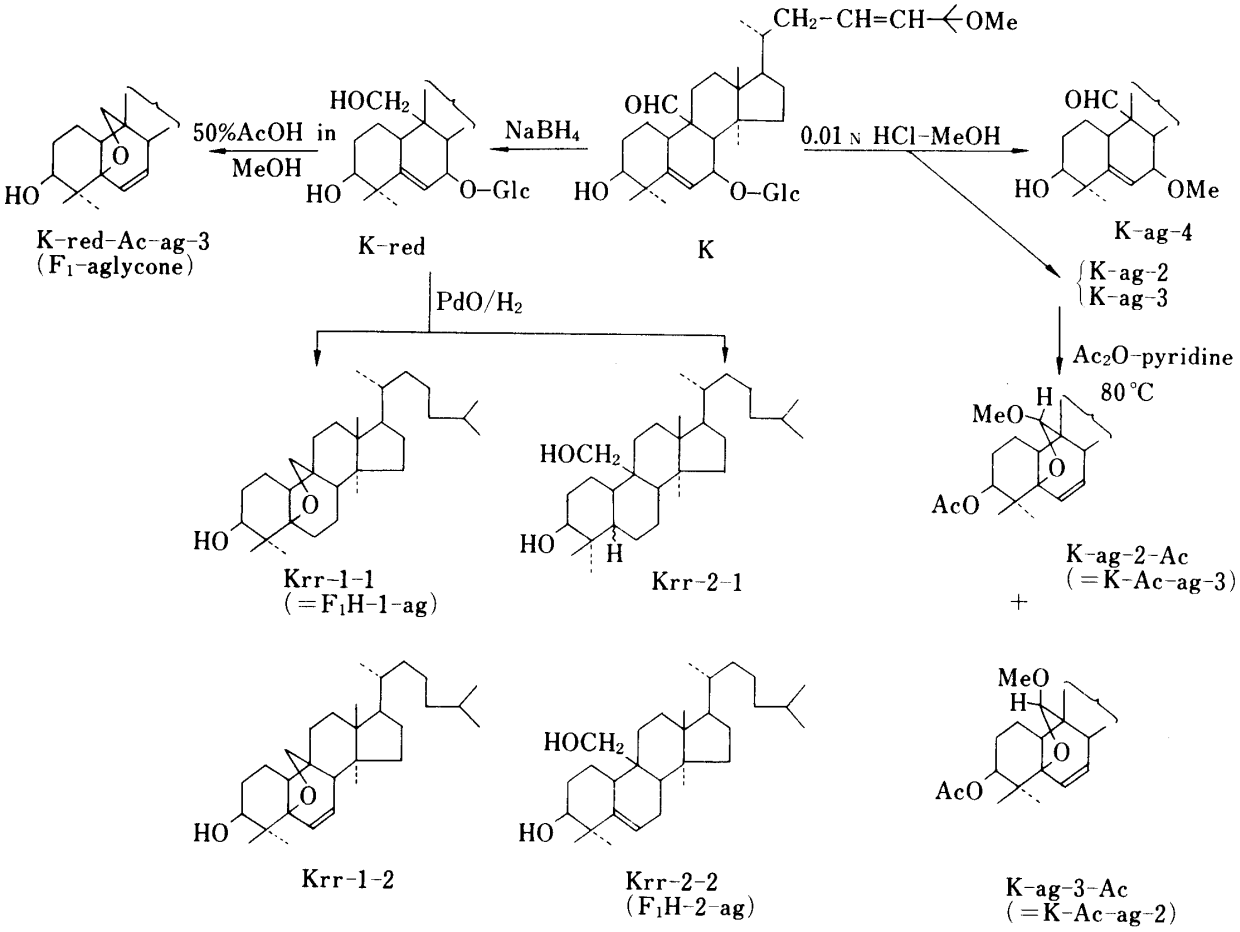


Chart 2

On treatment with $NaBH_4$, K gave a corresponding primary alcohol (K-red). It showed no signal of a formyl group but a signal of a new hydroxymethylene group was observed (^{13}C-NMR: δ 65.5, t). When K-red was hydrogenated over PdO, four compounds (Krr-1-1, -1-2, -2-1 and -2-2) were formed, none of which has a methoxyl group and a glucosyl group. Krr-1-1 and -2-2 were proved to be identical with 5,19-epoxy-5β-cucurbitan-3β-ol (F_1H-1-ag) and cucurbit-5-ene-3β, 19-diol (F_1H-2-ag),[1] respectively.

Krr-1-2, $C_{30}H_{50}O_2$, showed in its ^{1}H-NMR spectrum the signal of an oxymethine proton at δ 3.56 as a multiplet overlapped by the signals of the two oxymethylene protons (δ 3.55, d, J=9 Hz and δ 3.68, d, J=9 Hz), the signal of a hydroxyl proton (δ 4.21, H, d, J=10 Hz) which is obviously coupled with an oxymethine proton, and the signals of two olefinic protons on a *cis* double bond (δ 5.61, H, dd, J=10, 4 Hz; δ 6.14, H, dd, J=10, 2 Hz). These spectral data indicated Krr-1-2 to be 5,19-epoxy-5β-cucurbit-6-en-3β-ol.

The ^{1}H-NMR spectrum of Krr-2-1 showed signals of an oxymethine proton (δ 3.63, H, br s) and hydroxymethylene protons (δ 3.68, H, d, J=10 Hz; δ 4.27, H, d, J=10 Hz). No olefinic proton signal was observed. From this evidence, Krr-2-1 was considered to be cucurbitane-3β, 19-diol, but the configuration of C_5-H was not further examined.

When K-red was treated in 50% acetic acid in MeOH at 38°C, a compound having no glucosyl group was obtained. The ^{1}H-NMR, ^{13}C-NMR and IR spectra were identical with those of 5,19-epoxy-25-methoxy-5β-cucurbita-6,23-dien-3β-ol (F_1-aglycone).[1] This chemical conversion indicated that the hydroxymethylene group in K-red, and thus the formyl group in K, is located at C_9.

K was methanolyzed under mild conditions to give D-glucose (not methyl D-glucosides), a methyl ether (K-ag-4) of allyl alcohol and a mixture of two methylacetals (K-ag-2 and -3. ^{1}H-NMR: δ 3.38 and 3.40, acetal methyls. ^{13}C-NMR: δ 57.2, q and 57.6, q, acetal methyl carbons. δ 112.1, d and 114.9 ,d, acetal carbons).

K-ag-4 showed a ^{1}H-NMR spectrum similar to that of K except for one additional methoxyl signal and for the lack of the signals due to the sugar moiety. K-ag-4 showed a circular dichroism (CD) spectrum similar to that of K. Thus, K-ag-4 was concluded to have the structure in which the glucopyranosyl group of K has simply been substituted by a methoxyl group; ^{13}C–NMR spectrum of K-ag-4 was consistent with the proposed structure.

K-ag-2 and -3 were separated and purified by acetylation to give the corresponding acetates (K-ag-2-Ac and -3-Ac). The hydroxyl groups resisted acetylation with Ac_2O–pyridine at room temperature, and heating on a boiling water bath for 20 h for K-ag-3, and for two d for K-ag-2, was required for complete acetylation. Later, the same acetates were obtained by methanolysis of K acetate under mild conditions. The resistance of the hydroxyl groups in K-ag-2 and -3 to acetylation suggested that the methylacetal group is located in close proximity to the hydroxyl group.

K-ag-3-Ac, $C_{34}H_{54}O_5$, showed in its ^{1}H-NMR spectrum the signals of an acethyl methyl group (δ 2.11), methoxyl groups (δ 3.24 and 3.51), an acetal proton (δ 4.83), an acetylated methine proton (δ 4.97, br s) and olefinic protons (δ 5.60, 2H, br s, δ 5.63, H, dd, J=10, 4 Hz; δ 6.14, H, dd, J=10, 2.5 Hz). The ^{13}C-NMR spectrum exhibited the signals of two methoxyl carbons (δ 50.3, q and 57.5, q, the latter being the signal due to the acetal methyl carbon), three oxygenated carbon atoms (δ 74.8, s, 76.8, d and 84.1, s) and an acetal carbon atom (δ 112.3, d). From these spectral data, K-ag-3 was supposed to be a compound having the original side chain, a methylacetal ring bridged on two quaternary carbon atoms and a disubstituted double bond in the nucleus, *viz.* 5,19-epoxy-19,25-dimethoxy-5β-cucurbita-6,23-dien-3β-ol.

K-ag-2-Ac, $C_{34}H_{54}O_5$, the minor product, showed a ^{1}H-NMR spectrum similar to that of K-ag-3-Ac, but the signal of the acetal proton and the signal of one of the olefinic protons on the *cis* double bond appeared a little upfield at δ 4.62 (s) and δ 5.53 (dd, J=10, 3 Hz), respectively, indicating K-ag-2-Ac to be the isomer in which the configuration of the acetal proton differs from that of K-ag-3-Ac.

In view of the facts that the yield of K-ag-2 was smaller than that of K-ag-3, and that K-ag-2 required more drastic conditions for acetylation than those used for K-ag-3, the hydroxyl group of K-ag-2 appears to be more sterically hindered than that of K-ag-3. Consequently, the absolute configuration of C_{19} of K-ag-2 seems to be *S*, and thus K-ag-3 is the *R* isomer.[3]

K-ag-2 and -3 are not the genuine aglycones of K, nor is F_1-aglycone the genuine aglycone of K-red. Formation of the methylacetal ring by methanolysis of K, and of the ether ring from K-red can only be explained by a concerted process involving the fission of the linkage between the aglycone-C and the glucose-O to give an intermediate C_7 carbonium cation, migration of the double bond from $C_{5,6}$ to $C_{6,7}$, followed by ring closure between C_5 and C_{19}-oxygen. Krr-1-1 and Krr-1-2 are also considered to be formed by initial cleavage of the C_7-O bond and by the same mechanism.

From the above-mentioned chemical and spectral evidence, the structures of momordicosides K and L were determined as 7-*O*-β-D-glucopyranosides of 3β,7β-dihyroxy-25-methoxycucurbita-5,23-dien-19-al and its C_{25}-OH derivative, respectively. The configuration of the C_7-hydroxyl group was deduced from the positive CD curve[4] of K-red and also from the *J*-values (5 Hz) of C_6-H and C_7-H of L-ag-1.[5]

Experimental[6]

Isolation of Momordicosides K and L——Described in the preceding paper.[1]

Momordicoside K: Colorless needles from MeOH. mp 236—237°C. $[\alpha]_D^{20°}$ +63.3° (*c*=1.00, $CHCl_3$–MeOH (1: 1)). CD: $[\theta]_{208}^{11°}$ +58500° ($c=2.05\times10^{-4}$ g/ml, MeOH). *Anal.* Calcd for $C_{37}H_{60}O_9\cdot1/2H_2O$: C, 67.58; H, 9.28. Found: C, 67.45; H, 9.56. ^{1}H-NMR: 0.78, 0.87, 0.95, 1.12, 1.31 (×2), 1.42 (CH_3), 3.21 (3H, s, $-OCH_3$), 3.79 (H, br s, C_3–H), 5.57 (2H, br s, C_{23}–H, C_{24}–H), 6.17 (H, br d, *J*=6 Hz, C_6–H), 10.43 (H, s, –CHO). ^{13}C-NMR: Table.

Momordicoside K Acetate: Obtained by acetylation with Ac_2O–pyridine at room temperature: colorless needles from MeOH–H_2O. mp 166—169°C. $[\alpha]_D^{20°}$ +40.4° (*c*=1.00, $CHCl_3$). *Anal.* Calcd for $C_{47}H_{70}O_{14}\cdot1/2H_2O$: C, 65.05; H, 8.19. Found: C, 65.05; H, 8.42. ^{1}H-NMR: 0.81, 0.96, 1.07, 1.32 (×2) (CH_3), 1.84, 1.98, 2.01, 2.08, 2.32 (OAc), 3.23 (3H, s, OCH_3), 5.61 (2H?, br s, C_{23}–H, C_{24}–H), 6.07 (H, br d, *J*=5 Hz, C_6–H), 10.09 (H, s, –CHO). ^{13}C-NMR: quaternary C, 40.1, 45.6 (×2?), 47.8; oxygenated C (sugar moiety), 62.5, t, 69.1, 72.1, 72.3, 73.6, 98.6, d, (aglycone moiety), 50.1, q (OCH_3), 72.6, d (C_3), 74.7, s (C_{25}), 77.9, d (C_7), 206.2, d (CHO), olefinic C, 121.6, d, 128.2 d, 137.7, d, 146.9, s (C_5); acetyl carbonyl C, 169.7 (×2?), 170.1, 170.4.

Momordicoside L: Colorless needles from $CHCl_3$–MeOH. mp 227—232°C. $[\alpha]_D^{20°}$ +57.3° (*c*=1.00, $CHCl_3$–MeOH (1: 1)). CD: $[\theta]_{210}^{11°}$ +42900° ($c=1.92\times10^{-4}$ g/ml, MeOH). *Anal.* Calcd for $C_{36}H_{58}O_9\cdot2H_2O$: C, 64.48; H, 9.25. Found: C, 64.60; H, 9.32. ^{1}H-NMR: 0.77, 0.88, 1.15, 1.45, 1.53 (×2) (CH_3), 3.81 (br s, C_3–H), 4.99 (H, d, *J*=7 Hz, C_1–H of glucopyranosyl group), 5.92 (br s, C_{23}–H, C_{24}–H), 10.42 (H, s, –CHO). ^{13}C-NMR: Table.

Momordicoside L Acetate: Colorless needles from MeOH–H_2O. mp 148 K152°. $[\alpha]_D^{20°}$ +42.0° (*c*= 1.00, $CHCl_3$). IR ν_{max}^{KBr} cm^{-1}: 3450 (OH). *Anal.* Calcd for $C_{46}H_{68}O_{14}$: C, 65.40; H, 8.06. Found: C, 65.20; H, 8.28. ^{1}H-NMR: 0.76, 0.91, 1.07, 1.53 (×2) (CH_3), 1.83, 1.98, 2.01, 2.07, 2.30 (OAc), 5.92 (2H?, br s, C_{23}–H, C_{24}–H), 10.07 (H, s, –CHO).

Catalytic Hydrogenation of K Acetate——K acetate (100 mg) and PdO (75 mg) were added to MeOH (10 ml) and shaken in an H_2 atmosphere until the spot of K acetate could no longer be detected on thin-layer chromatography (TLC). PdO was removed by filtration. The filtrate was concentrated under reduced pressure to give a white powder (K-Ac-red) (95 mg). Crystallization from MeOH–$CHCl_3$ gave colorless needles. mp 202—208°C. CD: $[\theta]_{207}^{11°}$ +60800° ($c=0.25\times10^{-3}$ g/ml, MeOH). ^{1}H-NMR ($CDCl_3$): 0.79, 0.84, 0.89, 1.10, 1.18 (CH_3), 1.99, 2.01, 2.03, 2.08, 2.09 (OAc), 4.07 (d, *J*=5 Hz, C_7–H), 4.53 (H, d, *J*=8 Hz, C_1–H of glucopyranosyl group), 5.84 (H, br d, *J*=5 Hz, C_6–H), 9.59 (H, s, –CHO).

Catalytic Hydrogenation of L Acetate——L acetate (70 mg) was hydrogenated in the same way as described above to give L-Ac-red (65 mg): colorless needles from MeOH. mp 203—206°C. The IR and ^{1}H-NMR spectra were identical with those of K-Ac-red.

Methylation of K and L——K (100 mg) was added to freshly distilled tetrahydrofuran (THF) (5 ml) containing NaH (60 mg) and the mixture was sonicated for a few min. MeI (1 ml) was added to the mixture and the whole was stirred at room temperature for 20 h. The reaction mixture was diluted with MeOH, and concentrated under reduced pressure below 30°C. The residue was extracted with $CHCl_3$, and the $CHCl_3$-soluble material was chromatographed on silica gel (38 g) with 15% AcOEt in hexane to yield K permethylate (95 mg). Crystallization from MeOH gave colorless needles. mp 140—144°C. ^{1}H-NMR: 0.77, 0.92, 0.98, 1.05, 1.24, 1.31 (×2) (CH_3), 3.13, 3.21, 3.38, 3.53 (×2), 3.60 (OCH_3), 4.29 (H, br d, *J*=5 Hz, C_7–H), 4.58 (H, d, *J*=8 Hz, C_1–H of glucopyranosyl group), 5.58 (2H, br s, C_{23}–H, C_{24}–H), 6.11 (H, br d, *J*=5 Hz, C_6–H).

L (30 mg) was dissolved in THF (0.3 ml), then NaH (40 mg) was added to the solution and the whole was sonicated for a few min. MeI (0.3 ml) was added, and the reaction mixture was heated in a sealed tube at 60°C, then worked up as described above. The product (19 mg) was crystallized from MeOH. mp 142—144°C. The IR spectrum was superimposable on that of K permethylate.

Enzymatic Hydrolysis of L——L (200 mg) and crude hesperidinase (390 mg) were suspended in 20% EtOH (20 ml) and incubated at 38°C for 3 d. The precipitates were filtered off and extracted with $CHCl_3$. The residue (97 mg) after evaporation of the $CHCl_3$ was chromatographed twice on silica gel (40 g) using 4% MeOH in $CHCl_3$ to give crude L-ag-1 (66 mg). Crystallization from $CHCl_3$ gave colorless needles. mp 190—193°C. CD: $[\theta]_{204}^{11°}$ +61600° ($c=0.92\times10^{-4}$ g/ml, MeOH). ^{1}H-NMR: 0.84, 0.87, 0.98 (d, *J*=6 Hz), 1.17, 1.47, 1.51 (CH_3), 3.81 (H, br s, C_3–H), 4.36 (H, br d, *J*=5 Hz, C_7–H), 5.91 (2H, br s, C_{23}–H, C_{24}–H), 6.25 (H, br d, *J*=5 Hz, C_6–H), 10.58 (H, s, –CHO).

L-ag-1 Acetate: L-ag-1 was acetylated with Ac_2O–pyridine at room temperature. Colorless needles from MeOH. mp 102—105°C. IR ν_{max}^{KBr} cm^{-1}: 3450 (OH), 1745 (OAc), 1710 (–CHO). ^{1}H-NMR: 0.76, 0.84, 0.96 (d, J=5 Hz), 1.11, 1.20, 1.51 (CH_3), 1.84, 1.89 (OAc), 4.96 (C_3–H?, overlapped by H_2O signal), 5.41 (H, br d, J=6 Hz, C_7–H), 5.90 (2H, br s, C_{23}–H, C_{24}–H), 6.03 (H, br d, J=6 Hz, C_6–H), 10.23 (H, s, –CHO).

$NaBH_4$ Reduction of K——K (200 mg) was dissolved in EtOH–THF (4: 1) (5 ml), then $NaBH_4$ (11 mg) was added and the mixture was stirred at room temperature for 1 h. The reaction mixture was acidified by adding 1% acetic acid in MeOH. The solvent was evaporated off under reduced pressure, and the residue was chromatographed on silica gel (20 g) with 15% MeOH in $CHCl_3$ to give K-red (202 mg). Colorless platelets from MeOH. mp 195—197°C. $[\alpha]_D^{31°}$ +60.1° (c=1.05, $CHCl_3$–MeOH (1: 1)). CD: $[\theta]_{206}^{30°}$ +87900° (c=1.1×10^{-4} g/ml, MeOH). ^{1}H-NMR: 0.82, 1.01, 1.13, 1.20, 1.31 (×2) (CH_3), 3.21 (3H, s, OCH_3), 5.16 (H, br d, J=7 Hz, C_7–H), 5.60 (br s, C_{23}–H, C_{24}–H). ^{13}C-NMR: quaternary C, 38.9, 42.0, 46.1, 47.6; oxygenated C (sugar moiety), 62.8, t, 71.7, d, 76.0, d, 78.9, d, 79.0, d, 101.2, d, (aglycone moiety), 50.1, q (OCH_3), 65.5, t (C_{19}), 72.8, d, 74.9, d, 74.9, s (C_{25}); olefinic C, 120.9, d, 128.5, d, 137.5, d. The 4 th olefinic carbon signal was masked by the pyridine signal.

Catalytic Hydrogenation of K-red——K-red (300 mg) and PdO (350 mg) were added to MeOH (60 ml) and the mixture was shaken in an H_2 atmosphere for 1 h. PdO was removed by filtration and the filtrate was concentrated to dryness. The residue (254 mg) was suspended in water and extracted with $CHCl_3$. The $CHCl_3$-soluble material (181 mg) was chromatographed on silica gel (30 g). From the fractions eluted with $CHCl_3$, Krr-1 (67 mg) was obtained. Further elution with 1% MeOH in $CHCl_3$ gave Krr-2 (105 mg). Krr-1 was repeatedly chromatographed on silica gel (500 times the weight of material) using 2% acetone in hexane to give Krr-1-1 (31 mg) and Krr-1-2 (19 mg) in order of increasing polarity. Krr-2 was also repeatedly chromatographed on silica gel (500 times) using 20% hexane in AcOEt to give Krr-2-1 (22 mg) and Krr-2-2 (29 mg).

Krr-1-1: Colorless needles from MeOH. mp 130—133°C. The IR and ^{1}H-NMR spectra were the same as those of F_1H-1-ag.[1]

Krr-1-2: Colorless needles from MeOH. mp 148—151°C. EI-MS m/z: 442.386 (M^+). Calcd for $C_{30}H_{50}O_2$: 442.381. ^{1}H-NMR: 0.81, 0.87, 0.91, 0.93, 1.37 (CH_3), 3.56 (m, C_3–H), 3.55 (d, J=9 Hz, C_{19}–H), 3.68 (d, J=9 Hz, C_{19}–H), 4.21 (H, d, J=10 Hz, C_3–OH), 5.61 (H, dd, J=10, 4 Hz, C_7–H), 6.14 (H, dd, J=10, 2 Hz, C_6–H).

Krr-2-1: Colorless needles from AcOEt. mp 162—163°C. ^{1}H-NMR: 0.86, 0.92, 0.98, 1.20 (CH_3), 3.63 (H, br s, C_3–H), 3.68 (H, d, J=10 Hz, C_{19}–H), 4.27 (H, d, J=10 Hz, C_{19}–H).

Krr-2-2: Colorless needles from AcOEt. mp 162°C. The IR and ^{1}H-NMR signals were superimposable on those of F_1H-2-ag.[1]

Methanolysis of K-red——K-red (330 mg) was dissolved in 50% AcOH in MeOH (10 ml) and shaken at 38°C for 2 d. When the reaction mixture was neutralized by adding 5 N NaOH solution, precipitates were deposited. The precipitates were separated by centrifugation, washed with water and dried under reduced pressure. The supernatant and washings were combined and extracted with $CHCl_3$. $CHCl_3$ was evaporated off to give an oily material (68 mg). The precipitates (205 mg) and the $CHCl_3$ extract (68 mg) were combined and repeatedly chromatographed on silica gel (160 times the weight of material) using 10% AcOEt in hexane as a solvent to give two compounds (K-red-ag-2 and -3, in order of decreasing polarity).

K-red-ag-2: 74 mg. Colorless needles from acetone. mp 160—162°C. This compound seemed to be a mixture and was not characterized.

K-red-ag-3: 86 mg. Colorless leaflets from acetone–hexane. mp 139—140°C. The IR spectrum was superimposable on that of F_1-aglycone.[1]

Methanolysis of K——K (300 mg) was dissolved in 0.01 N HCl (MeOH) (5 ml) and the solution was stirred at room temperature for 1.5 h. The reaction mixture was neutralized with Ag_2CO_3 and filtered. The filtrate was treated with H_2S and concentrated to dryness, then the residue was extracted with $CHCl_3$. The $CHCl_3$-insoluble material was checked by TLC ($CHCl_3$–MeOH–H_2O (7: 5: 1)); Rf 0.19 (D-glucose, Rf 0.19; methyl α-D-glucopyranoside, Rf 0.36). The $CHCl_3$-soluble material (250 mg) was chromatographed on silica gel (76 g). Elution with 15% AcOEt in hexane gave a mixture of K-ag-2 and -3 (103 mg), and elution with 40% AcOEt in hexane gave K-ag-4 (64 mg).

K-ag-4: Colorless needles from hexane. mp 149—153°C. IR ν_{max}^{KBr} cm^{-1}: 3450 (OH), 1718 (–CHO). CD: $[\theta]_{207}^{11°}$ +55000° (c=0.9×10^{-4} g/ml, MeOH). *Anal.* Calcd for $C_{32}H_{52}O_4$: C, 76.80; H, 10.40. Found: C, 76.72; H, 10.38. ^{1}H-NMR: 0.83, 0.94, 1.16, 1.33 (×2), 1.49 (CH_3), 3.22 (3H, s, OCH_3), 3.28 (3H, s, OCH_3), 3.54 (H, br d, J=6 Hz, C_7–H), 3.80 (H, br s, C_3–H), 5.60—5.72 (2H, m, C_{23}–H, C_{24}–H), 6.13 (H, br d, J=6 Hz, C_6–H). ^{13}C-NMR: quaternary C, 41.9, 45.8, 47.9; oxygenated C, 50.2, q (C_{25}–OCH_3), 55.9, q (C_7–OCH_3), 74.8, s (C_{25}), 75.6, d (probably C_3 and C_7), 207.0, d (–CHO); olefinic C, 121.0, d, 128.3, d, 137.6, d, 147.6, s (C_5).

The mixture of K-ag-2 and -3 was repeatedly chromatographed on silica gel (500 times the weight of the materials) using 15% AcOEt in hexane to give crude K-ag-2 (25 mg) and K-ag-3 (60 mg). The crude K-ag-2 was acetylated in Ac_2O–pyridine (1: 1) (0.3 ml) with heating at 80°C for 52 h. The product was purified by column chromatography and by crystallization from MeOH to give K-ag-2-Ac (1 mg). mp 120—122°C. The IR spectrum was superimposable on that of K-Ac-ag-3 obtained by methanolysis of K acetate (see below).

K-ag-3 (15 mg) was acetylated with Ac_2O–pyridine (1: 1) (0.2 ml) with heating at 80°C for 20 h. The reaction product (20 mg) was purified by column chromatography (silica gel: 10 g, 10% AcOEt in hexane) and by crystallization from hexane to give K-ag-3-Ac (2 mg). mp 131—133°C. The IR spectrum was superimposable on that of K-Ac-ag-2 obtained by methanolysis of K acetate (see below).

Methanolysis of K Acetate——K acetate (1.45 g) was dissolved in 0.01 N HCl (MeOH) and the solution was stirred at room temperature overnight. The reaction mixture was neutralized with Ag_2CO_3 and filtered. The filtrate was treated with H_2S and concentrated to dryness. The residue was treated with hot hexane–AcOEt (9: 1) and the soluble material was subjected to chromatography on silica gel (300 times the weight of materials) using 10 % AcOEt in hexane as the eluting solvent to give K-Ac-ag-2 (92 mg) and K-Ac-ag-3 (33 mg).

K-Ac-ag-2 (=K-ag-3-Ac): Colorless needles from hexane. mp 130—133°C. CD: $[\theta]_{203}^{20°}$ −92400° ($c=0.73\times10^{-4}$ g/ml, MeOH). *Anal.* Calcd for $C_{34}H_{54}O_5$: C, 75.28; H, 9.96. Found: C, 75.11; H, 9.96. ^{1}H-NMR: 0.91, 0.94, 1.21, 1.33 (CH_3), 2.11 (OAc), 3.23 (3H, s, C_{25}–OCH_3), 3.51 (3H, s, C_{19}–OCH_3), 4.83 (s, C_{19}–H), 4.97 (H, br s, C_3–H), 5.60 (br s, C_{23}–H, C_{24}–H), 5.63 (dd, $J=10$, 4 Hz, C_7–H), 6.14 (dd, $J=10$, 2 Hz, C_6–H). ^{13}C-NMR: quaternary C, 37.9, 45.3, 48.2 (×2?); oxygenated C, 50.1, q (C_{25}–OCH_3), 57.5, q (C_{19}–OCH_3), 74.8, s (C_{25}), 76.8, d (C_3), 84.1, s (C_5), 112.3, d (C_{19}); olefinic C, 128.3, d, 131.5, d, 133.0, d, 137.5, d; acetyl carbonyl C, 170.7, s.

K-Ac-ag-3 (=K-ag-2-Ac): Colorless needles from MeOH. mp 124—124.5°C. CD: $[\theta]_{203}^{20°}$ −89400° ($c=0.67\times10^{-4}$ g/ml, MeOH). *Anal.* Calcd for $C_{34}H_{54}O_5$: C, 75.28; H, 9.96. Found: C, 75.30; H, 9.98. ^{1}H-NMR: 0.87, 0.92, 0.93, 1.23, 1.34 (×2) (CH_3), 2.11 (OAc), 3.25, 3.51 (OCH_3), 4.62 (H, s, C_{19}–H), 4.95 (br s, C_3–H), 5.53 (dd, $J=10$, 3 Hz, C_7–H), 5.60 (br s, C_{23}–H, C_{24}–H), 6.25 (dd, $J=10$, 2 Hz, C_6–H). ^{13}C-NMR: quaternary C, 37.6, 45.3, 48.2, 49.1; oxygenated C, 50.1, q (C_{25}–OCH_3), 55.2, q (C_{19}–OCH_3), 74.8, s (C_{25}), 76.9, d (C_3), 83.1, s (C_5), 113.3, d (C_{19}); olefinic C, 128.3, d, 129.4, d, 134.8 d, 137.6, d; acetyl carbonyl C, 170.6.

Methanolysis of K Permethylate. Identification of the Methylated Sugar——K permethylate (110 mg) was dissolved in 0.01 N HCl (MeOH) (6 ml) and the solution was stirred for 2 h at room temperature. The reaction mixture was treated in the usual manner and the product was chromatographed on silica gel (40 g). The aglycones were eluted with 30% AcOEt in hexane and the sugar (38 mg) was eluted with 50% AcOEt in hexane. TLC of the methylated sugar (Kieselgel F_{254}, hexane–AcOEt (1: 2)): *Rf* 0.13, 0.16 (methyl 2,3,4,6-tetra-*O*-methyl α-D-glucopyranoside, *Rf* 0.31). The sugar was refluxed in 2 N HCl (MeOH) (0.5 ml) for 1 h and treated with Ag_2CO_3 then H_2S. The product showed two spots on TLC: *Rf* 0.33, 0.23 (methyl 2,3,4,6-tetra-*O*-methyl α-D-glucopyranoside, *Rf* 0.23; β-anomer, 0.33). The methylation product was purified by column chromatography on silica gel (9 g) with hexane–AcOEt (1: 1). A thin-layer-chromatographically homogeneous oil (13 mg) was obtained. Its *Rf* value was identical with that of methyl 2,3,4,6-tetra-*O*-methyl α-D-glucopyranoside. $[\alpha]_D^{32°}$ +124.2° ($c=0.65$, $CHCl_3$).

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References and Notes

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3) The configuration of C_{19} of K-ag-2 and -3 will be discussed in the next paper in connection with the structures of momordicosides E_1 and e′.
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6) Instruments and materials employed in this work were the same as those described in the preceding paper.[1] Melting points are uncorrected. ^{1}H-NMR and ^{13}C-NMR spectra were measured in pyridine-d_5 and chemical shifts are expressed in the δ-scale with tetramethylsilane as an internal standard (s, singlet; br, broad; d, doublet; dd, double doublet; t, triplet; q, quartet; m, multiplet).