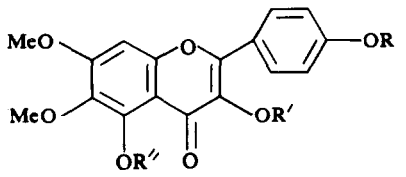

1 R = R″ = H, R′ = Glc

2 R = R″ = Me, R′ = H

3 R = Me, R′ = R″ = H

4 R = R′ = R″ = H

aglycone with acetobromoglucose and potassium hydroxide in acetone [5]. The second constituent isolated from *S. portulacastrum* was established as eupalitin from its physical and spectral behaviour [4].

EXPERIMENTAL

The plant material was identified by Miss Vir Bala Shah of Hoechst, Bombay and a voucher specimen has been deposited in the Herbarium of R. R. L. Jammu under accession No. 16091. A concd MeOH extract of dried whole plant material was successively triturated with petrol, $CHCl_3$, EtOAc and *n*-BuOH. The *n*-BuOH extract residue was charged on a Si gel column and graded elution with $CHCl_3$–MeOH mixtures yielded eupalitin (**4**) followed by the glycoside (**1**). The constituents were purified by crystallization from MeOH–EtOAc.

UV data for **1**, **4** *and* **2**. UV λ_{max}^{MeOH} nm: **1** 265, 360, 380 sh; +$AlCl_3$ 270, 300 sh, 418; +$AlCl_3$–HCl 270, 300 sh, 418; +NaOMe 272, 415; +NaOAc 265, 360, 380 sh. **4** 265, 360; +$AlCl_3$270, 368 sh, 415; +$AlCl_3$–HCl 270, 368 sh, 415; + NaOMe 272, 415. **2** 252, 362; +$AlCl_3$ 262, 421; +$AlCl_3$–HCl 260, 420, +NaOMe 263, 400.

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BERGENIN DERIVATIVES FROM *MALLOTUS JAPONICUS**

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Key Word Index—*Mallotus japonicus*; Euphorbiaceae; polyphenols; 11-*O*-galloylbergenin; 4-*O*-galloylbergenin; 11-*O*-galloyldemethylbergenin; geraniin; methylellagic acid; ^{13}C NMR.

Abstract—Three new bergenin derivatives were isolated from the bark of *Mallotus japonicus* and determined to be 11-*O*-galloylbergenin, 4-*O*-galloylbergenin and 11-*O*-galloyldemethylbergenin. The ^{13}C resonances of bergenin were fully assigned.

The polyphenol-rich bark and leaf of *Mallotus japonicus* have been used in Japan as remedies for gastric ulcer. Although we isolated two new tannins, mallotusinic acid and mallotinic acid [1], along with geraniin [2], from the leaf extract, bergenin has been the only compound isolated from the bark of this tree [3, 4]. The present investigation on the aqueous acetone extract of bark afforded three new bergenin derivatives **1**–**3**. Five known polyphenols, 3,3′,4-tri-*O*-methylellagic acid, ellagic acid, geraniin, corilagin, and (−)-epigallocatechin gallate, in addition to bergenin (**4**), were also isolated.

Compounds **1** and **2** analysed as $C_{21}H_{20}O_{13}$ were shown to be monogalloyl esters of bergenin by their ^1H, ^{13}C NMR and mass spectral data, and also by acid hydrolysis, which gave bergenin and gallic acid. The position of the galloyl group in **1** and **2** was elucidated by ^{13}C NMR. The carbon signals other than that of C-11 in the glucose moiety of bergenin, which have not been previously assigned [5], have now been assign-

*Part 2 in the series "Studies on Tannins and Polyphenols of *M. japonicus*". For Part 1 see ref. [1].

Table 1. ^{13}C NMR spectral data of bergenin and its derivatives (22.6 MHz, TMS as int. standard)

	Solvent A*			Solvent B†		
	4‡	5	6	1	2	4
C-2	81.7	81.5	81.6	81.0	83.3	83.5
C-3	70.7	70.9	70.1	72.3	70.3	72.4
C-4	73.9	74.3	84.1	75.8	76.6	76.1
C-4a	79.8	80.3	80.0	81.7	79.3	81.8
C-6	163.4	163.7	163.4	166.6	166.1	166.6
C-6a	117.9	126.5	126.4	120.1	119.7	120.1
C-7	109.8	109.4	109.4	112.1	111.9	111.8
C-8	150.9	153.2	153.2	153.2	152.9	153.2
C-9	140.6	148.2	148.3	143.2	143.1	143.1
C-10	148.0	151.1	151.3	150.0	150.0	150.3
C-10a	115.6	119.1	119.0	117.8	117.5	118.1
C-10b	72.6	71.7	71.6	74.8	74.5	74.7
C-11	61.3	61.2	61.3	65.2	62.6	63.2
OMe	59.9	60.7	60.7	61.4	61.4	61.3
—	—	61.3	61.3	—	—	—
—	—	56.0	56.1	—	—	—
—	—	—	60.4	—	—	—
C-1′	—	—	—	121.9	121.9	—
C-2′, C-6′	—	—	—	111.2	111.2	—
C-3′, C-5′	—	—	—	147.4	147.2	—
C-4′	—	—	—	140.8	140.7	—
C-7′	—	—	—	169.3	168.6	—

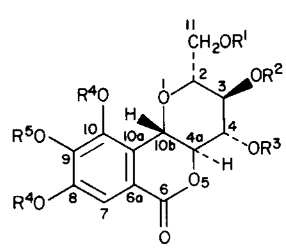
*Solvent A, $CDCl_3$–DMSO-d_6 (1:1).
†Solvent B, MeOH-d_4–DMSO-d_6 (15:1).
‡Data from ref. [5].

ed as follows. The carbons carrying free hydroxyl groups (C-3, C-4 and C-11) were unequivocally distinguished from the others (C-2, C-4a and C-10b) by the deuterium-induced differential isotope shift (DIS) measurement [6] of **4**. The former showed dual peaks of definite DIS (0.16 ppm), while the latter appeared as single peaks. Distinction of C-3 and C-4 and assignment of C-4a were based on the methylation shift [7] in the ^{13}C NMR spectra of 8, 10-di-*O*-methylbergenin (**5**) and tri-*O*-methylbergenin (**6**), which were obtained by methylation of **4** with diazomethane. Physical and spectral data of **6** were different from those of 3,8,10-tri-*O*-methylbergenin (**7**) and 8,10,11-tri-*O*-methylbergenin (**8**) [5]. Structure **6** was supported by the mass spectrum, in which a *m/z* 279 peak due to the fragment ion **A** [5] was exhibited. Confirmation of this structure was also made by the diagnostic chemical shift (δ 3.79) of the methoxyl signal on the glucose moiety in the ^1H NMR spectrum, as the C-3 and C-11 methoxyl signals in the ^1H NMR spectra of **7**, **8** and permethylated bergenin (**9**) [5] appear at δ 3.57 and 3.41, respectively, while the C-4 methoxyl signal resonates at δ 3.73. Assignment of C-2 was made referring to the reported ^{13}C NMR data of **8** [5]. Comparison of the ^{13}C NMR spectra of **1** and **2** with that of **4** based on these assignments (Table 1), with consideration of the acylation shift [8], has led to the formation of **1** and **2** as 11-*O*-galloylbergenin and 4-*O*-galloylbergenin respectively.

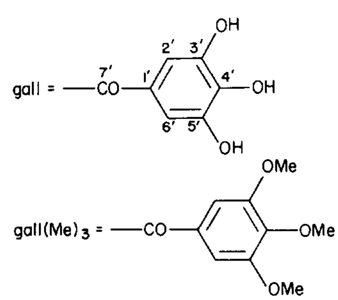
Compound **3**, $C_{20}H_{18}O_{13}$, was isolated as a minor component. The ^1H NMR spectrum of **3**, which is similar to that of **1** except for the absence of the methoxyl signal, indicates that **3** could be the demethyl derivative of **1**. Upon methylation with diazomethane, **3** gave a heptamethyl derivative (**10**), which was identified with the specimen obtained by methylation of **1**. The structure of **3** was thus elucidated as 11-*O*-galloyldemethylbergenin.

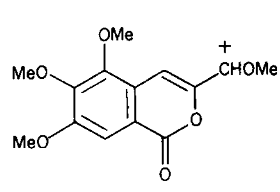
EXPERIMENTAL

Extraction and isolation. Fresh bark (1.6 kg) of *M. japonicus* Muell. Arg. collected in Okayama in June 1980 was homogenized in aq. Me_2CO (11 l.). The homogenate was filtered through Celite, the Me_2CO evaporated and the resulting aq. soln extracted with Et_2O (1 l. × 6). Evaporation of the Et_2O



1 R¹=gall, R²=R³=R⁴=H, R⁵=Me
2 R¹=R²=R⁴=H, R³=gall, R⁵=Me
3 R¹=gall, R²=R³=R⁴=R⁵=H
4 R¹=R²=R³=R⁴=H, R⁵=Me
5 R¹=R²=R³=H, R⁴=R⁵=Me
6 R¹=R²=H, R³=R⁴=R⁵=Me
7 R¹=R³=H, R²=R⁴=R⁵=Me
8 R²=R³=H, R¹=R⁴=R⁵=Me
9 R¹=R²=R³=R⁴=R⁵=Me
10 R¹=gall(Me)$_3$, R²=H, R³=R⁴=R⁵=Me





A (*m/z* 279)

extract, followed by crystallization from Me_2CO gave 3,3',4-tri-*O*-methylellagic acid (115 mg). Analyses by TLC (Si gel, C_6H_6–HOAc–dioxane, 18:1:5) and GC (TMSi ether, 1.5% SE-30, column temp. 170°, 240°) revealed the presence of gallic acid and 3,3'-di-*O*-methylellagic acid in the Et_2O extract. The aq. layer was extracted with EtOAc (1 l. × 8). The combined EtOAc layers were evaporated to give a brown residue (28 g), from which bergenin (5.3 g) was deposited on addition of MeOH. A portion (2 g) of the MeOH-soluble fraction was submitted to droplet counter-current chromatography (ascending method, *n*-BuOH–HOAc–H_2O, 4 : 1 : 5). Fractions (10 ml) were collected and every fifth fraction was monitored by A_{280} and by TLC (cellulose, 7% HOAc). Fractions 1–25 (0.16 g) gave crude ellagic acid. Fractions 26–50 (0.57 g) contained **1**, **2** and (−)-epigallocatechin gallate, amongst which **1** was crystallized from aq. MeOH and the others were separated by Sephadex LH-20 CC eluting with EtOH. Sephadex LH-20 CC of fractions 51–77 (0.42 g) gave geraniin. Fractions 78–120 (0.76 g) yielded bergenin. Evaporation of the stationary phase afforded crude corilagin. Yields of polyphenols from the EtOAc extract were **1**, 4.3%; **2**, 1.9%; ellagic acid, 3%; geraniin, 4.8%; bergenin, 33.7%; corilagin, 0.7%; (−)-epigallocatechin gallate, 1.3%. The air-dried bark (1 kg) was also treated in the same manner to give an Et_2O and an EtOAc extract (2.4 and 36.8 g respectively). Yields of polyphenols from the EtOAc extract of the dried bark were **1**, 5.2%; **2**, 4.7%; **3**, 0.4%; ellagic acid, 5%; geraniin, 1.9%; bergenin, 23.9%; corilagin, 4.5%.

1. Mp 179° (dec.) (from aq. MeOH), $[\alpha]_D^{15}$ +37.6° (EtOH; *c* 1.2), R_f 0.15 (TLC, cellulose, 7% HOAc) (Found: C, 48.55; H, 4.39. $C_{21}H_{20}O_{13} \cdot 2H_2O$ requires: C, 48.84; H, 4.68%.) UV λ_{max}^{EtOH} nm (log ϵ): 277 (4.22); IR ν_{max}^{KBr} cm^{-1}: 3300, 1710, 1630, 1610, 1510, 1230; ^1H NMR (90 MHz, Me_2CO-d_6): δ 7.22 (2H, *s*, gall-H), 7.12 (1H, *s*, H-7), 3.92 (3H, *s*, OMe); MS *m/z* (rel. int.): 328 (34), 208 (100), 237 (7), 170 (30).

2. Amorphous powder, $[\alpha]_D^{15}$ −51° (MeOH; *c* 1.0), R_f 0.36 (TLC, cellulose, 7% HOAc) (Found: C, 50.41; H, 4.31. $C_{21}H_{20}O_{13} \cdot H_2O$ requires: C, 50.61; H, 4.45%.) UV λ_{max}^{EtOH} nm (log ϵ): 277 (4.17); IR ν_{max}^{KBr} cm^{-1}: 3300, 1710, 1610, 1530, 1510, 1210; ^1H NMR (90 MHz, Me_2CO-d_6): δ 7.22 (2H, *s*, gall-H), 7.10 (1H, *s*, H-7), 3.95 (3H, *s*, OMe); MS *m/z* (rel. int.): 328 (43), 237 (10), 208 (100), 170 (12).

3. Mp 217° (dec.) (from aq. MeOH), $[\alpha]_D^{15}$ +63° (EtOH; *c* 0.7), R_f 0.04 (TLC, cellulose, 7% HOAc) (Found: C, 47.57; H, 4.04. $C_{20}H_{18}O_{13} \cdot 2H_2O$ requires: C, 47.81; H, 4.41%.) UV λ_{max}^{EtOH} nm (log ϵ): 283 (4.24); IR ν_{max}^{KBr} cm^{-1}: 3300, 1690, 1610, 1535, 1230; ^1H NMR (90 MHz, Me_2CO-d_6): δ 7.21 (2H, *s*, gall-H), 7.15 (1H, *s*, H-7); MS *m/z* (rel. int.): 314 (33), 223 (7), 194 (100), 170 (40).

All of the known polyphenols were identified by direct comparison with authentic samples.

Acid hydrolysis of **1** *and* **2**. To **1** (20 mg) in H_2O (2 ml) was added CF_3CO_2H (0.2 ml) and the mixture was refluxed for 30 hr. The residue obtained after evaporation was methylated with CH_2N_2–Et_2O at 4° for 2.5 hr. Prep. TLC (Si gel, C_6H_6–Me_2CO, 1 : 1) afforded **5** (4 mg), mp 205–207°, and methyl tri-*O*-methylgallate (4 mg), mp 79–81°, which were identified with authentic samples (mmp, ^1H NMR, IR and MS). Analogous hydrolysis of **2** gave the same products.

Preparation of 4,8,10-*tri*-*O*-*methylbergenin* (**6**). A soln of **4** (200 mg) in MeOH (20 ml) was treated with CH_2N_2–Et_2O for 4 hr at room temp. Evaporation of solvent, followed by repeated crystallization from Me_2CO–ligroin, gave colourless needles of 4,8,10-tri-*O*-methylbergenin (**6**) (140 mg), mp 154–155°, $[\alpha]_D^{15}$ −75° ($CHCl_3$; *c* 1.0) (Found: C, 55.06; H, 5.82. $C_{17}H_{22}O_9$ requires: C, 55.13; H, 5.99%.) UV λ_{max}^{EtOH} nm (log ϵ): 221 (4.44), 270 (3.88); ^1H NMR (90 MHz, $CDCl_3$): δ 7.45 (1H, *s*, H-7), 3.96, 3.93, 3.87, 3.79 (3H each, *s*, 4 OMe); MS *m/z* (rel. int.): 370 $[M]^+$ (100), 279 (11), 236 (43). This compound was also prepared in high yield (90%) by methylation with CH_2N_2 in the presence of catalytic amount of $SnCl_2$ [9].

Methylation of **3**. To a soln of **3** (18 mg) in MeOH (1 ml) was added an excess of CH_2N_2–Et_2O and the mixture kept at 4° for 15 min. The resulting residue after evaporation of solvent was purified by prep. TLC (Si gel, C_6H_6–Me_2CO, 1 : 1) to give colourless needles of **10** (10 mg), mp 204–206°, $[\alpha]_D^{15}$ −21° ($CHCl_3$; *c* 1.0) (Found: C, 55.86; H, 5.95. $C_{27}H_{32}O_{13} \cdot H_2O$ requires: C, 55.67; H, 5.88%.) UV λ_{max}^{EtOH} nm (log ϵ): 217 (4.69), 268 (4.18); ^1H NMR (90 MHz, $CDCl_3$): δ 7.33 (2H, *s*, gall-H), 7.45 (1H, *s*, H-7), 3.92 (9H, *s*), 3.90 (6H, *s*) 3.80 (6H, *s*); MS *m/z* (rel. int.): 564 $[M]^+$ (85), 279 (11), 236 (24), 212 (64), 195 (100). This product was identified with a sample prepared by treating **1** (20 mg) in Me_2CO (1 ml) with CH_2N_2–Et_2O at 4° for 15 min.

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