purchased from Aldrich Chemical Co. and **10** and **12** were prepared according to the literature.[19] ^{13}C NMR data were obtained with 50% solutions in $CDCl_3$. ^{13}C NMR data (Me_4Si standard) not in Scheme I; 1-chloro-*trans*-2-butene (**9**), C_2 127.5, C_3 130.7; 1-chloro-3-methyl-2-butene (**10**), C_2 121.0, C_3 139.0; 1-bromo-*trans*-2-butene (**11**), C_2 127.8, C_3 131.0; 1-bromo-3-methyl-2-butene (**12**), C_2 121.0, C_3 139.5; 2-methyl-1-pentene (**13**), C_1 110.1 (t), C_2 145.7 (s), C_3 40.6 (t), C_4 21.2 (t), C_5 13.9 ppm (q).

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Registry No.—**8a**, 62743-07-1; **8b**, 62743-08-2; **8c**, 62743-09-3; **9**, 4894-61-5; **10**, 503-60-6; **11**, 29576-14-5; **12**, 870-63-3; **13**, 763-29-1.

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Majusculamides A and B, Two Epimeric Lipodipeptides from *Lyngbya majuscula* Gomont

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Majusculamides A and B are major lipophilic constituents of the blue-green alga *Lyngbya majuscula* Gomont. Detailed spectral analysis, chemcial degradation, and x-ray crystallographic studies show that majusculamide A is *N*-[(2*R*)-2-methyl-3-oxodecanoyl]-D-*N*,*O*-dimethyltyrosyl-L-*N*-methylvalinamide (**6a**) and that majusculamide B is *N*-[(2*S*)-2-methyl-3-oxodecanoyl]-D-*N*,O-dimethyltyrosyl-L-*N*-methylvalinamide (**6b**). Majusculamide B is epimerized into a mixture of majusculamides A and B and then degraded into D-*N*,O-dimethyltyrosyl-L-*N*-methylvaline lactam (**2**) and racemic 2-methyl-3-oxodecanoic amide (**1**) at 140 °C in anhydrous dimethyl sulfoxide.

The blue-green alga *Lyngbya majuscula* Gomont is responsible for sporadic outbreaks of a contact dermatitis known in Hawaii as "swimmers' itch".[2] Not all varieties of this seaweed show dermonecrotic activity. *L. majuscula* from Laie Bay, Oahu, however, is frequently dermatitis-producing during the summer months. The causative agent, which is found in the lipid extract of the seaweed, may be debromoaplysiatoxin,[3] a poisonous substance that was first isolated from the digestive tract of the gastropod mollusk *Stylocheilus longicauda*.[4]

We have found that *L. majuscula* is characterized chemotaxonomically by the presence of two major lipophilic constituents which we have named majusculamides A and B. Majusculamides A and B are constituents of both the dermatitis- and nondermatitis-producing varities, but are not found in *L. gracilis*.[5a] In this report we describe the structure determination of these two nontoxic[5b] compounds.

Isolation. The alga was collected in shallow water (0.5–2 m) from several points around the island of Oahu, but mainly from Kahala Beach for the structure work. Extraction of the wet seaweed with methanol and chloroform or the freeze-dried seaweed with chloroform gave an oily extract which after chromatography and gel filtration resulted in a crystalline mixture of majusculamides A and B. Separation of the two epimeric compounds was readily achieved by high-pressure liquid chromatography.

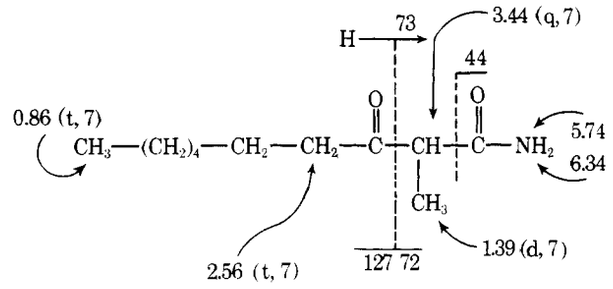
Structure Determination. Majusculamides A and B both crystallized from aqueous methanol and analyzed for $C_{28}H_{45}N_3O_5 \cdot H_2O$. Molecular ions could not be observed in their electron-impact (EI) mass spectra, but fragment ions appeared at *m/e* 486 and 487 for loss of NH_3 and NH_2 from the molecular ions and high-resolution mass measurements gave elemental compositions of $C_{28}H_{42}N_2O_5$ for the *m/e* 486 peaks. Molecular ions, however, were easily seen at *m/e* 503 in the field desorption (FD) mass spectra. The loss of NH_3 and NH_2 from the molecular ions in the EI mass spectra suggested that the majusculamides were primary amides and this was confirmed by IR and ^1H NMR.

The ^1H NMR spectra of majusculamides A and B in chloroform-*d* and dimethyl-d_6 sulfoxide at room temperature were rather complex due to the presence of two slowly interconverting conformers for each compound. At 140 °C in anhydrous dimethyl-d_6 sulfoxide, however, the ^1H NMR spectra were greatly simplified and each one clearly showed the presence of a para-substituted anisole ring, two *N*-methyl groups, three secondary methyl groups, two adjacent methine

groups with one bearing two of the secondary methyl groups, a methylene attached to a methine group, and an *n*-alkyl group attached to a carbonyl group.

Similarly the ^{13}C NMR spectra were also complex at room temperature. The 28 carbon atoms of majusculamide B, however, could be easily distinguished by determining the spectrum in anhydrous dimethyl-d_6 sulfoxide at 140 °C. One carbon was a ketone carbonyl and three carbons were amide carbonyls, in agreement with IR data and the neutrality of the compound. During the ^{13}C NMR experiments, which required over 15 h of continuous heating, appreciable darkening of the sample occurred. Gel filtration resulted in only a 50% recovery of majusculamide, which proved to be a mixture of the two epimers rather than the pure majusculamide B, and two new substances **1** and **2**, apparently products of a pyrolysis that had ensued during the heating period.

Compound **1** was optically inactive, had the molecular composition $C_{11}H_{21}NO_2$ as shown by combustion analysis and high-resolution mass spectrometry, and was easily deduced to be 2-methyl-3-oxodecanoic amide from spectral data.



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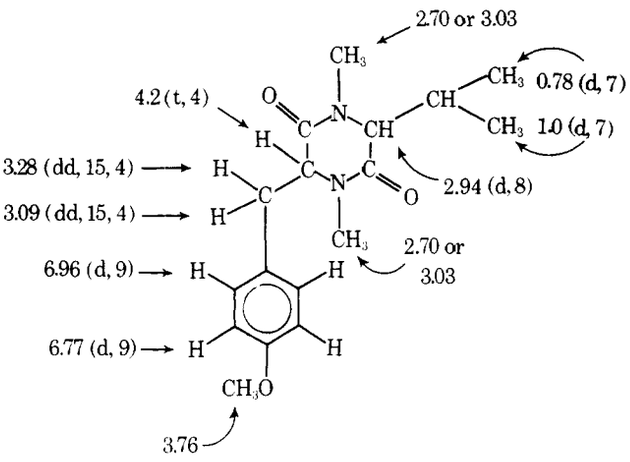
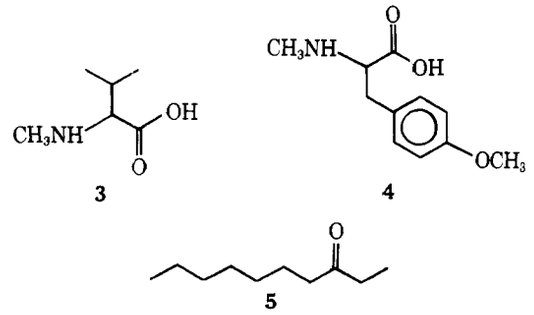
Compound **2** was optically active and analyzed for $C_{17}H_{24}N_2O_3$. Its structure was also readily elucidated from



2

spectral data as a cyclic dipeptide. Acid hydrolysis of the majusculamides to *N*-methylvaline (**3**) and *N,O*-dimethyltyrosine (**4**) confirmed the amino acid composition of **2**.

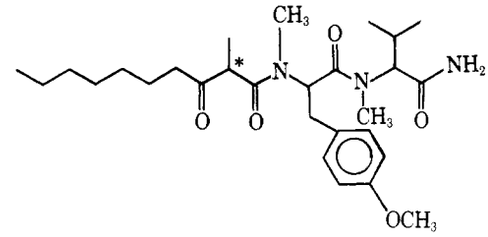


3 **4**

5

When the pyrolysis was repeated in wet dimethyl sulfoxide, only the dipeptide **2** was formed. Compound **1** could not be detected at all. Instead a volatile substance, having the same fruity odor as the acid hydrolysate of the majusculamides, was produced. The odoriferous compound, which formed a crystalline 2,4-dinitrophenylhydrazone, was shown to be 3-decanone (**5**).

Two possible skeletal structures could be written for the majusculamides and an unequivocal choice of **6** was made when partial acid hydrolysis of the majusculamides in aqueous methanol resulted in *N*-methylvaline and a mixture of **7** and the corresponding methyl ester **8**. *N,O*-Dimethyltyrosyl-*N*-methylvaline was not detected.



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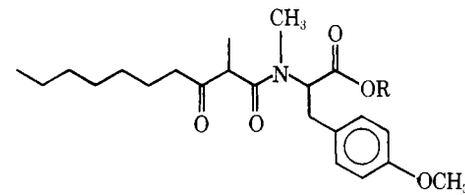
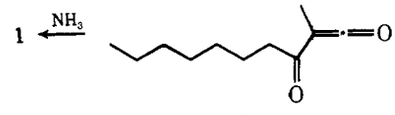
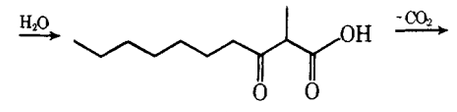


7, R = H
8, R = CH_3

To rationalize the pyrolysis in dimethyl sulfoxide, however, majusculamide B had to decompose to the cyclic dipeptide **2**, an intermediate ketene **9**, and ammonia. Combination of **9** and ammonia in the anhydrous solvent resulted in **1** whereas addition of water to the ketene in the wet solvent yielded the β-keto acid **10** which then decarboxylated to 3-decanone (**5**). Hydrolysis of majusculamide to **10** directly in the wet solvent appeared to be a less likely pathway.



9



10

Majusculamides A and B had to be α and β isomers involving the asymmetric carbon in the 3-oxodecyl side chain (* in **6**). When either majusculamide A or B was heated in dimethyl sulfoxide at 140 °C for several hours, tautomeric epimerization of this carbon occurred. The epimerization process was somewhat slow as the ^1H NMR spectra of the pure amides in Me_2SO-d_6 at 140 °C, which required about 10 min of heating, showed no visible amounts of the epimeric counterparts.

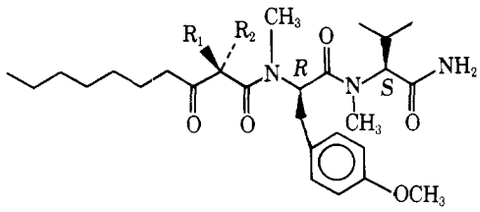
Relative Stereochemistry and X-Ray Crystallographic Studies. Crystals of majusculamide B suitable for single-crystal x-ray analysis were obtained by slow evaporation of MeOH–H_2O solutions at 0 °C. The crystal that was selected was 0.40 × 0.19 × 0.14 mm. Preliminary x-ray photographs showed orthorhombic symmetry and accurate cell constants were determined by a least-squares fit of 15 high angle re-

flections, where a = 9.027 (3), b = 12.102 (3), and c = 26.512 (9) Å. The systematic extinctions conformed to the common chiral space group $P2_12_12_1$. A density of ~1.16 g/cm^3 (flotation in aqueous zinc iodide solution) indicated one molecule per asymmetric unit.

Intensity data were collected at −100 °C with graphite monochromated Cu Kα radiation (1.54178 Å) and using an ω-scan technique and a nitrogen cold stream. All crystal data were recorded at −100 °C. A total of 2183 unique diffraction maxima with $2\theta \leq 114°$ were recorded and after correction for Lorentz, polarization, and background effects 1823 (84%) were judged observed [$I \geq 3\sigma(I)$]. The structure was solved by direct methods using a multisolution weighted tangent formula approach.[6] Full-matrix least-squares refinement with anisotropic temperature factors for the nonhydrogen atoms and isotropic temperature factors for hydrogen atoms have converged to a standard crystallographic residual of 0.031 for the observed reflections.[7] The weight of each observation is based on estimated error using counting statistics with a 2% instrumental instability factor. Figure 1 is a perspective drawing of the final x-ray model less hydrogen atoms. The single-crystal x-ray diffraction experiment could not resolve the question of absolute configuration. Further crystallographic details are available in the supplementary material.

In general all bond distances and angles agree well with generally accepted values for given bond types. The chiralities of the two amino acid α carbons [C(2) and C(7)] are opposite. The torsional angles of C(1)–C(2)–N(2)–C(6), N(2)–C(6)–C(7)–N(3), and C(6)–C(7)–N(3)–C(15) are 134.1, 61.7, and −127.8°, respectively. Therefore, C(2)-H is trans to N(2)–C(26); C(6)–O(2) is cis to C(7)–C(8); and C(7)-H is trans to N(3)–C(28). The torsional angle of C(6)–C(7)–C(8)–C(9) is −179°. There are two intermolecular hydrogen bonds between N(1)···O(1) of 2.840 Å and N(1)···O(2) of 2.890 Å. All other intermolecular approaches correspond to van der Waals interactions.

Absolute Configuration. Hydrolysis of a mixture of majusculamides A and B with refluxing 0.7 N hydrochloric acid in 30% aqueous methanol produced (+)-*N*-methylvaline and (−)-*N,O*-dimethyltyrosine. The *N*-methylvaline had an optical rotation that was identical with that reported in the literature for the L enantiomer.[8] The *N,O*-dimethyltyrosine, on the other hand, had an optical rotation that was equal in magnitude but opposite in sign to that of synthetic *N,O*-dimethyl-L-tyrosine.[9] Majusculamides A and B therefore had to have structures **6a** and **6b**, respectively.



6a, $R_1 = CH_3$; $R_2 = H$
b, $R_1 = H$; $R_2 = CH_3$

It is noteworthy to mention that when the hydrolysis of the majusculamides was carried out in stronger acid (3 N HCl), considerable racemization of the *N,O*-dimethyltyrosine, but not of the *N*-methylvaline, occurred.

Experimental Section

^{1}H and ^{13}C NMR spectra were obtained on a Varian XL-100 spectrometer equipped with a Digilab Fourier transform system; chemical shifts are reported in δ units (ppm) relative to Me_4Si (δ 0). Electron impact (EI) mass spectra were determined at 70 eV on a Varian MAT 311 high-resolution mass spectrometer; field desorption (FD) mass spectra were measured by Dr. D. Brent at the Wellcome Research Laboratories, Burroughs-Wellcome Co., Research Triangle Park, N.C. High-performance liquid chromatography was carried out on a Du Pont 830 liquid chromatograph equipped with a UV monitor and a Waters Associates R401 differential refractometer. Optical rotations were determined on a ETL-NPL (Ericsson Telephone Unlimited) automatic polarimeter. UV spectra were measured on a Cary 14 spectrophotometer and IR spectra on a Beckman IR-10 spectrophotometer.

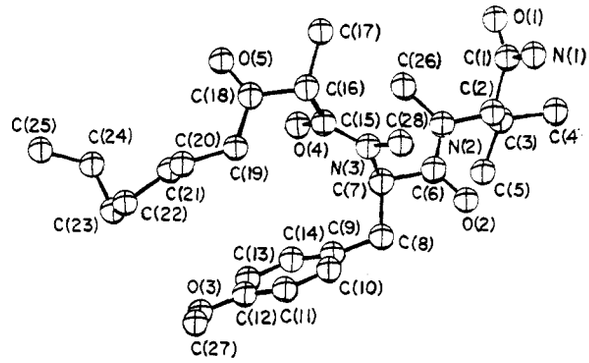


Figure 1. A computer-generated drawing of majusculamide B. Hydrogen atoms are omitted.

Isolation. *Lyngbya majuscula* was collected at Kahala Beach, Oahu, in Jan 1976 and dried at 50 °C for 20 h. The dried seaweed (915 g) was extracted twice with chloroform and twice with methanol. After evaporation of the solvents, both extracts were combined and partitioned between chloroform and water. The chloroform-soluble material, a viscous brown oil, amounted to 22.8 g.

Twenty grams of the crude extract was applied to a 5 × 55 cm column of Florisil and chromatographed with a hexane/chloroform/methanol gradient. The fraction eluted with 90% chloroform/10% methanol (6.41 g) was rechromatographed on a 3.5 × 45 cm column of silica gel using a chloroform/methanol gradient. Elution with 60% methanol/40% chloroform removed a 2.57-g fraction, 1.1 g of which was subjected to gel filtration on a 2 × 120 cm column of Sephadex LH-20 with 1:1 chloroform/methanol to give 510 mg of a black oil. This fraction was further chromatographed on neutral alumina (60 g, chloroform/methanol gradient) and finally on silica gel G (10 g, TLC grade, chloroform/methanol gradient) to yield 140 mg of a crystalline mixture of majusculamides A and B. Final separation of the two epimers was achieved by HPLC on a 1-ft column of μ-Porasil using chloroform/methanol (99.5/0.5) as eluent at a pressure of 500 psi.

Majusculamide A (**6a**) crystallized from aqueous methanol as white needles (36 mg): mp 96–97 °C; $[\alpha]^{26}D$ +19.3° (EtOH, *c* 1.14); IR (KBr) ν_{max} 3360, 3190, 1720, 1620 cm^{-1}; UV (EtOH) λ_{max} 224 nm (ϵ 7400), 277 (900), 283 (720); EI mass spectrum *m/e* 486 ($M^+ - NH_3$), 374, 290, 246, 183, 164 (base peak, $C_{10}H_{14}NO^+$), 161 ($C_{10}H_9O_2^+$), 121 ($C_8H_9O^+$); high-resolution mass measurement *m/e* 486.307978 ($C_{28}H_{42}N_2O_5$ requires 486.309381); FD mass spectrum *m/e* 503 (M^+); ^{1}H NMR (Me_2SO-d_6, 142 °C) δ 7.16 (d, J = 8.6 Hz, 2 H), 6.82 (d, J = 8.6 Hz, 2 H), 6.50 (b, 1 H), 5.65 (t, J = 7.5 Hz, 1 H), 4.45 (d, J = 10.5 Hz, 1 H), 3.79 (quartet, J = 7.0 Hz, 1 H), 3.76 (s, 3 H), 3.15 (dd, J = 14 and 7.5 Hz, 1 H), 2.99 (s, 3 H), 2.94 (s, 3 H), 2.84 (dd, J = 14 and 7.5 Hz, 1 H), 2.39 (t, J = 7.0 Hz, 2 H), 2.20 (m, 1 H), 1.50 (m, 2 H), 1.28 (m, 8 H), 1.08 (d, J = 7.0 Hz, 3 H), 0.96 (d, J = 6.5 Hz, 3 H), 0.90 (t, J = 7 Hz, 3 H), 0.72 (d, J = 6.5 Hz, 3 H).

Anal. Calcd for $C_{28}H_{45}N_3O_5 \cdot H_2O$: C, 64.5; H, 9.1; N, 8.1. Found: C, 64.1; H, 8.9; N, 8.4.

Majusculamide B (**6b**) crystallized from aqueous methanol as white needles (84 mg): mp (102–103 °C; $[\alpha]^{26}D$ +14.6° (EtOH, *c* 0.82); IR (KBr) ν_{max} 3380, 3210, 1725, 1620 cm^{-1}; UV (EtOH) λ_{max} 223 nm (ϵ 8100), 276 (840), 283 (760); EI mass spectrum identical with that of majusculamide A; ^{1}H NMR (Me_2SO-d_6, 140 °C) δ 7.16 (d, J = 8.6 Hz, 2 H), 6.82 (d, J = 8.6 Hz, 2 H), 6.50 (b, 1 H), 5.71 (t, J = 7.5 Hz, 1 H), 4.43 (d, J = 10.5 Hz, 1 H), 3.79 (quartet, J = 7.0 Hz, 1 H), 3.76 (s, 3 H), 3.17 (dd, J = 14 and 7.5 Hz, 1 H), 3.02 (s, 3 H), 2.90 (s, 3 H), 2.86 (dd, J = 14 and 7.5 Hz, 1 H), 2.21 (t, J = 7.0 Hz, 2 H), 2.19 (m, 1 H), 1.45 (m, 2 H), 1.29 (m, 8 H), 1.18 (d, J = 7.0 Hz, 3 H), 0.95 (d, J = 6.5 Hz, 3 H), 0.90 (t, J = 7 Hz, 3 H), 0.71 (d, J = 6.5 Hz, 3 H); ^{13}C NMR (Me_2SO-d_6, (140 °C) δ 205.3 (C), 171.2 (C), 171.0 (C), 170.5 (C), 158.3 (C), 130.0 (2 CH), 129.6 (C), 114.1 (2 CH), 63.2 (CH), 55.4 (CH_3), 54.6 (CH), 50.2 (CH), 39.6 (CH_2), 34.5 (CH_2), 31.0 (CH_3), 30.9 (CH_3), 30.2 (CH), 28.6 (2 CH_2), 26.3 (CH), 23.4 (CH_2), 22.0 (CH_2), 19.5 (CH_3), 18.6 (CH_3), 13.6 (CH_3), 13.0 (CH_3).

Anal. Calcd for $C_{28}H_{45}N_3O_5 \cdot H_2O$: C, 64.5; H, 9.1; N, 8.1. Found: C, 64.0; H, 8.9; N, 8.3.

Pyrolysis of the Majusculamides. A solution of 350 mg of majusculamide B in 2 mL of anhydrous Me_2SO-d_6 (100 atom %) was heated at 140 °C for 15 h to obtain a simplified ^{13}C NMR spectrum that was free of complexity due to two slowly interconverting conformers. Evaporation of the solvent gave a brown gum that was separated on a 2 × 120 cm column of Sephadex LH-20 with 1:1 $CHCl_3$/MeOH into three main fractions (monitored by UV).

Fraction 1 contained 190 mg of a crystalline mixture of majusculamides A and B.

Fraction 2 contained 90 mg of an oil, compound **2**, which slowly crystallized on standing at room temperature: mp 74–76 °C (recrystallization attempts were unsuccessful); $[\alpha]^{25}D$ −8.1° (EtOH, *c* 1.4); IR (thin film) ν_{max} 1650 cm^{-1}; UV (EtOH) λ_{max} 200 nm (ϵ 18 700), 225 (12 500), 275 (1700), 282 (1600); EI mass spectrum *m/e* 304 (M^+), 183 ($C_9H_{15}N_2O_2^+$), 121 ($C_8H_9O^+$, base peak); ^1H NMR ($CDCl_3$) δ 6.96 (d, J = 9 Hz, 2 H), 6.77 (d, J = 9 Hz, 2 H), 4.20 (t, J = 4 Hz, 1 H), 3.76 (s, 3 H), 3.28 (dd, J = 15 and 4 Hz, 1 H), 3.09 (dd, J = 15 and 4 Hz, 1H), 3.03 (s, 3 H), 2.94 (d, J = 8 Hz, 1 H), 2.70 (s, 3 H), 2.14 (m, 1 H), 1.00 (d, J = 7 Hz, 3 H), 0.78 (d, J = 7 Hz, 3 H); ^{13}C NMR ($CDCl_3$) δ 165.2 (C), 165.0 (C), 158.7 (C), 130.5 (2 CH), 126.3 (C), 113.6 (2 CH), 66.0 (CH), 62.3 (CH), 55.1 (CH_3), 36.4 (CH_2), 33.2 (CH_3), 32.4 (CH_3), 31.7 (CH), 19.1 (CH_3), 16.6 (CH_3).

Fraction 3 contained 60 mg of racemic 2-methyl-3-oxodecanoic amide (**1**) which crystallized from aqueous methanol as white needles: mp 100–101 °C; no $[\alpha]_D$ observed; IR (KBr) ν_{max} 3370, 3180, 1705, 1645 cm^{-1}; UV (EtOH) λ_{max} 199 nm (ϵ 3500), 222 sh (1100), 285 (280); EI mass spectrum *m/e* 200, 199 (M^+), 181 ($M^+ - H_2O$), 127 ($C_8H_{15}O^+$), 73 ($C_3H_7NO^+$, base peak), 72, 44 ($CONH_2^+$); high-resolution mass measurement *m/e* 199.1577 ($C_{11}H_{21}NO_2$ requires 199.1572); ^1H NMR ($CDCl_3$) δ 6.34 (b, 1 H), 5.74 (b, 1 H), 3.44 (quartet, J = 7 Hz, 1 H), 2.56 (t, J = 7 Hz, 2 H), 1.6 (bm, 2 H), 1.39 (d, J = 7 Hz, 3 H), 1.25 (bs, 8 H), 0.86 (t, J = 7 Hz, 3 H); ^{13}C NMR ($CDCl_3$) δ 209.4 (C), 172.0 (C), 54.0 (CH), 41.7 (CH_2), 31.6 (CH_2), 29.0 (2 CH_2), 23.4 (CH_2), 22.5 (CH_2), 15.1 (CH_3), 14.1 (CH_3).

Anal. Calcd for $C_{11}H_{21}NO_2 \cdot H_2O$: C, 60.6; H, 10.6; N, 6.3. Found: C, 60.8; H, 10.7; N, 6.5.

The pyrolysis was repeated by heating a solution of a 190-mg mixture of majusculamides A and B in 5 mL of wet dimethyl sulfoxide at 140 °C for 84 h. Fractionation of the reaction mixture on Sephadex LH-20 as described above gave 80 mg of impure **2**. Starting material and **1**, however, were not found. In a later fraction was found 18 mg of *p*-methoxybenzoic acid.

Hydrolysis. A. With 1 N Hydrochloric Acid. A 230-mg sample of majusculamides A and B was dissolved in 5 mL of methanol, 12 mL of 1 N HCl was added, and the solution was refluxed for 14 h. Extraction of the reaction mixture, which possessed a fruity odor, with CH_2Cl_2 yielded, after evaporation of the solvent, 80 mg of an odorless oil which was chromatographed on a column of Sephadex LH-20 (2 × 120 cm, 1:1 $CHCl_3$/MeOH) to give 18 mg of a crystalline acid **7** and 43 mg of a colorless, oily ester **8**. The odorless aqueous layer was evaporated to give 100 mg of crystalline material which was separated on a column of Sephadex G-15 (2 × 105 cm, 0.2 N acetic acid[10]) into 30 mg of *N*-methyl-L-valine (**3**) and 40 mg of *N,O*-dimethyl-D-tyrosine (**4**).

The acid, *N*-(2-methyl-3-oxodecanoyl)-*N,O*-dimethyl-D-tyrosine (a mixture of epimers), was recrystallized three times from $CHCl_3$/hexane to give white crystals: mp 93–103 °C; ^1H NMR ($CDCl_3$) δ 7.2–6.7 (m, 4 H), 5.4–4.9 (bs, 1 H), 5.38–5.14 (m, 1 H), 3.67 (s, 3 H), 3.6–2.9 (m, 3 H), 2.82 (s, 3 H), 2.5–1.8 (m, 2 H), 1.60–0.99 (bm, 13 H), 0.83 (t, 3 H); IR (KBr) ν_{max} 3300–2400 (broad), 1725, 1585 cm^{-1}; UV (EtOH) λ_{max} 225 nm (ϵ 9400), 276 (1250), 283 (1100); EI mass spectrum *m/e* 391 (M^+), 347 ($M^+ - CO_2$), 178 ($C_{10}H_{10}O_3^+$, base peak), 121 ($C_8H_9O^+$).

N-(2-Methyl-3-oxodecanoyl)-(*N,O*-dimethyl)-D-tyrosine methyl ester was a colorless oil and analysis by HPLC (1-ft μ-Porasil, 1:1 hexane/$CHCl_3$, 400 psi) showed the presence of two epimers: ^1H NMR ($CDCl_3$) δ 7.2–6.7 (m, 4 H), 5.48–5.18 (m, 1 H), 3.76 (s, 3 H), 3.72 (s, 3 H), 3.60–2.87 (m, 3 H), 2.83 (s, 3 H), 2.50–1.70 (m, 2 H), 1.6–0.99 (bm, 10 H), 1.07 (d, J = 7 Hz, 3 H), 0.84 (t, 3 H); IR (KBr) ν_{max} 1725 (broad), 1635 cm^{-1}; UV (EtOH) λ_{max} 225 nm (ϵ 11 000), 277 (1700), 284 (1600); EI mass spectrum *m/e* 405 (M^+), 346 ($M^+ - CO_2CH_3$), 192 ($C_{11}H_{12}O_3^+$, base peak), 121 ($C_8H_9O^+$).

The *N*-methyl-L-valine (**3**) had the following properties: $[\alpha]^{27}D$ +16° (H_2O, *c* 1.0) [reported[8] $[\alpha]^{15}D$ +17.5° (H_2O)]; ^1H NMR (D_2O) δ 3.15 (d, J = 5 Hz, 1 H), 2.71 (s, 3 H), 2.22 (m, 1 H), 1.03 (d, J = 6 Hz, 3 H), 1.00 (d, J = 6 Hz, 3 H); EI mass spectrum *m/e* 132 (M + 1), 131 (M^+), 88 ($C_5H_{14}N^+$), 86 ($C_5H_{12}N^+$, base peak).

The *N,O*-dimethyl-D-tyrosine (**4**) had the following properties: $[\alpha]^{25}D$ −17.1° (1 N HCl, *c* 0.82); ^1H NMR (D_2O + HCl) δ 7.25 (d, J = 9 Hz, 2 H), 6.99 (d, J = 9 Hz, 2 H), 4.27 (t, J = 7 Hz, 1 H), 3.80 (s, 3 H), 3.27 (d, J = 7 Hz, 2 H), 2.74 (s, 3 H); EI mass spectrum *m/e* 209 (M^+), 164 ($M - CO_2$), 121 ($C_8H_9O^+$, base peak), 88 ($C_3H_6NO_2^+$).

B. With 3 N Hydrochloric Acid. A 250-mg mixture of majusculamides A and B was hydrolyzed in 5 mL of refluxing 3 N HCl for 20 h. The hydrolysate was extracted with ether and the dried ethereal layer was carefully evaporated in the cold. The residual oil was treated with 2,4-dinitrophenylhydrazine to give an orange semicrystalline solid. Gel filtration on Sephadex LH-20 with 1:1 methanol/chloroform yielded 17 mg of 3-decanone 2,4-dinitrophenylhydrazone: orange needles from aqueous methanol, mp 53–54 °C (reported[11] 55.5–56.5 °C); EI mass spectrum[12] *m/e* (rel intensity) 336 (31), 301 (13), 252 (15), 178 (22), 83 (100); high-resolution mass measurement *m/e* 336.1789 ($C_{16}H_{24}N_4O_4$ requires 336.1797; ^1H NMR ($CDCl_3$) δ 11.2 (b, 1 H), 9.13 (d, J = 3 Hz, 1 H), 8.31 (dd, J = 10 and 3 Hz, 1 H), 7.95 (dd, J = 10 and 1.5 Hz, 1 H), 2.45 (q, J = 7 Hz, 2 H), 2.41 (t, J = 7 Hz, 2 H), 1.8–1.2 (b, 10 H), 1.22 (t, J = 7 Hz, 3 H), 0.88 (bt, J = 7 Hz, 3 H).

The aqueous phase was evaporated, redissolved in water, and reevaporated to remove excess HCl. The crystalline residue was applied to a column of Sephadex G-15 (2 × 105 cm) and eluted with 0.2 N acetic acid to give 20 mg of *N*-methyl-L-valine, $[\alpha]^{25}D$ +16° (H_2O, *c* 1.0), and 28 mg of partially racemized *N,O*-dimethyl-D-tyrosine, $[\alpha]^{25}D$ −8.1° (1 N HCl, *c* 2.7).

When the hydrolysis of majusculamide was carried out in refluxing 6 N HCl for 12 h, a small amount of *p*-hydroxybenzaldehyde, an orange solid that was identified by its ^1H NMR spectrum, was identified in the reaction mixture. Chromatography on silica gel G achieved the separation of the amino acids from the *p*-hydroxybenzaldehyde.

Synthesis of *N,O*-Dimethyl-L-tyrosine. L-Tyrosine was acetylated[13] and the *N*-acetyl-L-tyrosine was converted to *N*-acetyl-*O*-methyl-L-tyrosine with dimethyl sulfate[14] using previously described procedures. The *N*-acetyl protecting group was removed[14] and the resulting *O*-methyl-L-tyrosine was methylated to *N,O*-dimethyl-L-tyrosine following the procedure of Corti.[15] After purification on Sephadex G-15 as described above, it showed an optical rotation of $[\alpha]^{25}D$ +17.5° (1 N HCl, *c* 4.85).[9]

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Registry No.—**1**, 62758-03-6; **2**, 62758-04-7; **3**, 2480-23-1; **4**, 62758-05-8; **6a**, 62758-06-9; **6b**, 62840-08-8; **7** epimer 1, 62796-01-4; **7** epimer 2, 62796-02-5; **8** epimer 1, 62758-07-0; **8** epimer 2, 62758-08-1; 2,4-dinitrophenylhydrazine, 119-26-6; 3-decanone DNP, 62758-09-2; L-tyrosine, 60-18-4; *N*-acetyl-L-tyrosine, 537-55-3; *N*-acetyl-*O*-methyl-L-tyrosine, 28047-05-4; *N,O*-dimethyl-L-tyrosine, 52939-33-0.

Supplementary Material Available. The fractional coordinates (for unnatural enantiomer) and temperature factors (Table I), bond distances (Table II), and bond angles (Table III) for majusculamide B and the 100-MHz ^1H NMR spectra of majusculamides A and B in chloroform-*d* at 30 °C (Figure 2, A and B) and in dimethyl-d_6 sulfoxide at 140 °C (Figure 2, C and D) (4 pages). Ordering information is given on any current masthead page.

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1,2-Diphenylmaleyl, a Protecting Group for Amino Functions

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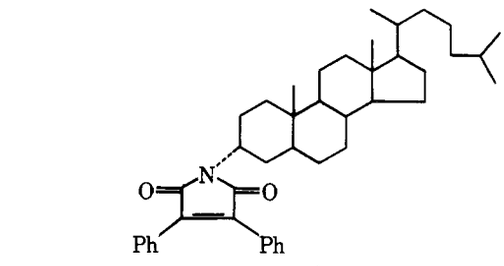
1,2-Diphenylmaleyl (DPM) is described as a protecting group for amino functions and was applied to carbohydrates and steroids. DPM derivatives are prepared through the condensation of the corresponding amines with 1,2-diphenylmaleic anhydride and removed by ethanolic hydrazine. DPM proves to be compatible with and a useful protecting group in glycoside synthesis.

Phthaloyl has been a particularly advantageous protecting group for amino functions in peptide chemistry,[1] penicillin chemistry,[2] and in many areas of natural product chemistry. Diphenylmaleyl (DPM) derivatives, introduced here as a protecting group for amino functions, bear many similarities to the corresponding phthaloyl derivatives, including, to some extent, the protection and deprotection steps; DPM derivatives differ, however, in that they are yellow and fluorescent and thus can be easily determined quantitatively and followed chromatographically; their increased volume compared to many other protecting groups for amino functions can affect adjacent functional groups and they can be conveniently modified into reactive protecting groups as will be described in the subsequent paper.

Compounds **1**, **2**, and **3** were conveniently prepared by heating 1,2-diphenylmaleic anhydride[3] and the corresponding amine in dimethylformamide or in dimethylformamide–toluene. Compound **3** in turn was acetylated in pyridine to give compound **4** ($J_{1,2}$ = 8 Hz) in a very good yield. The selective formation of the β-tetraacetate **4** could be explained by the possibility that compound **3**, owing to the steric effect of the DPM group, could be mostly the β anomer (this is supported by the low optical rotation) and by the less hindered approach of the acetylating reagent from the β (e) side. In the Fischer glycoside synthesis both the α and the β anomers (compounds **5**, and **6**, respectively) are formed, the β being the more abundant. It is pertinent to note that in this case the α anomer moves faster in TLC and is less polar than the β anomer. In addition, the NMR spectrum of compound **5** suggests that it might be present in chloroform solution as an equilibrium mixture of conformers rather than the $^4C_1$ chair conformer.

Compound **4** was treated with hydrobromic acid in acetic acid to yield the intermediate 1-bromo derivative (the DPM group appears to be unaffected even after 48 h under these conditions). Subsequent condensation of the bromo intermediate with methanol in the presence of mercuric cyanide afforded the β-glycoside **7** ($J_{1,2}$ = 9 Hz). A number of factors could govern the anomeric nature of compound **7**. At present, the anomeric composition of the bromo intermediate is not known and also it is not clear whether neighboring group participation of the DPM groups could take place; obviously, the easier approach for a nucleophile would be from the β side.

Compound **7** was correlated with compound **6** by deacetylation and by acetylation of compound **6**.



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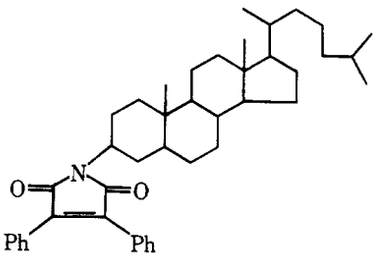

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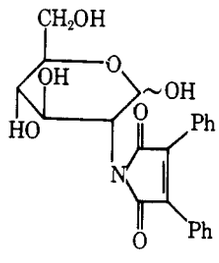

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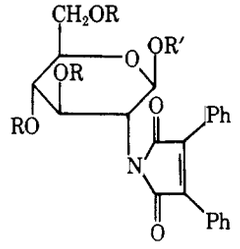
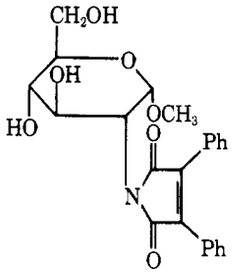


4, R = R′ = CH_3CO
6, R = H; R′ = CH_3
7, R = CH_3CO; R′ = CH_3



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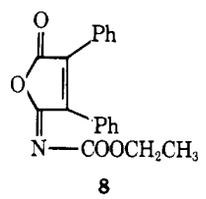

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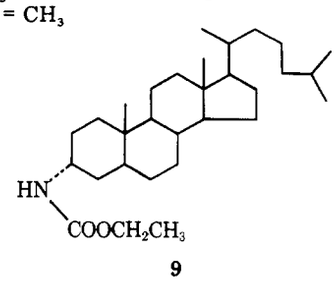

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