Tetrahedron Letters,Vol.24,No.20,pp 2087-2090,1983 0040-4039/83/202087-04$03.00/0
Printed in Great Britain

INDOLE ALKALOIDS FROM THE MARINE RED ALGA MARTENSIA FRAGILIS

Michael P. Kirkup and Richard E. Moore*

Department of Chemistry, University of Hawaii,
Honolulu, Hawaii 96822

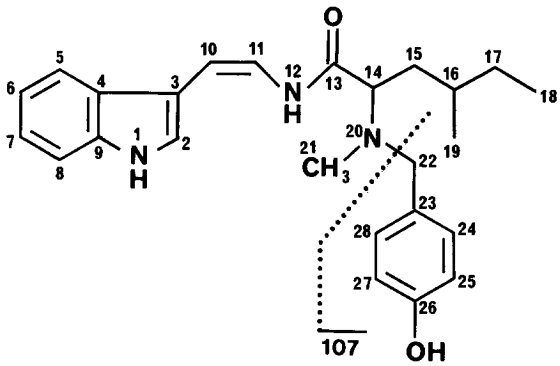
Abstract. The gross structures of fragilamide and martensines A and B, the first basic indole alkaloids to be isolated from a marine eukaryotic plant, are reported.

Martensia fragilis, a red alga belonging to the family Delesseriaceae, has a foul odor reminiscent of skatole. The odoriferous principle has not been identified, but three constituents of the essential oil are indole-3-carboxaldehyde, p-(hydroxymethyl)phenol, and p-(methoxymethyl)phenol.[1] The lipid extract of this alga, however, contains several novel indole alkaloids. Three of the compounds are fragilamide (1), martensine A (2), and martensine B (3) and represent the first basic indole alkaloids to be isolated from a eukaryotic marine alga.[2]

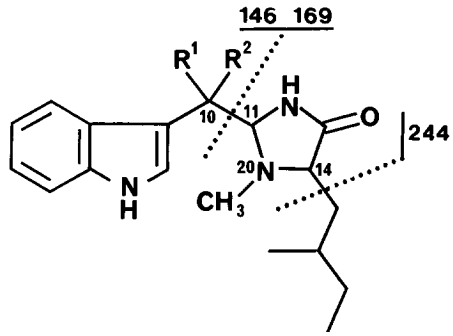
The seaweed was collected at Black Point and Mokuleia, Oahu. The algal extract (6.4 g) was partitioned between hexane and MeOH/H_2O (9:1). The MeOH/H_2O layer was adjusted in concentration to 2:1 and extracted with $CHCl_3$. Gel filtration of the $CHCl_3$ extract on Sephadex LH-20 with $CHCl_3$/MeOH (3:2) gave alkaloidal fractions A and B. Chromatography of the faster-moving fraction A on silica gel (1:7 acetone/CH_2Cl_2) followed by HPLC on partisil 10 (1:1 EtOAc/CH_2Cl_2) gave 540 mg of fragilamide A (1). Reverse-phase HPLC of the slower-moving fraction B on partisil 10 ODS (2:9 H_2O/MeOH) gave 25 mg of martensine B (3), mp 184-186°, and 140 mg of martensine A (2).

Fragilamide,[3] $[\alpha]_D$ -32° (c 0.62, MeOH), was a labile 3° amine that rapidly autoxidized in solution. The high resolution MS indicated a molecular formula $C_{25}H_{31}N_3O_2$ and the IR spectrum showed OH and NH bands (3385, 3290, 3120 cm^{-1}) and an amide carbonyl band (1640 cm^{-1}). Catalytic hydrogenation (30% Pd/C, MeOH, 2 hr) gave 10,11-dihydrofragilamide[4] (4) which underwent hydrogenolysis to p-cresol and N-methylhomoisoleucyltryptamine (5) after 24 hr. Acid hydrolysis of 5 (6N HCl, reflux 24 hr, N_2) produced tryptamine and (-)-N-methylhomoisoleucine (6)[5] which were separated on silica gel with n-BuOH/AcOH/H_2O (4:1:1).

Inspection of the 100 MHz ^1H NMR spectrum of fragilamide showed signals for a 3-substituted indole unit, a N-methylhomoisoleucyl unit, and a p-hydroxybenzyl group. The broad singlet at δ 10.52 was assigned to the phenolic OH and the broad doublet at δ 9.35 to an amide NH; the signal for the indole NH, however, was obscured by the aromatic signals. Spin-spin decoupling experiments indicated that the amide NH was connected to a cis disubstituted carbon-carbon double bond (δ 6.90 and 5.94, J = 8 Hz), which in turn was connected to the

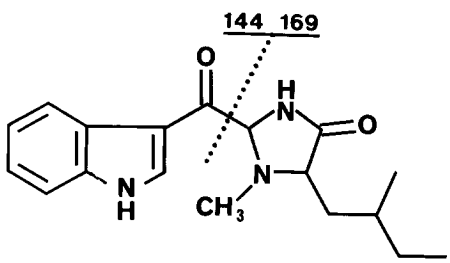


1

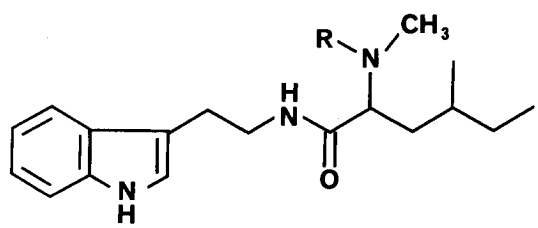


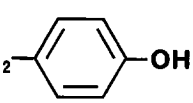
2 R^1 = OH ; R^2 = H

7 R^1 = H ; R^2 = OH

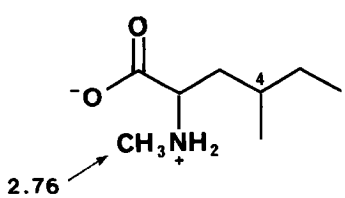


3



4 R = CH_2– 

5 R = H



6

indole unit at C-3 (br d at δ 5.94, assigned to H-10, shows 1 Hz coupling to br s at δ 7.48, assigned to H-2).

Chemical evidence for the enamide group was obtained by photooxidation[6] of fragilamide. When the alkaloid in MeOH was exposed to oxygen, strong sunlight, and the photosensitizer eosin bound to Amberlite IR 400 ion exchange resin for 2 hr, indole 3-carboxyaldehyde and p-(methoxymethyl)phenol were produced. The structure of fragilamide was therefore 1.

Martensine A[7], $[\alpha]_D$ +42° (c 0.34, MeOH), had an elemental composition, $C_{18}H_{25}N_3O_2$, by high resolution MS. The UV showed absorptions at 260, 280, and 288 nm, typical for an indole. Comparison of the ^{13}C and ^1H NMR spectra with those of skatole showed that martensine A was also a 3-substituted indole. When the alkaloid was treated with MeOH/HOAc at RT for several hours, an O-methyl derivative was obtained (m/e 329, 3H s at δ 3.22), indicating the presence of a benzylic OH (3300 cm^{-1}). Hydrolysis with 1N HCl (reflux, N_2) produced N-methylhomoisoleucine. The IR showed a carbonyl band at 1720 cm^{-1}, attributed to a 5-membered lactam ring. These data indicated that the structure of martensine A was 2.

Martensine B[8], $[\alpha]_D$ -18° (c 1.1, acetone), exhibited a small molecular ion peak at m/z 313 in its field desorption MS, but huge fragment ion peaks at m/z 169 and 144. The UV was similar to that of indole-3-carboxaldehyde and the IR showed two carbonyl bands at 1710 (γ-lactam) and 1675 cm^{-1} (aryl ketone). These data suggested that the structure of martensine B was 3. The structure was confirmed when $NaBH_4$ reduction led to 2 and 10-epimartensine A (7)[9] which could be separated by LC on silica gel (EtOAc). Compound 7 is a minor alkaloid in the alga.

Acknowledgement. This research was supported by NSF Grants CHE76-82517 and CHE79-25416. ^1H NMR studies at 360 MHz were carried out at Stanford Magnetic Resonance Laboratory under the auspices of NSF Grant GP-23633 and NIH Grant RR00711.

REFERENCES AND NOTES

1. R. E. Moore, Acc. Chem. Res., 10, 40 (1977).
2. Martensine A shows antibiotic activity against Bacillus subtilis, Staphylococcus aureus, and Mycobacterium smegmatis.
3. Spectral data for 1. UV (EtOH): λ_{max} 229 nm (ε 15,500), 280 (10,200). EIMS m/z (rel. int.): 405.242 (0.8, M^+), 299 (59), 220 (18), 158 (66), 130 (40), 114 (100). ^{13}C NMR (CD_3OD) δ 172.96 (s, C-13), 157.46 (s, C-26), 137.56 (s, C-9), 131.25 (s, C-24 and C-28), 130.95 (s, C-23), 128.85 (s, C-4), 123.64 (d), 123.15 (d), 120.47 (d), 120.10 (d), 119.94 (d), 116.07 (d, C-25 and C-27), 112.55 (d, C-8), 111.49 (s, C-3), 104.18 (d, C-10), 64.64 (d, C-14), 59.36 (t, C-22), 37.17 (q, C-21), 32.30 (d, C-16), 32.28 (t, C-15), 29.26 (t, C-18), 19.46 (q, C-19), 11.89 (q, C-18). ^1H NMR (acetone-d_6): δ 10.52 (br s, OH), 9.35 (br d, H-12), 7.65 (dd, H-5), 7.53 (dd, H-8), 7.48 (br s, H-2), 7.16 (m, H-6 and H-7), 6.94 (d, H-24 and H-28), 6.90 (dd, H-11), 6.62 (d, H-25 and H-27), 5.94 (dd, H-10), 3.49 (s, 2H on C-22), 3.32 (dd, H-14), 2.12 (s, N-CH_3), 2.0-1.0 (m, 5H on C-15, C-16, C-17), 0.90 (t, 3H on C-18), 0.86 (d, 3H on C-19); 'H-'H J (Hz) 2-10 = 1, 10-11 = 8, 11-12 = 10, 14,15 = 8.5, 14-15' = 3, 16-19 = 6.5, 17-18 = 6.5, 24-25 = 27-28 = 8.

4. Spectral data for 4. ^{1}H NMR (acetone-d_6): δ 8.32 (s, OH), 3.64 (q, 2H-11), 2.98 (t, 2H-10); J (Hz) 10-11 = 6.
5. (-)-Homoisoleucine has been isolated from the seeds of Aesculus californica [L. Fowden and A. Smith, Phytochemistry, 8, 809 (1967)]. It is an L amino acid, but its stereochemistry at C-4 is unknown.
6. C. S. Foote and J. W-P. Lin, Tetrahedron Lett., 3267 (1968); J. E. Huber, ibid., 3271 (1968).
7. Spectral data for 2. UV (EtOH): λ_{max} 223 nm (ε 20,900), sh 257 (5200), sh 268 (5500), 280 (5800), 288 (5200). IR (CCl_4): ν_{max} 3300, 1720 cm^{-1}. EIMS m/z (rel. int.): 315 (2), 297.1838 ($C_{18}H_{23}N_3O$, 1), 244 (5), 169.1342 ($C_9H_{17}N_2O$, 100), 146 (6), 145 (13), 144 (15). ^{13}C NMR (CD_3OD): δ 177.63 (s), 137.99 (s), 127.43 (s), 123.99 (d), 122.41 (d), 120.47 (d), 119.68 (d), 115.36 (d), 112.19 (d), 81.55 (d), 72.74 (d), 66.84 (d), 40.95 (q), 39.47 (t), 31.62 (d), 30.45 (t), 19.90 (q), 11.45 (q). ^{1}H NMR (acetone-d_6): δ 10.01 (br s, H-12), 7.732 (dt, H-5), 7.412 (dt, H-8), 7.353 (s, H-2), 7.115 (ddd, H-7), 7.019 (ddd, H-6), 6.92 (br s, H-1), 4.883 (br d, H-10); 4.288 (dd, H-11), 3.007 (ddd, H-14), 2.231 (s 3H-21); 1.810 (br m, H-16), 1.715 (dt, H-15); 1.45 (dqd, H-17), 1.41 (ddd, H-15'), 1.17 (dq, H-17'), 0.915 (d, 3H-19), 0.898 (t, 3H-18); 'H-'H J (Hz) 1-2 = 0, 2-10 = 1, 5-6 = 8.0, 5-7 = 1.4, 5-8 = 1, 6-7 = 7.0, 6-8 = 1, 10-11 = 4.8, 11,12 = 0, 11-14 = 1.4, 14-15 = 4.8, 14-15' = 6.6, 15-15' = -13.8, 15-16 = 7.8, 15'-16 = 5.8, 16-17 = 5.0, 16-17' = 7.4, 17-17' = -13.8, 16-19 = 6.7, 17-18 = 7.4.
8. Spectral data for 3. UV: λ_{max} 215 nm (ε 10,000), 244 (11,000), 261 (8400), 303 (10,000) in MeOH → λ_{max} 267 nm (10,000), 335 (10,000) in 1N NaOH in MeOH. IR (KBr): ν_{max} 3190, 1710, 1675 cm^{-1}. FDMS m/z (rel. int.): 313 (5), 295 (8), 169 (100), 144 (40). ^{1}H NMR (acetone-d_6): δ 8.58 (s, H-2), 8.37 (dd, H-5), 7.64 (br s, H-1), 7.58 (dd, H-8), 7.28 (m, H-6 and H-7), 4.69 (d, H-11), 3.00 (td, H-14), 2.55 (s, 3H-21), 2.0-1.0 (m, 5H on C-15, C-16, C-17), 0.90 (d, 3H-19), 0.88 (t, 3H-18).
9. Spectral data for 7. ^{1}H NMR (acetone-d_6): δ 7.697 (dt, H-5), 7.386 (dt, H-8), 7.336 (s, H-2), 7.095 (ddd, H-7), 7.004 (ddd, H-6), 6.93 (br, H-1), 4.906 (dd, H-10), 4.388 (dd, H-11), 2.942 (ddd, H-14), 2.544 (s, 3H-21), 1.647 (br m, H-16), 1.555 (dt, H-15), 1.375 (dqd, H-17), 1.18 (ddd, H-15'), 1.07 (dq, H-17'), 0.821 (t, 3H-18), 0.727 (d, 3H-19); 'H-'H J (Hz) essentially same as for 2, even 10-11 = 4.9.

(Received in USA 21 February 1983)