A New Apocarotenoid from the Marine Shellfish *Mytilus coruscus*

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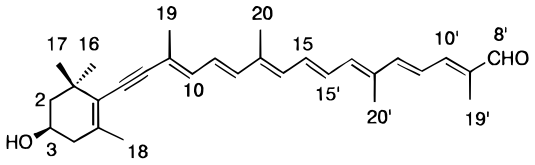
A new apocarotenoid (**1**) isolated from the marine shellfish *Mytilus coruscus* was determined to be (3*R*)-3-hydroxy-7,8-didehydro-8′-apo-*β*-caroten-8′-al by chemical and spectroscopic data.

During carotenoid studies on marine shellfishes,[1,2] the new acetylenic apocarotenoid **1** was isolated from the Japanese sea mussel *Mytilus coruscus* Gould (Mytilidae). This paper reports the isolation and structure elucidation of **1**.

The Me_2CO extract from *M. coruscus* was chromatographed on Si gel using increasing percentages of Me_2CO in *n*-hexane. Successive purification by HPLC on ODS afforded the new carotenoid (**1**).



Apoalloxanthinal (1)

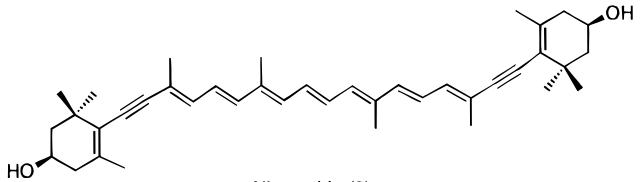
An HREIMS of **1** showed a molecular ion peak at *m*/*z* 430.2860, compatible with the formula $C_{30}H_{38}O_2$. Compound **1** showed absorption maxima at 427 (sh), 450, and 470 nm. On reduction with $NaBH_4$ in MeOH, **1** gave a more polar product, having absorption maxima at 405, 427, and 454 nm. These spectral properties were compatible with the presence of a halocynthiaxanthin-type chromophore in **1**.[3] The IR spectrum of **1** was consistent with the presence of a hydroxy group (3300 cm^{-1}), an acetylenic group (2160 cm^{-1}), and a conjugated carbonyl group (1660 cm^{-1}). Acetylation of **1** in dry pyridine with Ac_2O at room temperature produced a monoacetate. The presence of an aldehydic proton was revealed by a ^{1}H-NMR signal at δ 9.46.

The ^{1}H-NMR data for **1** in $CDCl_3$ are presented in Table 1. Assignments were made by a ^{1}H−^{1}H COSY experiment and by comparing these data with those of alloxanthin (**2**)[4] and *β*-apo-8′-carotenal.[5] The ^{1}H-NMR data for **1** indicated the presence of the partial structure of alloxanthin (H-2 to H-20) and *β*-apo-8′-carotenal (H-8′ to H-20′). Furthermore, the COSY experiment revealed the following proton−proton connectivities: H-2 to H-4, H-19 to H-12, H-20 to H-20′, and H-12 to H-19′ as shown in Table 1. Thus, the structure of **1** was determined as 3-hydroxy-7,8-didehydro-8′-apo-*β*-caroten-8′-al and was designated apoalloxanthinal. The CD spectrum of **1** showed almost the same Cotton effect as that of alloxanthin (**2**),[1] which has 3*R* and 3′*R* chiralities. Therefore, the chirality at C-3 of **1** was postulated to be *R*.

Apoalloxanthinal (**1**) was also isolated from the Japanese oyster *Crassostrea gigas* Thunberg (Ostreidae). It is assumed to be an oxidative cleavage metabolite of alloxanthin (**2**).

Table 1. ^{1}H-NMR Data of Apoalloxanthinal (**1**) in $CDCl_3$

H	δ	mult.	*J* (Hz)	^{1}H−^{1}H COSY correlated H
H-2ax	1.46	d,d	12.5, 12.5	H2eq, H3
H-2eq	1.84	d,d,d	12.5, 4, 1.5	H2ax, H3, H4eq
H-3	3.99	m		H2ax, H2eq, H4ax, H4eq
H-4ax	2.09	d,d	18.5, 9	H3, H4eq
H-4eq	2.43	d,d,d	18.5, 5.5, 1.5	H4ax, H3, H2eq
H-10	6.47	d,d	11.5, 1.0	H11, H19
H-11	6.57	d,d	15, 11.5	H10, H12
H-12	6.38	d	15	H11
H-14	6.30	d	11	H15, H20
H-15	6.77	d,d	14, 11	H14, H15′
CH_3-16	1.15	s		
CH_3-17	1.20	s		
CH_3-18	1.93	s		
CH_3-19	2.01	s		H10
CH_3−20	1.99	s		H14
H-15′	6.69	d,d	11.5, 5.5	H15, 14′
H-14′	6.46	d	11	H15′, 20′
H-12′	6.74	d	15	H11′
H-11′	6.68	d,d	15, 10	H-12′, 10′
H-10′	6.95	d	10	H-11′, H-19′
H-8′	9.46	s		
CH_3-19′	1.91	s		H-10′
CH_3-20′	2.01	s		H-14′



Alloxanthin (2)

Experimental Section

General Experimental Procedures. The UV−vis and CD spectra were recorded in Et_2O at room temperature with a Shimadzu UV-240 spectrophotometer and a JASCO J-500C spectropolarimeter, respectively. The IR spectra were recorded with a Hitachi 260−30 on KBr disk. The EIMS spectra were recorded using a Hitachi M-80 mass spectrometer with a direct inlet system of ionization energy of 70 eV at 190−200 °C. The ^{1}H-NMR spectra were measured with a Nicolet NT-360 (360 MHz) and Varian XL-300 (300 MHz) instruments in $CDCl_3$ with TMS as internal standard. HPLC was performed on a Shimadzu LC-6AD instrument with a Shimadzu SPD-6AV spectrophotometer set at 450 nm. The column used was a Shim-Pack PREP-ODS (Shimadzu, 20 mm × 250 mm, 5 μm) with a mobile phase of CH_3CN−CH_2Cl_2 (8:2).

Animal Material. *M. corusucus* and *C. gigas* were purchased at the fish market in Toba City, Mie, Japan. They were identified by Dr. Katsura Ooyama for Toba

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Aquarium. Voucher specimens are deposited at Kyoto Pharmaceutical University.

Extraction and Isolation of Carotenoids. The Me_2CO extract of muscle (5 kg) of *M. coruscus* was partitioned between *n*-hexane−Et_2O (1:1) and aqueous NaCl. The organic layer was dried over Na_2SO_4, then concentrated to dryness. The residue was subjected to column chromatography on Si gel using an increasing percentage of Me_2CO in *n*-hexane. Compound **1** was eluted with Me_2CO−*n*-hexane (2:3) from a Si gel column and was further purified by HPLC on ODS with CH_3CN−CH_2Cl_2 (8:2) to yield 2.0 mg (3% of the total carotenoid).

The following additional carotenoids were identified from *M. coruscus*: astaxanthin (0.5% of the total carotenoid); 7,8-didehydroastaxanthin (0.2%); 7,8,7′,8′-tetradehydroastaxanthin (0.2%); 4-ketoalloxanthin (0.5%); pectenolone (1%); diatoxanthin (9%); alloxanthin (20%); mytiloxanthin (23%); halocynthiaxanthin (4%); pyrrhoxanthinol (2%); pectenol A (16%); pectenol B (4%); 4-hydroxyalloxanthin A (2%); 4-hydroxyalloxanthin B (1%); crassostreaxanthin B (4%), heteroxanthin (2%), and peridininol (1%).

In a similar manner to that described above, compound **1** (1.0 mg) was also obtained from 5 kg of the edible part of *C. gigas*.

Apoalloxanthinal (1): HREIMS *m/z* [M^+] 430.2860 ($C_{30}H_{38}O_2$ requires 430.2872); EIMS (70 eV) *m/z* [M^+] 430 (100), 368 (17), 255 (20), 119 (42), 95 (35), 43 (51); IR (KBr) 3300, 2160, 1660, 960 cm^{-1}; vis (Et_2O) λ_{max} 427 (sh), 450, 470 nm (% III/II = 5); CD (Et_2O) λ_{ext} 360 nm ($\Delta\epsilon$ 0), 330 (0.4), 310 (0), 280 (−1.6), 255 (0), 240 (−1.5); ^1H NMR ($CDCl_3$, 360 MHz), see Table 1.

Reduction of 1. A solution of **1** (0.3 mg) in 5 mL MeOH was treated with $NaBH_4$ (5 mg) for 30 min at room temperature to give a product (0.4 mg) that showed vis (Et_2O) λ_{max} 405, 427, 454 nm (% III/II = 70); EIMS *m/z* [M^+] 432 (100), 414 (20), 221 (40), 95 (30), 43 (60).

Acetylation of 1. A solution of **1** (0.3 mg) in dry pyridine (3 mL) was treated with 3 mL of Ac_2O for 60 min at room temperature to give a product that showed vis (Et_2O) λ_{max} 427 (sh), 450, 470 nm (%III/II = 5); EIMS *m/z* [M^+] 472 (100), 412 (20), 221 (40), 119 (50), 95 (45), 43 (70).

Alloxanthin (2): isolated from *M. coruscus*; CD (Et_2O) λ ext 360 nm ($\Delta\epsilon$ 0), 350 (0.5), 325 (0), 285 (−2.0), 255 (0), 240 (−3.5).[1]

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References and Notes

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