

Review

## New and Rare Carotenoids Isolated from Marine Bacteria and Their Antioxidant Activities

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**Abstract:** Marine bacteria have not been examined as extensively as land bacteria. We screened carotenoids from orange or red pigments-producing marine bacteria belonging to rare or novel species. The new acyclic carotenoids with a C<sub>30</sub> aglycone, diapolycopenedioid acid xylosylesters A–C and methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate, were isolated from the novel Gram-negative bacterium *Rubritalea squalenifaciens*, which belongs to phylum Verrucomicrobia, as well as the low-GC Gram-positive bacterium *Planococcus maritimus* strain iso-3 belonging to the class Bacilli, phylum Firmicutes, respectively. The rare monocyclic C<sub>40</sub> carotenoids, (3*R*)-saproxanthin and (3*R*,2'*S*)-myxol, were isolated from novel species of Gram-negative bacteria belonging to the family Flavobacteriaceae, phylum Bacteroidetes. In this review, we report the structures and antioxidant activities of these carotenoids, and consider relationships between bacterial phyla and carotenoid structures.

**Keywords:** diapolycopenedioid acid xylosylesters A–C; methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate; (3*R*)-saproxanthin; (3*R*,2'*S*)-myxol; antioxidant activity

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## 1. Introduction

Some species of bacteria, yeast, and fungi, as well as algae and higher plants, synthesize a large number of carotenoids with different molecular structures, and more than 750 carotenoids with different structures have been isolated from natural sources [1]. Many beneficial pharmaceutical effects of carotenoids have recently been reported. Therefore, evaluating the pharmaceutical potentials of various carotenoids may represent an interesting field in medical research. However, the number of carotenoid species that have been examined for this purpose has been limited, and has included C<sub>40</sub> carotenoids possessing skeletons composed of 40 carbon atoms, such as dicyclic carotenoids, e.g.,  $\beta$ -carotene,  $\alpha$ -carotene,  $\beta$ -cryptoxanthin, zeaxanthin, lutein, canthaxanthin, astaxanthin, and fucoxanthin, and the acyclic carotenoid lycopene [2–8]. Difficulties have been associated with identifying natural sources to supply sufficient amounts of new or rare carotenoids, with the exception of carotenoids that can be isolated from a species of higher plants or algae or chemically synthesized. It has therefore been desirable to find cultivable bacteria that produce new or rare carotenoids, since they can easily be reproduced.

Marine bacteria have not been examined as extensively as land bacteria. Thus, the Marine Biotechnology Institute Co., Ltd. (MBI, Kamaishi, Japan) was established in December, 1988, and continued to isolate novel or rare marine bacteria until March, 2008, the number of which reached more than ten thousand [9–12]. Many bacteria have been shown to produce dicyclic or monocyclic C<sub>40</sub> carotenoids, in addition to some acyclic C<sub>30</sub> carotenoids with a 30 carbon skeleton [1,13]. The MBI isolated new or rare dicyclic C<sub>40</sub> carotenoids with the  $\beta$ -carotene ( $\beta,\beta$ -carotene) skeleton from Gram-negative marine bacteria belonging to the class  $\alpha$ -Proteobacteria, phylum Proteobacteria, e.g., astaxanthin glucoside from *Paracoccus* sp. strain N81106 (re-classified from *Agrobacterium aurantiacum*) [14], 2-hydroxyastaxanthin from *Brevundimonas* sp. strain SD212 [15], and 4-ketonestoxanthin 3'-sulfate from *Erythrobacter* sp. strain. PC6 (re-classified from *Flavobacterium* sp. PC-6; MBIC02351) [16]. These marine bacteria were also able to produce astaxanthin [17]. The carotenoid biosynthesis gene clusters of these marine bacteria have been elucidated in detail [17–19].

The generation of free radicals has been suggested to play a major role in the progression of a wide range of pathological disturbances, including myocardial and cerebral ischemia [20], atherosclerosis [21], renal failure [22], inflammation [23], and rheumatoid arthritis [24]. The subsequent peroxidative disintegration of cells and organelle membranes has also been implicated in various pathological processes [25]. Carotenoid pigments, which have been shown to possess strong antioxidant activities, have been attracting increasing attention due to their beneficial effects on human health, e.g., their potential to prevent diseases such as cancer and cardiovascular diseases [26].

We have attempted to identify novel or rare types of carotenoids from yellow or red pigment-producing marine bacteria that were classified to belong to rare or novel species by 16S rRNA analyses since 2002. The results of this screening led to the isolation of diapolyycopenediols xylosylesters A–C (new carotenoids) from *Rubritalea squalenifaciens* [27,28], methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate (a new carotenoid) from *Planococcus maritimus* [29], and (3*R*)-saproxanthin and (3*R*,2'*S*)-myxol (rare carotenoids) from a novel species belonging to the family

Flavobacteriaceae [30]. In this review, we report the structures and antioxidant activities of these carotenoids, and consider relationships between bacterial phyla and carotenoid structures.

## 2. Results

### 2.1. Diapolycopenediolic Acid Xylosylesters A–C from *Rubritalea Squalenifaciens* [27,28]

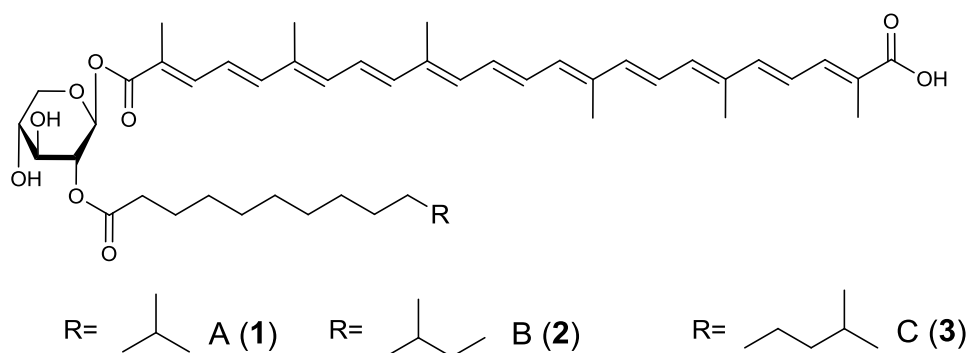
A yellow pigment-producing bacterium (strain HOact23<sup>T</sup>) that was found to produce squalene was isolated from the homogenate of the marine sponge *Halicondria okadai*, which had been collected from the Miura peninsula (Kanagawa, Japan), and was subsequently classified as a novel species in the genus *Rubritalea*, belonging to phylum Verrucomicrobia, based on 16S rRNA gene sequence data. The name proposed for the new taxon was *Rubritalea squalenifaciens* [31], with the type strain HOact23<sup>T</sup> (=MBIC08254<sup>T</sup> = DSM 18772<sup>T</sup>).

*R. squalenifaciens* was cultured in 100 mL of medium (1.0% starch, 0.4% yeast extract, and 0.2% peptone in seawater) in a 500 mL Erlenmeyer flask at 30 °C on a rotary shaker at 120 rpm for 2 days, and the carotenoids produced were purified from the cells using chromatographic methods (EtOAc/H<sub>2</sub>O partition → silica gel column chromatography CH<sub>2</sub>Cl<sub>2</sub>–MeOH (20:1) → preparative silica gel HPLC CH<sub>2</sub>Cl<sub>2</sub>–MeOH (15:1) → preparative ODS HPLC (MeOH)). Three carotenoids were purified from cells in the 42-liter culture (diapolycopenediolic acids xylosylesters A (**1**) 10.2 mg, B (**2**) 3.0 mg, and C (**3**) 2.2 mg, respectively). The structures of compounds **1–3** were determined by HRESI-MS and spectroscopic (UV-Vis, NMR (1D and 2D investigations on <sup>1</sup>H and <sup>13</sup>C nuclei), and [α]<sub>D</sub>) analyses as shown in Figure 1. Compounds **1–3** were all new carotenoids.

Compounds **1–3** possessed diapolycopenediolic acid (C<sub>30</sub> carotenoid) [32,33] as their aglycone. Diapolycopenediolic acid glucosyl ester and diapolycopenediolic acid diglucosyl were previously shown to be carotenoids that possessed diapolycopenediolic acid as the aglycone [32]. Compounds **1–3** were the first carotenoids to include 2-acetyl-D-xylose in their structures.

The antioxidant activity of compound **1** was evaluated using <sup>1</sup>O<sub>2</sub> suppression activity. Its IC<sub>50</sub> was 5.1 μM (the IC<sub>50</sub> values of astaxanthin and β-carotene were 8.9 μM and >100 μM, respectively).

**Figure 1.** The structures of diapolycopenediolic acids A (**1**), B (**2**) and C (**3**).



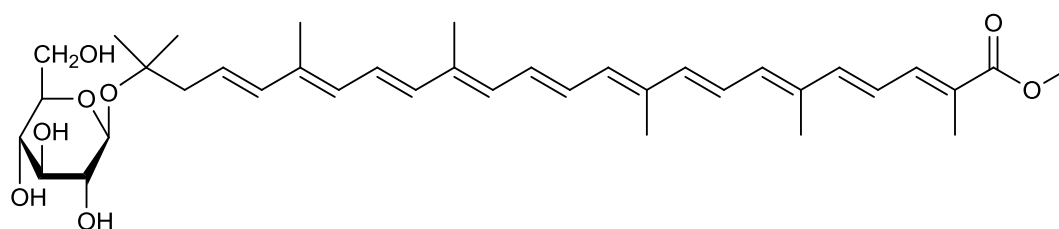
### 2.2. Methyl 5-Glucosyl-5,6-Dihydro-Apo-4,4'-Lycopenoate from *Planococcus Maritimus* [29]

A yellow pigment-producing bacterium (strain iso-3), which was found to be solvent-tolerant, was isolated as an orange-pigmented colony from the microbial analysis of a sample derived from an

intertidal sediment from the Clyde estuary, UK. The 16S rRNA gene sequence of strain iso-3 was the most similar to that of type strain *Planococcus maritimus* (99.5 as a similarity score, and 96.4 as an s\_ab score, from the Sequence match analysis of RDP), which belongs to the class Bacilli, phylum Firmicutes, and was identified as *Planococcus maritimus* strain iso-3.

Strain iso-3 was cultured in 100 mL of medium (Marine Broth 2216, Difco, Sparks, MD, USA) in a 500 mL Erlenmeyer flask at 30 °C on a rotary shaker at 120 rpm for 1 day, and the carotenoid produced was purified from the alkaline-digested cells using chromatographic methods (EtOAc/H<sub>2</sub>O partition → silica gel column chromatography (CH<sub>2</sub>Cl<sub>2</sub>–MeOH (10:1) → preparative silica gel HPLC CH<sub>2</sub>Cl<sub>2</sub>–MeOH (10:1) → preparative ODS HPLC (96% MeOH)). A total of 2.5 mg of pure methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate (**4**) was obtained from the cells in the 18-liter culture, and the structure of compound **4** was determined by HRESI-MS and spectroscopic (UV-Vis, NMR (1D and 2D investigations on <sup>1</sup>H and <sup>13</sup>C nuclei), and [α]<sub>D</sub>) analyses, as shown in Figure 2. Compound **4** was a new carotenoid. Compound **4** possessed 5,6-dihydro-5-hydroxy-apo-4,4'-lycopene-4'-oic acid (C<sub>30</sub> carotenoid) as its aglycone. Although 4,4'-diapocarotene-4-oic acid [32] was previously reported to be a related C<sub>30</sub> carotenoid aglycone, 5,6-dihydro and 5-hydroxy functions in the aglycone of compound **4** were demonstrated for the first time. The antioxidant activity of compound **4** was evaluated using <sup>1</sup>O<sub>2</sub> suppression activity, and its IC<sub>50</sub> value was 5.1 μM. We previously described the isolated carotenoid as methyl glucosyl-3,4-dihydro-apo-8'-lycopenoate [29], but confirmed its structure as methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate, as shown in this review. Corrigenda is currently being prepared for the previous study.

**Figure 2.** The structure of methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate (**4**).



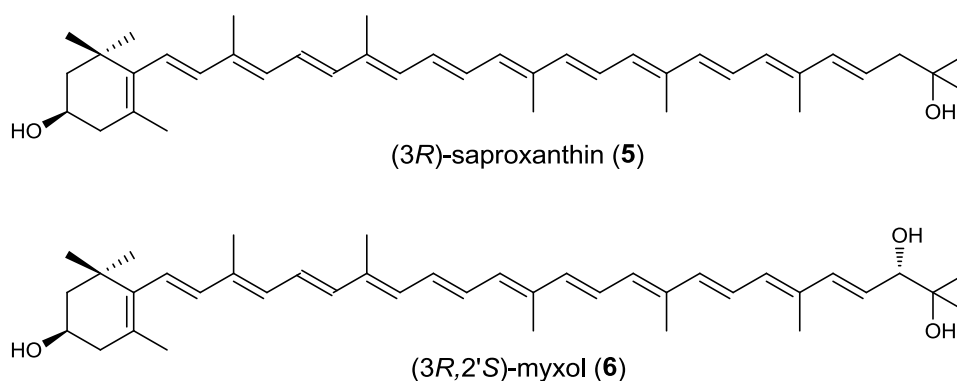
### 2.3. (3R)-Saproxanthin and (3R,2'S)-Myxol [30]

Strain 04OKA-13-27 (MBIC08261) was isolated from the dense mats of filamentous algae from within the territory of damselfish (*Stegastes nigricans*). Strain YM6-073 (MBIC06409) was isolated from a sediment sample collected 0.1 m below the surface of the sea by cultivating for 30 days on an HSV medium. The two marine bacteria, which had been collected off the coast of Okinawa Prefecture, were classified on the basis of this 16S rRNA gene sequences. A similarity search in the databases of the DNA Data Bank of Japan (DDBJ) and RNA Database Project II (RDPII) showed the 16S rRNA gene sequences of the both strains (04OKA-13-27 and YM6-073) to be 96.5% (1408 bp/1459 bp) similar to *Stanierella latercula* ATCC 23177<sup>T</sup>, 95.5% (1324 bp/1386 bp) similar to *Gaetbulimicrobium brevivittae* strain SMK-19<sup>T</sup>, and 94.2% (1306 bp/1386 bp) similar to *Robiginitalea biformata* strain HTCC2501<sup>T</sup>. The phylogenetic relationship between these strains was deduced with already known species in the family Flavobacteriaceae. The result obtained revealed that the two bacterial strains should be classified as novel species of the family Flavobacteriaceae.

Both 04OKA-13-27 and YM6-073 were cultured in 100 mL of medium (Marine Broth 2216, Difco) in a 500 mL Sakaguchi flask at 30 °C on a rotary shaker at 100 rpm for 1 day, and the carotenoids produced were each purified from the cells using chromatographic methods (EtOAc/H<sub>2</sub>O partition → silica gel column chromatography hexane–ethyl acetate (2:1) → preparative silica gel high performance thin layer chromatography (HPTLC; Merck, Darmstadt, Germany) CH<sub>2</sub>Cl<sub>2</sub>–MeOH (10:1) → preparative ODS HPLC (MeOH)). A total of 0.3 mg (04OKA-13-27) and 0.5 mg (YM6-073) of pure carotenoids were obtained from the cells of each 2 liter culture, and the carotenoids were identified as (3*R*)-saproxanthin (04OKA-13-27) (**5**) and (3*R*,2'*S*)-myxol (YM6-073) (**6**) by MS, <sup>1</sup>H-NMR, and CD analyses, respectively (Figure 3).

The antioxidative activities of compounds **5** and **6** were examined using rat brain homogenate model. Compounds **5** and **6** showed potent antioxidant activities (IC<sub>50</sub> 2.1 μM (**5**) and 6.2 μM (**6**)) (IC<sub>50</sub> 10.9 μM (β-carotene)).

**Figure 3.** The structures of (3*R*)-saproxanthin (**5**) and (3*R*,2'*S*)-myxol (**6**).



### 3. Discussion

The MBI has isolated approximately 1000 pigment-producing marine bacteria. We selected 10 strains, which were identified as rare or novel species by 16S rRNA, including strain HOact23<sup>T</sup> (*Rubritalea squalenifaciens* sp. nov., phylum Verrucomicrobia), strain iso-3 (*Planococcus maritimus*, the class Bacilli, phylum Firmicutes), strain 04OKA-13-27 (novel species of the family Flavobacteriaceae), and strain YM6-073 (novel specie of the family Flavobacteriaceae) from these isolated bacteria.

We found two-type new C<sub>30</sub> carotenoids diapolyconedioc acid xylosylesters (compound **1–3**) from HOact23<sup>T</sup> and methyl 5-glucosyl-5,6-dihydro-apo-4,4'-lycopenoate (compound **4**) from iso-3 through the isolation and structural analyses of carotenoids produced by these strains. Acyclic C<sub>30</sub> carotenoids were previously shown to be contained in land bacteria including *Staphylococcus aureus*, belonging to the class Bacilli, and the methanotrophs *Methylobacterium rhodium* (formerly *Pseudomonas rhodos*), belonging to the class α-Proteobacteria, and *Methylomonas* sp., belonging to the class γ-Proteobacteria [17,34]. Thus, acyclic C<sub>30</sub> carotenoids are likely to widely exist in domain bacteria (prokaryotes), *i.e.*, they are present not only in some low-GC Gram-positive bacteria, but also in some phyla in Gram-negative bacteria. The strong singlet-oxygen-quenching activities of our C<sub>30</sub>

carotenoids also indicated that such C<sub>30</sub> carotenoids are promising as functional carotenoids, although these *in vivo* functional analyses have not yet been conducted.

We isolated two rare monocyclic C<sub>40</sub> carotenoids with one 3-hydroxy- $\beta$ -ring ((3*R*)-saproxanthin (compound **5**) from 04OKA-13-27 and (3*R*,2'*S*)-myxol (compound **6**) from YM6-073), which belong to the family Flavobacteriaceae, phylum Bacteroidetes. (3*R*)-Saproxanthin has only previously been detected from *Saprospira grandis*, which belongs to the family Saprospiraceae, phylum Bacteroidetes [35]. Hence, marine bacterial strain 04OKA-13-27 was the second species to produce saproxanthin. (3*R*,2'*S*)-Myxol has only previously been detected in marine bacterial strain P99-3 (MBIC03313), belonging to the family Flavobacteriaceae [15], and in the cyanobacterium *Anabaena variabilis* ATCC 29413, phylum Cyanobacteria [36]. Therefore, marine bacterial strain YM6-073 was the third species to produce myxol. Myxoxanthophyll (myxol 2'-fucoside), which is widely distributed in phylum Cyanobacteria, contains myxol as its aglycone. These findings indicated that such monocyclic C<sub>40</sub> carotenoids with one 3-hydroxy- $\beta$ -ring exist in phylum Bacteroidetes as well as phylum Cyanobacteria.

The carotenoids produced by the six other strains isolated were all zeaxanthin, which is a common carotenoid in domain bacteria. Our study may be effective for identifying rare and new carotenoids based on its ratio (4/10). In addition, all the rare and new carotenoids (**1–6**) isolated possessed potent antioxidant activities.

#### 4. Conclusions

Marine bacteria are likely to produce carotenoids to protect themselves from activated oxygen produced by sunlight (mainly <sup>1</sup>O<sub>2</sub>); therefore, their potent antioxidant activities were expected and reasonable. Therefore, the techniques performed in our study effectively identified new antioxidant carotenoids.

#### Author Contributions

Kazutoshi Shindo performed the experiments and wrote the text; Norihiko Misawa supervised the project and corrected the manuscript.

#### Conflicts of Interest

The authors declare no conflict of interest.

#### References

1. Britton, G.; Liaaen-Jensen, S.; Pfander, H. *Carotenoids Handbook*; Birkhäuser Verlag: Basel, Switzerland, 2004.
2. Nishino, H.; Tokuda, H.; Murakoshi, M.; Satomi, Y.; Masuda, M.; Onozuka, M.; Yamaguchi, S.; Takayasu, J.; Tsuruta, J.; Okuda, M.; *et al.* Cancer prevention by natural carotenoids. *Biofactors* **2000**, *13*, 89–94.

3. Iwamoto, T.; Hosoda, K.; Hirano, R.; Kurata, H.; Matsumoto, A.; Miki, W.; Kamiyama, M.; Itakura, H.; Yamamoto, S.; Kondo, K. Inhibition of low-density lipoprotein oxidation by astaxanthin. *J. Atheroscler. Thromb.* **2000**, *7*, 216–222.
4. Krinsky, N.I.; Landrum, J.T.; Bone, R.A. Biological mechanism of the protective role of lutein and zeaxanthin in the eye. *Annu. Rev. Nutr.* **2003**, *23*, 171–201.
5. Naito, Y.; Uchiyama, K.; Aoi, W.; Hasegawa, G.; Nakamura, N.; Yoshida, N.; Maoka, T.; Takahashi, J.; Yoshikawa, T. Prevention of diabetic nephropathy by treatment with astaxanthin in diabetic db/db mice. *Biofactors* **2004**, *20*, 49–59.
6. Hosokawa, M.; Kudo, M.; Maeda, H.; Kohno, H.; Tanaka, T.; Miyashita, K. Fucoxanthin induces apoptosis and enhances the antiproliferative effect of PPARgamma ligand, troglitazone, on colon cancer cells. *Biochim. Biophys. Acta* **2004**, *1675*, 113–119.
7. Talegawkar, S.A.; Johnson, E.J.; Carithers, T.C.; Taylor, H.A., Jr.; Bogle, M.L.; Tucker, K.L. Carotenoid intakes, assessed by food-frequency questionnaires (FFQs), are associated with serum carotenoid concentrations in the Jackson Heart Study: Validation of the Jackson Heart Study Delta NRI Adult FFQs. *Public Health Nutr.* **2008**, *11*, 989–997.
8. Sugiura, M.; Nakamura, M.; Ogawa, K.; Ikoma, M. High serum carotenoids associated with lower risk for bone loss and osteoporosis in post-menopausal Japanese female subjects: Prospective cohort study. *PLoS One* **2012**, *7*, e52643.
9. Katsuta, A.; Adachi, K.; Matsuda, S.; Shizuri, Y.; Kasai, H. *Ferrimonas marina* sp. nov. *Int. J. Syst. Evol. Microbiol.* **2005**, *55*, 1851–1855.
10. Tao, L.; Yao, H.; Kasai, H.; Misawa, N.; Cheng, Q. A carotenoid synthesis gene cluster from *Algoriphagus* sp. KK10202C with a novel fusion-type lycopene- $\beta$ -cyclase gene. *Mol. Genet. Genomics* **2006**, *276*, 79–86.
11. Peng, X.; Adachi, K.; Chen, C.; Kasai, H.; Kanoh, K.; Shizuri, Y.; Misawa, N. Discovery of a marine bacterium producing 4-hydroxybenzoate and its alkyl esters, parabens. *Appl. Environ. Microbiol.* **2006**, *72*, 5556–5561.
12. Matsuo, Y.; Katsuta, A.; Matsuda, S.; Shizuri, Y.; Yokota, A.; Kasai, H. *Mechercharimyces mesophilus* gen. nov., sp. nov. and *Mechercharimyces asporophorigenens* sp. nov., antitumor substance-producing marine bacteria, and description of *Thermoactinomycetaceae* fam. nov. *Int. J. Syst. Evol. Microbiol.* **2006**, *56*, 2837–2842.
13. Goodwin, T.M. *The Biochemistry of Carotenoids*, 2nd ed.; Plant Chapman and Hall: London, UK, 1980; Volume 1, pp. 291–319.
14. Yokoyama, A.; Adachi, K.; Shizuri, Y. New carotenoid glucosides, astaxanthin glucoside and adonixanthin glucoside, isolated from the astaxanthin-producing marine bacterium, *Agrobacterium aurantiacum*. *J. Nat. Prod.* **1995**, *58*, 1929–1933.
15. Yokoyama, A.; Miki, W.; Izumida, H.; Shizuri, Y. New trihydroxy-keto-carotenoids isolated from an astaxanthin-producing marine bacterium. *Biosci. Biotechnol. Biochem.* **1996**, *60*, 200–203.
16. Yokoyama, A.; Izumida, H.; Shizuri, Y. New carotenoid sulfates isolated from a marine bacterium. *Biosci. Biotechnol. Biochem.* **1996**, *60*, 1877–1878.
17. Misawa, N. Carotenoids. In *Comprehensive Natural Products II Chemistry and Biology*; Mander, L., Liu, H.-W., Eds.; Elsevier: Oxford, UK, 2010; Volume 1, pp. 733–753.

18. Misawa, N.; Satomi, Y.; Kondo, K.; Yokoyama, A.; Kajiwarra, S.; Saito, T.; Ohtani, T.; Miki, W. Structure and functional analysis of a marine bacterial carotenoid biosynthesis gene cluster and astaxanthin biosynthetic pathway proposed at the gene level. *J. Bacteriol.* **1995**, *177*, 6575–6584.
19. Nishida, Y.; Adachi, K.; Kasai, H.; Shizuri, Y.; Shindo, K.; Sawabe, A.; Komemushi, S.; Miki, W.; Misawa, N. Elucidation of a carotenoid biosynthesis gene cluster encoding a novel enzyme, 2,2'- $\beta$ -hydroxylase, from *Brevundimonas* sp. strain SD212 and combinatorial biosynthesis of new or rare xanthophylls. *Appl. Environ. Microbiol.* **2005**, *71*, 4286–4296.
20. Traysman, R.J.; Kirsch, J.R.; Koehler, R.C. Oxygen radical mechanisms of brain injury following ischemia and reperfusion. *J. Appl. Physiol.* **1991**, *71*, 1185–1195.
21. Palinski, W.; Rosenfeld, M.E.; Yla, H.S.; Gurtner, G.C.; Socher, S.S.; Butler, S.W.; Parthasarathy, S.; Carew, T.E.; Steinberg, D.; Witztum, J.L. Low density lipoprotein undergoes oxidative modification *in vivo*. *Proc. Natl. Acad. Sci. USA* **1989**, *86*, 1372–1376.
22. Erdogan, C.; Unlucerci, Y.; Turkmen, A.; Kuru, A.; Cetin, O.; Bekpinar, S. The evaluation of oxidative stress in patients with chronic renal failure. *Clin. Chim. Acta* **2002**, *322*, 157–161.
23. Cheeseman, K.H.; Forni, L.G. An investigation of the novel anti-inflammatory agents ONO-3144 and MK-447. Studies on their potential antioxidant activity. *Biochem. Pharmacol.* **1988**, *37*, 4225–4233.
24. Bodamyali, T.; Kanczler, J.M.; Millar, T.M.; Stevens, C.R.; Blake, D.R. Free radicals in rheumatoid arthritis: Mediators and modulators. *Oxid. Stress Dis.* **2004**, *10*, 591–610.
25. Mylonas, C.; Kouretas, D. Lipid peroxidation and tissue damage. *In Vivo* **1999**, *13*, 295–309.
26. Van den Berg, H.; Faulks, R.; Granado, H.F.; Hirschberg, J.; Olmedilla, B.; Sandmann, G.; Southon, S.; Stahl, W. The potential for the improvement of carotenoid levels in foods and the likely systemic effects. *J. Sci. Food Agric.* **2000**, *80*, 880–912.
27. Shindo, K.; Mikami, K.; Tamesada, E.; Takaichi, S.; Adachi, K.; Misawa, N.; Maoka, T. Diapolyycopendioic acid xylosyl ester, a novel glycol-C<sub>30</sub>-carotenoic acid produced by a new marine bacterium *Rubritalea squalenifaciens*. *Tetrahedron Lett.* **2007**, *48*, 2725–2727.
28. Shindo, K.; Asagi, E.; Sano, A.; Hotta, E.; Minemura, N.; Mikami, K.; Tamesada, E.; Misawa, N.; Maoka, T. Diapolycopenedioic acid xlosyl esters A, B, and C, novel antioxidative glycol-C<sub>30</sub>-carotenoic acids produced by a new marine bacterium *Rubritalea squalenifaciens*. *J. Antibiot.* **2008**, *61*, 185–191.
29. Shindo, K.; Endo, M.; Miyake, Y.; Wakasugi, K.; Morritt, D.; Bramley, M.P.; Fraser, D.P.; Kasai, H.; Misawa, N. Methyl glucosyl-3,4-dehydro-apo-8'-lycopenoate, a novel antioxidative glycol-C<sub>30</sub>-carotenoic acid produced by a marine bacterium *Planococcus maritimus*. *J. Antibiot.* **2008**, *61*, 729–735.
30. Shindo, K.; Kikuta, K.; Suzuki, A.; Katsuta, A.; Kasai, H.; Yasumoto-Hirose, M.; Matsuo, Y.; Misawa, N.; Takaichi, S. Rare carotenoids, (3R)-saproxanthin and (3R,2'S)-myxol, isolated from novel marine bacteria (*Flavobacteriaceae*) and their antioxidative activities. *Appl. Microbiol. Biotechnol.* **2007**, *74*, 1350–1357.
31. Kasai, H.; Katsuta, A.; Sekiguchi, H.; Matsuda, S.; Adachi, K.; Shindo, K.; Yoon, J.; Yokota, A.; Shizuri, Y. *Rubritalea squalnifaciens* sp. nov., a squalene-producing marine bacterium belonging to subdivision 1 of the phylum “Verrucomicrobia”. *Int. J. Syst. Evol. Microbiol.* **2007**, *57*, 1630–1634.



32. Kleing, H.; Schmitt, R.; Meister, W.; Englert, G.; Thommen, H. New carotenoic acid glucosyl esters from *Pseudomonas rhodos*. *Z. Naturforsch C* **1979**, *34*, 181–185.
33. Osawa, A.; Iki, K.; Sandmann, G.; Shindo, K. Isolation and identification of 4,4'-diapolycopene-4, 4'-dioic acids produced by *Bacillus firmus* GB1 and its singlet oxygen quenching activity. *J. Oleo Sci.* **2013**, *62*, 955–960.
34. Tao, L.; Schenzle, A.; Odom, J.M.; Cheng, Q. Novel carotenoid oxidase involved in biosynthesis of 4,4'-diapolycopene dialdehyde. *Appl. Environ. Microbiol.* **2005**, *71*, 3294–3301.
35. Aasen, A.J.; Liaaen-Jensen, S. The carotenoids of flexibacteria: II. A new xanthophyll from *Saprospira grandis*. *Acta Chem. Scand.* **1966**, *20*, 811–819.
36. Takaichi, S.; Mochimaru, M.; Maoka, T. Presence of free myxol and 4-hydroxymyxol and absence of myxol glycosides in *Anabaena variabilis* ATCC 29413, and proposal of biosynthetic pathway of carotenoids. *Plant Cell Physiol.* **2006**, *47*, 211–216.

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