[S4-3] [4/20/2001(Fri) 11:00-11:30/Hall B]

Marine Microorganisms From The Chemical Viewpoint: A New Biomedical Resource For New Drug Discovery

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1. Introduction

Marine organism comprise over half a million species. Due to their living environment very unusual as compared with terrestrial organisms, marine organisms metabolite and produce a variety of substances which often have various unprecedented chemical structures. In recent years, an increasing number of marine natural products have been reported and reviewed.**1** Some of the marine natural products isolated have not only served as potential lead compounds for clinically useful drugs but actually used a chemical probes useful for basic studies in the fields of life sciences.**2**

Marine microorganisms are receiving increasing attention as sources of bioactive compounds, and expanded research can be expected in this area. Marine natural product research is now focusing more on marine microorganisms, mainly bacteria and fungi that can be cultured.**3,4** The marine fungi, particularly those associated with marine animals and plants, appear to be an unusually reach resource for secondary metabolites.

In this paper, I will present our recent investigations on bioactive metabolites isolated from the marine microalgi and the algicolous marine fungi.

2. Bioactive metabolites of marine microalgi.

(1) Marine diatom *Nitzschia* sp.

In order to screen new radical scavenging principle which is expected to be antiaging drug lead, we have investigated 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity of the marine microalgi, green algae (10 species), diatom (10 species) and cyanobacteria (10 species). The significant activities (IC50: < 100 g/mL) were observed in 4 species of green algae (MA002, 006, 009, 010), 1 species of diatom (MA015) and 5 species of cyanobacteria (MA017, 018, 019, 024, 025). Within the scope of family tested, MA009 (IC50 = 78 g/mL), MA015 (IC50 = 38 g/mL) and MA019 (IC50 = 41 g/mL) displayed the most significant activity. Among the marine microalgi tested at family level, *Cyanophycean* microalgae was shown to be the most active family on screening of new bioactive compounds.**5**

Allenic and epoxycarotenoid, fucoxanthin (**1**) was isolated from the marine bacillariophycean microalga *Nitzschia sp.* and the structure was assigned on the basis of comprehensive spectroscopic analyses. Fucoxanthin was detected only from diatom among three families (green algae, diatom and cyanobacterium) of the marine microalgi tested. Therefore, the fucoxanthin (**1**) is a good chemotaxonomic marker for three families tested. Fucoxanthin (**1**) showed a radical scavenging activity against 1,1-diphenyl-2-picrylhydrazyl (DPPH), peroxynitrite (ONOO-), and nitric oxide radical (.NO) with IC_{50} values of 32 M, 60 M, and 0.5 M respectively.**6**

Two new diacyl galactolipids I (**2**) and II (**3**) have been isolated from the marine

bacillariophycean microalga *Nitzschia* sp. and their structures were determined as (2*S*)-3-*O*-β -D-galactopyranosyl-1,2-di-*O*-(9Z-hexadecenoyl)glycerol (**2**) and (2*S*)-3-*O*-β -D-galactopyranosyl-1-*O*-(5Z,8Z,11Z,14Z,17Z-eicosapenta-enoyl)-2-*O*-(9Z-hexa decenoyl) glycerol (**3**) by physicochemical evidence.

Diacyl galactolipids II (**3**) has the 3-polyunsaturated fatty acid, eicosapentaenoic acid (EPA) which plays important roles in biological functions of life-processes. Therefore, the biological significance of these metabolites is of interest.**7**

(2) Marine cyanobacterium *Oscilaltoria* sp.

In a study on the bioactive metabolites from the marine microalgae, we have investigated the secondary metabolites from the marine cyanobacterium, *Oscillatoria* sp. (strain #, MA25) and isolated 2-*O*-(α -D-glucopyranosyl)glycerol (**4**), which was detected only from cyanobacterium among three families of the marine microalgae. Therefore, it might be used as a chemotaxonomic marker.**8**

From the other species of cyanobacterium, *Oscillatoria* (strain #, MA18), we recently isolated new glycolipids, **5**, **6**, and inseparable **7**/**8**.**9**

A new diacylgalactolipid **9** has been isolated from the another marine cyanobacterium *Oscillatoria* sp. (strain #, MA20), and the structure was elucidated as (2*S*)-3-*O*--D-galactopyranosyl-1-*O*-(9Z,12Z-octadecadienoyl)-2-*O*-(4Z-hexadecenoyl)glycerol by enzymatic partial hydrolysis using lipase and physicochemical evidence which includes determining double bond position in hexadecenoic acid moiety.**10**

3. Bioactive metabolites of marine fungi

(1) Radical scavenging activity

As part of our search to find new radical scavenging active compounds from the marine fungus, we have investigated the metabolites of 180 strains of the marine fungi. The significant activities (% Inhibition : DPPH 40% >, ONOO- 80% >, ROS 80% >) were observed in 12 strains of marine fungi (MFA 002, MFA 006, MFA 014, MFA 038, MFA 055, MFA 089, MFA 124, MFA 133, MFA 143, MFA 153, MFA 158, MFA 175). Among them, MFA153 (*Aspergillus parasiticus*) displayed the most significant activity (DPPH : IC50 = 2 ㎍/㎖).

(2) Marine algicolous fungus, *Aspergillus parasiticus*

According to an assay-guided isolation from the MFA 153 (*Aspergillus parasiticus*), we have isolated MFA 153B4, as the most active metabolite (DPPH : IC50 = 0.2 ㎍/ml). The elucidation of the stereostructure is now in progress.

A new ergosteryl myristate (**10**) and ergosterol (**11**) have been isolated from the organic extract of the mycelium of marine algicolous fungus, *Aspergillus parasiticus* (strain #, MFA 153). The structure of a new compound (**10**) was assigned on the basis of comprehensive spectroscopic analyses and chemical synthesis.**11**

(3) Marine algicolous fungus, *Penicillium* sp.

Two new cytotoxic dioxopiperazine dimers, 11,11'-dideoxyverticillin A (**12**) and

11'-deoxyverticillin A (**13**), and the previously described verticillin A (**14**), have been isolated from the mycelium of a marine-derived fungus of the genus Penicillium (strain #, CNC 350) . An inactive, but new *bis*dethio-*bis*(methylthio)-dioxopiperazine (**15**), was also obtained by extraction of the culture broth. The structures and absolute stereochemistries of the new compounds were assigned on the basis of NMR and CD experiments. Compounds **12** and **13** exhibit potent in *vitro* cytotoxicity against HCT-116 human colon carcinoma (IC50 = 30 ng/mL).**12**

4. References

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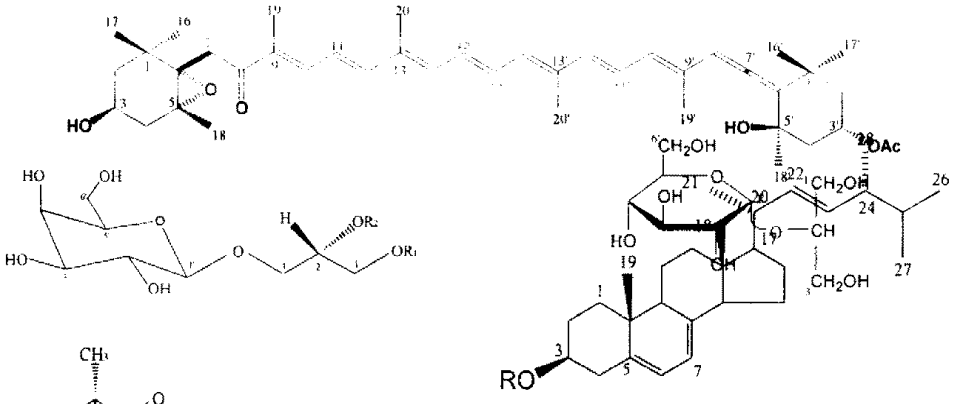
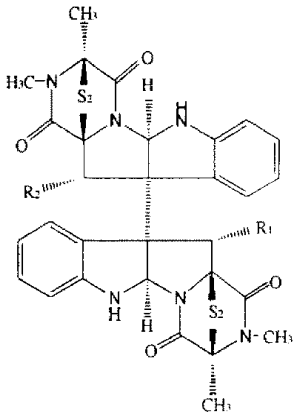
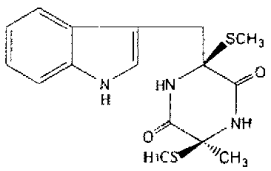


1



2 : R_1, R_2 = 9*z*-hexadecenoyl
3 : R_1 = 5*z*,8*z*,11*z*,14*z*,17*z*-eicosapentaenoyl (EPA)
R_2 = 9*z*-hexadecenoyl
5 : R_1 = 9Z,12Z-octadecadienoyl, R_2 = 9Z-hexadecenoyl
6 : R_1 = 9Z,12Z,15Z-octadecatrienoyl, R_2 = 9Z-hexadecenoyl
7/8 : R_1 = 9Z-hexadecenoyl, R_2 = hexadecanoyl
R_1 = 9Z-octadecenoyl, R_2 = 9Z-hexadecenoyl
9 : R_1 = 9Z,12Z-octadecadienoyl, R_2 = 4Z-hexadecenoyl



10 : R = $OCCH_2(CH_2)_{10}CH_2CH_3$
11 : R = H

4

15

12 : R_1, R_2 = H
13 : R_1 = OH, R_2 = H
14 : R_1, R_2 = OH