

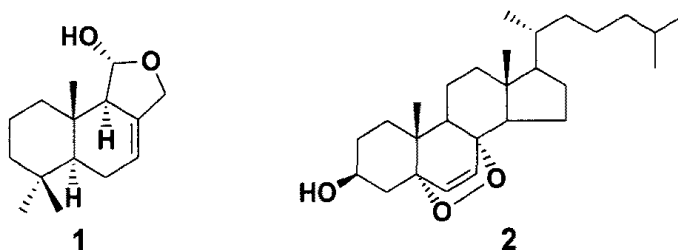
Search for Bioactive Natural Products from Marine Organisms Collected from Boso Peninsula in Japan

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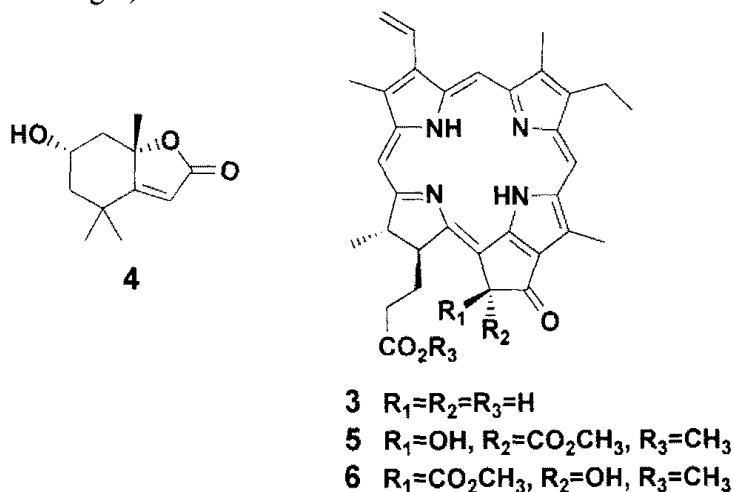
During our investigations on search for bioactive substances from marine and terrestrial natural resources, we recently initiated a program on search for bioactive natural products from marine organisms collected around Boso Peninsula, Chiba prefecture, in Japan. Here we describe a result of our recent study on the chemical constituents of nudibranch and Corallinaceous red algae as well as our preliminary results on isolation of field-collected microalgae and their laboratory cultivation.

- 1) Nudibranch and sea hare: Fourteen new sesquiterpenes of the drimane series were obtained from the ethanol extracts of a big-size nudibranch *Dendrodoris carbunculosa*, found at the south coast of Boso Peninsula, in November 1999. Most of these sesquiterpenes exhibited cytotoxicity against murine leukemia P388 cells, and isodrimeninol (**1**), the major sesquiterpene of this animal, proved to have sharp peppery taste. Constituents of sea hare *Aplysia juliana*, collected from the south coast of Boso Peninsula, were also investigated to isolate 5,8 α -epidioxy-5 α -cholest-6-en-3 β -ol (**2**) and pyropheophorbide a (**3**).



- 2) Corallinaceous red algae: Chemical constituents of three Corallinaceous algae *Corallina pilulifera*, *Amphiroa zonata*, and *Marginisporum crassissimum*, collected at Tainoura, Amatsu-Kominato, Chiba, in May, 1999 were studied by thin-layer chromatography screening as well as antimicrobial activity assay against *Bacillus subtilis*. These three algae were revealed to have very similar TLC profiles to each other, and EtOAc layers of three algae showed weak antimicrobial activity. The ethyl acetate-soluble fraction of the methanol extract of *Corallina pilulifera* was revealed to contain pyropheophorbide a (**3**) and monoacylglycerol.
- 3) Microalgae cultivation: Exploitation of new medicinal resources is quite important for the discovery of new natural molecules. "Microalgae" may be strongly expected to be potentially useful as new medicinal resources. Isolation of microalgae such as diatoms or dinoflagellates from sea-water samples collected in Chiba and other places in Japan is currently investigated in our laboratory to use these purified microalgae for mass culture in the laboratory and preliminary bioactivity-test screenings. On the other hand, we studied the cultivations of 13 species of microalgae (dinoflagellates and diatoms) obtained

from NIES or CCMP. Among them, the diatom *Triceratium dubium* (NIES 556) was uniaxially cultured at 25 °C for 2 weeks statically in a seawater medium enriched with f/2 supplement. The MeOH extract of the harvested algal cells exhibited inhibition effect on norepinephrine-induced contraction in rat thoracic aorta at the concentration of 3×10^{-4} g/mL. The EtOAc-soluble fraction of the MeOH extract was subjected to silica gel column chromatographies, followed by gel filtration on Sephadex LH-20. Further purification using reversed-phase HPLC on ODS afforded epiloliolide (**4**; 0.008% yield, wet weight) and a 1:1 mixture of 13²-hydroxyphaeophorbide a methyl esters (**5** and **6**; 0.01% yield, wet weight).¹



(1) Sakio, Y.; Ishibashi, M.; Okuyama, E. *Nat. Med.* **2000**, *54*, 104.