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# Partial purification of naturial antifouling agents from the marine coralline alga *Lithophyllum yessoense* Foslie

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## Introduction

One of the most dominant species causing the algal whitening in Korea and Japan is *Lithophyllum yessoense* Foslie (Tokuda et al. 1994; Kim 2000). Even though biological grazing or physical sloughing is sufficient to prevent recruitement of fleshy seaweed, allelopathic substances also may contribute to prevent spore settlement or germination of various fleshy seaweed. The coralline alga has allelopathic activity against the spore settlement and germination of various different fleshy seaweed taxa(Kim 2004). Isolation of the active allelopathic compounds will lead discovery of antifouling agents in marine environment. Antifouling activities of the *L. yessoense* extracts were tested against *Enteromorpha linza*, a representative fouling seaweed and the *Mytilus edulis*, a representative fouling shellfish. The natural antifouling substances were isolated by several separation techniques such as MeOH-$H_2O$(4:1) extraction, fractionating by polarity, silica gel chromatography and Sephadex LH-20 gel filtraction chromatography. Further purification is carring out using HPLC.

## Material and methods

### Algal and mussel materal

*Lithophyllum yessoense* and *Enteromorpha linza* were collected at Daebo, Pohang and Chungsapo, Busan respectively. Aquacultured mussel of *Mytilus edulis*, sized 4.5±0.2cm in shell length in repulsive experiments.

### Fractionating by polarity

The fractions were separated into 5 different classes according to polarity (Harborne 1998).

Measurement of antifouling activity

Antifouling activity was measured by the method of Cho et al.(2001).

Silica gel chromatography

The material adsorbed on the column was eluted using different ratio of hexane, diethyl ether, acetone, ethyl acetate, acetonitrile, and methanol.

Sephadex LH-20 gel filtraction chromatography

Gel filtraction chromatography was carried out using a 2.0×110 cm column of Sephadex LH-20. Algal extracts(4ml) were passed through the column at a flow rate of 0.5ml $min^{-1}$, and 2ml fractions were collected. Fractions were measured for dry weight and antifouling activity.

## Results

The isolation of antifouling agent was started with preparation of MeOH-$H_2O$ (4:1) extract of *Lithophyllum yessoense*. Fractions were then separated into 5 different classes according to polarity. Further purification of antifouling agents in Fraction Ⅲ(chloroform phase) by silica gel chromatography revealed that SS, SG and SF fractions had the inhibition activities of algal spore settlement and germination and the foot repulsion activity of mussel, respectively. Then further purification was carried out Sephadex LH-20 gel filtration chromatography. Each fraction showing antifouling activity separated by gel filtration was pooled and then further purification is carring out using HPLC.

## References

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