IDENTIFICATION AND CHARACTERIZATION OF GROWTH-PROMOTING SUBSTANCES FOR THE MARINE DIATOM *Navicula* sp.

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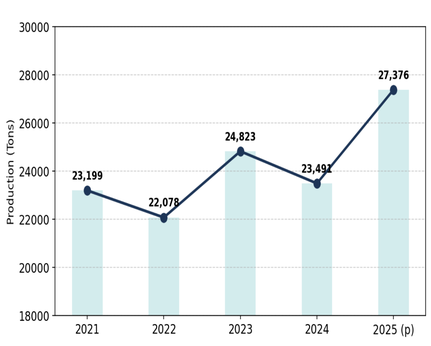
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Abstract

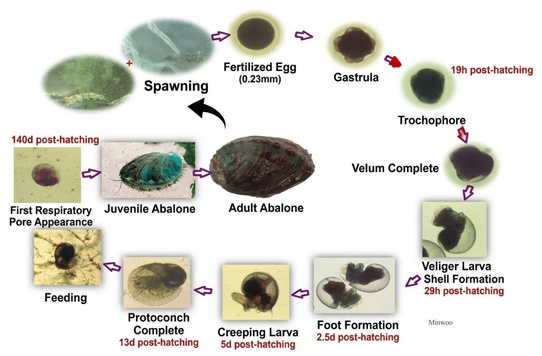
Abalone aquaculture represents a major component of the marine aquaculture industry in Korea, with annual production exceeding 20,000 metric tonnes in recent years. Wando, located on the southern coast, serves as the primary farming region, supported by extensive seaweed cultivation. However, high mortality rates during the early developmental stages of abalone remain a critical challenge, largely due to environmental variability and insufficient availability of benthic diatoms, which serve as an essential food source for abalone post-larvae. Ensuring a stable and sufficient supply of benthic diatoms remains technically challenging under conventional cultivation conditions. Therefore, this study aimed to develop growth-promoting substances to enhance the proliferation of the benthic diatom Navicula sp. Various candidate materials, including alginic acid, fucoidan, Scytosiphon lomentaria, Ecklonia cava, boiled extracts derived from Ecklonia cava processing byproducts (EBE), and Porphyra tenera, were evaluated for their growth-enhancing effects. Additionally, EBE was combined with inorganic nutrient components, including phosphate fertilizer (PF), nitrogenous manure (NM), and sodium silicate (SS), to determine an optimal growth-promoting formulation. Among the tested materials, EBE exhibited the most pronounced stimulatory effect on diatom growth. Further optimization demonstrated that a mixture of EBE, PF, NM, and SS at a ratio of 1:1:1:1 resulted in the highest growth enhancement. The optimized Navicula growth-promoting substance (NGPS) increased the growth of Navicula sp. by approximately 150% compared with commercially available products. These findings suggest that the developed formulation can effectively enhance benthic diatom production and may contribute to improving abalone seed survival and aquaculture productivity.

Introduction

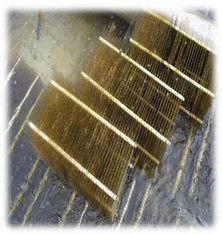
❖ *Korea abalone aquaculture production*



❖ *Life cycle of abalone*



- **Significance of diatom growth → Shortage of diatom, which are utilized to feed for abalone spat, increase mortality.**



- **3~5cm stages of growth as mortality about 40%**
- **Abalone spat is oxygen consumption more than abalone**

Growth



- **Growth-promoting of benthic diatom for sufficient feeding**

Navicula sp.



Scientific classification	
Kingdom	Chromalveolata
Division	Heterokontophyta
Class	Bacilariophyceae
Order	Naviculales
Family	Naviculaceae
Genus	Navicula

❖Temperature : 20˚C

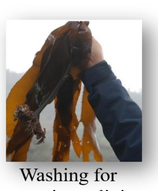
❖Lux : 5000lux

❖Light & dark : 12h / 12h

❖Culture medium : f/2

Material & Methods

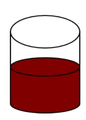




Washing for removing salinity



Steaming of *E. cava* at 150°C



Steaming liquid



Vacuum evaporation



Freeze drier

***Ecklonia cava* boiled extracts**

Ethanol sedimentation method

DW : Ethanol = 1 : 2

Supernatant liquid

Ethyl acetate

Polyphenols

Minerals

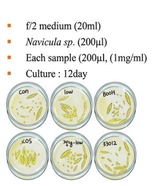
Sediment

Ammonium sulfate

Proteins

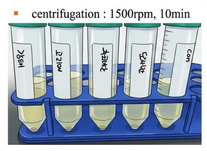
Carbohydrates

- f/2 medium (20ml)
- *Navicula sp.* (200µl)
- Each sample (200µl, (1mg/ml)
- Culture : 12day



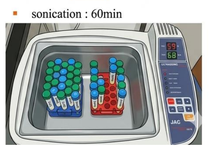
Scraper

- centrifugation : 1500rpm, 10min



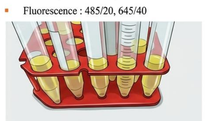
Chlorophyll-a extraction

- sonication : 60min

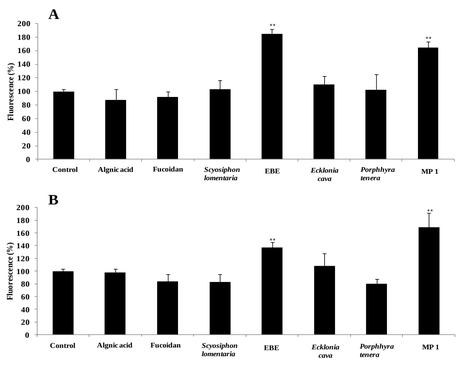


Chlorophyll-a

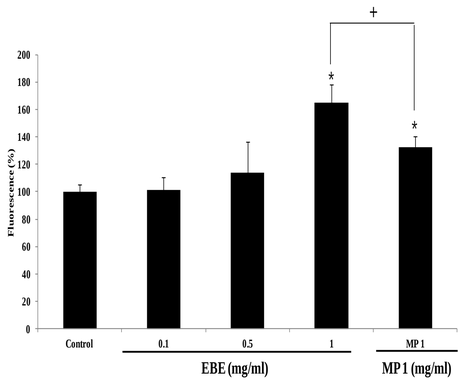
- Black well plate each 100µl transfer
- Centrifugation : 1500rpm, 10min
- Fluorescence : 485/20, 645/40



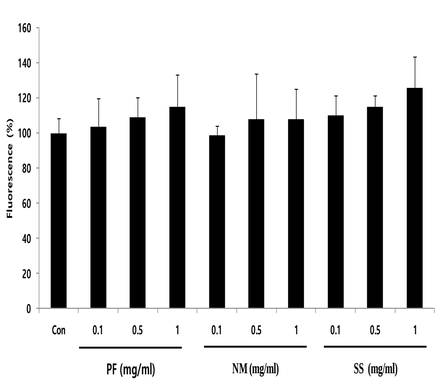
Result



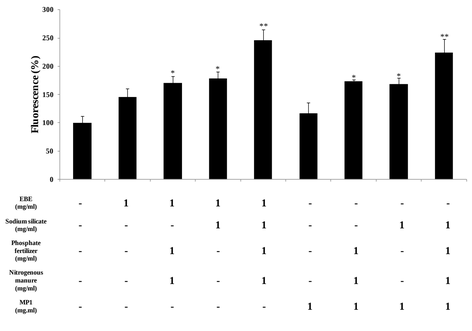
Each sample was added to Petri dishes in the following quantities: 200 µg of each sample (1 mg l^{-1}), 20 mL of f/2 medium and 200 µL of diatoms. Values with different superscripts show significant differences at $P < 0.05$.



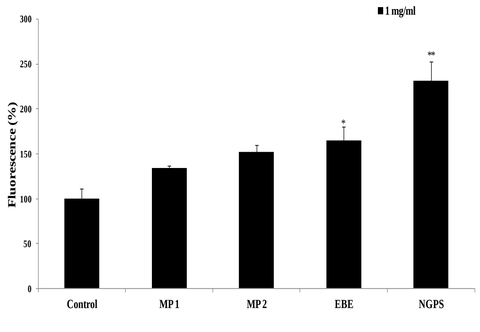
Chlorophyll-a analyzes confirming effects on the growth of *Navicula* sp. of different concentrations of EBE. Values with different superscripts show significant differences at $P < 0.05$.



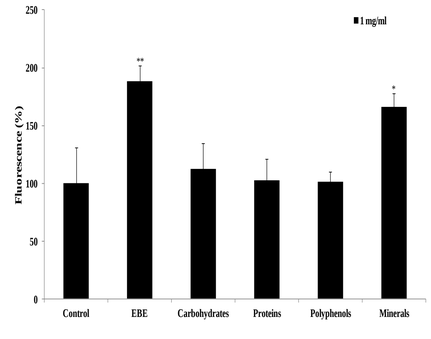
Chlorophyll-a analyzes confirming effects on the growth of *Navicula* sp. of different concentrations of PF, NM and SS. Standard error bars are shown.



To estimate growth rates of diatoms, various concentrations of mixtures were added to each sample. Standard error bars are shown.



Effects of NGPS on the growth of *Navicula* sp., compared with control (non-treated), MP 1, MP 2 and EBE.



Growth of diatoms treated with carbohydrate, protein, polyphenol and minerals purified from EBE. Effects of EBE on the growth of *Navicula* sp., compared with control (non-treated), carbohydrate, protein, polyphenol and minerals.

Conclusion

NGPS developed from EBE effectively promoted the growth of Navicula sp. and outperformed commercial products. Therefore, NGPS has potential as a cost-effective growth enhancer for abalone aquaculture.