Effect of a marine algal constituent on the growth of lettuce and rice seedlings

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<u>Abstract</u> - A dibromocatechol, the methyl ether of lanosol was isolated from the methanol extract of <u>Odonthalia washingtoniensis</u> and <u>O. floccosa</u>. It showed the stimulating effect on the growth elongation of certain terrestrial plants.

INTRODUCTION

Seaweeds have been reported from Roman times as a source as an agricultural fertilizer, a practice still used today in some areas of the world. While some of their utility is no doubt due to organic nutrients and inorganic minerals, algal constituents have been isolated that exhibit specific control over terrestrial plant growth, with both of gibberellins and cytokinins being isolated from algal sources. During our continuing search for natural products as a resource for biologically active compounds, we examined an extract of the red algae, \underline{O} . washingtoniensis Kylin and \underline{O} . floccosa (Esper) Falk. (Rhodophyta) for phytoactive components. Preliminary bioassays using lettuce and rice plants revealed that the methanol extract of \underline{O} . washingtoniensis and \underline{O} . floccosa possessed potent growth stimulatory activity. We report the isolation, identification and biological activity of α - \underline{O} -methyllanosol (3,4-dibromo-5-methoxymethyl-1,2-benzenediol).

RESULTS AND DISCUSSION

Isolation and identification of plant growth stimulator substance

Biologically active compound was isolated from benzene as colorless needles, m.p. 130°C, the mass spectrum indicated to be a dibromocompound, $C_8H_8O_3Br_2$, [M¹] at 310, 312, 314, (1:2:1). The $^{13}C\text{-NMR}$ spectrum indicated a penta substituted benzene with absorption at δ 145.1 (s), 143.8 (s), 129.2 (s), 114.6 (d), 113.5 (s) and 113.0 (s), a methoxy group at δ 57.7 (q), and a benzylic methylene group at δ 74.0 ppm (t). The $^{14}\text{-NMR}$ spectrum shows the signals for an aromatic proton at δ 6.95 (s, 1H), a methoxy group at δ 3.32 (s, 3H), and a benzylic methylene group at δ 4.38 ppm (s, 2H) respectively. Also, a 10.72% NOE of the aromatic proton was observed when irradiated at δ 4.38 ppm (benzylic methylene group) shows the relationship of these two groups. In spite of the possible proton exchange with the D_4 -methanol solution, two signals from two hydroxy protons were observed at δ 10.1 (s) and δ 9.58 ppm (s). This implies 1,2-relationship of the diols with strong intramolecular hydrogen bond keeping the protons from exchanging with the solvent. From these spectra this compound was identified as the α -Q-methyllanosol, a compound previously isolated from other red algae, e.g., Odonthaliaceae 1 , 2 Polysiphoniaceae 3 , and Rhodomelaceae 1 , 2,4.

Methyllanosol

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Growth stimulating effect of methyllanosol on terrestrial plants

The presence of methyllanosol was observed to have little effect on rice seedlings even at the concentration of 100 ppm. However, as exhibited in Figure 1, and Figure 2, the growth of lettuce hypocotyl and root were stimulated at this concentration. The overall appearance of greater health among the seedlings exposed to methyllanosol was striking. These seedlings were much larger than the seedlings in the control group and they appeared to be normal in all proportions. No inappropriate elongation was apparent in either the hypocotyl or the root of the lettuce seedlings. In addition, the hypocotyl of the exposed seedlings were of a much richer and deeper green color, and the roots had more root hairs, than the control group.

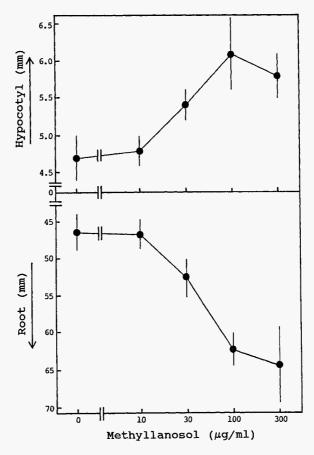
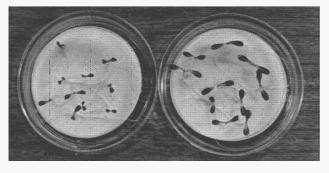


Fig. 1. Effect of methyllanosol on the growth of lettuce seedlings. Vertical lines represent standard errors.



Control

Treated

Fig. 2. Effect of methyllanosol on the seedlings of <u>Lactuca</u> <u>sativa</u>. The treated group with 100 ppm of methyllanosol for 5 days shows seedlings with greener and healthier leaves.

This is the first report on the effect of methyllanosol on terrestrial It is well known that lettuce seedlings are inactive to indolacetic acid (IAA) but active to gibberellins (GAs). Although the structure of methyllanosol seems to resemble synthetic auxins such as 2,4-D, the results from the growth experiments with lettuce seedlings show gibberellin-like activity. Further work is now in progress to elucidate the mechanism of this growth regulator.

BIOASSAY

Growth experiments were carried out according to the method of Kamisaka⁵. Seeds of lettuce (L. sativa L., cv. Grand Rapids) were germinated on 2 layers of filter paper, moistened with distilled water, and kept for 2 days under continuous fluorescent light (3,000 lux at plant level) at 25.0 ± 0.5 °C. Rice seeds (Oryza sativa L., cv. Norin 20) were allowed to germinate in the dark at 30°C. Ten seedlings, selected for uniformity, were placed on 2 layers of filter paper in a 9 cm petri dish containing 4 ml of test solution. Seedlings were allowed to grow under the same light and temperature conditions used for their germination. After 3 days of cultivation, the length of the hypocotyls and roots were measured and an average was taken of 30 seedlings from 3 petri dishes.

CONCLUSION

These effects of methyllanosol on plants, has several explanations. Methyllanosol may be a new class of plant growth regulator showing gibberellin-like activity. It is possible that some phenolic substances may be involved as algal growth regulators. The free benzyl alcohol, lanosol and its sulfonate as well as other simple phenols, have been observed to both stimulate and depress the growth of marine algae. Specially, it had been reported that lower concentrations of lanosol strongly stimulated growth of red algae. The presence of excreted phenolics has been implicated as necessary for the normal morphology and completion of their life cycle in algal species such as <u>Ulva</u> and <u>Monostroma</u>⁷. There is also the potential that these or similiar compounds may be used as an antifouling agent. Many simple phenols exhibit antibacterial and antifungal activity, and it has been suggested that phenols may be involved in regulating the growth of epiphytes and parasites in algae8.

Acknowledgements

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REFERENCES

- N. Katsui, Y. Suzuki, S. Kitamura and T. Irie. Tetrahedron, 23, 1185 1. (1967)
- 2. M. Pederson, P. Saenger and L. Fries. Phytochemistry, 13, 2273 (1974)
- W. Fenical, <u>J. Phycol.</u>, <u>11</u>, 245 (1975)
- K. Kurata and T. Amiya, Chem. Lett., 1435 (1977)
- 5. S. Kamisaka, <u>Plant and Cell Physiol</u>., <u>14</u>, 747 (1973)
 L. Fries., <u>Experentia</u>, <u>29</u>, 1436 (1973)
- L. Provasoli, Publ. 1700 Natn. Acad. Sci. Washington (1969) 7.
- J. MeLachlan and J.S. Craigie, <u>J. Phycol</u>. <u>2</u>, 133 (1966)