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Isolation and purification of selenoproteins from *Emiliania huxleyi* (Coccolithophorid)

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We found coccolithophrids, unicellular calcifying algae, require essentially selenium for their growth. To analyze the function of selenium (Se) in algae, we intended to purify, identify and characterize selenoproteins of coccolithophrids.

Emiliania huxleyi was grown in a medium containing ⁷⁵Se and/or ³⁵S to label selenoproteins. Fractionation was disturbed by the high viscidity of crude extract. Anion exchange chromatography was effective to fractionate proteins and to remove the viscidity. Selenoproteins were fractionated using DEAE-toyopearl (anion exchange chromatography) and Diol-150 (gel-filtration). ⁷⁵Se-labeled proteins were discerned as bands of SDS-PAGE with the molecular masses of 60, 31, 29, 25 and 20kDa, and these proteins were brought to the determination of N-terminal amino-acid sequences. However, the meaningful homology with proteins in the database was not observed.

739(S827)

INDUCTION OF HYDROCARBON-SYNTHESIS IN BOTRYOCOCCUS BRAUNII

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The unicelullar green alga Botryococcus braunii synthesizes a large amount of hydrocarbon (30-60% of cell dry weight). It is C_{29} or C_{31} straight chain with two double bonds. We have clarified that the hydrocarbonsynthesis occurred on the plasma membrane of daughter protoplasts just after cell division. In this study, we attempt to induce the hydrocarbon-synthesis in interphase cells. Interphase cells were treated with various kinds of enzymes for cell wall-lysis (cellulase, macerozyme, hemicellulase etc.) and then cultured in medium containing of $[1-^{14}C]$ acetate (2.5 μ Ci/ml) for 2hr. Lipids were extracted with n-hexane and subjected to thin laver chromatography. The incorporation of ¹⁴C into hydrocarbons was measured. Spheroplasts synthesized hydorcarbons 10-15 times as much as those produced by untreated interphase cells (control). In the spheroplasts, trans-Golgi networks produced the special vesicles that were observed in untreated cells just after cell division.

740(S828)

CYTOPLASMIC CALCIUM CONTENT IN CHARACEAE Masashi Tazawa, Munehiro Kikuyama¹ and Yoshiji Okazaki² Department of Applied Physics and Chemistry, Fukui University of Technology, Fukui 910-8505; ¹Laboratory of Biology, University of the Air, Chiba 261-0014; ²Department of Biology, Osaka Medical College, Osaka 569-0084

Internodal cells of three species of Characeae, Nitella flexilis, Nitella axilliformis and Chara corallina, were analyzed for the content of Ca2+ and Mg2+ in the whole cytoplasm, chloroplast-free cytoplasm and chloroplasts. To isolate the cytoplasm without contamination of Ca2+ from both cell wall and vacuole, the vacuolar sap was replaced with a slightly hypertonic sorbitol solution containing Sr²⁺ by the vacuolar perfusion method, after the cell had been treated with 5 mM SrCl₂ No significant difference in the content of Mg²⁺ (7 - 8 mM) was found between three speices of Characeae in both whole cytoplasm and chloroplst-free cytoplasm. But significant differences were observed in the Ca2+ content of whole cytoplasm and chloroplst-free cytoplasm (N. flexilis 18.8 and 16.3 mM, N. axilliformis 9.6 and 8.3 mM, C. corallina. 5.7 and 7.0 mM). Chloroplasts of N. flexilis contained about seven times more Ca2+ (31 mM) and about two time more Mg2+ (17 mM) than those of C. corallina.

s220