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# Production of unsaturated fatty acids from marine microalga, *Chlorella* sp. by utilizing the oyster shells

**Hyeon Yong Lee**
Seowon University, Korea

Thousand kilo tons of oyster shells have been produced from the oyster processing industries in Korea. However, efficient ways of removing or treating the oyster shell wastes have not been developed yet, only by breaking them down to be buried or spread on the ground of the farms, which have caused serious environmental problems even though they contain lots of calcium and potentially can be converted into carbon dioxide under certain conditions. Therefore, conversion into carbon dioxide from the oyster shells can be utilized to grow marine alga for economically obtaining unsaturated fatty acids, especially DHA (docosahexaenoic acid) or EPA (eicosapentaenonic acid).

The oyster shells were crashed and broken down to ca. 250 nm diameter size nano particles by a high pressure homogenizer at 20,000 psi, which can be easily dissolved into acetate solution, not strong acids such as HCl or $H_2SO_4$. The marine microalga, *Chlorella* sp. was grown up to 10 g of dry wt./L with enforced sea water of trace amounts of key minerals and 25-35% (v/v) of filtered oyster shell solution as inorganic carbon sources without supplying any extra carbon dioxide. It was found that the nano sized oyster shell can be easily converted into carbon dioxide up to 350-500 uatm of pCO2 not by adding strong acids, possibly due to high surface areas and easy penetration into the hard shell membranes. However, *Chlorella* sp. could not grow in the medium extracted from the oyster shells powder (ca. 1-100 um diameter) dissolved with strong acids because of the residues of strong acids and very low carbon dioxide concentration in the solution. The biomass from the culture broth also contained ca. 35% of total lipids with 20-25% of DHA and EPA depending on the mixing ratio of the oyster shell solution and light intensity, whose concentrations were relatively higher than those from artificial sea water mediums as 15-18% of DHA, due to enough high carbon dioxide and essential minerals from oyster shells. The optimal culture condition of producing maximum 25% of DHA and EPA from 10 g-dry wt./L of biomass was determined as conventional sea water with 30% (v/v) of oyster shell solution and 12:12 hour cyclic illumination of 10 w/m$^2$ light intensity. These results tell that the unique cheap source of carbon dioxide can be used to obtain functionally valuable biomass, especially for high amounts of EPA, DHA and/or protein as SCP (Single Cell Protein) by directly utilizing sea water without supplying any inorganic carbon sources. This process can have great advantages of easy scaling-up the outdoor culture system for economic production of functional biomass.

hyeonl@seowon.ac.kr