

## ***Interactive comment on “Change in coccolith morphology by responding to temperature and salinity in coccolithophore *Emiliana huxleyi* (Haptophyta) isolated from the Bering and Chukchi Seas” by K. Saruwatari et al.***

**Anonymous Referee #1**

Received and published: 27 November 2015

The authors reported interesting observations; clonal culture strains of *E. huxleyi* changed morphology and size of coccoliths in relation to change in temperature and salinity in laboratory culture experiments. Their observations are very interesting, however, I cannot evaluate accuracy of their experiments at this moment, since they did not describe details of their experiments in the Materials and Methods. Authors did not describe timing of measurements of cell growth rate and of size of coccoliths in their culture experiments, despite it is well known that growth rate of culture strain usually differs greatly between exponentially (logarithmic) and stationary growth phases, and size

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of coccoliths of *E. huxleyi* changes in relation to growth phase (growth rate); *E. huxleyi* make smaller coccoliths in the exponentially growth phase and make larger coccoliths in the stationary phase (Young and Westbroek, 1991). So I am unsure whether the observations on change in coccolith size in this study actually reflect change in temperature and/or salinity, or just reflect change in growth phase. Another problem; there are too many mistakes in citations. I would recommend authors add detailed information of experiments to materials and methods, reread related papers, and rewrite manuscript with correct references for resubmission.

Followings are my other comments;

Abstract has too much detailed information. The information on the name of the ship used for sampling (line 2), latitude and longitude of sampling localities (line 8), and explanations of classification of morphotype (lines 15-18, the sentence started from According. . .) are unnecessary here.

Lines 23-25 of the page 17752. Authors wrote “This indicates that subarctic and arctic coccolithophore strains can survive in a wide range of seawater temperatures and at lower salinities due to their marked morphometric adaptation ability” without explaining how ‘morphometric adaptation’ helps adaptation of *E. huxleyi* to various temperature/salinity conditions. Please explain it in the Discussion.

Lines 14-16 of the page 17753; Please describe the definition of ‘warm water’ and ‘cold water’ in the studied area (Arctic Ocean and Bering Sea).

Line 18 of the page 17753; Prymnesiophyceae not Prymneophyceae. More correctly, *E. huxleyi* belongs to the Family Noelaerhabdaceae, Order Isochrysidales, Class Prymnesiophyceae not to Prymneophyceae family.

Lines 1-9 of the page 17754; Citations in these sentence are wrong. Authors wrote “Hagino et al. (2011) classified coccolith morphotype into four groups: (1) Type A and Type R with. . . .”. Correctly, Hagino et al. (2011) classified *E. huxleyi* into seven groups!

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Therefore, the all explanations concerning morphotypes of Hagino et al. (2011) in these sentences are inaccurate.

Line 4 of the page 17754; 'corona' should be written in italic.

Line 14 of the page 17754; 'McIntyre and Bé' not 'McIntyre and Be'

Lines 14-16 of the page 17754; Citations in these sentences are completely wrong. Authors wrote "According to McIntyre and Be (1967), Type A and Type C likely correspond to warm- and cold-water types, respectively, although Hagino et al. (2011) reported that Type C has not always been reported in cold-water environments". Correctly, McIntyre and Bé (1967) just described warm and cold types of *E. huxleyi*. Young and Westbroek (1991) renamed warm and cold types of McIntyre and Bé (1967) as Types A and C, respectively. They renamed the morphotypes of *E. huxleyi* since Winter (1987) mentioned cold type (= Type C in Young and Westbroek 1991) was not always related to low temperature. Hagino et al. (2000) and Hagino et al. (2006) reported type C from tropical area, but Hagino et al. (2011) did not. Hagino et al. (2011) just introduced observation by Winter (1987) and interpretation by Young and Westbroek (1991).

2. Materials and methods; Please provide more detailed information on materials and methods of experiments.

Line 12 of the page 17755; How did you collect 'samples' that yielded your culture strains?

Please show in situ seawater temperature and salinity of the water samples that yielded culture strains used in this study.

Line 17 of the page 17755; How did you establish clonal culture strain from your 'samples'?

Lines 20-21 of the page 17755; Authors wrote "The growth rate at each temperature was calculated as the average value of triplicate experiments, and the error bars indicated the minimum and maximum values." I think the growth rate of *E. huxleyi* is usu-

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ally changes during culture experiments. Please describe the detailed method used for monitoring of growth rate in this study, and provide information of growth phase of each culture strain at the timing of sampling for the studies of growth rate.

Line 26 of the page 17755; How did you know the strain MS1 is type A?

Line 26 of the page 17755; The strain code of MS1 in the Roscoff culture collection is RCC 1226 not D2801-5.

Line 1 of the page 17756; How did you know the NIES 1311 is type O?

Line 24 of the page 17756; How did you prepare sample for measurement of cell density in a polarized microscope?

Lines 26-29 of the page 17756; Please describe pore size, diameter, and product name of the polycarbonate filter.

Young and Westbroek (1991) reported size of coccoliths of *E. huxleyi* changes in culture experiments in relation to growth phase. Please provide information of growth phase of each culture strain at the timing of sampling for morphometric studies under SEM.

Lines 2-10 of the page 17759; Young and Westbroek (1991) and Young et al. (2003) mentioned that central area of Type A consists of 'curved elements', while that of Type B (and B/C) consists of 'lath-like elements'. Authors classified their culture strains into type B/C without description of morphology of central area elements. So I am unsure if their strains are actually type B/C or not. Please describe morphology of central area elements of the culture strains used in this study.

Lines 10-11 of the page 17762; What is the 'scaling low'?

Lines 22-23 of the page 17762 "On the other hand, Types A and B were found around the Southern Subtropical Front in a warm-water areas." Please provide information on papers that reported Type B from warm-water area.

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