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硕 士 学 位 论 文

颗石藻 *Emiliania huxleyi* 光合、呼吸及钙化作用对海洋酸化的响应初探

Responses of photosynthesis, respiration and calcification of the coccolithophorid *Emiliania huxleyi* to ocean acidification -a preliminary study

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摘　要

工业革命以来，人类活动导致大气中二氧化碳浓度不断升高。海洋作为碳的一个重要储库吸收 CO_2，导致海水酸度增加、pH 值降低，并引起碳酸钙饱和度不断降低，这种现象被称为“海洋酸化”。颗石藻是海洋浮游钙化生物中的主要类群，它通过光合、呼吸及钙化作用对海洋调节大气 CO_2 的两类生物泵过程均有贡献，颗石藻的这些生物过程对海洋酸化的响应因而也成为研究大气 CO_2 升高对海洋碳循环影响的核心问题之一。本论文通过对 *Emiliania huxleyi* 颗石藻进行批次（Batch culturing）与半连续培养（Semi-continuous culturing），探讨了不同培养方式下大气 CO_2 浓度对 *E. huxleyi* 颗石藻的光合、呼吸及钙化作用的影响。本论文还研究了东海海区夏季的群落呼吸速率，为研究海洋酸化对该海区碳循环特别是呼吸作用的影响提供背景知识。

在批次培养中，我们向培养液中通入高浓度 CO_2，并通过 pH 控制器调控培养液的 pH 于 8.2、8.0 和 7.8，用以模拟大气二氧化碳分压（pCO_2）水平于 380、720、1100 ppm，并研究了不同水平 CO_2 对 *E. huxleyi* 颗石藻细胞生长、颗粒有机碳（POC）生产速率、钙化速率、PIC/POC 比值的影响。实验结果表明，未经过适应的颗石藻在高浓度 CO_2 下生长受到抑制，相对于 pH=8.2，单位细胞钙化速率分别下降 13.5%（pH=8.0）和 46%（pH=7.8），而 PIC/POC 则分别降低了 11.1% 和 38.9%；但是，单位细胞 POC 生产速率在不同 pH 条件下无明显差异；通过对三种钙化速率测定方法的比较发现，三种方法所得出的结果相近，其中，用总碱度变量计算的方法精度高于用 Ca^{2+}和颗粒无机碳（PIC）。

在半连续培养中，通过植物生长气候箱调节空气中 CO_2 分压，分别为 520 ppm 和 1100 ppm。经过充分适应后，细胞生长速率不受 CO_2 分压影响，但 POC 生产速率在高浓度 CO_2 下得到促进，增幅为 20%，不同 CO_2 分压条件下钙化速率差异不大，在测定误差范围内。半连续培养中颗石藻的 C/N 比值高于 Redfield 比值且随 pH 的降低而升高。根据暗培养前后溶氧的变化计算得出的颗石藻呼吸速率在 pH 由 8.0 变为 7.8 时升高 38.6%。

以上表明，不同培养方式下，颗石藻生物过程对海洋酸化的响应差异明显。在批次培养中，pCO_2 升高抑制细胞生长及钙化作用，对光合作用的促进不显著；

在半连续培养中， pCO_2 升高对细胞生长和钙化作用无影响，光合作用及呼吸作用得到明显促进。这些差异说明在进行酸化研究时细胞对 pH 变化的适应情况对实验的结果影响很大。

通过对现场海水进行暗培养，测定培养瓶中溶解氧变化的方法测定了夏季东海海区 22 个站位表、中、底层的群落呼吸速率。表层群落呼吸速率的变化范围是 1.2-27.8 μmol $L^{-1}d^{-1}$，平均值为 9.0 μmol $L^{-1}d^{-1}$。表层高值区出现在长江口附近海域及浙江沿岸，其值随离岸距离的增加而降低。群落呼吸速率的垂直分布特征受长江冲淡水影响显著。在受到长江冲淡水影响(盐度小于 32)的站位群落呼吸速率随深度的增加而降低，在该区域以外（盐度大于 32），群落呼吸速率随深度变化不明显。夏季东海群落呼吸的主要贡献者是细菌与浮游植物。表层海水中，长江冲淡水区域群落呼吸速率与叶绿素 a 存在显著的相关性（R^2=0.66），浮游植物的呼吸作用对群落呼吸耗氧的贡献占 58.4%，而不受长江冲淡水影响的区域细菌的贡献较大占 62.9%。真光层以下群落呼吸速率与细菌丰度相关性显著（R^2=0.53），细菌的呼吸作用是微型生物耗氧的主要因素，占群落呼吸的 89.3%。真光层中水柱积分的初级生产力与呼吸作用（P/R）的比值变化范围为 0.2-0.9，平均值为 0.5，表明东海夏季呈现出异养特征。呼吸速率消耗的有机碳除了由初级生产提供，还可能由河流输入或者沉积物的再悬浮提供。

总之，*E. huxleyi* 颗石藻光合、呼吸及钙化作用对海洋酸化的响应，随着细胞对 pH 变化的适应情况的不同而有所差异。东海群落呼吸作用是东海碳循环中的一个重要环节，是研究海洋酸化对东海碳循环影响的重要因素。

关键词：海洋酸化；*Emiliania huxleyi*；钙化作用；光合作用；东海；呼吸速率

Abstract

Atmospheric CO_2 concentration has been increasing due to antropogenic CO_2 emissions since the industrial revolution. Ocean as an important "sink" of atmospheric CO_2 has uptaken roughly 30% of the anthropogenic CO_2. One of the consequences of the penetration of anthropogenic CO_2 into the ocean is the increase in the acidity of the ocean, and the reduction in pH and the saturation state of calcium carbonate in the ocean. Such a phenomenon has been identified as "ocean acidification". Coccolithophorid species *Emiliania huxleyi* is a major phytoplanktonic calcifier in the ocean, and the responses of photosynthesis and calcification of Coccolithophorid *Emiliania huxleyi* to ocean acidification have gained wide attention when researching the impact of elevated pCO_2 on ocean carbon cycling. Through batch and semi-continuous culturing of *Emiliania huxleyi*, we examined the impacts of pCO_2 on *Emiliania huxleyi* in different culturing methods. In addition, we investigated the community respiration rates in the East China Sea (ECS) in summer, in attempting to provide baselines for the future researches on response of carbon cycling through respiration to ocean acidification.

During the batch culturing, we simulated three levels of pCO_2 (380, 720 and 1100 ppm) using a pH controller to sustain 3 pH levels (8.2, 8.0 and 7.8) through bubbling high concentration CO_2 into the culture. We examined the effects of increased pCO_2 on the growth rate, POC production rate and calcification rate, as well as the PIC/POC ratio of *Emiliania huxleyi*. Without pre-adaption to pH changes, the growth of *Emiliania huxleyi* was inhibited at low pH. However, no significant changes were observed to the cellular POC production rates although the cellular calcification rates declined by 13.5% at pH=8.0 and 46% at pH=7.8 as compared to the control (pH=8.2). Consequntly, PIC/POC ratios were also reduced by 11.1% and 38.9%. We found that using different methods in the calculation of calcification rates had similar results but using the approach of alkalinity budgeting had higher precision than Ca^{2+} and PIC budgeting.

During the semi-continuous culturing, CO_2 concentrations under 520 and 1100 ppm

were manipulated by using a plant growth chamber. After fully adapted to pH changes, CO_2 concentration had no impact on the growth of *Emiliania huxleyi*, while POC production rate increased at high pCO_2, the variation of calcification rate in different pH lever was within the measurement precision. As a result, PIC/POC ratio reduced by 11.5% from pH= 8.0 to pH=7.8. C/N ratios of *Emiliania huxleyi* under both pH levels were higher than Redfield ratio and decreased with increasing pH. The respiration rate, determined from in vitro changes in dissolve O_2 during dark incubation, was elevated by 38.6% when pH decreased from 8.0 to 7.8.

The responses of biological processes of *Emiliania huxleyi* were distint under different incubation conditions. During the batch culturing, elevated pCO_2 inhibited their cell gowth and calcification, but had no effects on photosynthesis. During the semi-continuous culturing, elevated pCO_2 had no impact on the cell growth and calcification, but significantly promoted photosynthesis.These discrepancies imply that adapthion of cells to the pH change has an important effect on the results of acidification researches.

We investigated the community respiration rates in the ECS in summer by measuring the changes in dissolved O_2 in dark incubation. In the surface layer, community respiration rates changed from 1.2 to 27.8 μmol $L^{-1}d^{-1}$ and the average value was 9.0 μmol $L^{-1}d^{-1}$. At surface, the area of highest values was near to the mouth of the Changjiang Estuary and the nearshore region off Zhejiang. Community respiration rates generally decreased offshore. There were no obvious changes in community respiration rates vertically at stations not influented by Changjiang plume (salinity>32). But at stations influenceed by the Changjiang plume (salinity<32), community respiration rates decreased with depth. Community respirations were overall dominated by bacteria and phytoplankton. In the surface layer, community respiration rate was highly correlated with chlorophyll a (R^2=0.66) and phytoplankton contributed to 58.4% of the community O_2 consumption in the Changjiang plume area, while in the area not influented by the Changjiang plume the contribution of bacterial respiration was 62.9%. Below euphotic zone, community respiration rate was corrrelated to the bacteria abundance (R^2=0.53) and the bacterial respiration

contributed to 89.3% of the community respiration. In euphotic zone, the ratio of integrated primary production to respiration (P/R) varied within 0.2-0.9 and the average value was 0.5, suggesting that the ECS was predominantly heterotrophic in summer. Organic carbon to fuel the heterotrophic respiration must be therefore added from the river plum or resuspended sediments.

In summary, when *Emiliania huxleyi* was in different conditions of adaptation to pH changes, its responses of photosynthesis and calcification to ocean acidification were distinct. Community respiration plays an important role in carbon cycle in the East China Sea and is an important factor when we research the impact of ocean acidification on carbon cycle in this area.

Keywords: ocean acidification; *Emiliania huxleyi*; calcification; photosynthesis;

the East China Sea; community respiration rate