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Biomass productivity of native algal communities in Minamisoma city, Fukushima Prefecture, Japan

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ABSTRACT

Coastal areas in the Tohoku region were severely damaged by the tsunami that followed the March 2011 earthquake. Consequently, many lands have become non-arable. To assess the use of these non-arable lands for the production of algal biomass, we carried out experimental cultivations of native algal communities in Minamisoma city, Fukushima Prefecture, located in the cool temperate zone of Japan. Productivity of the native algal community was measured using 1-m^2 raceway ponds (120 L) and open vessel-type bioreactors (50 L–500 L), with different hydraulic retention times (HRTs) and in the presence or absence of sodium acetate. Maximum productivity in 1-m^2 raceway ponds (10-cm water depth) was observed with an HRT of 4 days and sodium acetate supplementation in June 2015 (13.2 g/m^2/day); average productivity throughout the year was 10.6 g/m^2/day. In the case of deep-water cultivation (80-cm water depth) in vessel-type cultures, the highest productivity (> 29 g/m^2/day) was obtained between January and September 2015. The dominant native microalgal species in the cultivations were *Desmodesmus* sp. and *Scenedesmus acuminatus*. Heterotrophic organisms, native bacteria, and protista, such as ciliates and amoeba, were also observed. Species composition and abundance varied depending on the medium content and seasonal conditions.

1. Introduction

Coastal areas in Fukushima Prefecture, including Minamisoma city, were severely damaged by the tsunami and the nuclear accident that followed the March 2011 earthquake. Many lands have been made non-arable by the tsunami. Basic guidelines for reconstruction of the area devastated by the earthquake, tsunami, and nuclear disaster were approved by the cabinet on 29 July 2011. One of the priorities is to promote the development of a world class advanced research center for renewable energy, around which to cluster related industries [1]. To meet the guidelines' indications, a R&D project for algae-based fuel production has been launched with financial support from the Fukushima Prefectural Government, the Japanese Reconstruction Agency, and the Ministry of Economy, Trade, and Industries.

The coastal area of Fukushima Prefecture is situated in the cool temperate zone of North East Japan. Average annual irradiance between 1986 and 2010 was ca. 1800 h, about half of that observed in North West Australia (> 3600 h) [2], and the mean temperature is 12.3 °C [3]. Specifically, temperatures range from 1.9 °C to 10.0 °C between November and April, and from 14.9 °C to 23.9 °C from May to October. Thus, the temperature fluctuation in this area is much higher than that in Karratha, North West Australia [4]. Similarly, wide annual temperature ranges are observed in cool temperate zones in Europe, Canada, China, Korea, Central Asia, Russia, etc. To enable commercial production of algae-based fuels in such cool temperate areas with large temperature fluctuations and relatively low irradiance, it is important to identify ways to efficiently produce algal biomass. Moreover, two-thirds of the island of Japan consists of mountain area; the flat land is mostly occupied by cities, industrial zones, and agriculture, and is very expensive. Thus, it is important to solve land issues for future commercialization of algae-based fuels.

In most cases, monocultures of oil-rich and high-growth microalgae have been used for algal fuel production. However, good growth under optimal and sub-optimal environmental conditions is limited by the

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tolerance level of the cultured species. In addition, algal monoculture is sometimes subjected to total algal biomass loss (pond crash) due to natural introduction of predator or pest species [5]. Without appropriate environmental controls, it is not possible to establish stable year-round biomass production in monoculture in temperate regions with large climate fluctuations. In contrast, the cultivation of native algal communities is suitable for large-scale biomass production and presents several benefits compared to monocultures of selected strains [5,6]: 1) stable culture and production owing to the absence of pond crash, 2) efficient use of resources (nutrients) by niche complementarity, 3) cheap and easy operation and maintenance. In addition, the culturing of native algal communities has been used for wastewater treatment in high-rate algal ponds (HRAPs) [7–9].

Considering the climate of coastal regions in the Fukushima Prefecture and the advantages brought by the use of native algal communities, we assessed the possibility of algae-based biofuel commercialization in this area by evaluating algae biomass production in Minamisoma city using raceway pond and vessel-type cultures and a native algal community. Based on the results, we here discuss future prospects for algal fuel production and commercialization in cool temperate regions.

2. Materials & methods

2.1. Location of the experimental facility

Cultivation experiments were conducted at the Minamisoma R&D center for algal biomass production, Minamisoma city, Fukushima Prefecture (37.644673°N, 141.009490°E, Fig. 1), managed by the Algae Industry Incubation Consortium, Japan (https://algae-consortium.jp/).

2.2. Native algal communities for cultivation

We collected native algal communities (25–100 μm in size) from three ponds located near the experimental facility in Minamisoma city. Pond water was first filtered through a 100-μm-mesh plankton net to remove large-size zooplankton and then passed through a 25-μm-mesh plankton net. The concentrated samples were mixed and seeded in 1-m^2 raceway ponds. The dominant microalgal genera in the samples were identified to be *Desmodesmus* spp., *Scenedesmus* spp., *Dictyosphaerium* spp., and *Klebsormidium* sp. based on light microscopy.

2.3. Cultivation in 1-m^2 raceway ponds

The productivity of native algal communities between October 2014 and September 2015 was investigated using 1-m^2 raceway ponds (120 L, 10-cm water depth, Fig. 2) placed in a greenhouse. However, because of heavy rains in the middle of September 2015, experimental facilities were flooded and we were not able to obtain data on algal productivity for that month. The greenhouse had ventilation windows equipped with a mesh curtain to keep out birds and large insects. Native algal community cultivations were conducted under hydraulic retention times (HRTs), over 4, 7, and 14 days, and in the presence or absence of sodium acetate (1 g/L). Medium K [10] was used as a standard inorganic medium and air containing 1% (v/v) CO_2 was aerated with a 20-cm-long sparger at a flow rate of 1 L/min. Rotation speed of the paddle wheel was set to 15 rpm. Flow rate was approximately 30 cm/s at the center of the waterway. Apart from CO_2 aeration, pH was not controlled and varied from 8.6 to 9.2 during the experiments. All experiments were carried out in triplicate for each condition.

A semi-continuous culture technique was used in the experiments. We harvested 50% (60 L for HRT = 4 days), 33.3% (40 L for HRT = 7 days), or 16.7% (20 L for HRT = 14 days) of culture suspension every two days, and replaced it with new medium (60 L, 40 L, and 20 L, respectively). All semi-continuous cultures were started in the middle of September 2014 and reached an apparent steady state of growth by October 2014, when the investigations on algal productivity were launched.

The harvested culture suspension was divided in two samples. One sample (10–20 mL) was filtered through a GF/C filter (Whatman, Little Chalfont, UK) and then dried in a dehydrator (105 °C) for several days to measure dry cell weight (DCW). The other sample (20 mL) was fixed with glutaraldehyde (2.5%) and used for species identifications and for evaluating their frequencies under a light microscope equipped with Nomarski differential interference contrast optics (Leica DM2500; Leica, Tokyo, Japan). For identification of one of the dominant alga *Scenedesmus acuminatus*, we established unialgal strains, observed on their morphological characteristics, and analyzed 18S rDNA sequences according to the previous methods [11]. The identified species or taxa were classified into four categories based on their frequencies (0, 1, 2, 3, 4). For this purpose, the fixed sample was concentrated 10 times by moderate centrifugation (10 × *g*, 20 min), the supernatant was discarded, and 10 μL of concentrated sample was placed under the microscope (400 ×). After checking the entire preparation area, cells present in < 10% of viewing fields resulted in the organism being ranked as “1.” When cells were present in approximately 10% of viewing fields, the organism was ranked as “2.” When cells appeared in approximately 50% of viewing fields, the organism was ranked as “3.” Maximum frequency of appearance was ranked as “4” and corresponded to cells being present in every viewing field. Undetected taxa

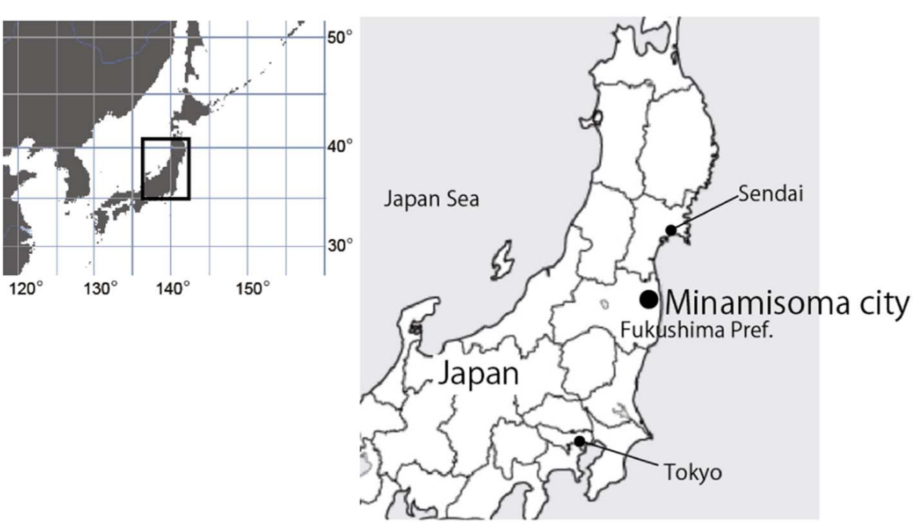

Fig. 1. Location of Minamisoma city, Fukushima Prefecture.