

## Summary

Entrance Year : 2014

United Graduate School of Agricultural Sciences

Course : Resources and Environmental Science of Agriculture, Forestry and Fisheries

Name : Yuki Watanabe

<b>Title</b>	The photosynthetic response of a few Japanese species of <i>Pyropia</i> (Bangiales, Rhodophyta) to an environmental gradient
--------------	--

Key word (Algae) (**Photosynthesis**) (**Pulse amplitude-modulated (PAM)-chlorophyll fluorometry**)

### Chapter 1: General Introduction

The species of red algal genus *Pyropia* is distributed widely in the subarctic, temperate, and tropical waters in the world. These species are regarded as an edible resource, and a few cultivars including *Pyropia yezoensis* f. *narawaensis* are cultivated throughout Japan. Currently, around 60,000 tons of dry weight (= 340,000 tons of wet weight) of gametophyte has been harvested annually in Japan.

The optimum growth environment and photosynthetic characteristics of the Japanese macro algae have been well-studied since the 1950s and 1960s, and have contributed to the development of cultivation systems for increased production. In addition, the discovery of the heteromorphic life history of *Pyropia* has also to improve the techniques of Japanese *Nori* cultivation. However, these reports regarding the ecophysiology of *Pyropia* have mostly focused on the cultivars or commercial species. Modern knowledge of their ecophysiology as well as of other native species, is essential for the optimization of mariculture of these species and conservation of natural communities.

In recent years, several photosynthesis-related investigations have utilized the pulse amplitude-modulated (PAM)-chlorophyll fluorometer to elucidate photosynthetic responses of whole intact plants and macroalgae. This instrument can quickly and efficiently evaluate their photosynthetic responses, particularly the photochemical efficiency of photosystem II, to various stresses. While reports regarding the physiological responses of Chinese species of *Pyropia* under various environmental stresses (e.g. temperature, salinity and dehydration) are well-documented, those of Japanese species remain limited.

In this study, I focused on elucidating the photosynthetic responses of some Japanese species of *Pyropia*, including the cultivated and endangered ones, to various optimal and stressful environments by using PAM-chlorophyll fluorometry. Knowledge on these aspects shall lead to continued advances in cultivation to ensure sufficient supply of these important fisheries species.

Chapter 2: The effect of irradiance and temperature on the photosynthesis of a cultivated red alga, *Pyropia tenera* (= *Porphyra tenera*), at the southern limit of distribution in Japan

The effect of irradiance and temperature on the photosynthesis of the red alga, *Pyropia tenera*, was determined for maricultured gametophytes and sporophytes collected from Yatsushiro Bay, Kyushu, Japan, using both PAM-chlorophyll fluorometry and dissolved oxygen sensors.

A model of the net photosynthesis–irradiance ( $P$ - $E$ ) relationship of the gametophytes at 12°C revealed that the net photosynthetic rate quickly increased at irradiances below the estimated saturation irradiance of 46  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ , and the compensation irradiance was 9  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ . Gross photosynthesis rates (GP) and dark respiration rates (DR) for the macroscopic gametophytes were determined over a range of temperatures (8–34°C), revealing that the GP of 46.3  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1}$  was highest at 9.3°C, and the DR increased at a rate of 0.93  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1} \text{°C}^{-1}$ . The measured DR ranged from -0.06  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1}$  at 6°C to -25.2  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1}$  at 34°C. The highest value of the maximum quantum yield ( $F_v/F_m$ ) for the gametophytes occurred at 22.4°C and was 0.48, although those of the sporophyte occurred at 12.9°C and was 0.52. This species may be considered well-adapted to the current range of seawater temperatures in this region. However, since the gametophytes have such a low temperature requirement, they are most likely close to their tolerable temperatures in the natural environment.

Chapter 3: Photosynthetic response of *Pyropia yezoensis* f. *narawaensis* (Bangiales, Rhodophyta) to a thermal and PAR gradient vary with the life-history stage

The effect of PAR and temperature on photosynthesis of the cultivated red alga, *Pyropia yezoensis* f. *narawaensis* (Saga-#5 Strain), was determined for microscopic sporophytes and macroscopic gametophytes with PAM-chlorophyll fluorometry and dissolved oxygen sensors.

A clear difference in the temperature response of the  $F_v/F_m$  and GP was revealed between the two life-history stages. The microscopic sporophyte  $F_v/F_m$  was not sensitive to temperature, although the model revealed an  $F_v/F_m$  maximum (0.60) at 16.7°C. In contrast, the macroscopic gametophyte  $F_v/F_m$  was clearly sensitive to temperature and attained a maximum mean value of 0.55 at 14.7°C. The relationship between GP and temperature was also different. The maximum GP of the sporophyte occurred at 30.7°C and was 17.1  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1}$ ; however, the maximum GP of the gametophyte were much higher (110  $\mu\text{g O}_2 \text{mg}_{\text{chl-a}}^{-1} \text{min}^{-1}$ ) and occurred at a lower temperature (14.4°C). The response of oxygenic net photosynthesis to PAR also varied, and the initial slope ( $a$ ), the saturation PAR ( $E_k$ ) and the maximum photosynthetic rate ( $P_{\text{max}}$ ) of the gametophyte were much higher than the sporophyte. Therefore, I hypothesized that in species with a heteromorphic life history, such as those in the genus *Pyropia*, physiological responses to environmental gradients would be dissimilar.

Chapter 4: The effect of irradiance and temperature on the photosynthesis of two red algal gametophytes, *Pyropia dentata* and *Pyropia seriata*, from Kyusyu Island, Japan

The effects of irradiance and temperature on the photosynthesis of the red algae, *Pyropia dentata* and

*Pyropia seriata*, were determined for naturally occurring gametophytes collected from Kyushu Island, Japan. Photosynthetic efficiency was measured by using both PAM-chlorophyll fluorometry and dissolved oxygen sensors.

The  $F_v/F_m$  was determined over a range of temperatures (8–36°C), revealing that the highest values in *P. dentata* and *P. seriata* occurred at 11.9°C and 12.2°C, respectively. Additionally, the highest GP in the two species were 40.4  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 26.3°C and 66.6  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 20.7°C, respectively. Furthermore, the DR increased from 0.91  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 8°C to 15.5  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 36°C for *P. dentata*, and increased from 2.52  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 8°C to 16.7  $\mu\text{g O}_2 \text{ g}_{\text{w.w.}}^{-1} \text{ min}^{-1}$  at 36°C for *P. seriata*. The response of oxygenic net photosynthesis to PAR was different between them. The estimated  $E_k$  of *P. dentata* and *P. seriata* was 105  $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$  and 209  $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$ , respectively.

## Chapter 5: Chronological change and the potential of recovery on the photosynthetic efficiency of *Pyropia yezoensis* f. *narawaensis* (Bangiales) during the sporelings frozen storage treatment in the Japanese *Nori* cultivation

The chronological change of photosynthetic efficiency in a frozen storage treatment of the Japanese *Nori* cultivation industry was examined in the cultivated red alga, *Pyropia yezoensis* f. *narawaensis* (Saga-#5 Strain) by using PAM-chlorophyll fluorometry.

During the desiccation process that was conducted after the nursery cultivation season in November, the  $F_v/F_m$  of the gametophytic sporelings growing on the *Nori*-net decreased monotonically with decreasing absolute water content (AWC), and was around 0.1 at 20 % AWC. During frozen storage of the *Nori*-net, the  $F_v/F_m$  of the frozen gametophyte was low but stable, and ranged between  $0.10 \pm 0.02 \text{ SD}$  and  $0.14 \pm 0.05 \text{ SD}$ . The magnitude of  $F_v/F_m$  for the gametophyte of the freezing treatment, after 10 min and 3 h of immersion in seawater, recovered quickly. After 10 min and 3 h of immersion, these values were  $0.29 \pm 0.12 \text{ SD}$  and  $0.47 \pm 0.05 \text{ SD}$  during the 14 days of freezing treatment, and  $0.15 \pm 0.02 \text{ SD}$  and  $0.29 \pm 0.04 \text{ SD}$  after 71 days of freezing treatment, and suggest that the ability to recover gradually decreased as the storage duration increased. The response of  $F_v/F_m$  from general cultivation (i.e. directly cultivated from the nursery cultivation season) and those after 47 days of freezing were almost identical, suggesting that the current *Nori*-net frozen storage period was not detrimental to the gametophyte.

## Chapter 6: General Consideration

In the heteromorphic life history of *Pyropia*, maximal rates of photosynthetic activity for each of the life-history stages seemed to coincide with what each stage experienced in their natural environment. Indeed, optimum temperature range of photosynthetic efficiency for the macroscopic gametophyte and microscopic sporophyte was most likely related to the temperatures occurring during the life-history

stages in their natural habitat. In the present study, temperature optima of the photosynthetic efficiency of macroscopic gametophytes and microscopic sporophytes were elucidated for the first time in *P. tenera* and *P. yezoensis* f. *narawaensis*. Furthermore, PAR optima and the photosynthetic characteristics were also different in the two life-history stages in *P. yezoensis* f. *narawaensis*. Especially, the  $E_k$  and the  $P_{max}$  of macroscopic gametophyte were greater than those of microscopic sporophyte. It is likely to reflect the peculiar low-PAR environment of the sporophytes, which occur in the dead oyster shells. Adaptation to low-PAR conditions and resistance to relatively high temperature conditions might be one of the mechanisms that enable the sporophytes to survive the summer period.

Characteristic temperature and PAR optima on photosynthesis was also observed in two wild gametophytes of *P. dentata* and *P. seriata*. These two species showed a similar trend of photosynthesis under temperature or PAR gradients. However, optimal temperature,  $E_k$  and  $P_{max}$  was different, suggesting that adaptation to the environment is closely related to the distribution and habitat of each species.

Characteristic tolerance to the desiccation and freezing on the photosynthesis of *P. yezoensis* f. *narawaensis* was also confirmed. During the desiccation process, the  $F_v/F_m$  of the gametophytes decreased monotonically with decreasing AWC, however, it had still potential of recovery. During the frozen storage period, the  $F_v/F_m$  of the frozen gametophytes were low but stable, and after 10 min and 3 h of immersion in seawater, recovered quickly; however these values were decreasing with prolonged freezing. These results suggest, that the relevant period of storage in Japanese *Nori* cultivation is not detrimental for the cultivation. However, it is necessary to examine longer freezing periods for further study.

Finally, PAM-chlorophyll fluorometry was a powerful non-destructive measurement tool to elucidate the quick response of photosynthetic efficiency to various optimal and stressful environments including temperature, irradiance and desiccation. The use of such equipment has definitely provided satisfactory information regarding the photosynthetic performance of several seaweeds, which will in some way lead to the development and proper management of seaweed mariculture in Japan.