Performance of the Ammonia-nitrogen Uptake by Macroalga Sterile *Ulva* sp. (Chlorophyta)

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Introduction

Water quality has been identified to be one of the most important factors concerning the sustainability of the shrimp culture industry. The major problem associated with the closed intensive shrimp culture system is the accumulating of the ammonia-nitrogen (ammonia-N) resulting from the decomposition of the feeding. The ammonia-N not only can cause the eutrophication but also has the acute toxicity. Even existing in very low concentration (8.6 μM-N), it can also retard the growth of the shrimp. How to remove the ammonia-N in the closed environment has become the key point. On the other hand, for seaweed the ammonia-N is indispensable nutrition. So, as an economical and feasible proposal, using macroalgae to remove the ammonia-N has been studied in recent years.

In this study, using the sterile *Ulva* sp., a kind of easily obtained macroalga, the performance of the ammonia-N uptake was investigated.

1. Experimental

The collecting and preconditioning of the sterile *Ulva* sp. were as same as the previous work [1]. The algae used in this study were in ammonia-N starved condition. The experimental apparatuses are as shown in **Figure 1(a)** and **(b)**. While the run① with different starting ammonia-N concentrations were carried out at different time, the run② were conducted at the same time in order to cancel the effect of algae nutrition condition's difference and so on [2]. The experimental conditions are shown in **Table 1**. Selecting these conditions was to make observing the uptake performance in the respective uptake phase easier.

2. Results and Discussion

2.1 Uptake phases in ammonia-N uptake

The example of the time courses of the ammonia-N concentration and uptake rate is shown in **Figure 2**. The concentration of ammonia-N in the culture medium decreased with time and so did the uptake rate. The three distinct, succeeding phases of the uptake rate, which is called surge uptake, internally controlled uptake and externally controlled uptake, were observed. In surge uptake phase lasting for the first 1h, the rate was relatively high. This phenomenon seemed to support the theory proposed by Rosenberg (1984) and Fujita (1985) that the presence of surge uptake constitutes an adaptive response to nitrogen limitation in macroalgae[4][5]. For the wide fluctuations of ammonia-N concentration in shrimp culture ponds, the ability of surge ammonia uptake would give this macroalga the competitive advantage and make it the optimum plant for removing ammonia-N from the shrimp pond. Besides, in this phase, with the ammonia transported into the algae's cells and assimilation rate inside the cells increasing, the uptake rate declined abruptly. It suggests that the uptake rate not only be connected with the ammonia-N concentration in culture medium but also the ammonia-N status of the alga itself. When the rate of the membrane transport equals to the assimilation rate, the uptake rate kept relative constant, which is called internally controlled uptake phase. After this phase, with the Ammonia-N concentration in the culture medium dropped to a very low

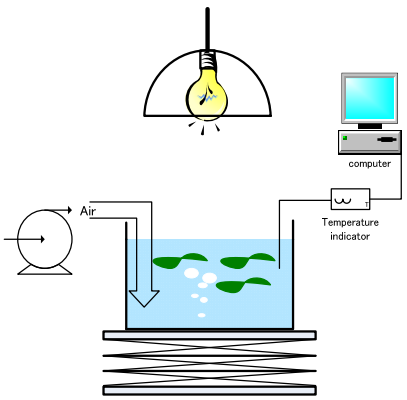

Figure 1(a) Apparatus for run①

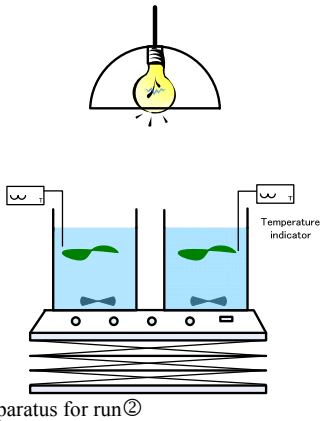

Figure 1(b) Apparatus for run②

Table 1. Principal experimental conditions

	run①	run②
Apparatus	A	B
Light [μ mol・m^{-2}・s^{-1}]	1800	1800
T[℃]	30±2	25±2、30±2
$C_{TAN,0}$ [μ M]	16～66	36～161
V [L]	5	2
M_{alga}[g]	4.2～4.9	2.7～3.4
Acclimation time [min]	30	30
Aeration	○	×
Light : Dark cycle	14 : 10	14 : 10
Culture medium	artificial seawater	
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level, the uptake rate also decreased, which is called externally controlled uptake.

2.2 Effect of ammonia-N concentration on uptake

Figure 3(A) and **(B)** present the effect of ammonia-N concentration in the culture medium on the time courses of the ammonia-N concentration and the uptake rate. The surge uptake appeared across a wide range of initial ammonia-N concentration. Moreover, the uptake rate of the run with high starting concentration trended to be higher. On the other hand, in the internally controlled uptake phase, the rate dropped to the identical level. This result indicated that the uptake rate in this phase had no relation with the ammonia-N content in tissue. Further, with the decrease of the starting concentration, the lasting time of the internally controlled uptake turned short.

2.3 Initial uptake rate at different temperatures

The results show that at 25°C the performance of the ammonia-N uptake was similar with that at 30°C. That is to say at the lower temperature, the sterile *Ulva* sp. also had the capability of the ammonia-N uptake. The differences were appeared in the initial uptake stage, which is also called membrane transport process (see **Figure 4**). The initial uptake rate at 30°C was higher. In this process, the behavior of the carrier protein in the cell membrane has the same mechanism with the enzyme-catalyzed reactions and the kinetics can be described as the Michaelis-Menten equation [1]. The experiments were carried out on different days and the ammonia-N contents in tissue were different. Although as mentioned above in surge uptake phase uptake rate is also connected with the ammonia-N content in tissue, the plotting of the initial transport rates vs. initial ammonia-N concentration still quit fitted to the Michaelis-Menten plotting. This indicates that at the same temperature the most important factor affecting on the uptake rate in the membrane transport process should be the initial ammonia-N concentration in culture medium but not the ammonia-N content in tissue.

Conclusion

The surge uptake during the uptake process was observed across a wide range of initial ammonia-N concentrations. In internally controlled uptake phase, the uptake rate has no relation with the ammonia-N content in tissue. The ammonia-N uptake capability of sterile *Ulva* sp. at different temperatures was confirmed. The initial uptake rate increased with temperature. At the same temperature, the most important factor affecting the uptake rate in the membrane transport process is the initial ammonia-N concentration in culture medium.

Nomenclature

0 = initial

C_{TAN} = concentration of ammonia-N in the culture media [μM-N]

$\pi_{t,TAN}$ = membrane transport rate of ammonia-N [μmol-N·g-DW^{-1}·h^{-1}]

$\pi_{u,TAN}$ = uptake rate of ammonia-N [μmol-N·g-DW^{-1}·h^{-1}]

References

[1] Sato, K., Eksangsri, T., Egashira, R.; "Ammonia-Nitrogen Uptake by Seaweed for water quality Control in Intensive Mariculture Ponds," J. Chemical Engineering of Japan, 39, 247-255 (2006)

[2] Morten Foldager Pedersen; "Transient Ammonium Uptake in the Macroalga *Ulva* Lactuca (Chlorophyta): Nature, Regulation, and the Consequences for Choice of Measuring Technique" J. Phycol 30, 980-986 (1994)

[3] Onishi M; "The Approach to the Experiment on Enzyme Reaction Kinetics" ISBN 4-7622-0518-4 (1987)

[4] Rosenberg, G., Probyn, T.A.&Mann, K.H.; "Nutrient uptake and growth kinetics in brown seaweeds: response to continuous and single additions of ammonium," *J. Exp. Mar. Biol. Ecol.,* 80, 125-46 (1984)

[5] Fujita, R. M.; "The Role of Nitrogen Status in Regulating Transient Ammonium Uptake and Nitrogen Storage by Macroalgae," *J. Exp. Mar. boil. Ecol.*, 92, 283-301 (1985)

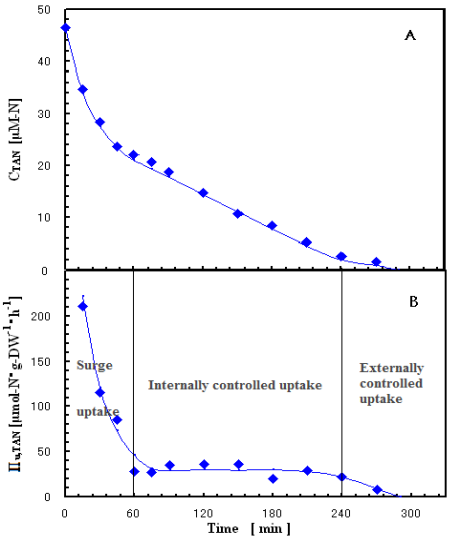

Figure 2 The time courses of ammonia-N concentration (A) and uptake rate (B) in run①

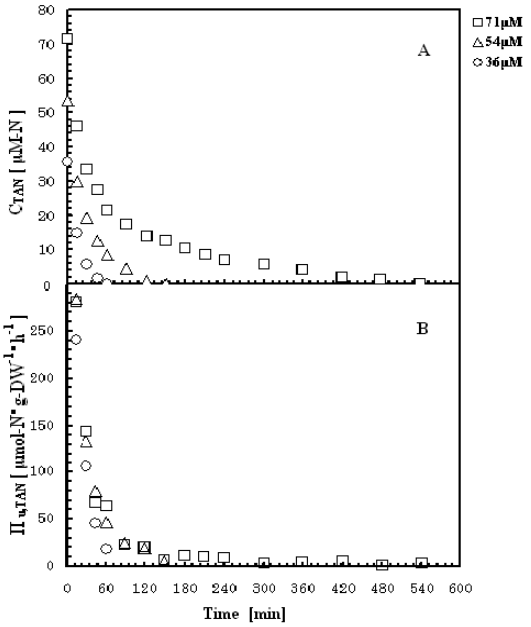

Figure 3 The time courses in run②: (A) ammonia-N concentration; (B) ammonia-N uptake rate

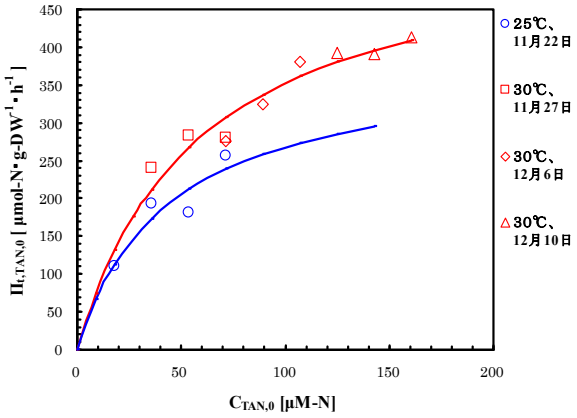

Figure 4 The effect of ammonia-N concentration on the initial uptake rate at different temperatures