

## The Effects of Nitrogen as $\text{NO}_3^-$ and $\text{NH}_4^+$ on the Growth and Symbiont (*Anabaena azollae*) of *Azolla pinnata* R. Brown

Arunothai Jampeetong<sup>1,2</sup>, Thunyachol Sripakdee<sup>1</sup>, Tanaporn Khamphaya<sup>1</sup> and Sutthathorn Chairuangstri<sup>1,2\*</sup>

<sup>1</sup>Department of Biology, Faculty of Science, Chiang Mai University, Chiang Mai 50200, Thailand

<sup>2</sup>Environmental Science Program, Faculty of Science, Chiang Mai University, Chiang Mai 50200, Thailand

\*Corresponding author. E-mail: s.suwann@gmail.com

### ABSTRACT

*The growth, morphology, and symbiont (Anabaena azollae) of Azolla pinnata R. Brown were investigated under different external N-supply regimes to inform the plant's potential in wastewater treatment. Azolla pinnata plants were supplied with nitrogen as  $\text{NO}_3^-$  or  $\text{NH}_4^+$  at four different concentrations (0, 0.5, 1, and 5 mM) and incubated in a greenhouse for 14 days. The relative growth rates of  $\text{NO}_3^-$ -fed plants were not significantly different between treatments, but decreased significantly at the highest  $\text{NH}_4^+$  concentration. Moreover, the  $\text{NO}_3^-$  concentration did not affect root number. The highest  $\text{NH}_4^+$  concentration (5 mM) decreased both the root length and number of symbionts (Anabaena azollae) in the mature leaves of Azolla pinnata. Because Azolla pinnata continued to grow well with supplied  $\text{NO}_3^-$  and  $\text{NH}_4^+$ , and retained their ability to absorb nitrogen, they offer potential for treating wastewater, except at the highest  $\text{NH}_4^+$  concentration, which led to toxicity.*

**Keywords:** *Azolla pinnata*, *Anabaena azollae*, Heterocyst,  $\text{NH}_4^+$  toxicity, Symbiont

### INTRODUCTION

*Azolla pinnata* R. Brown is a free-floating aquatic fern belonging to the family Azollaceae. It is widely distributed in Asia and along the coast of tropical Africa (Wagner, 1997). The plant consists of alternately arranged leaves on a prostrate, floating rhizome, with one or two roots hanging in the water column. The leaf is bilobed, consisting of a chlorophyllous floating dorsal lobe and a colorless and partially submerged ventral lobe. A cavity in the ventral leaves houses symbiotic cyanobacteria, *Anabaena azollae* (Pabby et al., 2003). This symbiont fixes  $\text{N}_2$  from the atmosphere and produces a high N level in the plant tissue of *Azolla pinnata*, making the plant useful as green manure for rice fields, where it has been used for several centuries (Shi and Hall, 1988; Peters and Meeks, 1989; Forni et al., 2001; de Macale and Vlek, 2004).