



RESEARCH PAPER

Effects of water turbulence on variations in cell ultrastructure and metabolism of amino acids in the submersed macrophyte, *Elodea nuttallii* (Planch.) H. St. John

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ABSTRACT

The interactions between macrophytes and water movement are not yet fully understood, and the causes responsible for the metabolic and ultrastructural variations in plant cells as a consequence of turbulence are largely unknown. In the present study, growth, metabolism and ultrastructural changes were evaluated in the aquatic macrophyte *Elodea nuttallii*, after exposure to turbulence for 30 days. The turbulence was generated with a vertically oscillating horizontal grid. The turbulence reduced plant growth, plasmolysed leaf cells and strengthened cell walls, and plants exposed to turbulence accumulated starch granules in stem chloroplasts. The size of the starch granules increased with the magnitude of the turbulence. Using capillary electrophoresis–mass spectrometry (CE-MS), analysis of the metabolome found metabolite accumulation in response to the turbulence. Asparagine was the dominant amino acid that was concentrated in stressed plants, and organic acids such as citrate, ascorbate, oxalate and γ -amino butyric acid (GABA) also accumulated in response to turbulence. These results indicate that turbulence caused severe stress that affected plant growth, cell ultrastructure and some metabolic functions of *E. nuttallii*. Our findings offer insights to explain the effects of water movement on the functions of aquatic plants.

INTRODUCTION

The array of functions provided by aquatic plants is essential to maintain the ecological balance of aquatic ecosystems. Some of the important functions of aquatic plants include primary production, provisioning of food, habitat and refuge for different aquatic fauna, contributions to biogeochemical cycles and regulation of sediment transport, among others (Bornette & Puijalon 2011; Folkard 2011; Nepf 2012). Aquatic plants experience various disturbances in aquatic systems for two reasons: the highly dynamic nature of the systems and the sedentary nature of the plants (Chehab *et al.* 2009). The productivity of plants is highly dependent on environmental stresses and a wide range of fluctuations in specific abiotic factors, such as water movement, sediment properties, macro- and micronutrient levels in the water column, availability of light, UV radiation, toxic metals and temperature, as reported in the literature (Bornette & Puijalon 2011). Among the abiotic factors, water movement is the most important for submersed aquatic plants because the flow-driven drag and lift forces have either a positive or a negative effect depending on their magnitude. For example, low flow is beneficial for photosynthesis (Westlake 1967), whereas flow-driven mechanical stresses alter growth, physiology and morphology in aquatic plants (Asaeda *et al.* 2010; Bornette & Puijalon 2011).

In addition to plant growth, biochemical evidence is also widely applied to detect plant stress. For example, the accumulation of reactive oxygen species (ROS) is one of the primary

signals of stress (Apel & Hirt 2004; Gill & Tuteja 2010), and plants have evolved defence mechanisms to counter the damage caused by excess ROS in stressful environments. One mechanism is the activation of ROS scavenging antioxidant enzymes such as catalase, peroxidase, superoxide dismutase and ascorbic peroxidase (Apel & Hirt 2004; Gill & Tuteja 2010). Another defence mechanism is to accumulate multiple compatible solutes to protect the plant cell against oxidative damage (Liu *et al.* 2011). The osmolytes produced under stress are regarded as an osmoregulation mechanism that protects cells from damage, and in addition to osmoregulatory protection, these osmolytes are also ROS scavengers (Bohnert & Shen 1998). In this context, the accumulation of amino acids is widely thought to be a direct protective mechanism in many terrestrial plants (Rhodes *et al.* 1999).

In the natural environment, turbulence is one of the primary causes of mechanical stress exerted on submersed aquatic plants (Champika Ellawala *et al.* 2011; Atapaththu & Asaeda 2015). Although the stress responses of submersed plants to turbulence were reported previously (Champika Ellawala *et al.* 2011; Ellawala *et al.* 2013), information on the variation in amino acid metabolism with exposure to turbulence is scarce, to our knowledge. Furthermore, the ultrastructural changes in cells of submersed plants in response to turbulence are largely unknown. Ingber (2003) proposed that understanding the mechanisms behind the mechanical forces that regulate and integrate cellular activities in plants should be one of the new

