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Characteristics of transmembrane proton transport in the cells of *Lupinus polyphyllus*

Research Article

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Abstract: The aim of this work was to examine H^+-ATPase hydrolytic activity and the stability of transmembrane electrochemical gradients in membrane vesicles isolated from seedlings and leaf cells of an invasive Washington lupine (*Lupinus polyphyllus* Lindl.) and compare them with non-invasive yellow lupine (*Lupinus luteus* L.). Temperatures of 25 and 30°C, keeping in mind possible climate warming, were used. For harder stress conditions, short term cold treatment (-8°C *in vivo*) was used. Plasmalemma-, tonoplast-, and endoplasmic reticulum-enriched membrane fractions were obtained from a sucrose density gradient and identified. Differences in ATPase hydrolytic activity were significant only between lupine species and were more obvious in plasmalemma-enriched fractions. Preincubation of seedlings and leaves at -8°C for 15 min to 24 h showed that microsomic fraction membranes of invasive lupine were more stable (according to Na^+-diffusive potential) at low temperature compared to yellow lupine ones. The level of transmembrane electrochemical potential, mainly evoked by ATP-dependent active proton transport, was almost equal in both lupine species. Supposedly, the cells of invasive lupine are able to maintain transmembrane electrochemical potential by the employment of lower hydrolytic activity of H^+-ATPase, thus saving energy for growth.

Keywords: *H^+-ATPase • Cold stress • Invasiveness • Transmembrane electrochemical gradient • Lupinus polyphyllus*

1. Introduction

The Washington lupine (*Lupinus polyphyllus* Lindl.) is one of the most aggressive invasive plants in Lithuania, capable of forcing out local species from their habitats. What are the cellular characteristics of such an invasive plant? In terms of plant response reactions, some of them might include responses controlled by the hormonal system, stability and functioning of roots, seed spread and ripening features, and the production of specific chemical substances that influence neighbouring plants. Any of these processes might contribute to the survival of an aggressive plant in non-native conditions; nevertheless, one of the essential processes in a cell of an invasive plant must be tolerance of new stress conditions. The secondary invasion to less favourable conditions for invasive plants is also mentioned: P_i uptake pattern with the highest affinity at submicromolar range was supposed to be of critical importance for the capacity of *Hakea sericea* to invade and proliferate throughout vast areas of nutrient-deprived soils [1]; a shift in life-history strategy during expansion in two most stressful habitats was also described for *Acer negundo* [2].

Plant responses to environmental influences are strongly determined by their capability to support cell homeostasis. One of the key molecular players in cell homeostasis is plasmalemma H^+-ATPase, which establishes a proton-electrochemical gradient across the plasma membrane, as well as the tonoplast H^+-ATPase and H^+-PPase, which are involved in such intricate physiological processes as intracellular pH regulation, cell elongation, and adaptation to different temperature stresses [3-8]. In our earlier work dealing with H^+-ATPase activity and proton transport peculiarities in freezing-tolerant and non-tolerant genotypes of meadow fescue (*Festuca pratensis* Huds.), we showed that stress responses varied in different genotypes according to acclimation and freezing temperatures [9].

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