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SEASONAL CHANGES IN POLAR LIPID FATTY ACID COMPOSITION COINCIDE WITH CHANGES IN FREEZING STRESS RESISTANCE IN PINE NEEDLES

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INTRODUCTION: Seasonal changes in the freezing stress resistance of woody plants have been observed to be parallel to the variation in the amount of phospholipids(1). Some researchers have correlated changes, of individual phospholipids (4) and the increase in unsaturation of the fatty acids in these lipids, to an increase in freezing stress resistance (3,10). However, in some studies this relationship has not been found (5,6). On pine species, only limited reports are available. In the present study, seasonal changes in the fatty acid composition of polar lipids and freezing stress resistance of pine needles were examined.

MATERIALS AND METHODS: The needles from five different Red pine (*Pinus resinosa*) and Austrian pine (*Pinus nigra*) trees were collected at 2-4 week intervals between March, 1988 and May, 1989. The youngest age class of needles was used for the analyses.

Excised needles were slowly frozen to -20 °C (summer samples) or to liquid nitrogen (winter samples). After thawing, the viability of the needles was assessed by ion leakage and visual water soaking (7). The frost killing temperature was calculated according to the relationship developed by Sutinen and Palta (8).

Membrane polar lipids from 1 gram of needles were extracted using a modified procedure of Bligh and Dyer (9). This polar lipid fraction was then methylated under alkaline conditions to obtain fatty acid methyl esters. Fatty acid composition was determined by using a Shimadzu GC-9AM gas chromatograph.

RESULTS AND DISCUSSION: The amount of linoleic acid (18:2) and stearic acid (18:1) was highest at the end of January (Fig. 1). At that time, the needles were most hardy and could survive the liquid nitrogen temperature (Fig. 2). A dramatic increase in freezing stress resistance in November was accompanied by an increase in 18:2 fatty acid. Decrease in freezing stress resistance during spring and summer was accompanied by a decrease in 18:2. Throughout the year, seasonal changes in freezing stress resistance paralleled changes in 18:2. The changes in 18:1 were similar to 18:2 but to a lesser extent. The changes in palmitic acid (16:0) were opposite to the changes in 18:1 and 18:2 (Fig. 1). Similar seasonal trends were observed in the case of Austrian pine needles (data not shown).

Hellergren et al. (6) did not observe any connection between the level of unsaturation of fatty acids of plasma membranes and freezing stress resistance in pine needles. However, in that study, needles of artificially acclimated pine seedlings, which gained the freezing stress resistance of only -32 °C, were used. In thylakoid membranes of spruce needles, an increase in the unsaturation of fatty acids has been reported in winter needles when compared to summer needles (3).

In plasma membranes of mulberry bark cells, the ratio of unsaturated to saturated fatty acids increased parallel to freezing stress resistance (10). In this study, an increase in the amount of 18:2 fatty acid and a slight decrease in 16:0 during cold acclimation was found. However, no changes in the amount of 18:1 fatty acid were observed (10). In another study, an increase of 18:2 and decrease of 16:0 fatty acids accompanied by an increase of Mg²⁺-ATPase activity in plasma membranes of potato leaves have been found (11). This suggests that changes in membrane lipids may be important in mediating the membrane protein function.

An increase in unsaturation level has been interpreted to mean an increase in the "fluidity" of the membranes.

Further studies are under way to determine specific lipid changes and ATPase activity changes in plasma membranes of pine needles.

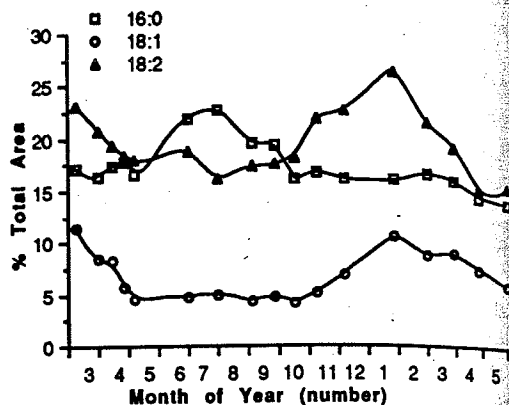


Fig. 1 Seasonal changes in fatty acid composition of total polar lipid in Red pine needles. Values are mean of 5 reps (SD<10%).

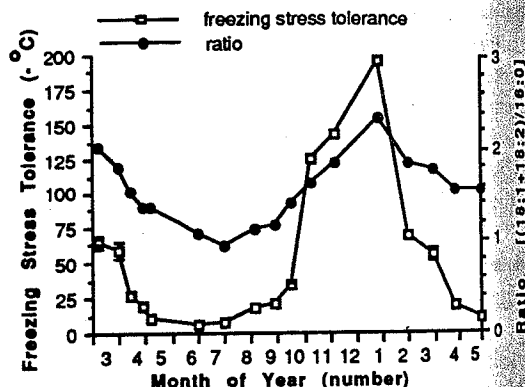


Fig. 2 Seasonal changes in freezing stress tolerance and the ratio of 18:1 and 18:2 to 16:0 fatty acids of polar lipids of Red pine needles.

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