

Survival and physiological parameters of embryogenic tissues of *Picea abies* cultured *in vitro*

E. Nawrocka*, H. Fuchs, T. Hazubska-Przybył, E. Ratajczak

Institute of Dendrology, Polish Academy of Sciences, Parkowa 5, 62-035 Kórnik, Poland,

*enawrocka@man.poznan.pl

Keywords: cellular respiration, *in vitro* culture, plant regulators, reactive oxygen species

This study aimed to evaluate the survival of selected embryogenic tissue lines of *Picea abies* and to explore potential markers of genotype-specific viability. Tissue physiology was assessed using a combination of established and modern approaches: measurement of hydrogen peroxide (H₂O₂), Seahorse analysis of cellular respiration (OCR and OCR/ECAR), and quantification of plant regulators using ultra-high pressure liquid chromatography with tandem mass spectrometry (UHPLC–MS/MS). These methods allow the investigation of oxidative status, metabolic activity, and regulatory signaling in embryogenic tissues, providing a multi-layered perspective on factors influencing survival.

H₂O₂ levels differed significantly among lines and sampling time points. In some lines, moderate H₂O₂ at early stages increased over time, whereas in others, high initial H₂O₂ may have contributed to oxidative stress and tissue death, suggesting that fine-tuned regulation of oxidative balance is important for tissue viability. Preliminary observations indicate that variability in plant regulator contents could influence cell division, tissue growth, and maintenance in general, while initial Seahorse analyses reveal metabolic differences among genotypes, highlighting the heterogeneity of energetic profiles even within surviving lines. Although no direct causal link between metabolic indicators and survival can yet be established, these preliminary data provide a valuable reference for understanding genotype-specific physiological traits in *P. abies* embryogenic cultures and may inform the optimization of *in vitro* culture protocols.

This study demonstrates how a combination of physiological and biochemical approaches can uncover subtle differences among *P. abies* lines, providing preliminary insights into mechanisms underlying tissue viability and offering a foundation for further studies aimed at identifying reliable markers of survival and developmental potential. The findings may also support the development of clonal propagation and breeding strategies in conifers.

We thank Prof. Jacek Kęsy (Nicolaus Copernicus University, Toruń, Poland) for his assistance with UHPLC–MS/MS analyses, and B.Sc. Karol Lesiakowski for technical support.