

Title Nitrate reductase-mediated nitric oxide generation is associated with chilling injury in maize (*Zea mays* L.) cultured cells

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Abstract

Physiological disorders including chilling injury are one of the major concerns dealing with the quality of postharvest commodities. Here, we reported that nitric oxide (NO) accumulation was involved in the cell death process induced by chilling in maize (*Zea mays* cv. Black Mexican Sweet) cultured cells. NO has been previously reported as a signalling molecule involved in plant growth, development, adaptation and defence to environmental stress. We found that chilling caused an increase in NO in maize cells. Treatment of cells with NO scavenger, 2-phenyl-4,4,5,5-tetramethylimidazole-1-oxyl-3-oxide (PTIO), significantly reduced chilling injury. N omega -nitro-L-arginine, an inhibitor of nitric-oxide synthase (NOS), did not reduce chilling injury and NOS activity determined by the conversion of L-arginine to L-citrulline, was undetectable in maize cells, suggesting that NO accumulation during chilling was unlikely mediated by NOS activity. Addition of 20 mM KNO₃ to a complete MS medium resulted in severe loss of cell viability after 5 days of chilling, which was in parallel with higher nitrate reductase activity and increased nitrite as well as NO concentrations. However, removal of any nitrogen sources from the medium led to an undetectable nitrite concentration during chilling and a significant increase in cell's chilling survival. Viability of cells cultured in the 20 mM KNO₃-added medium could be improved by adding PTIO. These findings suggest that the increased NO content in maize cells during chilling may be generated from nitrite by nitrate reductase activity and is likely involved in the chilling-induced cell death.