BMC Plant Biology

Meeting abstract

Production of nitric oxide and reactions with plant hemoglobins under hypoxic stress

Christos Dordas*[1], Brian B Hasinoff[3], Jean Rivoal[2] and Robert D Hill[2]

Address: [1]Laboratory of Agronomy, School of Agriculture, Aristotle University of Thessaloniki, 54124 Thessaliniki, Greece, [2]Department of Plant Science, University of Manitoba, Winnipeg, MB R3T 2N2, Canada and [3]Faculty of Pharmacy, University of Manitoba, Winnipeg, MB R3T 2N2, Canada

Email: Christos Dordas* - chdordas@agro.auth.gr

* Corresponding author

from Cell Biology of Nitric Oxide and Cell Death in Plants
Yalta, Ukraine, 8–11 September 2004

Published: 31 May 2005

BMC Plant Biology 2005, **5**(Suppl 1):S11 doi:10.1186/1471-2229-5-S1-S11

Nitric oxide (NO) is a reactive gas involved in many biological processes of animals, plants and microbes. The objectives of this study were: to examine whether NO is produced in transgenic maize suspension cell cultures and transgenic alfalfa roots exposed to hypoxic growth conditions, to determine whether there is a relationship between a class 1 hemoglobin and the amount of NO detected under hypoxic conditions, to estimate the route of formation and breakdown of NO in the tissue and to determine whether there is a relationship between NO production and aerenchyma formation. Maize suspension cell cultures and alfalfa roots, transformed to express the sense or antisense strands of barley hemoglobin were used to overexpress or underexpress class 1 hemoglobin. Up to 500 nmol NO/g fresh weight were detected in maize cells exposed to low oxygen tensions for 24 h. The steady state levels of NO in the different cell lines under hypoxic conditions had an inverse relationship to the level of hemoglobin in the cells. There was no detectable NO produced under aerobic growth conditions. Moreover EPR spectra showed evidence of nitrosylated heme complexes in denatured samples of hypoxically-exposed maize cells from both a wild type cell culture and a culture transformed to overexpress hemoglobin. No EPR signal characteristic of nitrosylated heme complexes was evident under aerobic conditions or in treated maize cells transformed to reduce hemoglobin expression. Spectroscopic data demonstrated that recombinant maize hemoglobin reacted with NO to form methemoglobin and NO_3^-. Nitrate was shown to be a precursor of NO in hypoxic maize suspension cell cultures by using $^{15}NO_3^-$ and EPR spectroscopy, suggesting that NO is formed via nitrate reductase during hypoxia. There was an inverse relationship between the expression of hemoglobin and the formation of aerenchyma under hypoxic conditions. The Hb^- line displayed strong evidence of aerenchyma formation under hypoxia, whereas an Hb^+ line showed only slight evidence of cell breakdown characteristic of aerenchyma formation. The levels of NO expressed distal to the root tip were, at least, five fold greater than those found in the tip. The results demonstrate that NO is produced in plant tissues grown under low oxygen tensions and suggest that class 1 hemoglobins have a significant function in regulating NO levels.