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Biological Nitrogen Fixation for the 21st Century

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NATURAL AND DARK-INDUCED SENESCENCE IN LUPIN ROOT NODULES

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Physiological, biochemical and ultrastructural changes in naturally and dark-stressed lupin nodules were examined to stablish if the mechanisms underlying both processes are related.
Exposure of lupins to darkness (4 to 7 days) induced decreases in N_2 fixation, leghemoglobine and total cytosolic proteins that mimic natural senescence. Under dark-stress and natural senescence several changes were observed in the electrophoretic protein pattern: a decrease of a polypeptide of molecular weight higher than 92 kDa and an increase of a polypeptide of 24 kDa and 16 kDa. The component of 16 kDa cross-reacted with an antibody against a polypeptide of the same molecular weight, found in yelow lupin roots exposed to various stressing factors, that was related with a Cu-Zn/SOD. However, in nodules the polypeptide of 16 kDa did not correspond to any SOD isoenzymes.
Darkness increased levels of a Mn-SOD isoenzyme and total SOD activity by 22% after 4 d, 40% after 5 d and 70% after 7 d similar to the increase detected in nodules of 9 weeks-old plants. Also there was a significant decline of catalase activity and increased level of total peroxidases, although ascorbate-peroxidase remained constant.
The increases in the antioxidant enzymes SOD and peroxidases under dark-stress and natural senescence did not keep active oxygen species (AOS) bellow damaging levels, resulting in oxidative damage to proteins. These results suggest that natural and dark-induced senescence in nodules might be the result of the imbalance between the increased production of AOS and the capacity of the scavenger-enzymes.
Darkness also caused severe ultrastructural alterations: bacteroid cytosol almost disappeared and immunocytochemical changes were detected on the bacteroid cell wall protein; however, the peribacteroid membrane stayed unruptured until a very advanced senescence stage. Same alterations were observed in naturally senescent nodules.

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