The rate of nitrite reduction in leaves as indicated by O₂ and CO₂ exchange during photosynthesis

Hillar Eichelmann, Vello Oja and Agu Laisk

Institute of Molecular and Cell Biology, Tartu University, Estonia, yeti@kodu.ee

Photosynthetically reduced ferredoxin (Fd⁻) in leaf chloroplasts is used for the reduction of NO_2^- to NH_4^+ and 1.5 O_2 are evolved. This offers a possibility for the *in vivo* measurement of NO₂⁻ reduction in intact leaves as the surplus of O₂ evolution over CO₂ fixation. Light response (at 300 ppm CO₂ and 10 - 50 ppm O₂ in N₂) and CO₂ response curves (at absorbed photon fluence rate, PAD of 550 μ mol m⁻² s⁻¹) of O₂ evolution and CO₂ uptake were measured in leaves of different species. Tobacco (Nicotiana tabacum L.) leaves were grown on NO₃⁻ and NH₄⁺ as N source. Potato (Solanum tuberosum L.), sorghum (Sorghum bicolor L. Moench) and amaranth (Amaranthus cruentus L.) leaves were grown on NH4NO3 nutrient. The surplus of photosynthetic O_2 evolution in excess of the photosynthetic CO_2 uptake was measured with the help of a zirconium cell O₂ and an infrared absorption CO₂ analyzers and interpreted to be the rate of electron flow to acceptors alternative to CO₂, mainly to NO_2^- , SO_4^{-2-} and oxaloacetate. In the NO_3^- -nutritioned tobacco, as well as in sorghum and amaranth and young potato, the photosynthetic O₂ - CO₂ flux difference increased at very low light intensities (PAD) to the value of about 0.5 - 1 μ mol m⁻² s⁻¹ and the rate saturated already at PAD of 50 μ mol quanta m⁻² s⁻¹. At higher PADs another component of the O₂ - CO₂ excess was observed, which increased about proportionally with the photosynthetic rate to the maximum of about 1 μ mol m⁻² s⁻¹. In the NH₄⁺ -fed tobacco, as well as in potato during tuber filling, the low-PAD component of the surplus O₂ evolution was virtually missing. The results show that the photoreduction of NO_2^- and CO_2 reduction compete for one and the same pool of Fd⁻ with rather similar affinities. NO₂⁻ reduction saturates at a low light intensity at rate of about 9% of the maximum O₂ evolution rate. The measured nitrite reduction rate is in agreement with leaf N/C molar ratio. At higher PADs oxaloacetate reduction is superimposed on the NO₂⁻ reduction. The NH₄⁺ "toxicity" and role of NO₂⁻ and oxaloacetate reduction in regulation of ATP/NADPH balance are discussed.