# Redox homeostasis by alternative V-nitrogenase improves growth

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Biological nitrogen fixation, the microbial conversion of $N_2$ gas into ammonia, is an essential nitrogen input to the biosphere. This process is catalyzed by three isoforms of the metalloenzyme nitrogenase ("nase"). The canonical Mo-nase, which produces the least $H_2$ by-product during $N_2$ reduction, is considered the most efficient isoform and the key enzyme responsible for nitrogen fixation in the environment. The 'alternative' V- and Fe-only nitrogenases are thought to be backup enzymes, useful when Mo is limiting or possibly at cold temperatures. However, genes for the alternative nitrogenases are present in diverse taxa and environments, including Mo-replete, temperate ecosystems, suggesting that these isoforms have additional physiological roles. Uncovering these roles, and thereby elucidating the factors that control the presence and activity of the different nitrogenases, is necessary to understand nitrogen fixation fluxes in the environment. Here we explore whether the higher $H_2$ production by the V-nase may be beneficial to growth under reducing conditions compared to the canonical Mo-nase. To test the effect of reductant availability on nitrogen fixation, we grew nitrogenase mutants of the metabolically versatile *Rhodopseudomonas palustris* photoheterotrophically on carbon substrates with different oxidation states. On a relatively oxidized carbon substrate, succinate, *R. palustris* consistently grows slower with the alternative nitrogenases than the Mo-nase. In stark contrast, on the more reduced carbon substrates acetate and butyrate, *R. palustris* grows up to ~30% faster when using the alternative V-nase instead of the Mo-nase. $H_2$ production and $N_2$ reduction rates demonstrate that this growth rate advantage is due to the co-benefits of simultaneous nitrogen acquisition and electron disposal by the V-nase. Our results uncover a redox balancing function for alternative nitrogenases that can lead to superior growth, implying that cellular redox homeostasis is an important constraint on the function and enzymology of nitrogen fixation. We suggest that, in addition to cold or Mo-limited environments, alternative isoforms like the V-nase may be active and beneficial for growth in reducing environments as well.