## Abstract

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## Does the vacuolar H<sup>+</sup>-pyrophosphatase contribute to vacuolar acidification?

In plant cells two proton pumps are assumed to be responsible for acidification of the lytic vacuole. Both pumps are thought to act synergistically to establish a transmembrane proton gradient ( $\Delta pHVac$ ) which drives solute transport across the tonoplast (Maeshima, 2001; Gaxiola et al., 2007). The presence of both proton pumps has been implicated in all types of vacuoles (Martinoia et al., 2007). Activity of the vacuolar H<sup>+</sup>-pyrophosphatase (V-PPase) is supposed to dominate in growing tissues, whereas the vacuolar H<sup>+</sup>-ATPase (V-ATPase) might represent the major tonoplast proton pump in mature tissues (Maeshima, 2000). Not much is known about the individual contribution of the pumps to the  $\Delta pHVac$ . We found that in mutants lacking the tonoplast-specific V-ATPase (vha-a2 vha-a3), vegetative growth is decreased and vacuolar pH is elevated, leading us to the question whether the V-PPase is able to compensate at least to some extend the loss of V-ATPase activity (Krebs et al., 2010). Interestingly, plants overexpressing the V-PPase in the tonoplast V-ATPase mutant background were not able to restore the reduced growth phenotype but indeed displayed an increased protein abundance and H<sup>+</sup>-PPase activity. Additionally, knock-out mutant plants of the V-PPase are only slightly reduced in their growth and the vacuolar pH of those plants does not differ from the wild type. This indicates that the V-PPase is not important for vacuolar acidification. Furthermore, we have hints that the remaining pH difference between the cytosol and the vacuolar lumen in vha-a2 vha-a3 is achieved due to TGN/EE-derived vesicles acidified by VHA-a1 containing V-ATPase complexes. In agreement with our results, it was recently shown that the main function of AVP1 is in the hydrolysis of cytosolic PPi, rather than vacuolar acidification (Ferjani et al., 2011).