VPH1 is one of two yeast genes encoding isoforms of the a subunit of the yeast V-ATPase V0 domain. Vacuolar-ATPasesare ATP-dependent proton pumps that acidify intracellular vacuolar compartments. Vacuolar acidification is important for many cellular processes, including endocytosis, targeting of newly synthesized lysosomal enzymes, and other molecular targeting processes. The V-ATPase consists of two separable domains. The V1 domain has eight known subunits, is peripherally associated with the vacuolar membrane, and catalyzes ATP hydrolysis. The V0 domain is an integral membrane structure of five subunits, and transports protons across the membrane. The structure, function, and assembly of V-ATPases are reviewed in references 9, 6, 10 and 11. The vph1 null mutant is viable but lacks V-ATPase activity and ATP-dependent proton pumping, and is defective in vacuolar acidification. The nucleotide-binding subunits of the V1 domain are present but not associated with the vacuolar membrane in vph1-1 mutant cells. The vph1-1 mutation also alters cellular phosphate trafficking. Point mutations have identified amino acid residues in Vph1p that are likely to be involved in proton transport, ATPase activity, and V-ATPase holoenzyme assembly. The vph1 null mutant shows a modest growth defect at neutral pH or in the presence of excess calcium; deletion of both VPH1 and STV1, which encodes the second a subunit isoform, causes a more severe growth defect, similar to the phenotypes of other V-ATPases subunit nulls. Overproduction of Stv1p partially restores vacuolar acidification in the vph1 null mutant. Vph1p and Stv1p show different localization patterns in indirect immunofluorescence assays, suggesting that they may be equivalent subunits for V-ATPases located on different organelles. Vph1p interacts directly with two V-ATPase assembly factors, Vma12p and Vma22p. In the absence of Vma22p, Vph1p is degraded in the ER.Stv1p and Vph1p are 55% identical and proteins similar to Vph1p have also been identified in rat, mouse, C. elegans and humans. Mutations in the isoforms of human V-ATPase most similar to Vph1p, a3 and a4, result in osteopetrosis and distal renal tubular acidosis, respectively.