VMA4 encodes the E subunit of the yeast V-ATPase V1 domain. Vacuolar-ATPasesare ATP-dependent proton pumps that have been identified in many eukaryotes, where they 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 4, 5, 6 and 7. The vma4 null mutant is viable but lacks vacuolar-ATPase activity and cannot grow at neutral pH or on nonfermentable carbon sources. A temperature sensitive vma4 mutant, which loses V-ATPase activity and vacuolar acidification upon incubation at high temperatures, shows abnormal bud morphology and delocalization of actin and chitin at the restrictive temperature. Vma4p is required for the V1 domain to assemble onto the vacuolar membrane.CC,PH, PI,