

**FIGURE LEGENDS****Figure 1.** Structure of yeast Vps4.

(a) Domain organization of the human and yeast Vps4 proteins. Numbering scheme corresponds to the yeast Vps4 protein.

(b) Stereoview of the Vps4<sub>ΔMIT</sub> structure. This structure corresponds to molecule A in the ATP<sub>γ</sub>S bound state in crystal form 2. Domains are color coded as in part (a). Note that the small helix spanning residues 199-206 is not numbered in order to maintain consistency with previous conventions.

(c) Superposition of yeast Vps4<sub>ΔMIT</sub> (in crystal form 1) and human VPS4B<sub>ΔMIT</sub> (lighter shades) in their sulfate-bound states. The two proteins overlay with an RMSD of 1.2 Å over 225 C<sub>α</sub> positions within the AAA ATPase cassette.

**Figure 2.** Vps4 nucleotide binding pockets.

(a) Stereoview of the Vps4-ATP<sub>γ</sub>S nucleotide binding site of molecule A in crystal form 2. Active site Vps4 residues are color coded according to their functional roles in Mg<sup>2+</sup> coordination/ATP hydrolysis (cyan, S180, D232 and E/Q233), adenine ring stacking (magenta, Y181 and M307) and phosphate sensing (yellow, K179 and N277) with the “arginine finger” residue R288 from an adjacent molecule in the modeled hexamer shown in green. Note that the E233Q mutant was used here and throughout to allow ATP/ATP<sub>γ</sub>S binding while inhibiting hydrolysis.

(b) Electron density for the ATP<sub>γ</sub>S nucleotides in molecule A of Vps4<sub>ΔMIT</sub> crystal form 2. The densities show (F<sub>0</sub> – F<sub>C</sub>) omit maps contoured at 2.5 σ.