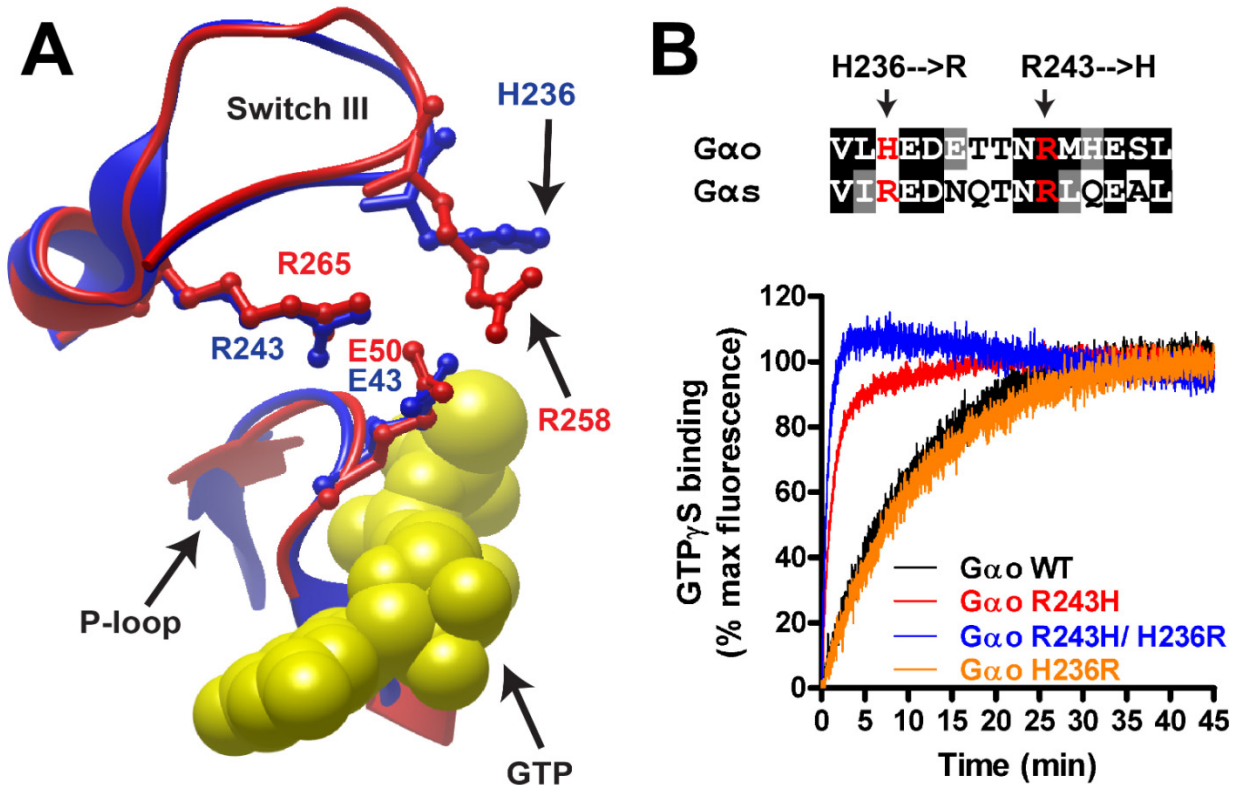


FIGURE S1

**Figure S1. Mutation of H236 to R in *Gao* WT or *Gao* R243H does not affect GTP γ S binding.**

(A) *Gas* and *Gao* differ in a critical residue of the SwIII. Overlay of the P-loop and SwIII/ α 3 of active *Gao* (blue, PBD: 3C7K) and active *Gas* (red, PBD: 1A2T) with a ball and stick representation of H236/R265, R243/R265 E43/E50. Nucleotide is shown in yellow. *Gas* R258 is close to the P-loop residue E50 (suggesting an electrostatic interaction) but the corresponding position in *Gao* is not conserved (H236).

(B) Top, alignment of *Gao*/*Gas* sequences and mutant design. *Gao* H236 was mutated to R alone or in combination with the R243H mutation to mimic the sequence of *Gas* WT or *Gas* R265H, respectively. **Bottom**, *Gao* H236R (orange) and *Gao* H236R/ R243H (blue) bind GTP γ S at the same rate as *Gao* WT (black) and *Gao* R243H (red), respectively. GTP γ S binding was determined by intrinsic fluorescence measurements as described in *Experimental procedures*. One experiment representative of 3 is shown.