

## SUPPLEMENTAL DATA

Fig. S1. Nucleotide-loaded state analysis of recombinant Arf4 and Arf6. 33  $\mu\text{g}$  of Arf4 and 43  $\mu\text{g}$  of Arf6 have been denatured at 95°C and centrifuged. Supernatant was subjected to reversed-phase HPLC analysis. Pure solution of GDP and GTP were used as standard.

Fig. S2. No vesicle formation in absence of Arf. COPI vesicles were generated *in vitro* from purified rabbit liver coatamer with or without recombinant myristoylated Arf1 and rat liver Golgi-enriched membranes in the presence of GTP $\gamma$ S, and purified by centrifugation through two sucrose cushions. 1% of the initial reaction mix (I for Input) and 50% of the vesicle fraction (V) were subjected to SDS-page and western-blot analysis.