

## MOLECULE PAGE

# Rab6

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Gene symbols: [Rab6a](#), [Rab6b](#)

## 1. General Function

Rab6 proteins are the mammalian homolog of yeast Ypt6. There are three forms: 6a, 6a' (also known as 6c) and 6b (6). Rab6a and 6a' are splice variants differing in only 3 amino acids and are expressed in most tissues. Rab6b is expressed mainly in brain (14). Rab6 localizes primarily to the Golgi apparatus with increasing amounts in the trans Golgi and trans Golgi network (TPN). As with other Rab proteins, Rab6 is geranylgeranylated on two cysteine residues at its carboxy terminal and this prenylation is essential for its localization to cellular membranes. Little is known of GEFs that activate Rab6 although two GAPs with activity towards Rab6 are known (6).

Functionally Rab6 is believed to be involved in a type of retrograde intra-Golgi transport that is independent of the primary form which utilizes COP I (6,17). Evidence suggests that Rab6a regulates the link between membranes and microtubule-based motor proteins. A second potential function for Rab6 is the budding off from the TGN of constitutive exocytotic vesicles which associate with kinesin 1 and move to the plasma

membrane (9). Recently Rab 6 and 6a were shown to recruit myosin II to induce fission of vesicles from tubules that bud off the TGN (13). There is also evidence that Rab6a' is involved in transport from early/recycling endosomes to the TGN (11). A plethora of potential Rab6 effector proteins have been identified with many being either Golgins or kinesins (1,3,16) most of which bind to Rab6 in the GTP-liganded form. Both Rab6a and 6a' have two PKC phosphorylation sites and phosphorylation may affect their membrane association (8). Rab 6 has been crystallized and its structure solved (2).

## 2. Rab6 in Pancreas

Rab6 has been reported on pancreatic zymogen granules by Western blotting (10) and mass spectrometry (5,15). In one of these studies it was reported as Rab6a (14). When immunohistochemistry was used, however, only a small fraction of isolated ZG were stained and the primary cellular localization was on the Golgi and TGN (5). As shown in the figure below Rab6

staining is largely overlapping with TGN38 staining, a marker for the TGN.

**c. Mouse lines**

None.

Rab6 has also been studied in rat AR42J and human SOJ-6 pancreatic cell lines (4). Rab6 mRNA and protein were identified in both cells and the localization of Rab6 protein was primarily over the Golgi by immunohistochemistry. In AR42J cells Rab6 partitioned between cytosol and microsomal membrane fractions. However, in SOJ-6 cells it was only present associated with membranes. The authors showed that different forms of Rab GDI were absent or altered in SOJ-6 cells and suggested that these changes could underly the failure of SOJ-6 cells to secrete Bile Salt Dependent Lipase (BSDL). In AR42J cells treatment with antisense oligonucleotide specific for Rab6 decreased BSDL secretion after 70 hours indicating that Rab6 is involved in the secretory process in AR42J cells (4).

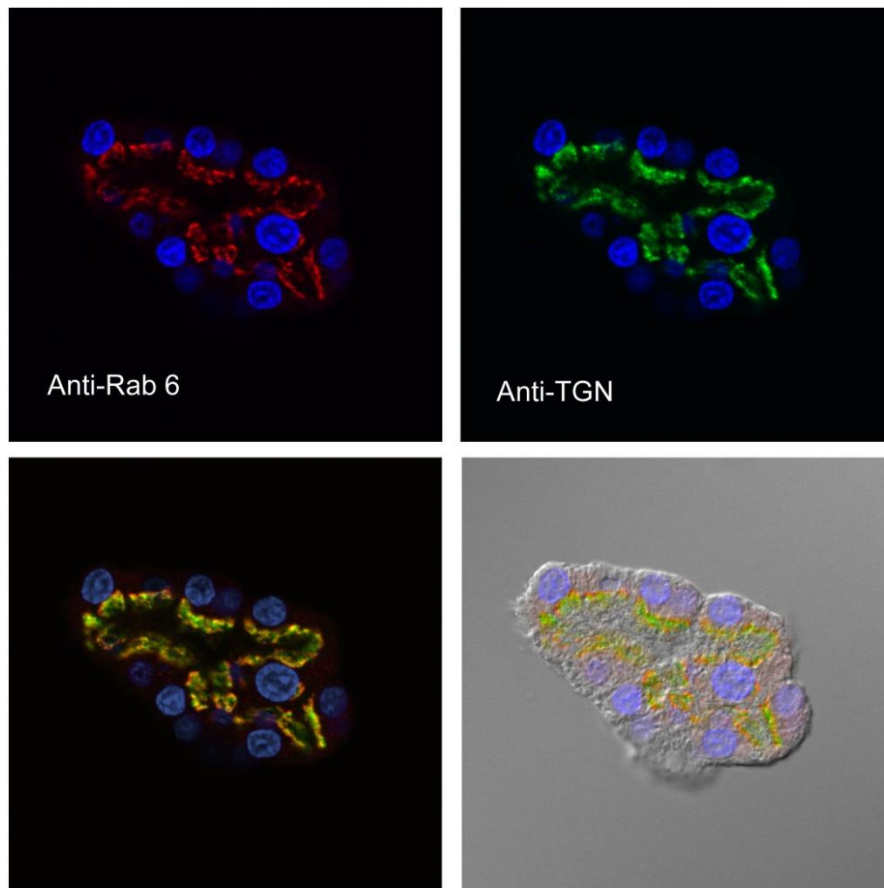
### **3. Tools for study of Rab6**

**a. cDNA**

cDNA clones for human Rab6a, 6b, and 6a' (6c) in pcDNA 3.1 are available from the Missouri S & T cDNA Resource ([www.cdna.org/](http://www.cdna.org/)). Several studies have been published using constitutively active (Rab6 Q72L) or dominant negative (Rab6 T27N) mutant plasmids based on mutating residues known to be important in Ras (7,12).

**b. Antibodies**

Rabbit antibodies raised against the C-terminal of human Rab6 (Ser 184 to Glu203) have been used to identify Rab6 by western blotting and immunohistochemistry in AR42J cells (4) and Rat pancreas (5). A Ab against this region is sold by Santa Cruz Biotechnology as a IgG (sc-310) and stated to detect all isoforms of Rab6 of mouse, rat, and human origin. We used this Ab for immunohistochemistry (Fig 1) at a dilution of 1:50 -1:100 (5). A mouse monoclonal against Rab6a is available from Abcam (ab55660) but has not been checked by us on pancreas.



**Figure 1. Confocal immunohistochemistry** supplied by S.A. Ernst (University of Michigan) and was carried out on frozen sections of isolated rat acini with rabbit Anti-Rab6 (red) from Santa Cruz (sc-310) and mouse monoclonal Ab against TGN38 (green) from Abcam (ab2809). DAPI (blue) was used to stain nuclei. In the bottom right panel the fluorescence is superimposed on a Nomarski Image.

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