Comparative Analysis of Rab GTPases and SNAREs in Eukaryotic Genomes

Akiyasu C. Yoshizawa acyshzw@kuicr.kyoto-u.ac.jp
Shuichi Kawashima shuichi@kuicr.kyoto-u.ac.jp
Minoru Kanehisa kanehisa@kuicr.kyoto-u.ac.jp

Bioinformatics Center, Institute for Chemical Research, Kyoto University, Gokasho, Uji, Kyoto 611-0011, Japan

Keywords: intracellular vesicular transport, Rab, small GTPase, SNARE, cluster analysis

1 Introduction

The intracellular vesicular transport is indispensable function for eukaryotic cells to maintain the intracellular membrane compartments. In order that the transport vesicles fuse only to the precise target membranes and that the macromolecules are selectively delivered, unique mutual recognitions are required between the vesicles and the membranes. These unique recognitions and membrane fusions are mediated by the SNARE (Soluble N-ethyl maleimide sensitive factor Attachment protein REceptor) superfamily proteins, which are well conserved throughout the eukaryotic evolution.

Recently ongoing eukaryotic genome projects have revealed a number of SNARE molecules in the whole genomes, but the number was fewer than expected suggesting that the same SNARE molecules mediate different transport pathways. For example, sec22p of *S.cerevisiae* carries both directions of transport between the endoplasmic reticulum (ER) and *cis*-Golgi apparatus. Therefore, additional proteins should be involved in the roles of mediating unique recognitions.

Rab proteins belong to the Ras small GTPase superfamily, and have been considered to be involved in the vesicular transport since the identification of the first Rab molecule in 1983. In 1999, mammalian Rab5, whose localization is early endosomes, was revealed to bind to the tethering factor EEA1 (Early Endosome Antigen 1), which is required before the SNARE mutual recognition, and also binds to Syntaxin6 and 13, t-SNAREs on early endosome. Therefore, SNARE systems and Rab GTPases are considered to be functionally related. For example, the localizations of both of mammalian Rab1 and Rab2 are between ER and *cis*-Golgi apparatus, while Rab1 mediates the transport from ER to Golgi and Rab2 mediates from Golgi to ER. Thus, it is probable that Rab molecules mediate the vesicular transport processes cooperating with the SNARE system.

We formerly performed a hierarchical cluster analysis of SNARE molecules, and showed that SNAREs form clusters according to their intracellular localizations [4]. Here, we performed the hierarchical cluster analysis of eukaryotic Rab GTPases stored in the KEGG/GENES database by considering their intracellular localizations and also the clusters of SNAREs in order to examine a possible increase of the numbers of Rab paralogs through the evolution.

2 Method

(1) The collection of known SNAREs and Rabs was created with the methods as described before [4] including PSI-BLAST search (E-value < 0.01), sequence similarity examination with SSEARCH 3.4t05 program and the complete-linkage hierarchical cluster analysis. The KEGG/GENES database was used for all operations. The clustering results were examined manually with published articles [1, 2, 3] and annotations in the database entries.

(2) The "best-hit" homolog of each obtained sequence against all the eukaryotic proteins contained in KEGG/GENES were searched using KEGG/SSDB database.

3 Results and Discussion

Generally, there are small numbers of SNAREs on the pathways near ER, and large numbers on the pathways near plasma membrane and endosomal membranes, and similar tendency was observed in case of Rabs. There are small numbers of t-SNAREs of ER and v-SNAREs on the pathway between ER and Golgi, and the number of Rab paralogs is also small. SNAREs of animal plasma membrane and of plant endosomal membranes have many paralogs, and so was Rabs (examples shown in Table 1).

Table 1: The numbers of SNAREs and Rabs in KEGG/GENES database. The entries of *S.pombe*, *S.cerevisiae*, and *D.melanogaster* are of the complete genomes, and others are of the partial genomes.

	S.pombe	*S.cerevisiae*	*C.elegans*	*D.melanogaster*	*R.norvegicus*	*M.musculus*	*H.sapiens*	*A.thaliana*
	fission yeast	*baker's yeast*	*worm*	*fly*	*rat*	*mouse*	*human*	*weed*
Endosomal								
Syntaxins (t-SNARE)	1	2	1	2	2	1	1	3
VAMP7 (v-SNARE)	0	0	0	1	1	0	1	4+7?
ykt6 (v-SNARE)	1	1	1	1	1	1	1	2
Rab11	1	2	1	1	2	2	2	13
from ER to Golgi								
Syntaxins (t-SNARE)	1	1	1	1	1	1	1	2
sec22 (v-SNARE)	1	1	1	1	1	1	3	2
Rab1	1	1	1	1	1	1	2	3

In most cases where multiple Rab molecules exist on a pathway, one of them has strong orthologous relations within animals and another one of the others has relations within plants and fungi (Fig.1). These observations may represent the evolutional process of Rab GTPases.

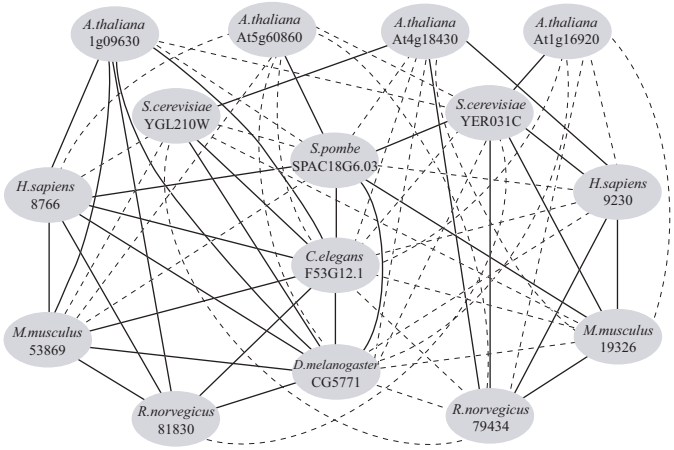

Figure 1: Orthologous relations of Rab11 family(endosomal membranes). Solid lines mean bi-directional best hits between the molecules.Dotted lines mean one-directional best hits(the directions are not shown).

Acknowledgments

This work was supported by grants from the Ministry of Education, Science, Sports and Culture, the Science and Technology Agency, and the Japan Society for the Promotion of Science.

The computational resource was provided by the Bioinformatics Center, Institute for Chemical Research, Kyoto University.

References

[1] Martinez, O. and Goud, B., Rab proteins, *Biochimica et Biophysica Acta*, 1404:101–112, 1998.

[2] Pereira-Leal, J. and Seabra, M.C., Evolution of the Rab family of small gtp-binding proteins, *J. Mol. Biol.*, 313:889–901, 2001.

[3] Segev, N., Ypt and Rab GTPases: Insight into functions through novel interactions, *Curr. Opin. Cell Biol.*, 13:500–511, 2001.

[4] Yoshizawa, A.C., Kawashima, S., and Kanehisa, M., The construction of a database on the intracellular vesicular transport, *Genome informatics*, 11:276–277, 2000.