

3.2 ROP

Most ROP-GAPs and Cdc42/ Rac effectors contain a distinctive Cdc42/ Rac-interactive binding (CRIB) domain, where they mediate binding of active GTPase. The regulators of ROPs in plants are with the exception of three GDI homologs. Rho GTPases associated effector proteins include RhoGAPs, RhoGEFs, and RhoGDIs (Manneville and Hall, 2002; Kost, 2008). ROPs regulate various plant cellular responses including defense against pathogens linked with disease resistance. Similar to mammalian Rac, ROPs trigger hydrogen peroxide production, a crucial component associated with the cell death via activation of NADP oxidase in plants (Agrawal, 2003).

3.3 RAN

Targets like importin α , importin β , CAS and TPX2 are required for Ran GTP and chromatin induced spindle assemble. Interaction of γ -tubulin, microtubule associated proteins and dynein were found to be required for RAN GTP induced spindle formation. Divergent forms of TPX2 were found in plants like Arabidopsis and indicates that RAN GTP may have more targets in M phase (Oliver et al., 2001)

3.4 ARF

Vesicle transport is mediated by ARF GTPases as key regulators. They recruit vesicular coat complexes and transport vesicles forming effector proteins to vesicle budding membrane sites (Bonifacino and Lippincott Schwartz, 2003; Serafini et al., 1991). Activation of ARF by GEF is the limiting factor regulating the timing and location of vesicle budding (Mosessoova et al., 2003). ARF-GDP-GTP switching controls myristoylated N-terminal sequence of 12 residues that grabbed into a surface of the groove on ARF-GDP (Amor et al., 1994) and exposed to membrane binding on ARF-GTP (Pasqualato et al., 2001).

4. Key roles of small GTPases

Stress signals are sensed by plants and transduced to activate downstream targets through intricate signaling network and crosstalk, in which transcription factors and signal-transducing GTPases play key roles enabling them to overcome the stress. The earlier reports on legume plants (table 1) indicate that there is an explorable diversity among GTPases. Small GTPases feature play central roles in endomembrane trafficking. ROP (Rho of Plants) has emerged as plant-specific GTPases playing critical roles to orchestrate unique cellular function such as regulating cell motility, cell division, directional growth, polarity establishment, stress and plant hormonal signaling (Manneville and Hall, 2002). The functional activity of small GTPases favors the ON and OFF state of small GTPases, respectively. The expression of regulators and their spatial and temporal distribution pattern determine the activity and hence turn-on the switch of small GTPases. There is increasing evidence demonstrating that small GTPase are involved in mediation of numerous physiological processes, ranging from pollen growth, root hair development (Li et al., 2001; Yang, 2002; Boureux et

al., 2007) and cytoplasmic streaming (Yasin, 2013 and unpublished) was regulated by myosins and other motor proteins (Ueda et al., 1996 ; Yasin and Rajkumar, 2014) in response to abiotic stress. They play roles in membrane trafficking (Stenmark et al., 2001), act as interactors with motor proteins in signaling (Yasin, 2013, Yasin and Rajkumar, 2014, Zhang et al., 2014), involved in mediating proteins and RNA transport across the nuclear envelope (Nie et al., 2003). SsRab 2 water stress tolerance (Mahony and Oliver, 1999), OsRab7 to cold stress (Nahm et al., 2003), and AtRabG3e to salt / osmotic stress (Mazel et al., 2004, Yasin and Rajkumar, 2014) explaining that these small GTPases can play a major role as molecular switches in plant stress signaling. OsRab7 was differentially regulated under cold, salt, dehydration and ABA stress conditions regulating vesicular transport to the vacuole (Nahm et al., 2003). Thus the direct relationship of small GTPases with motor proteins in carrying out these events is established. Though we got confirming results, the contradictory retraction by Holweg and Peter (2004 and 2008) after deep sequencing of the material they used shows that, it is not only myosin XI K but other motor proteins are also involved in carrying out these functions. Further this could be concluded like the specific role of myosin XI K can be substituted by the presence of other myosins or kinesins (Fig. 1). When we observed the displacement of an organelle/ vesicle or cytoplasmic bulb it is very clear that only myosin is not sufficient to make this action faster but it needs other motor proteins also. Apart from that Ca²⁺ levels and Mg²⁺ levels along with pH flux plays a major in stress signalling (Yasin, 2015).

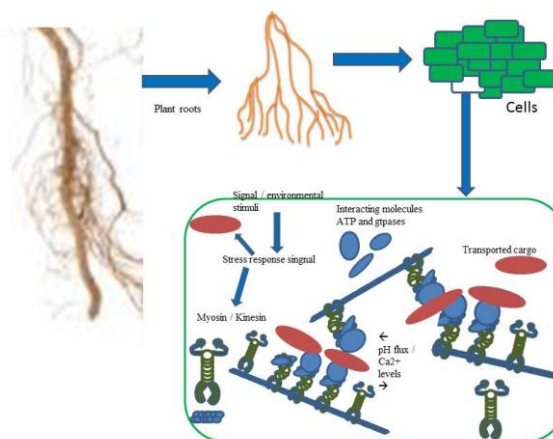


Figure 1: Perspective illustration of rab interaction in intracellular transport

5. Small GTPases in Stress Tolerance of Legumes

Some of the signal transduction pathways are mediated by GTP binding protein-coupled receptor. Sequence of Rho-like GTPase isolated from *Lotus japonicus* designated as LjROP6 is closest to *Arabidopsis thaliana* ROP6 and *Medicago truncatula* ROP6. Interaction of Rop6 with NFR5 was successfully established both in vitro and in planta but not with NFR1. Much of the information about the functions of plant Rab GTP-binding proteins has been derived from

complementation and expression studies in yeast (Ueda et al., 2004), subcellular localization data (Ueda et al., 1996), and transgenic approaches (Cheon et al., 1993).

A group of 10 small GTPases that are likely to be mainly expressed in nodules were identified. Detailed molecular genetic analyses through RNA interference silencing mutants confirmed the role of GTPases in nodulation. MfARL1, a small GTPase was isolated from *M. falcata*, through salt stress SSH cDNA library. The expression of MfARL1 in Arabidopsis seedlings conferred the transgenic plants more tolerant to salt stress. A Rho-like small GTPase from *Lotus japonicus* was identified as an NFR5-interacting protein. Detailed accounts on legume small GTPases were listed in table 1.

6. Conclusion

Exploitation of genetic variation and desirable traits among germplasm resources through molecular breeding and translational genomics, could facilitate in identifying tolerant genetic resources. The present manuscript is framed with an objective of reviewing with a view to exploit genomics tools in investigating the diversity and molecular responses of small GTPases and its interacting molecules to stress tolerance.

Table 1: List of small GTPase reported in legume plants

Gene / GTPase	Predicted role of GTPase	Crop	Reference
RAB 1 and RAB7	Rab 1 and Rab7p in the formation of the peribacteroid membrane in root nodules.	Soybean	Choong et al., 1993
G-Protein Genes	Dimers are active and trimeric proteins are inactive conformation.	Soybean	Choudhury and Pandey, 2013
RAB and ARF	Crucial role of small GTP-binding proteins in diverse cellular processes	<i>Medicago sativa</i> ,	Claudia et al., 1995
ROP GTPases / CAE1/MRH2	Activation of ROP2 reverses defective root hair tip growth caused by MRH2 Kinesin mutation	Arabidopsis	Guohua Yang et al., 2007
RAC proteins/RHO	ROS mediated defense mechanism	Soybean	Jumok Park et al., 2000
ROP-type GTPases	Silencing of MtROP9 reduced ROS production and suppressed induction of ROS-related enzymes in transgenic roots infected with pathogenic and symbiotic microorganisms facilitating alternative defense mechanism	<i>Medicago truncatula</i>	Kiirika et al., 2014
ROP GTPase	Rop proteins accumulate to high levels in rapidly growing tapetal cells of pea anthers.	Pea (<i>Pisum sativum</i>)	Lin et al., 2001
SAR1 and ARF 1	Small gtpases are involved in vacuolar sorting of proteins	Soybean	Maruyama, et al., 2006
RHO & ROP	Rheostat mechanism of GTPase signaling resulting in increased NADPH oxidase activity under hypoxia	<i>Arabidopsis thaliana</i>	Michael and Foyer, 2014
G-proteins	Monomeric G-proteins up-regulation by ethylene	Pea (<i>Pisum sativum</i>)	Moshkov et al., 2003
PSA2 gene/ YPT/rab	YPT/rab family is down regulated by light, mediated by phytochrome	Pea (<i>Pisum sativum</i>)	Takehito et al., 1999
ROP	Root-hair tip elongation requires NADPH-oxidase driven production of ROS, activated by small ROP-family GTPases which can bind to and activate NADPH oxidase	<i>Medicago truncatula</i>	Wong et al., 2007
NAP and PIR proteins	To investigate the role of the lotus NAP1 and PIR1 proteins during infection, we analyzed the actin structure in root hairs at three developmental stages.	<i>Lotus japonicus</i>	Yokota et al., 2009
ARF/SAR, RAB, ROP & RAN	Some homologues of these genes could have been subfunctionalized to perform lineage-specific regulatory roles such as nodulation in leguminous plants	<i>Medicago sativa</i> , <i>Pisum sativum</i> , <i>Glycine max</i> , <i>Vicia faba</i> , <i>Vigna radiata</i> , and <i>Cicer arietinum</i>	Yuksel and Memon, 2008
ARF/SAR, RAB, ROP RAN.	Nodule-specific functions including the regulation of cargo-trafficking of materials between symbionts , molecular switches coordinating intricate interplay of events, establishment and maintenance of symbiomes together with tens of already characterized nodular genes.	<i>Medicago truncatula</i>	Yuksel and Memon, 2009

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