Abstract

Polar cell growth is one of the most fundamental processes in plant development. Pollen tubes represent excellent experimental system to study basic rules of this phenomenon. Polar growth is governed by a precise spatiotemporal coordination of various molecules, such as small GTPases, actin cytoskeleton, protein kinases and reactive oxygen species, to achieve its establishment and maintenance. In this study, we utilized a combination of diverse experimental approaches together with advanced computational methods to investigate the role of two signaling phospholipids, phosphatidic acid (PA) and phosphatidylinositol 4,5-bisphophate (PIP_2) in the polar growth. We described the involvement of PA in the regulation of actin cytoskeleton, which dynamics is essential for the proper pollen tube growth. We found direct interaction of actin with the PA-producing enzyme, phospholipase D (PLDβ)1 and we showed that actin affects the activity of PLDβ. We further described structural details of the PA inhibition of actin-capping protein and we proposed the model of positive-feedback loop of the actin dynamics regulation by PA and PLDβ1. To get insight into the PA localization in pollen tubes, we prepared PA-binding domain of Spo20p as YFP-fusion protein and we used it as PA-biosensor. We found that PA is enriched in subapical region of growing pollen tubes and that it partially overlaps with PIP_2 near to the apex. During exocytosis, a fusion of the secretory vesicles carrying cell-wall building material with the target plasma membrane is mediated by evolutionary conserved tethering complex, the exocyst. We found that SEC3a subunits of the exocyst complex localizes in the subapical part of pollen tubes and we showed that this localization of SEC3a is mediated by its interaction with PIP_2. Altogether, our data strongly show that signaling phospholipids control several aspects crucial for the normal pollen tube growth.