



Article scientifique

Article

2013

Published version

Open Access

This is the published version of the publication, made available in accordance with the publisher's policy.

Growth control: function follows form

Loewith, Robbie Joséph

How to cite

LOEWITH, Robbie Joséph. Growth control: function follows form. In: Current Biology, 2013, vol. 23, n° 14, p. R607–609. doi: 10.1016/j.cub.2013.05.048

This publication URL: <https://archive-ouverte.unige.ch/unige:103233>

Publication DOI: [10.1016/j.cub.2013.05.048](https://doi.org/10.1016/j.cub.2013.05.048)

Dispatches

Growth Control: Function Follows Form

Cell division, intuitively, is often dependent upon increases in cellular mass and volume. Less obvious is the reciprocal regulation of growth by the cell division cycle. In budding yeast, this link is mediated by the cell-cycle-dependent polarization of actin.

Robbie Loewith

Organism size can vary by as much as 22 orders of magnitude ranging from ultra-small archaea found at the Richmond Mine at Iron Mountain near Redding in Northern California (mean volume of $\sim 0.03 \mu\text{m}^3$) [1] to a honey fungus, *Armillaria ostoyae*, in the Malheur National Forest in the Blue Mountains of eastern Oregon (covering an area of 8.9 km^2 and weighing an estimated mass of $5.5 \times 10^5 \text{ kg}$) [2,3]. Growth is thus a fundamental tenet of biology; but, how is it regulated? Organismal growth is dictated by both genetics and by environmental conditions (Chihuahua versus Saint Bernard dog breeds and Bonsai trees respectively provide illustrative examples). Studies, particularly those made with fission and budding yeasts, have additionally demonstrated that cellular growth is regulated by cell-intrinsic cues. In a recent issue of *Current Biology*, Goranov *et al.* [4] now demonstrate that, by antagonizing the activity of target of rapamycin complex 1 (TORC1), which is a widely conserved regulator of eukaryote growth, actin polarization — and by extension, cell morphology — is one such cell-intrinsic growth regulator.

Growth of budding yeast cells is usually highly polarized (Figure 1). Mass synthesized in the mother cell is targeted, via vesicular trafficking, to the bud for deposition (mother cells, by definition, have already attained the critical size permissive for division and thus their growth is normally minimal during the cell cycle). This targeted vesicular trafficking is supported by a polarization of actin cables towards the bud in early S phase. Concurrently, cortical actin patches, thought to be sites of endocytosis and cell-wall remodeling, concentrate within the bud (reviewed in [5]). Previously, by quantifying the growth rates of budding yeast cells arrested at various stages of the cell division cycle, the Hansen,

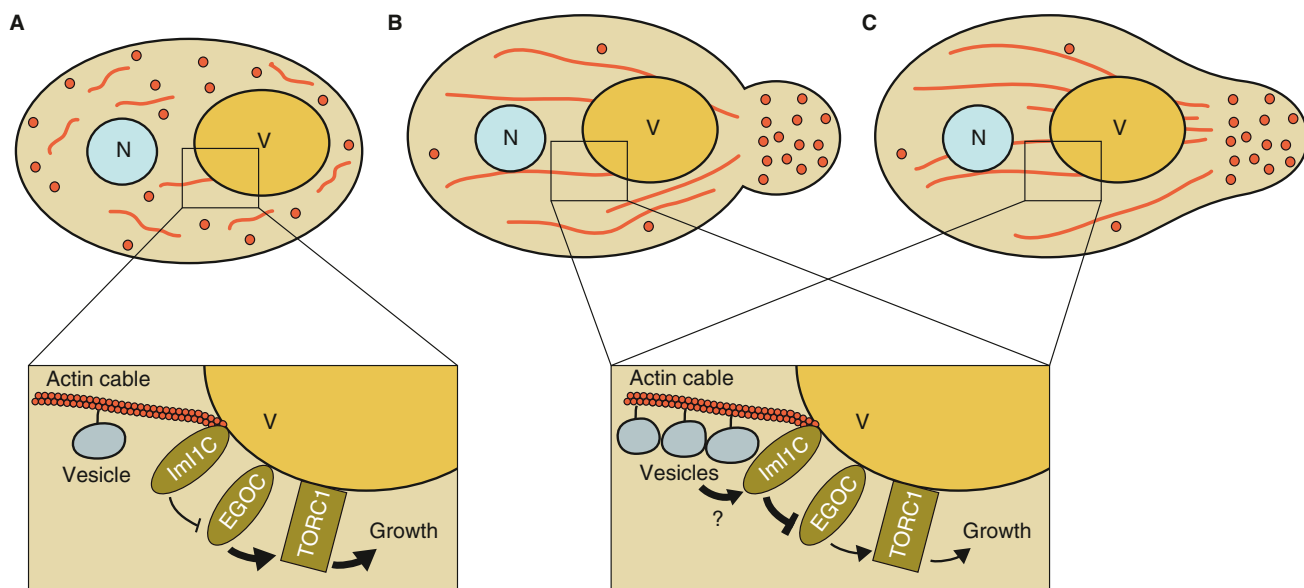
Tyers and Amon groups collaboratively found that growth is faster in anaphase- and G1-arrested cells than in cells arrested in S phase or early mitosis, and thus observed an intriguing anti-correlation between growth rate and extent of actin polarization [6]. Subsequent manipulations of actin dynamics, to either promote or prevent polarization, demonstrated conclusively that polarization of the actin cytoskeleton markedly attenuates growth. However, these studies did not illuminate the molecular mechanisms involved.

Now, having picked up where they left off, Goranov *et al.* [4] probed whether either RAS or TORC1 might be the molecular target through which actin polarization influences cellular growth. Both RAS and TORC1 are conserved regulators of eukaryote growth. RAS is a small GTPase, activated in yeast by poorly characterized signals emanating from carbon metabolism (reviewed in [7]). GTP-bound RAS binds adenyl cyclase and thereby stimulates production of cyclic AMP (cAMP) and subsequently activation of protein kinase A (PKA). PKA has many substrates that ultimately stimulate growth and simultaneously antagonize various stress responses. Hyperactivation of RAS signaling (via expression of constitutively active *RAS2^{V19}* or deletion of *BCY1*, encoding a PKA regulatory subunit) did not abrogate growth inhibition caused by actin polarization, suggesting that RAS/PKA is not the relevant target through which actin polarization regulates cell growth.

TORC1 is an assembly of several proteins, including the target of rapamycin (TOR) serine/threonine protein kinase (reviewed in [8]). TORC1 activity is stimulated by quality and quantity of nutrients (carbon, nitrogen, phosphate, amino acids, and likely others) and is antagonized by abiotic

stressors. With the exception of amino acids, how TORC1 activity is regulated by these various cues is also poorly understood. Amino-acid abundances are thought to be signaled to TORC1 via the small GTPases Gtr1 and Gtr2, components of the EGO complex [9] (known as the Ragulator complex in metazoans [10]). In the GTP-bound state, Gtr1 promotes TORC1 activation. GTP-loading of Gtr1 is mediated by the guanine-nucleotide exchange factor Vam6 [11] and the GTPase activity of Gtr1 is stimulated by the Iml1 complex, composed of Iml1, Npr2 and Npr3 [12]. TORC1 has three major effectors, a PKA-like kinase known as Sch9, a subset of type 2A protein phosphatases (regulated in part by the protein Tip41), and a putative transcription factor, Sfp1, which regulates ribosome biogenesis. Like RAS/PKA, TORC1 effectors also stimulate growth and simultaneously antagonize various stress responses. Importantly, Goranov *et al.* [4] found that, in cells in which these TORC1 effectors were engineered to be constitutively active, actin polarization no longer efficiently reduced cell growth. Furthermore, hyperactivation of Gtr1 signaling via expression of constitutively active *GTR1^{Q65L}*, or by deletion of *IML1*, *NPR2* or *NPR3*, similarly reduced growth inhibition caused by actin polarization. Lastly, it was found that actin polarization triggered phosphorylation changes in multiple TORC1 effectors in a manner consistent with TORC1 inhibition. Collectively, these observations strongly suggest that TORC1 is a relevant target through which actin polarization impinges upon cell growth. Although this signaling pathway was elucidated using pheromone treatment to induce strong actin polarization, it is likely active during normal cell cycles but with amplitudes that are too small to consistently measure.

Whether or not actin-polarization cues signal to TORC1 via the Iml1 and EGO complexes, as hinted at in Goranov *et al.* [4], is an interesting speculation that awaits biochemical experimentation for confirmation. In



Current Biology

Figure 1. Actin polarization in yeast impacts upon TORC1 and growth.

(A) Neither actin cables nor cortical actin patches (both in red) are polarized in freshly budded, G1 phase daughter cells. In contrast, actin structures are highly polarized in G2/M phase mother cells (B), or in G1 phase cells exposed to mating pheromone (C). In these polarized cells, actin cables orient towards and cortical actin patches accumulate in the bud (B) or shmoo tip (C). In non-polarized cells, growth, which is mediated largely by vesicular traffic, is thought to be isotropic, i.e. not limited to a particular place or volume. In polarized cells, growth is limited to a particular site, here the bud or shmoo tip. Goranov *et al.* [4] propose that polarized growth may trigger a backlog of vesicles along actin cables that — potentially through the Iml1 and EGO complexes (Iml1C and EGO) — antagonizes TORC1 signaling (zoomed images). Reduced TORC1 activity slows biomass production and this is necessary for cells to survive long periods of highly polarized growth. N, nucleus; V, vacuole.

yeast, polarization of growth is mediated by the transport of lipid vesicles on actin cables and the fusion of these vesicles with the plasma membrane to enlarge the cell. Goranov *et al.* [4] speculate that polarization of growth — which, by definition, confines the volume to which new mass can be deposited — slows the rate at which these vesicles can be ‘off-loaded’ and the subsequent vesicle ‘traffic jams’ could produce the growth-arrest cue. How vesicle traffic jams would be sensed by TORC1 remains a mystery. Curiously, the Iml1 complex is thought to share homology with the HOPS and CORVET complexes [13] involved in vesicle trafficking to and from the vacuole where, coincidentally, TORC1 resides. As the authors suggest, this raises the possibility that TORC1 is sensitive to vesicle traffic to or from the vacuole (Figure 1). Importantly, decreasing macromolecule synthesis in cells experiencing prolonged periods of highly polarized growth, for example, during protracted exposure to mating pheromone, appears to be physiologically relevant because loss of this feedback makes it much more difficult for cells to eventually re-enter

the cell cycle after pheromone withdrawal.

Growth of vertebrate cells has also been observed to be differentially regulated throughout the cell cycle (reviewed in [14]) and, as in yeast, appears to be fastest in G1 [15]. This suggests that yeast will once again punch above their weight class in the elucidation of the mechanisms by which eukaryote cells regulate their growth. Notably, cell-cycle-regulated growth control is often disrupted in transformed cells, and, in some cases, this contributes to aneuploidy and tumorigenesis [16]. Beyond its potential clinical relevance, the Goranov study also adds to a growing list of unexpected functions for polymerized actin, which, in vertebrates, now also includes nuclear roles, such as the regulation of transcription [17]. Lastly, this study provides another example of how TOR operates in homeostatic feedback loops to regulate growth. Said another way, there are now several reports demonstrating that TOR effectors are also upstream regulators. Goranov *et al.* [4], for instance, demonstrate that TORC1 is regulated downstream of actin polarization; but, reciprocally,

actin polarization has also been suggested to be regulated downstream of TORC1 [18]. Similarly, TORC1 and TORC2 function both upstream and downstream of ribosome biogenesis and sphingolipid production, respectively [8,19]. Although TOR signaling has historically been suggested to regulate growth in response to environmental cues, these studies suggest a second *leitmotiv* for TOR signaling, that is, cell-intrinsic or homeostatic control of cell growth. Understanding these feedback networks will be an important challenge, especially since TOR is a validated drug target in cancer amongst other diseases [20].

References

1. Comolli, L.R., Baker, B.J., Downing, K.H., Siegerist, C.E., and Banfield, J.F. (2009). Three-dimensional analysis of the structure and ecology of a novel, ultra-small archaeon. *ISME J.* 3, 159–167.
2. Ferguson, B.A., Dreisbach, T.A., Parks, C.G., Filip, G.M., and Schmitt, C.L. (2003). Coarse-scale population structure of pathogenic *Armillaria* species in a mixed-conifer forest in the Blue Mountains of northeast Oregon. *Canad. J. Forest Res.* 33, 612–623.
3. Smith, M.L., Bruhn, J.N., and Anderson, J.B. (1992). The fungus *Armillaria bulbosa* is among the largest and oldest living organisms. *Nature* 356, 428–431.

4. Goranov, A.I., Gulati, A., Dephoure, N., Takahara, T., Gygi, S.P., Manalis, S., and Amon, A. (2013). Changes in cell morphology are coordinated with cell growth through the TORC1 pathway. *Curr. Biol.* 23, 1269–1279.
5. Michelot, A., and Drubin, D.G. (2011). Building distinct actin filament networks in a common cytoplasm. *Curr. Biol.* 21, R560–R569.
6. Goranov, A.I., Cook, M., Ricicova, M., Ben-Ari, G., Gonzalez, C., Hansen, C., Tyers, M., and Amon, A. (2009). The rate of cell growth is governed by cell cycle stage. *Genes Dev.* 23, 1408–1422.
7. Broach, J.R. (2012). Nutritional control of growth and development in yeast. *Genetics* 192, 73–105.
8. Loewith, R., and Hall, M.N. (2011). Target of rapamycin (TOR) in nutrient signaling and growth control. *Genetics* 189, 1177–1201.
9. Dubouloz, F., Deloche, O., Wanke, V., Camerani, E., and De Virgilio, C. (2005). The TOR and EGO protein complexes orchestrate microautophagy in yeast. *Mol. Cell* 19, 15–26.
10. Gaubitz, C., and Loewith, R. (2012). Amino acid signaling in high definition. *Structure* 20, 1993–1994.
11. Binda, M., Péli-Gulli, M.P., Bonfils, G., Panchaud, N., Urban, J., Sturgill, T.W., Loewith, R., and De Virgilio, C. (2009). The Vam6 GEF controls TORC1 by activating the EGO complex. *Mol. Cell* 35, 563–573.
12. Panchaud, N., Péli-Gulli, M.P., and De Virgilio, C. (2013). Amino acid deprivation inhibits TORC1 through a GTPase-activating protein complex for the Rag family GTPase Gtr1. *Sci. Signal.* 6, ra42.
13. Dokudovskaya, S., Waharte, F., Schlessinger, A., Pieper, U., Devos, D.P., Cristea, I.M., Williams, R., Salamero, J., Chait, B.T., Sali, A., et al. (2011). A conserved coatomer-related complex containing Sec13 and Seh1 dynamically associates with the vacuole in *Saccharomyces cerevisiae*. *Mol. Cell. Proteomics* 10, M110.006478.
14. Goranov, A.I., and Amon, A. (2010). Growth and division—not a one-way road. *Curr. Opin. Cell Biol.* 22, 795–800.
15. Tzur, A., Kafri, R., LeBleu, V.S., Lahav, G., and Kirschner, M.W. (2009). Cell growth and size homeostasis in proliferating animal cells. *Science* 325, 167–171.
16. Wilker, E.W., van Vugt, M.A., Artim, S.A., Huang, P.H., Petersen, C.P., Reinhardt, H.C., Feng, Y., Sharp, P.A., Sonenberg, N., White, F.M., et al. (2007). 14-3-3sigma controls mitotic translation to facilitate cytokinesis. *Nature* 446, 329–332.
17. Baarlink, C., Wang, H., and Grosse, R. (2013). Nuclear actin network assembly by formins regulates the SRF coactivator MAL. *Science* 340, 864–867.
18. Aronova, S., Wedaman, K., Anderson, S., Yates, J., 3rd, and Powers, T. (2007). Probing the membrane environment of the TOR kinases reveals functional interactions between TORC1, actin, and membrane trafficking in *Saccharomyces cerevisiae*. *Mol. Biol. Cell* 18, 2779–2794.
19. Berchtold, D., Piccolis, M., Chiaruttini, N., Riezman, I., Riezman, H., Roux, A., Walther, T.C., and Loewith, R. (2012). Plasma membrane stress induces relocalization of Slim proteins and activation of TORC2 to promote sphingolipid synthesis. *Nat. Cell Biol.* 14, 542–547.
20. Benjamin, D., Colombi, M., Moroni, C., and Hall, M.N. (2011). Rapamycin passes the torch: a new generation of mTOR inhibitors. *Nat. Rev. Drug Discov.* 10, 868–880.

Department of Molecular Biology, University of Geneva, 1211 Geneva, Switzerland; and Swiss National Science Foundation National Centres for Competence in Research “Frontiers in Genetics” and “Chemical Biology”.
E-mail: robbie.loewith@unige.ch

<http://dx.doi.org/10.1016/j.cub.2013.05.048>

Marine Biology: New Light on Growth in the Cold

The recent collapse of the Antarctic Larson ice shelves revealed a slow growing benthic community on the seabed below. But a revisit just four years later revealed rapid growth of glass sponges. Antarctic continental shelves could become sites of significant carbon sequestration.

David K.A. Barnes

Antarctica's continental shelves are amongst the widest (and deepest) worldwide. What makes them even more different to elsewhere, though, is that about a third of their area is hidden from sight and light by huge sprawling ice shelves floating over them. Decades ago, it was with some surprise that macroscopic life was found surviving in the pitch black underneath the ice. This lightless life was sparse, depauperate, and it seemed that all activity was exemplary of 'life in the slow lane'. However, we know that during interglacial periods ice shelf margins retreat through break ups to expose the water above the seabed to light. The drastic increases in air and sea temperatures around the Antarctic Peninsula over the last half century have undoubtedly increased the rate and magnitude of ice shelf collapses. To see how life responded to such events was

always going to be a 'must see' for ecologists. In 2007, marine biologists aboard RV Polarstern were the lucky ones and reached the site of one of the most spectacular collapses in recent years around the Larson A and B ice shelves, off the East Antarctic Peninsula. In this issue of *Current Biology*, a new paper [1] by Laura Fillingner and colleagues describes the very surprising finds of a recent repeat trip to this same location.

The 2007 visit was a breakthrough [2], allowing scientists to take a first wide-scale look at a community which had emerged after developing under an ice shelf for tens or possibly even hundreds of thousands of years. Perhaps this was the closest we had come to envisaging life in a polar region during glacial periods — although there is evidence that, during glaciations, ice-shelves were actually touching the seabed in many areas despite the depth of the continental shelf around Antarctica. What the scientists found

on the Larson embayment seabed was a community characterised by slow growing sponges and some faster growing pioneers, such as stalked ascidians. The former were interpreted to represent what had been present underneath the ice shelf and the latter were assumed to be new arrivals [2,3] — it could be argued that neither was a huge surprise.

This exposure of vast areas of continental shelf provided more than just an opportunity to look at how life had coped under thick ice and how others had then recolonised. The finding had potential implications for the change in Earth's climate that brought about the collapse in the first place. What had been ice covered and dark was now coastal open water that could support phytoplankton blooms, which take up carbon dioxide, which if buried on the seabed reduces aerial carbon dioxide and ultimately temperature. Major new phytoplankton blooms were indeed observed in the regions of ice shelf collapse, leading to estimates that existing ice-shelf losses meant that per year roughly 3.5×10^6 tonnes of carbon were being added to animals as new growth [4]. This finding would generate only modest scientific interest unless the carbon was being genuinely sequestered rather than merely 'borrowed' and rapidly