Année de publication : 2020

François Dossin, Inês Pinheiro, Jan J Żylicz, Julia Roensch, Samuel Collombet, Agnès Le Saux, Tomasz Chelmicki, Mikaël Attia, Varun Kapoor, Ye Zhan, Florent Dingli, Damarys Loew, Thomas Mercher, Job Dekker, Edith Heard (2020 Feb 7)

SPEN integrates transcriptional and epigenetic control of X-inactivation.

Nature : 455-460 : DOI : 10.1038/s41586-020-1974-9

Résumé

Xist represents a paradigm for the function of long non-coding RNA in epigenetic regulation, although how it mediates X-chromosome inactivation (XCI) remains largely unexplained. Several proteins that bind to Xist RNA have recently been identified, including the transcriptional repressor SPEN, the loss of which has been associated with deficient XCI at multiple loci. Here we show in mice that SPEN is a key orchestrator of XCI in vivo and we elucidate its mechanism of action. We show that SPEN is essential for initiating gene silencing on the X chromosome in preimplantation mouse embryos and in embryonic stem cells. SPEN is dispensable for maintenance of XCI in neural progenitors, although it significantly decreases the expression of genes that escape XCI. We show that SPEN is immediately recruited to the X chromosome upon the upregulation of Xist, and is targeted to enhancers and promoters of active genes. SPEN rapidly disengages from chromatin upon gene silencing, suggesting that active transcription is required to tether SPEN to chromatin. We define the SPOC domain as a major effector of the gene-silencing function of SPEN, and show that tethering SPOC to Xist RNA is sufficient to mediate gene silencing. We identify the protein partners of SPOC, including NCoR/SMRT, the mA RNA methylation machinery, the NuRD complex, RNA polymerase II and factors involved in the regulation of transcription initiation and elongation. We propose that SPEN acts as a molecular integrator for the initiation of XCI, bridging Xist RNA with the transcription machinery-as well as with nucleosome remodellers and histone deacetylases-at active enhancers and promoters.

Année de publication : 2019

Cédric Delevoye, Michael S Marks, Graça Raposo (2019 Dec 14)

Corrigendum to « Lysosome related organelles as functional adaptations of the endolysosomal system », Current Opinion in Cell Biology, volume 59, 2019, 147-158.

Current opinion in cell biology : 141 : DOI : S0955-0674(19)30097-3

Résumé

Maddalena Nano, Simon Gemble, Anthony Simon, Carole Pennetier, Vincent Fraisier, Veronique Marthiens, Renata Basto (2019 Nov 12)

Cell-Cycle Asynchrony Generates DNA Damage at Mitotic Entry in Polyploid Cells.

Current biology : CB : 3937-3945.e7 : DOI : S0960-9822(19)31232-1

Résumé

Polyploidy arises from the gain of complete chromosome sets [1], and it is known to promote cancer genome evolution. Recent evidence suggests that a large proportion of human tumors experience whole-genome duplications (WGDs), which might favor the generation of highly abnormal karyotypes within a short time frame, rather than in a stepwise manner [2-6]. However, the molecular mechanisms linking whole-genome duplication to genetic instability remain poorly understood. Using repeated cytokinesis failure to induce polyploidization of Drosophila neural stem cells (NSCs) (also called neuroblasts [NBs]), we investigated the consequences of polyploidy in vivo. Surprisingly, we found that DNA damage is generated in a subset of nuclei of polyploid NBs during mitosis. Importantly, our observations in flies were confirmed in mouse NSCs (mNSCs) and human cancer cells after acute cytokinesis inhibition. Interestingly, DNA damage occurs in nuclei that were not ready to enter mitosis but were forced to do so when exposed to the mitotic environment of neighboring nuclei within the same cell. Additionally, we found that polyploid cells are cell-cycle asynchronous and forcing cell-cycle synchronization was sufficient to lower the levels of DNA damage generated during mitosis. Overall, this work supports a model in which DNA damage at mitotic entry can generate DNA structural abnormalities that might contribute to the onset of genetic instability.

Gaelle Boncompain, Nelly Gareil, Sarah Tessier, Aurianne Lescure, Thouis R Jones, Oliver Kepp, Guido Kroemer, Elaine Del Nery, Franck Perez (2019 Nov 5)

BML-265 and Tyrphostin AG1478 Disperse the Golgi Apparatus and Abolish Protein Transport in Human Cells.

Frontiers in cell and developmental biology : 232 : DOI : 10.3389/fcell.2019.00232

Résumé

The steady-state localization of Golgi-resident glycosylation enzymes in the Golgi apparatus depends on a balance between anterograde and retrograde transport. Using the Retention Using Selective Hooks (RUSH) assay and high-content screening, we identified small molecules that perturb the localization of Mannosidase II (ManII) used as a model cargo for Golgi resident enzymes. In particular, we found that two compounds known as EGFR tyrosine kinase inhibitors, namely BML-265 and Tyrphostin AG1478 disrupt Golgi integrity and abolish secretory protein transport of diverse cargos, thus inducing brefeldin A-like effects. Interestingly, BML-265 and Tyrphostin AG1478 affect Golgi integrity and transport in human cells but not in rodent cells. The effects of BML-265 are reversible since Golgi integrity and protein transport are quickly restored upon washout of the compounds. BML-265 and Tyrphostin AG1478 do not lead to endosomal tubulation suggesting that, contrary to brefeldin A, they do not target the -Golgi ARF GEF BIG1 and BIG2. They quickly induce COPI dissociation from Golgi membranes suggesting that, in addition to EGFR kinase, the -Golgi ARF GEF GBF1 might also be a target of these molecules. Accordingly, overexpression of

GBF1 prevents the effects of BML-265 and Tyrphostin AG1478 on Golgi integrity.

Gaelle Boncompain, Floriane Herit, Sarah Tessier, Aurianne Lescure, Elaine Del Nery, Pierre Gestraud, Isabelle Staropoli, Yuko Fukata, Masaki Fukata, Anne Brelot, Florence Niedergang, Franck Perez (2019 Oct 31)
Targeting CCR5 trafficking to inhibit HIV-1 infection.
Science advances : eaax0821 : DOI : 10.1126/sciadv.aax0821

Résumé

Using a cell-based assay monitoring differential protein transport in the secretory pathway coupled to high-content screening, we have identified three molecules that specifically reduce the delivery of the major co-receptor for HIV-1, CCR5, to the plasma membrane. They have no effect on the closely related receptors CCR1 and CXCR4. These molecules are also potent in primary macrophages as they markedly decrease HIV entry. At the molecular level, two of these molecules inhibit the critical palmitoylation of CCR5 and thereby block CCR5 in the early secretory pathway. Our results open a clear therapeutics avenue based on trafficking control and demonstrate that preventing HIV infection can be performed at the level of its receptor delivery.

Joseph C Boyd, Alice Pinheiro, Elaine Del Nery, Fabien Reyal, Thomas Walter (2019 Oct 15)
Domain-invariant features for mechanism of action prediction in a multi-cell-line drug screen.
Bioinformatics (Oxford, England) : 1607-1613 : DOI : 10.1093/bioinformatics/btz774

Résumé

High-content screening is an important tool in drug discovery and characterization. Often, high-content drug screens are performed on one single-cell line. Yet, a single-cell line cannot be thought of as a perfect disease model. Many diseases feature an important molecular heterogeneity. Consequently, a drug may be effective against one molecular subtype of a disease, but less so against another. To characterize drugs with respect to their effect not only on one cell line but on a panel of cell lines is therefore a promising strategy to streamline the drug discovery process.

Denis Krndija, Fatima El Marjou, Boris Guirao, Sophie Richon, Olivier Leroy, Yohanns Bellaiche, Edouard Hannezo, Danijela Matic Vignjevic. (2019 Aug 16)
Active cell migration is critical for steady-state epithelial turnover in the gut.
Science : 365(6454) : 705-710 : DOI : 10.1126/science.aau3429

Résumé

Steady-state turnover is a hallmark of epithelial tissues throughout adult life. Intestinal epithelial turnover is marked by continuous cell migration, which is assumed to be driven by mitotic pressure from the crypts. However, the balance of forces in renewal remains ill-defined. Combining biophysical modeling and quantitative three-dimensional tissue imaging with genetic and physical manipulations, we revealed the existence of an actin-related protein 2/3 complex-dependent active migratory force, which explains quantitatively the profiles of cell speed, density, and tissue tension along the villi. Cells migrate collectively with minimal rearrangements while displaying dual-apicobasal and front-back-polarity characterized by actin-rich basal protrusions oriented in the direction of migration. We propose that active migration is a critical component of gut epithelial turnover.

Anna Guadall, Sylvie Cochet, Olivier Renaud, Yves Colin, Caroline Le Van Kim, Alexandre G de Brevern, Wassim El Nemer (2019 Aug 16)

Dimerization and phosphorylation of Lutheran/basal cell adhesion molecule are critical for its function in cell migration on laminin.

The Journal of biological chemistry : 14911-14921 : DOI : 10.1074/jbc.RA119.007521

Résumé

Tumor cell migration depends on the interactions of adhesion proteins with the extracellular matrix. Lutheran/basal cell adhesion molecule (Lu/BCAM) promotes tumor cell migration by binding to laminin α5 chain, a subunit of laminins 511 and 521. Lu/BCAM is a type I transmembrane protein with a cytoplasmic domain of 59 (Lu) or 19 (Lu(v13)) amino acids. Here, using an array of techniques, including site-directed mutagenesis, immunoblotting, FRET, and proximity-ligation assays, we show that both Lu and Lu(v13) form homodimers at the cell surface of epithelial cancer cells. We mapped two small–small motifs in the transmembrane domain as potential sites for monomers docking and identified three cysteines in the cytoplasmic domain as being critical for covalently stabilizing dimers. We further found that Lu dimerization and phosphorylation of its cytoplasmic domain were concomitantly needed to promote cell migration. We conclude that Lu is the critical isoform supporting tumor cell migration on laminin 521 and that the Lu:Lu(v13) ratio at the cell surface may control the balance between cellular firm adhesion and migration.

Eric Peterman, Paulius Gibieža, Johnathon Schafer, Vytenis Arvydas Skeberdis, Algirdas Kaupinis, Mindaugas Valius, Xavier Heiligenstein, Ilse Hurbain, Graca Raposo, Rytis Prekeris (2019 Jul 20)

The post-abscission midbody is an intracellular signaling organelle that regulates cell proliferation.

Nature communications : 3181 : DOI : 10.1038/s41467-019-10871-0

Résumé

Once thought to be a remnant of cell division, the midbody (MB) has recently been shown to have roles beyond its primary function of orchestrating abscission. Despite the emerging roles of post-abscission MBs, how MBs accumulate in the cytoplasm and signal to regulate

cellular functions remains unknown. Here, we show that extracellular post-abscission MBs can be internalized by interphase cells, where they reside in the cytoplasm as a membrane-bound signaling structure that we have named the MBsome. We demonstrate that MBsomes stimulate cell proliferation and that MBsome formation is a phagocytosis-like process that depends on a phosphatidylserine/integrin complex, driven by actin-rich membrane protrusions. Finally, we show that MBsomes rely on dynamic actin coats to slow lysosomal degradation and propagate their signaling function. In summary, MBsomes may sometimes serve as intracellular organelles that signal via integrin and EGFR-dependent pathways to promote cell proliferation and anchorage-independent growth and survival.

Christin Bissig, Pauline Croisé, Xavier Heiligenstein, Ilse Hurbain, Guy M Lenk, Emily Kaufman, Ragna Sannerud, Wim Annaert, Miriam H Meisler, Lois S Weisman, Graça Raposo, Guillaume van Niel (2019 Mar 30)

Correction: The PIKfyve complex regulates the early melanosome homeostasis required for physiological amyloid formation (doi:10.1242/jcs.229500).

Journal of cell science : DOI : jcs231746

Résumé

Jamecna D, Polidori DJ, Mesmin B, Dezi M, Lévy D, Bigay J, Antonny B (2019 Mar 22)

An intrinsically disordered region in OSBP acts as an entropic barrier to control protein dynamics and orientation at membrane contact sites

Developmental cell * : * highlighted Trend in Cell Biology 2019 : DOI : 10.1016/j.devcel.2019.02.021

Résumé

Lipid transfer proteins (LTPs) acting at membrane contact sites (MCS) between the ER and other organelles contain domains involved in heterotypic (e.g. ER to Golgi) membrane tethering as well as domains involved in lipid transfer. Here, we show that a long ≈ 90 aa intrinsically unfolded sequence at the N-terminus of oxysterol binding protein (OSBP) controls OSBP orientation and dynamics at MCS. This Gly-Pro-Ala-rich sequence, whose hydrodynamic radius is twice as that of folded domains, prevents the two PH domains of the OSBP dimer from homotypically tethering two Golgi-like membranes and considerably facilitates OSBP in-plane diffusion and recycling at MCS. Although quite distant in sequence, the N-terminus of OSBP-related protein-4 (ORP4) has similar effects. We propose that N-terminal sequences of low complexity in ORPs form an entropic barrier that restrains protein orientation, limits protein density and facilitates protein mobility in the narrow and crowded MCS environment.

Anahi Capmany, Azumi Yoshimura, Rachid Kerdous, Valentina Caorsi, Aurianne Lescure, Elaine Del Nery, Evelyne Coudrier, Bruno Goud, Kristine Schauer (2019 Mar 16)

MYO1C stabilizes actin and facilitates the arrival of transport carriers at the Golgi complex.
Journal of cell science : DOI : jcs225029

Résumé

In this study, we aimed to identify the myosin motor proteins that control trafficking at the Golgi complex. In addition to the known Golgi-associated myosins MYO6, MYO18A and MYH9 (myosin IIA), we identified MYO1C as a novel player at the Golgi in a human cell line. We demonstrate that depletion of induces Golgi complex fragmentation and decompaction. MYO1C accumulates at dynamic structures around the Golgi complex that colocalize with Golgi-associated actin dots. depletion leads to loss of cellular F-actin, and Golgi complex decompaction is also observed after inhibition or loss of the actin-related protein 2/3 complex, Arp2/3 (also known as ARPC). We show that the functional consequence of depletion is a delay in the arrival of incoming transport carriers, both from the anterograde and retrograde routes. We propose that MYO1C stabilizes actin at the Golgi complex, facilitating the arrival of incoming transport carriers at the Golgi.This article has an associated First Person interview with the first author of the paper.

Varun Kapoor, William G Hirst, Christoph Hentschel, Stephan Preibisch, Simone Reber (2019 Mar 9)
MTrack: Automated Detection, Tracking, and Analysis of Dynamic Microtubules.
Scientific reports : 3794 : DOI : 10.1038/s41598-018-37767-1

Résumé

Microtubules are polar, dynamic filaments fundamental to many cellular processes. In vitro reconstitution approaches with purified tubulin are essential to elucidate different aspects of microtubule behavior. To date, deriving data from fluorescence microscopy images by manually creating and analyzing kymographs is still commonplace. Here, we present MTrack, implemented as a plug-in for the open-source platform Fiji, which automatically identifies and tracks dynamic microtubules with sub-pixel resolution using advanced objection recognition. MTrack provides automatic data interpretation yielding relevant parameters of microtubule dynamic instability together with population statistics. The application of our software produces unbiased and comparable quantitative datasets in a fully automated fashion. This helps the experimentalist to achieve higher reproducibility at higher throughput on a user-friendly platform. We use simulated data and real data to benchmark our algorithm and show that it reliably detects, tracks, and analyzes dynamic microtubules and achieves sub-pixel precision even at low signal-to-noise ratios.

Christin Bissig, Pauline Croisé, Xavier Heiligenstein, Ilse Hurbain, Guy M Lenk, Emily Kaufman, Ragna Sannerud, Wim Annaert, Miriam H Meisler, Lois S Weisman, Graça Raposo, Guillaume van Niel (2019 Feb 3)

The PIKfyve complex regulates the early melanosome homeostasis required for physiological amyloid formation.
Journal of cell science : DOI : jcs229500

Résumé

The metabolism of PI(3,5)P2 is regulated by the PIKfyve, VAC14 and FIG4 complex, mutations in which are associated with hypopigmentation in mice. These pigmentation defects indicate a key, but as yet unexplored, physiological relevance of this complex in the biogenesis of melanosomes. Here, we show that PIKfyve activity regulates formation of amyloid matrix composed of PMEL protein within the early endosomes in melanocytes, called stage I melanosomes. PIKfyve activity controls the membrane remodeling of stage I melanosomes, which regulates PMEL abundance, sorting and processing. PIKfyve activity also affects stage I melanosome kiss-and-run interactions with lysosomes, which are required for PMEL amyloidogenesis and the establishment of melanosome identity. Mechanistically, PIKfyve activity promotes both the formation of membrane tubules from stage I melanosomes and their release by modulating endosomal actin branching. Taken together, our data indicate that PIKfyve activity is a key regulator of the melanosomal import-export machinery that fine tunes the formation of functional amyloid fibrils in melanosomes and the maintenance of melanosome identity.This article has an associated First Person interview with the first author of the paper.

Beber A, Taveneau C, Nania M, Tsai FC, Di Cicco A, Bassereau P, Lévy D, Cabral JT, Isambert H, Mangenot S*, Bertin A* (2019 Jan 24)
Membrane reshaping by micrometric curvature sensitive septin filaments
Nature communications : DOI : 10.1038/s41467-019-08344-5

Résumé

Septins are cytoskeletal filaments that assemble at the inner face of the plasma membrane.They are localized at constriction sites and impact membrane remodeling. We report in vitro tools to examine how yeast septins behave on curved and deformable membranes. Septins reshape the membranes of Giant Unilamellar Vesicles with the formation of periodic spikes, while flattening smaller vesicles. We show that membrane deformations are associated to preferential arrangement of Septin filaments on specific curvatures. When binding to bilayers supported on custom-designed periodic wavy patterns displaying positive and negative micrometric radii of curvatures, septin filaments remain straight and perpendicular to the curvature of the convex parts, while bending negatively to follow concave geometries. Based on these results, we propose a theoretical model that describes the deformations and micrometric curvature sensitivity observed in vitro. The model captures the reorganizations of septin filaments throughout cytokinesis in vivo, providing mechanistic insights into cell division.