

Année de publication : 2019

Clémentine Villeneuve, Emilie Lagoutte, Ludmilla de Plater, Samuel Mathieu, Jean-Baptiste Manneville, Jean-Léon Maître, Philippe Chavrier, Carine Rossé (2019 Nov 9)

aPKC ζ triggers basal extrusion of luminal mammary epithelial cells by tuning contractility and vinculin localization at cell junctions.

Proceedings of the National Academy of Sciences of the United States of America : [DOI :](#)

[201906779](#)

Résumé

Metastasis is the main cause of cancer-related deaths. How a single oncogenic cell evolves within highly organized epithelium is still unknown. Here, we found that the overexpression of the protein kinase atypical protein kinase C ζ (aPKC ζ), an oncogene, triggers basally oriented epithelial cell extrusion in vivo as a potential mechanism for early breast tumor cell invasion. We found that cell segregation is the first step required for basal extrusion of luminal cells and identify aPKC ζ and vinculin as regulators of cell segregation. We propose that asymmetric vinculin levels at the junction between normal and aPKC ζ cells trigger an increase in tension at these cell junctions. Moreover, we show that aPKC ζ cells acquire promigratory features, including increased vinculin levels and vinculin dynamics at the cell-substratum contacts. Overall, this study shows that a balance between cell contractility and cell-cell adhesion is crucial for promoting basally oriented cell extrusion, a mechanism for early breast cancer cell invasion.

Année de publication : 2017

Catalina Lodillinsky, Katrina Podsypanina, Philippe Chavrier (2017 Mar 1)

Social networking in tumor cell communities is associated with increased aggressiveness.

Intravital : e1112476 : [DOI : 10.1080/21659087.2015.1112476](#)

Résumé

Extracellular vesicles (EVs) are lipid-bilayer-enclosed vesicles that contain proteins, lipids and nucleic acids. EVs produced by cells from healthy tissues circulate in the blood and body fluids, and can be taken up by unrelated cells. As they have the capacity to transfer cargo proteins, lipids and nucleic acids (mostly mRNAs and miRNAs) between different cells in the body, EVs are emerging as mediators of intercellular communication that could modulate cell behavior, tissue homeostasis and regulation of physiological functions. EV-mediated cell-cell communications are also proposed to play a role in disease, for example, cancer, where they could contribute to transfer of traits required for tumor progression and metastasis. However, direct evidence for EV-mediated mRNA transfer to individual cells and for its biological consequences in vivo has been missing until recently. Recent studies have reported elegant experiments using genetic tracing with the Cre recombinase system and intravital imaging that visualize and quantify functional transfer of mRNA mediated by EVs in

the context of cancer and metastasis.

G-A Franzetti, K Laud-Duval, W van der Ent, A Brisac, M Irondelle, S Aubert, U Dirksen, C Bouvier, G de Pinieux, E Snaar-Jagalska, P Chavrier, O Delattre (2017 Jan 31)

Cell-to-cell heterogeneity of EWSR1-FLI1 activity determines proliferation/migration choices in Ewing sarcoma cells.

Oncogene : DOI : [10.1038/onc.2016.498](https://doi.org/10.1038/onc.2016.498)

Résumé

Ewing sarcoma is characterized by the expression of the chimeric EWSR1-FLI1 transcription factor. Proteomic analyses indicate that the decrease of EWSR1-FLI1 expression leads to major changes in effectors of the dynamics of the actin cytoskeleton and the adhesion processes with a shift from cell-to-cell to cell-matrix adhesion. These changes are associated with a dramatic increase of in vivo cell migration and invasion potential. Importantly, EWSR1-FLI1 expression, evaluated by single-cell RT-ddPCR/immunofluorescence analyses, and activity, assessed by expression of EWSR1-FLI1 downstream targets, are heterogeneous in cell lines and in tumours and can fluctuate along time in a fully reversible process between EWSR1-FLI1(high) states, characterized by highly active cell proliferation, and EWSR1-FLI1(low) states where cells have a strong propensity to migrate, invade and metastasize. This new model of phenotypic plasticity proposes that the dynamic fluctuation of the expression level of a dominant oncogene is an intrinsic characteristic of its oncogenic potential. *Oncogene* advance online publication, 30 January 2017; doi:10.1038/onc.2016.498.

Année de publication : 2016

Antonio Castro-Castro, Valentina Marchesin, Pedro Monteiro, Catalina Lodillinsky, Carine Rossé, Philippe Chavrier (2016 Aug 9)

Cellular and Molecular Mechanisms of MT1-MMP-Dependent Cancer Cell Invasion.

Annual review of cell and developmental biology

Résumé

Metastasis is responsible for most cancer-associated deaths. Accumulating evidence based on 3D migration models has revealed a diversity of invasive migratory schemes reflecting the plasticity of tumor cells to switch between proteolytic and nonproteolytic modes of invasion. Yet, initial stages of localized regional tumor dissemination require proteolytic remodeling of the extracellular matrix to overcome tissue barriers. Recent data indicate that surface-exposed membrane type 1-matrix metalloproteinase (MT1-MMP), belonging to a group of membrane-anchored MMPs, plays a central role in pericellular matrix degradation during basement membrane and interstitial tissue transmigration programs. In addition, a large body of work indicates that MT1-MMP is targeted to specialized actin-rich cell protrusions termed invadopodia, which are responsible for matrix degradation. This review

describes the multistep assembly of actin-based invadopodia in molecular details. Mechanisms underlying MT1-MMP traffic to invadopodia through endocytosis/recycling cycles, which are key to the invasive program of carcinoma cells, are discussed. Expected final online publication date for the Annual Review of Cell and Developmental Biology Volume 32 is October 06, 2016. Please see <http://www.annualreviews.org/catalog/pubdates.aspx> for revised estimates.

Hiroaki Kajiho, Yuko Kajiho, Emanuela Frittoli, Stefano Confalonieri, Giovanni Bertalot, Giuseppe Viale, Pier Paolo Di Fiore, Amanda Oldani, Massimiliano Garre, Galina V Beznoussenko, Andrea Palamidessi, Manuela Vecchi, Philippe Chavier, Frank Perez, Giorgio Scita (2016 Jun 4)

RAB2A controls MT1-MMP endocytic and E-cadherin polarized Golgi trafficking to promote invasive breast cancer programs.

EMBO reports : [DOI : e201642032](https://doi.org/10.1038/e201642032)

Résumé

The mechanisms of tumor cell dissemination and the contribution of membrane trafficking in this process are poorly understood. Through a functional siRNA screening of human RAB GTPases, we found that RAB2A, a protein essential for ER-to-Golgi transport, is critical in promoting proteolytic activity and 3D invasiveness of breast cancer (BC) cell lines. Remarkably, RAB2A is amplified and elevated in human BC and is a powerful and independent predictor of disease recurrence in BC patients. Mechanistically, RAB2A acts at two independent trafficking steps. Firstly, by interacting with VPS39, a key component of the late endosomal HOPS complex, it controls post-endocytic trafficking of membrane-bound MT1-MMP, an essential metalloprotease for matrix remodeling and invasion. Secondly, it further regulates Golgi transport of E-cadherin, ultimately controlling junctional stability, cell compaction, and tumor invasiveness. Thus, RAB2A is a novel trafficking determinant essential for regulation of a mesenchymal invasive program of BC dissemination.

Emilie Lagoutte, Clémentine Villeneuve, Laurence Lafanechère, Claire M Wells, Gareth E Jones, Philippe Chavier, Carine Rossé (2016 Apr 28)

LIMK Regulates Tumor-Cell Invasion and Matrix Degradation Through Tyrosine Phosphorylation of MT1-MMP.

Scientific reports : 24925 : [DOI : 10.1038/srep24925](https://doi.org/10.1038/srep24925)

Résumé

During their metastatic spread, cancer cells need to remodel the extracellular matrix in order to migrate through stromal compartments adjacent to the primary tumor. Dissemination of breast carcinoma cells is mediated by membrane type 1-matrix metalloproteinase (MT1-MMP/MMP14), the main invadopodial matrix degradative component. Here, we identify MT1-MMP as a novel interacting partner of dual-specificity LIM Kinase-1 and -2 (LIMK1/2), and provide several evidence for phosphorylation of tyrosine Y573 in the cytoplasmic domain of MT1-MMP by LIMK. Phosphorylation of Y573 influences association of F-actin binding protein

cortactin to MT1-MMP-positive endosomes and invadopodia formation and matrix degradation. Moreover, we show that LIMK1 regulates cortactin association to MT1-MMP-positive endosomes, while LIMK2 controls invadopodia-associated cortactin. In turn, LIMK1 and LIMK2 are required for MT1-MMP-dependent matrix degradation and cell invasion in a three-dimensional type I collagen environment. This novel link between LIMK1/2 and MT1-MMP may have important consequences for therapeutic control of breast cancer cell invasion.

Nadège Gruel, Laetitia Fuhrmann, Catalina Lodillinsky, Vanessa Benhamo, Odette Mariani, Aurélie Cédenot, Laurent Arnould, Gaëtan Macgrogan, Xavier Sastre-Garau, Philippe Chavrier, Olivier Delattre, Anne Vincent-Salomon (2016 Feb 19)

LIN7A is a major determinant of cell-polarity defects in breast carcinomas.

Breast cancer research : BCR : 23 : [DOI : 10.1186/s13058-016-0680-x](https://doi.org/10.1186/s13058-016-0680-x)

Résumé

Polarity defects are a hallmark of most carcinomas. Cells from invasive micropapillary carcinomas (IMPCs) of the breast are characterized by a striking cell polarity inversion and represent an interesting model for the analysis of polarity abnormalities.

Année de publication : 2015

Valentina Marchesin, Antonio Castro-Castro, Catalina Lodillinsky, Alessia Castagnino, Joanna Cyrta, Hélène Bonsang-Kitzis, Laetitia Fuhrmann, Marie Irondelle, Elvira Infante, Guillaume Montagnac, Fabien Rey, Anne Vincent-Salomon, Philippe Chavrier (2015 Oct 28)

ARF6-JIP3/4 regulate endosomal tubules for MT1-MMP exocytosis in cancer invasion.

The Journal of cell biology : 339-58 : [DOI : 10.1083/jcb.201506002](https://doi.org/10.1083/jcb.201506002)

Résumé

Invasion of cancer cells into collagen-rich extracellular matrix requires membrane-tethered membrane type 1-matrix metalloproteinase (MT1-MMP) as the key protease for collagen breakdown. Understanding how MT1-MMP is delivered to the surface of tumor cells is essential for cancer cell biology. In this study, we identify ARF6 together with c-Jun NH2-terminal kinase-interacting protein 3 and 4 (JIP3 and JIP4) effectors as critical regulators of this process. Silencing ARF6 or JIP3/JIP4 in breast tumor cells results in MT1-MMP endosome mispositioning and reduces MT1-MMP exocytosis and tumor cell invasion. JIPs are recruited by Wiskott-Aldrich syndrome protein and scar homologue (WASH) on MT1-MMP endosomes on which they recruit dynein-dynactin and kinesin-1. The interaction of plasma membrane ARF6 with endosomal JIPs coordinates dynactin-dynein and kinesin-1 activity in a tug-of-war mechanism, leading to MT1-MMP endosome tubulation and exocytosis. In addition, we find that ARF6, MT1-MMP, and kinesin-1 are up-regulated in high-grade triple-negative breast

cancers. These data identify a critical ARF6-JIP-MT1-MMP-dynein-dynactin-kinesin-1 axis promoting an invasive phenotype of breast cancer cells.

C Lodillinsky, E Infante, A Guichard, R Chaligné, L Fuhrmann, J Cyrta, M Irondelle, E Lagoutte, S Vacher, H Bonsang-Kitzis, M Glukhova, F Reyal, I Bièche, A Vincent-Salomon, P Chavrier (2015 Apr 21)

p63/MT1-MMP axis is required for in situ to invasive transition in basal-like breast cancer.

Oncogene : 344-57 : [DOI : 10.1038/onc.2015.87](https://doi.org/10.1038/onc.2015.87)

Résumé

The transition of ductal carcinoma in situ (DCIS) to invasive breast carcinoma requires tumor cells to cross the basement membrane (BM). However, mechanisms underlying BM transmigration are poorly understood. Here, we report that expression of membrane-type 1 (MT1)-matrix metalloproteinase (MMP), a key component of the BM invasion program, increases during breast cancer progression at the in situ to invasive breast carcinoma transition. In the intraductal xenograft model, MT1-MMP is required for BM transmigration of MCF10DCIS.com breast adenocarcinoma cells and is overexpressed in cell clusters overlying focal BM disruptions and at the invasive tumor front. Mirrored upregulation of p63 and MT1-MMP is observed at the edge of MCF10DCIS.com xenograft tumors and p63 is required for induction of MT1-MMP-dependent invasive program in response to microenvironmental signals. Immunohistochemistry and analysis of public database reveal that p63 and MT1-MMP are upregulated in human basal-like breast tumors suggesting that p63/MT1-MMP axis contributes to progression of basal-like breast cancers with elevated p63 and MT1-MMP levels.

Valentina Marchesin, Guillaume Montagnac, Philippe Chavrier (2015 Mar 24)

ARF6 promotes the formation of Rac1 and WAVE-dependent ventral F-actin rosettes in breast cancer cells in response to epidermal growth factor.

PloS one : e0121747 : [DOI : 10.1371/journal.pone.0121747](https://doi.org/10.1371/journal.pone.0121747)

Résumé

Coordination between actin cytoskeleton assembly and localized polarization of intracellular trafficking routes is crucial for cancer cell migration. ARF6 has been implicated in the endocytic recycling of surface receptors and membrane components and in actin cytoskeleton remodeling. Here we show that overexpression of an ARF6 fast-cycling mutant in MDA-MB-231 breast cancer-derived cells to mimic ARF6 hyperactivation observed in invasive breast tumors induced a striking rearrangement of the actin cytoskeleton at the ventral cell surface. This phenotype consisted in the formation of dynamic actin-based podosome rosette-like structures expanding outward as wave positive for F-actin and actin cytoskeleton regulatory components including cortactin, Arp2/3 and SCAR/WAVE complexes and upstream Rac1 regulator. Ventral rosette-like structures were similarly induced in MDA-

MB-231 cells in response to epidermal growth factor (EGF) stimulation and to Rac1 hyperactivation. In addition, interference with ARF6 expression attenuated activation and plasma membrane targeting of Rac1 in response to EGF treatment. Our data suggest a role for ARF6 in linking EGF-receptor signaling to Rac1 recruitment and activation at the plasma membrane to promote breast cancer cell directed migration.

Kalthoum Ben M'Barek, Diana Molino, Sandrine Quignard, Marie-Aude Plamont, Yong Chen, Philippe Chavrier, Jacques Fattaccioli (2015 Mar 16)

Phagocytosis of immunoglobulin-coated emulsion droplets.

Biomaterials : 270-7 : [DOI : 10.1016/j.biomaterials.2015.02.030](https://doi.org/10.1016/j.biomaterials.2015.02.030)

Résumé

Phagocytosis by macrophages represents a fundamental process essential for both immunity and tissue homeostasis. The size of targets to be eliminated ranges from small particles as bacteria to large objects as cancerous or senescent cells. Most of our current quantitative knowledge on phagocytosis is based on the use of solid polymer microparticles as model targets that are well adapted to the study of phagocytosis mechanisms that do not involve any lateral mobility of the ligands, despite the relevance of this parameter in the immunological context. Herein we designed monodisperse, IgG-coated emulsion droplets that are efficiently and specifically internalized by macrophages through in-vitro FcγR-mediated phagocytosis. We show that, contrary to solid polymeric beads, droplet uptake is efficient even for low IgG densities, and is accompanied by the clustering of the opsonins in the zone of contact with the macrophage during the adhesion step. Beyond the sole interest in the design of the material, our results suggest that lateral mobility of proteins at the interface of a target greatly enhances the phagocytic uptake.

Année de publication : 2014

Mathieu Boissan, Guillaume Montagnac, Qinfang Shen, Lorena Griparic, Jérôme Guitton, Maryse Romao, Nathalie Sauvonnnet, Thibault Lagache, Ioan Lascu, Graça Raposo, Céline Desbourdes, Uwe Schlattner, Marie-Lise Lacombe, Simona Polo, Alexander M van der Bliet, Aurélien Roux, Philippe Chavrier (2014 Jun 28)

Membrane trafficking. Nucleoside diphosphate kinases fuel dynamin superfamily proteins with GTP for membrane remodeling.

Science (New York, N.Y.) : 1510-5 : [DOI : 10.1126/science.1253768](https://doi.org/10.1126/science.1253768)

Résumé

Dynamin superfamily molecular motors use guanosine triphosphate (GTP) as a source of energy for membrane-remodeling events. We found that knockdown of nucleoside diphosphate kinases (NDPKs) NM23-H1/H2, which produce GTP through adenosine triphosphate (ATP)-driven conversion of guanosine diphosphate (GDP), inhibited dynamin-

mediated endocytosis. NM23-H1/H2 localized at clathrin-coated pits and interacted with the proline-rich domain of dynamin. In vitro, NM23-H1/H2 were recruited to dynamin-induced tubules, stimulated GTP-loading on dynamin, and triggered fission in the presence of ATP and GDP. NM23-H4, a mitochondria-specific NDPK, colocalized with mitochondrial dynamin-like OPA1 involved in mitochondria inner membrane fusion and increased GTP-loading on OPA1. Like OPA1 loss of function, silencing of NM23-H4 but not NM23-H1/H2 resulted in mitochondrial fragmentation, reflecting fusion defects. Thus, NDPKs interact with and provide GTP to dynamins, allowing these motor proteins to work with high thermodynamic efficiency.

Carine Rossé, Catalina Lodillinsky, Laetitia Fuhrmann, Maya Nourieh, Pedro Monteiro, Marie Irondelle, Emilie Lagoutte, Sophie Vacher, François Waharte, Perrine Paul-Gilloteaux, Maryse Romao, Lucie Sengmanivong, Mark Linch, Johan van Lint, Graça Raposo, Anne Vincent-Salomon, Ivan Bièche, Peter J Parker, Philippe Chavrier (2014 Apr 21)

Control of MT1-MMP transport by atypical PKC during breast-cancer progression.

Proceedings of the National Academy of Sciences of the United States of America : E1872-9 : [DOI : 10.1073/pnas.1400749111](https://doi.org/10.1073/pnas.1400749111)

Résumé

Dissemination of carcinoma cells requires the pericellular degradation of the extracellular matrix, which is mediated by membrane type 1-matrix metalloproteinase (MT1-MMP). In this article, we report a co-up-regulation and colocalization of MT1-MMP and atypical protein kinase C iota (aPKC ι) in hormone receptor-negative breast tumors in association with a higher risk of metastasis. Silencing of aPKC in invasive breast-tumor cell lines impaired the delivery of MT1-MMP from late endocytic storage compartments to the surface and inhibited matrix degradation and invasion. We provide evidence that aPKC ι , in association with MT1-MMP-containing endosomes, phosphorylates cortactin, which is present in F-actin-rich puncta on MT1-MMP-positive endosomes and regulates cortactin association with the membrane scission protein dynamin-2. Thus, cell line-based observations and clinical data reveal the concerted activity of aPKC, cortactin, and dynamin-2, which control the trafficking of MT1-MMP from late endosome to the plasma membrane and play an important role in the invasive potential of breast-cancer cells.

Guillaume Montagnac, Philippe Chavrier (2014 Feb 28)

[When microtubules meet clathrin-coated pits].

Médecine sciences : M/S : 130-3 : [DOI : 10.1051/medsci/20143002005](https://doi.org/10.1051/medsci/20143002005)

Résumé

Année de publication : 2013

Pedro Monteiro, Carine Rossé, Antonio Castro-Castro, Marie Irondelle, Emilie Lagoutte, Perrine Paul-Gilloteaux, Claire Desnos, Etienne Formstecher, François Darchen, David Perrais, Alexis Gautreau, Maud Hertzog, Philippe Chavrier (2013 Dec 18)

Endosomal WASH and exocyst complexes control exocytosis of MT1-MMP at invadopodia.

The Journal of cell biology : 1063-79

Résumé

Remodeling of the extracellular matrix by carcinoma cells during metastatic dissemination requires formation of actin-based protrusions of the plasma membrane called invadopodia, where the trans-membrane type 1 matrix metalloproteinase (MT1-MMP) accumulates. Here, we describe an interaction between the exocyst complex and the endosomal Arp2/3 activator Wiskott-Aldrich syndrome protein and Scar homolog (WASH) on MT1-MMP-containing late endosomes in invasive breast carcinoma cells. We found that WASH and exocyst are required for matrix degradation by an exocytic mechanism that involves tubular connections between MT1-MMP-positive late endosomes and the plasma membrane in contact with the matrix. This ensures focal delivery of MT1-MMP and supports pericellular matrix degradation and tumor cell invasion into different pathologically relevant matrix environments. Our data suggest a general mechanism used by tumor cells to breach the basement membrane and for invasive migration through fibrous collagen-enriched tissues surrounding the tumor.