Année de publication : 2021

Ashley L Arthur, Amy Crawford, Anne Houdusse, Margaret A Titus (2021 May 27)
VASP mediated actin dynamics activate and recruit a filopodia myosin.
eLife : DOI : 10.7554/eLife.68082

Résumé

Filopodia are thin, actin-based structures that cells use to interact with their environments. Filopodia initiation requires a suite of conserved proteins but the mechanism remains poorly understood. The actin polymerase VASP and a MyTH-FERM (MF) myosin, DdMyo7 in amoeba, are essential for filopodia initiation. DdMyo7 is localized to dynamic regions of the actin-rich cortex. Analysis of VASP mutants and treatment of cells with anti-actin drugs shows that myosin recruitment and activation in requires localized VASP-dependent actin polymerization. Targeting of DdMyo7 to the cortex alone is not sufficient for filopodia initiation; VASP activity is also required. The actin regulator locally produces a cortical actin network that activates myosin and together they shape the actin network to promote extension of parallel bundles of actin during filopodia formation. This work reveals how filopodia initiation requires close collaboration between an actin binding protein, the state of the actin cytoskeleton and MF myosin activity.

Anne Houdusse, Margaret A Titus (2021 May 25)
The many roles of myosins in filopodia, microvilli and stereocilia.
Current biology : CB : R586-R602 : DOI : S0960-9822(21)00518-2

Résumé

Filopodia, microvilli and stereocilia represent an important group of plasma membrane protrusions. These specialized projections are supported by parallel bundles of actin filaments and have critical roles in sensing the external environment, increasing cell surface area, and acting as mechanosensors. While actin-associated proteins are essential for actin-filament elongation and bundling in these protrusions, myosin motors have a surprising role in the formation and extension of filopodia and stereocilia and in the organization of microvilli. Actin regulators and specific myosins collaborate in controlling the length of these structures. Myosins can transport cargoes along the length of these protrusions, and, in the case of stereocilia and microvilli, interactions with adaptors and cargoes can also serve to anchor adhesion receptors to the actin-rich core via functionally conserved motor-adaptor complexes. This review highlights recent progress in understanding the diverse roles myosins play in filopodia, microvilli and stereocilia.

Catalina Lodillinsky, Laetitia Fuhrmann, Marie Irondelle, Olena Pylypenko, Xiao-Yan Li, Hélène Bonsang-Kitzis, Fabien Reyal, Sophie Vacher, Claire Calmel, Olivier De Wever, Ivan Bièche, Marie-Lise Lacombe, Ana Maria Eiján, Anne Houdusse, Anne Vincent-Salomon, Stephen J Weiss,

Philippe Chavrier, Mathieu Boissan (2021 May 20)

Metastasis-suppressor NME1 controls the invasive switch of breast cancer by regulating MT1-MMP surface clearance.

Oncogene : DOI : 10.1038/s41388-021-01826-1

Résumé

Membrane Type 1 Matrix Metalloprotease (MT1-MMP) contributes to the invasive progression of breast cancers by degrading extracellular matrix tissues. Nucleoside diphosphate kinase, NME1/NM23-H1, has been identified as a metastasis suppressor; however, its contribution to local invasion in breast cancer is not known. Here, we report that NME1 is up-regulated in ductal carcinoma in situ (DCIS) as compared to normal breast epithelial tissues. NME1 levels drop in microinvasive and invasive components of breast tumor cells relative to synchronous DCIS foci. We find a strong anti-correlation between NME1 and plasma membrane MT1-MMP levels in the invasive components of breast tumors, particularly in aggressive histological grade III and triple-negative breast cancers. Knockout of NME1 accelerates the invasive transition of breast tumors in the intraductal xenograft model. At the mechanistic level, we find that MT1-MMP, NME1 and dynamin-2, a GTPase known to require GTP production by NME1 for its membrane fission activity in the endocytic pathway, interact in clathrin-coated vesicles at the plasma membrane. Loss of NME1 function increases MT1-MMP surface levels by inhibiting endocytic clearance. As a consequence, the ECM degradation and invasive potentials of breast cancer cells are enhanced. This study identifies the down-modulation of NME1 as a potent driver of the in situ-to invasive transition during breast cancer progression.

Anne Houdusse (2021 May 14)

Biological nanomotors, driving forces of life.

Comptes rendus biologies : 53-78 : DOI : 10.5802/crbiol.45

Résumé

Life is driven by awe-inspiring coordinated movements observed in cells and tissues. In each cell, nm-size molecular motor proteins contribute to these movements as they power numerous mechanical processes with precision and complex orchestration. For the multiple functions that an eukaryotic cell accomplish, motility is essential both at molecular and cellular scales. Tissue morphogenesis, cell migration, cell division or cell differentiation are all controlled by the precise action of such nanomotors that work on cytoskeletal tracks using ATP as fuel. The study of motility has a long history and scientists of all disciplines have contributed to its understanding. The first part of this review compares myosin and kinesin motors to describe the principles underlying how motors convert chemical energy into mechanical movement. In a second part, I will describe how sequence differences selected through evolution can lead to distinct force production output despite a common mechanism. Motors within a superfamily can thus carry out distinct functions in cells. Such differences give rise to their individual, specific motility properties, including reversal of directionality or ability to organize cytoskeletal tracks. The power of structural biology to reveal unexpected and surprising structures, with certainty when visualized at atomic resolution, has been a great advantage for this field. The critical insights gained from the

structures can be carefully tested with functional experiments, leading to progress in defining the role motors play in cells. Last, I will describe how targeting these motors can be beneficial for human health. Allosteric sites for specific small molecules can act as activators or inhibitors of the force produced by these nanomotors. While frequent sites of mutations in these motors can lead to disease phenotypes, high therapeutic potential of allosteric effectors is now established for heart muscle diseases and should be extended to treat other pathologies.

Julien Robert-Paganin, Xiao-Ping Xu, Mark F Swift, Daniel Auguin, James P Robblee, Hailong Lu, Patricia M Fagnant, Elena B Krementsova, Kathleen M Trybus, Anne Houdusse, Niels Volkmann, Dorit Hanein (2021 Mar 26)

The actomyosin interface contains an evolutionary conserved core and an ancillary interface involved in specificity.

Nature communications : 1892 : DOI : 10.1038/s41467-021-22093-4

Résumé

Plasmodium falciparum, the causative agent of malaria, moves by an atypical process called gliding motility. Actomyosin interactions are central to gliding motility. However, the details of these interactions remained elusive until now. Here, we report an atomic structure of the divergent Plasmodium falciparum actomyosin system determined by electron cryomicroscopy at the end of the powerstroke (Rigor state). The structure provides insights into the detailed interactions that are required for the parasite to produce the force and motion required for infectivity. Remarkably, the footprint of the myosin motor on filamentous actin is conserved with respect to higher eukaryotes, despite important variability in the Plasmodium falciparum myosin and actin elements that make up the interface. Comparison with other actomyosin complexes reveals a conserved core interface common to all actomyosin complexes, with an ancillary interface involved in defining the spatial positioning of the motor on actin filaments.

Année de publication : 2020

Dihia Moussaoui, James P Robblee, Daniel Auguin, Elena B Krementsova, Silvia Haase, Thomas Ca Blake, Jake Baum, Julien Robert-Paganin, Kathleen M Trybus, Anne Houdusse (2020 Oct 13)

Full-length myosin A and essential light chain PfELC structures provide new anti-malarial targets.

eLife : DOI : 10.7554/eLife.60581

Résumé

Parasites from the genus Plasmodium are the causative agents of malaria. The mobility, infectivity, and ultimately pathogenesis of rely on a macromolecular complex, called the glideosome. At the core of the glideosome is an essential and divergent Myosin A motor (PfMyoA), a first order drug target against malaria. Here, we present the full-length structure

of PfMyoA in two states of its motor cycle. We report novel interactions that are essential for motor priming and the mode of recognition of its two light chains (PfELC and MTIP) by two degenerate IQ motifs. Kinetic and motility assays using PfMyoA variants, along with molecular dynamics, demonstrate how specific priming and atypical sequence adaptations tune the motor's mechano-chemical properties. Supported by evidence for an essential role of the PfELC in malaria pathogenesis, these structures provide a blueprint for the design of future anti-malarials targeting both the glideosome motor and its regulatory elements.

Máté Gyimesi, Ádám I Horváth, Demeter Túrós, Sharad Kumar Suthar, Máté Pénzes, Csilla Kurdi, Louise Canon, Carlos Kikuti, Kathleen M Ruppel, Darshan V Trivedi, James A Spudich, István Lőrincz, Anna Á Rauscher, Mihály Kovács, Endre Pál, Sámuel Komoly, Anne Houdusse, András Málnási-Csizmadia (2020 Oct 9)

Single Residue Variation in Skeletal Muscle Myosin Enables Direct and Selective Drug Targeting for Spasticity and Muscle Stiffness.

Cell : 335-346.e13 : DOI : S0092-8674(20)31138-7

Résumé

Muscle spasticity after nervous system injuries and painful low back spasm affect more than 10% of global population. Current medications are of limited efficacy and cause neurological and cardiovascular side effects because they target upstream regulators of muscle contraction. Direct myosin inhibition could provide optimal muscle relaxation; however, targeting skeletal myosin is particularly challenging because of its similarity to the cardiac isoform. We identified a key residue difference between these myosin isoforms, located in the communication center of the functional regions, which allowed us to design a selective inhibitor, MPH-220. Mutagenic analysis and the atomic structure of MPH-220-bound skeletal muscle myosin confirmed the mechanism of specificity. Targeting skeletal muscle myosin by MPH-220 enabled muscle relaxation, in human and model systems, without cardiovascular side effects and improved spastic gait disorders after brain injury in a disease model. MPH-220 provides a potential nervous-system-independent option to treat spasticity and muscle stiffness.

H Lee Sweeney, Anne Houdusse, Julien Robert-Paganin (2020 May 27)

Myosin Structures.

Advances in experimental medicine and biology : 7-19 : DOI : 10.1007/978-3-030-38062-5_2

Résumé

Directed movements on actin filaments within the cell are powered by molecular motors of the myosin superfamily. On actin filaments, myosin motors convert the energy from ATP into force and movement. Myosin motors power such diverse cellular functions as cytokinesis, membrane trafficking, organelle movements, and cellular migration. Myosin generates force and movement via a number of structural changes associated with hydrolysis of ATP, binding to actin, and release of the ATP hydrolysis products while bound to actin. Herein we provide

an overview of those structural changes and how they relate to the actin-myosin ATPase cycle. These structural changes are the basis of chemo-mechanical transduction by myosin motors.

Année de publication : 2019

Julien Robert-Paganin, Olena Pylypenko, Carlos Kikuti, H Lee Sweeney, Anne Houdusse (2019 Nov 6)

Force Generation by Myosin Motors: A Structural Perspective.

Chemical reviews : 5-35 : DOI : 10.1021/acs.chemrev.9b00264

Résumé

Generating force and movement is essential for the functions of cells and organisms. A variety of molecular motors that can move on tracks within cells have evolved to serve this role. How these motors interact with their tracks and how that, in turn, leads to the generation of force and movement is key to understanding the cellular roles that these motor-track systems serve. This review is focused on the best understood of these systems, which is the molecular motor myosin that moves on tracks of filamentous (F-) actin. The review highlights both the progress and the limits of our current understanding of how force generation can be controlled by F-actin-myosin interactions. What has emerged are insights they may serve as a framework for understanding the design principles of a number of types of molecular motors and their interactions with their tracks.

Ashley L Arthur, Livia D Songster, Helena Sirkia, Akash Bhattacharya, Carlos Kikuti, Fernanda Pires Borrega, Anne Houdusse, Margaret A Titus (2019 Oct 16)

Optimized filopodia formation requires myosin tail domain cooperation.

Proceedings of the National Academy of Sciences of the United States of America : 22196-22204 : DOI : 10.1073/pnas.1901527116

Résumé

Filopodia are actin-filled protrusions employed by cells to interact with their environment. Filopodia formation in Amoebozoa and Metazoa requires the phylogenetically diverse MyTH4-FERM (MF) myosins DdMyo7 and Myo10, respectively. While Myo10 is known to form antiparallel dimers, DdMyo7 lacks a coiled-coil domain in its proximal tail region, raising the question of how such divergent motors perform the same function. Here, it is shown that the DdMyo7 lever arm plays a role in both autoinhibition and function while the proximal tail region can mediate weak dimerization, and is proposed to be working in cooperation with the C-terminal MF domain to promote partner-mediated dimerization. Additionally, a forced dimer of the DdMyo7 motor is found to weakly rescue filopodia formation, further highlighting the importance of the C-terminal MF domain. Thus, weak dimerization activity of the DdMyo7 proximal tail allows for sensitive regulation of myosin activity to prevent inappropriate activation of filopodia formation. The results reveal that the principles of MF

myosin-based filopodia formation are conserved via divergent mechanisms for dimerization.

Julien Robert-Paganin, James P Robblee, Daniel Auguin, Thomas C A Blake, Carol S Bookwalter, Elena B Krementsova, Dihia Moussaoui, Michael J Previs, Guillaume Jousset, Jake Baum, Kathleen M Trybus, Anne Houdusse (2019 Jul 25)

Plasmodium myosin A drives parasite invasion by an atypical force generating mechanism.

Nature communications : 3286 : DOI : 10.1038/s41467-019-11120-0

Résumé

Plasmodium parasites are obligate intracellular protozoa and causative agents of malaria, responsible for half a million deaths each year. The lifecycle progression of the parasite is reliant on cell motility, a process driven by myosin A, an unconventional single-headed class XIV molecular motor. Here we demonstrate that myosin A from Plasmodium falciparum (PfMyoA) is critical for red blood cell invasion. Further, using a combination of X-ray crystallography, kinetics, and in vitro motility assays, we elucidate the non-canonical interactions that drive this motor's function. We show that PfMyoA motor properties are tuned by heavy chain phosphorylation (Ser19), with unphosphorylated PfMyoA exhibiting enhanced ensemble force generation at the expense of speed. Regulated phosphorylation may therefore optimize PfMyoA for enhanced force generation during parasite invasion or for fast motility during dissemination. The three PfMyoA crystallographic structures presented here provide a blueprint for discovery of specific inhibitors designed to prevent parasite infection.

Marco Lucchino, Anne Billet, Antoine Versini, Harikrishna Bavireddi, Bhanu-Das Dasari, Sylvain Debieu, Ludovic Colombeau, Tatiana Cañeque, Alain Wagner, Géraldine Masson, Frédéric Taran, Philippe Karoyan, Muriel Delepierre, Christine Gaillet, Anne Houdusse, Sébastien Britton, Frédéric Schmidt, Jean-Claude Florent, Philippe Belmont, David Monchaud, Janine Cossy, Christophe Thomas, Arnaud Gautier, Ludger Johannes, Raphaël Rodriguez (2019 Feb 26)

2nd PSL Chemical Biology Symposium (2019): At the Crossroads of Chemistry and Biology.

Chembiochem : a European journal of chemical biology : 968-973 : DOI : 10.1002/cbic.201900092

Résumé

Chemical Biology is the science of designing chemical tools to dissect and manipulate biology at different scales. It provides the fertile ground from which to address important problems of our society, such as human health and environment.

Année de publication : 2018

Julien Robert-Paganin, Daniel Auguin, Anne Houdusse (2018 Oct 3)
Hypertrophic cardiomyopathy disease results from disparate impairments of cardiac myosin function and auto-inhibition.
Nature communications : 4019 : DOI : 10.1038/s41467-018-06191-4

Résumé

Hypertrophic cardiomyopathies (HCM) result from distinct single-point mutations in sarcomeric proteins that lead to muscle hypercontractility. While different models account for a pathological increase in the power output, clear understanding of the molecular basis of dysfunction in HCM is the mandatory next step to improve current treatments. Here, we present an optimized quasi-atomic model of the sequestered state of cardiac myosin coupled to X-ray crystallography and in silico analysis of the mechanical compliance of the lever arm, allowing the systematic study of a large set of HCM mutations and the definition of different mutation classes based on their effects on lever arm compliance, sequestered state stability, and motor functions. The present work reconciles previous models and explains how distinct HCM mutations can have disparate effects on the motor mechano-chemical parameters and yet lead to the same disease. The framework presented here can guide future investigations aiming at finding HCM treatments.

Alison G Tebo, Frederico M Pimenta, Martha Zoumpoulaki, Carlos Kikuti, Helena Sirkia, Marie-Aude Plamont, Anne Houdusse, Arnaud Gautier (2018 Aug 9)
Circularly Permuted Fluorogenic Proteins for the Design of Modular Biosensors.
ACS chemical biology : DOI : 10.1021/acschembio.8b00417

Résumé

Fluorescent reporters are essential components for the design of optical biosensors that are able to image intracellular analytes in living cells. Herein, we describe the development of circularly permuted variants of Fluorescence-Activating and absorption-Shifting Tag (FAST) and demonstrate their potential as reporting module in biosensors. Circularly permutated FAST (cpFAST) variants allow one to condition the binding and activation of a fluorogenic ligand (and thus fluorescence) to analyte recognition by coupling them with analyte-binding domains. We demonstrated their use for biosensor design by generating multicolor plug-and-play fluorogenic biosensors for imaging the intracellular levels of Ca in living mammalian cells in real time.

Léa Ripoll, Xavier Heiligenstein, Ilse Hurbain, Lia Domingues, Florent Figon, Karl J Petersen, Megan K Dennis, Anne Houdusse, Michael S Marks, Graça Raposo, Cédric Delevoye (2018 Jun 8)
Myosin VI and branched actin filaments mediate membrane constriction and fission of melanosomal tubule carriers.

The Journal of cell biology : 2709-2726 : DOI : 10.1083/jcb.201709055

Résumé

Vesicular and tubular transport intermediates regulate organellar cargo dynamics. Transport carrier release involves local and profound membrane remodeling before fission. Pinching the neck of a budding tubule or vesicle requires mechanical forces, likely exerted by the action of molecular motors on the cytoskeleton. Here, we show that myosin VI, together with branched actin filaments, constricts the membrane of tubular carriers that are then released from melanosomes, the pigment containing lysosome-related organelles of melanocytes. By combining superresolution fluorescence microscopy, correlative light and electron microscopy, and biochemical analyses, we find that myosin VI motor activity mediates severing by constricting the neck of the tubule at specific melanosomal subdomains. Pinching of the tubules involves the cooperation of the myosin adaptor optineurin and the activity of actin nucleation machineries, including the WASH and Arp2/3 complexes. The fission and release of these tubules allows for the export of components from melanosomes, such as the SNARE VAMP7, and promotes melanosome maturation and transfer to keratinocytes. Our data reveal a new myosin VI- and actin-dependent membrane fission mechanism required for organelle function.