## Phylogeny

– AGC kinase group → Rho-associated kinase (ROK) family → ROCK sub-family as mapped on the human kinome tree (feng2016rhokinase(rock) pages 1-5).  
– Closest paralogue: ROCK1; nearest AGC relatives: MRCK, DMPK and citron kinase (guan2013advancesinthe pages 1-2).  
– Conserved orthologs: Mus musculus Rock2, Danio rerio rock2a, Drosophila rok, Caenorhabditis elegans LET-502 (feng2016rhokinase(rock) pages 1-5).

## Reaction Catalyzed

ATP + protein-Ser/Thr → ADP + protein-O-phospho-Ser/Thr (kang2011peptidesubstratesfor pages 1-2).

## Cofactor Requirements

Catalytic turnover requires Mg²⁺ for ATP coordination (kang2011peptidesubstratesfor pages 4-4).

## Substrate Specificity

– High-throughput motif profiling: preference for basic residues at −3/−2; optimal consensus R/K-R/K-X-S/T (johnson2023anatlasof pages 4-5).  
– Classical biochemistry supports broader R/K-X-S/T or R/K-X-X-S/T recognition found on MYPT1, MLC and ERM proteins (liao2007rhokinase(rock) pages 1-2, hartmann2015thefunctionof pages 4-5).

## Structure

Domain organization  
1. N-terminal bilobed kinase domain, residues ~92–354, adopts active conformation without activation-loop phosphorylation (unknownauthors2023computationalexplorationof pages 9-11).  
2. Central ~600-aa coiled-coil dimerization segment containing the Rho-binding domain (RBD) (julian2014rhoassociatedcoiledcoilcontaining pages 1-3).  
3. C-terminal split PH domain interrupted by a cysteine-rich C1 domain; PH-C1 clamps the kinase in an autoinhibited state and binds PIP2/PIP3 uniquely in ROCK2 (hartmann2015thefunctionof pages 1-2).

3D structural features  
– Crystal structures of the kinase domain (PDB 2ETR, 6ED6) reveal ordered αC-helix, intact hydrophobic spine and Met172 gatekeeper; Y-27632 binds the ATP pocket via canonical hinge hydrogen bonds (johnson2023anatlasof pages 4-5, guan2013advancesinthe pages 10-10).

## Regulation

Autoinhibition and activation  
– PH-C1/kinase interaction keeps enzyme inactive; binding of GTP-loaded RhoA/B/C to the RBD relieves inhibition (sawada2014rhorhoassociatedcoiledcoilforming pages 3-4).

Post-translational modifications  
– PLK1 multisite phosphorylation enhances activity (hartmann2015thefunctionof pages 1-2).  
– PKA phosphorylates Ser1131, modulating function (feng2016rhokinase(rock) pages 1-5).  
– Autophosphorylation at Ser1366 marks the active enzyme (hartmann2015thefunctionof pages 1-2).  
– Tyr722 phosphorylation decreases RhoA affinity (hartmann2015thefunctionof pages 1-2).  
– Ubiquitination by SMURF1 drives proteasomal degradation (feng2016rhokinase(rock) pages 1-5).  
– Proteolytic cleavage by caspase-2/3 or granzyme B removes the C-terminus, generating constitutively active fragments (feng2016rhokinase(rock) pages 1-5, hartmann2015thefunctionof pages 1-2).

Additional regulators  
– Small GTP-binding proteins Gem and Rad bind adjacent to the RBD and inhibit kinase output (julian2014rhoassociatedcoiledcoilcontaining pages 3-4).  
– Circadian transcriptional control by BMAL1 drives daily oscillation of vascular ROCK2 levels (hartmann2015thefunctionof pages 4-5).

## Function

Expression  
– High mRNA/protein in brain, heart, skeletal and smooth muscle, lung and placenta; skeletal-muscle splice variant ROCK2m adds 57 aa (hartmann2015thefunctionof pages 1-2).

Upstream signals  
– Activation through Rho GTPases engaged by GPCR, RTK, cytokine and integrin pathways (narumiya2018rhosignalingresearch pages 7-10, sawada2014rhorhoassociatedcoiledcoilforming pages 3-4).

Principal substrates / downstream effects  
– MYL9 (MLC) Ser19 → Ca²⁺-sensitized smooth-muscle contraction (liao2007rhokinase(rock) pages 1-2).  
– MYPT1 Thr696/Thr853 → inhibition of myosin phosphatase (shah2016areviewon pages 1-2).  
– LIM kinases, ERM proteins, adducin, BRCA2, IRF4, VIM and others → stress-fiber formation, focal adhesion maturation, Th17 cytokine production, centrosome duplication (narumiya2018rhosignalingresearch pages 10-13, hartmann2015thefunctionof pages 4-5).

Physiological roles  
– Regulates vascular tone, cardiac hypertrophy/fibrosis, cytokinesis during development, keratinocyte differentiation, hippocampal spine morphology and circadian control of aortic contractility (hartmann2015thefunctionof pages 1-2, julian2014rhoassociatedcoiledcoilcontaining pages 4-5, narumiya2018rhosignalingresearch pages 10-13).

## Inhibitors

ATP-competitive active-site inhibitors  
– Y-27632, IC₅₀ ≈ 0.14 µM (narumiya2018rhosignalingresearch pages 10-13).  
– Fasudil, IC₅₀ ≈ 0.3 µM (feng2016rhokinase(rock) pages 1-5).  
– Ripasudil, nanomolar potency; approved for glaucoma (unknownauthors2023computationalexplorationof pages 2-5).  
– RKI-1447, sub-100 nM dual ROCK1/2 inhibitor (hobson2018identificationofselective pages 1-2).  
– KD025 (belumosudil), ROCK2-selective, cellular IC₅₀ ≈ 0.058 µM, suppresses Th17 signalling (narumiya2018rhosignalingresearch pages 10-13).  
– SLx-2119, enzymatic IC₅₀ = 51 nM with high kinome selectivity (shah2016areviewon pages 2-4).

## Other Comments

Hyper-activation or over-expression links ROCK2 to systemic and pulmonary hypertension, heart failure, psoriasis, rheumatoid arthritis, graft-versus-host disease, inflammatory bowel disease, glaucoma and metastatic cancer (hartmann2015thefunctionof pages 1-2, narumiya2018rhosignalingresearch pages 10-13, watanabe2024thespecificrock2 pages 1-2).

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