## Phylogeny

CHK1 orthologs are present from the fission yeast Schizosaccharomyces pombe to Caenorhabditis elegans, Xenopus laevis and Drosophila melanogaster, indicative of deep evolutionary conservation of the checkpoint kinase branch (gatei2003ataxiatelangiectasiamutated(atm)and pages 6-7).  
Manning’s kinome classification places CHK1 in the CMGC group within the Chk1/Chk2 family (matthews2013structurebaseddesigndiscovery pages 19-20), whereas an alternative analysis assigns it to the CAMK group, highlighting a classification discrepancy (gorecki2021clinicalcandidatestargeting pages 21-22).  
CHK1 and the ATM-responsive kinase CHK2 form a distinct checkpoint kinase lineage that maintains the canonical Ser/Thr catalytic core but differs in upstream activation mechanisms (dent2019investigationalchk1inhibitors pages 1-3, matthews2013structurebaseddesigndiscovery pages 1-2).

## Reaction Catalyzed

ATP + [protein]-Ser/Thr → ADP + [protein]-O-phospho-Ser/Thr (chen2000implicationsforchk1 pages 8-9, matthews2013structurebaseddesigndiscovery pages 17-19).

## Cofactor Requirements

Catalytic activity requires divalent metal ions, principally Mg²⁺, which coordinate the ATP phosphates in the active site (gorecki2021clinicalcandidatestargeting pages 21-22, matthews2013structurebaseddesigndiscovery pages 17-19).

## Substrate Specificity

CHK1 preferentially phosphorylates the consensus motif R-X-X-S/T (chen2000implicationsforchk1 pages 8-9, matthews2013structurebaseddesigndiscovery pages 17-19).  
Phosphoproteomic profiling shows preference for a basic or hydrophobic residue immediately N-terminal to the phosphoacceptor and selectivity for serine over threonine with additional sequence bias at the +1 and +3 positions (gorecki2021clinicalcandidatestargeting pages 21-22, matthews2013structurebaseddesigndiscovery pages 17-19).

## Structure

CHK1 consists of an N-terminal bilobal kinase domain (~residues 1-289) followed by a C-terminal KA1 regulatory domain that modulates kinase activity (matthews2013structurebaseddesigndiscovery pages 19-20, gorecki2021clinicalcandidatestargeting pages 21-22).  
A 1.7 Å crystal structure reveals the kinase domain in an open lobe conformation with an ordered activation loop in the absence of phosphorylation (chen2000implicationsforchk1 pages 8-9).  
The catalytic core contains the conserved DFG motif at the start of the activation segment and the HRD motif in the catalytic loop, positioning the catalytic aspartate for phosphotransfer (gorecki2021clinicalcandidatestargeting pages 21-22, matthews2013structurebaseddesigndiscovery pages 19-20).  
Hinge residues Glu85 and Cys87 form key hydrogen bonds with ATP and most inhibitors, while Asn59 and Leu84 create a buried pocket exploited for selective inhibitor binding (chen2000implicationsforchk1 pages 8-9, matthews2013structurebaseddesigndiscovery pages 14-16).  
Deletion of the C-terminal segment elevates catalytic activity ~20-fold, demonstrating an autoinhibitory mechanism mediated by intramolecular contacts (chen2000implicationsforchk1 pages 8-9).

## Regulation

ATR phosphorylates Ser317 and Ser345, generating full activation during replication stress (gorecki2021clinicalcandidatestargeting pages 21-22, goto2015novelinsightsinto pages 1-2).  
Autophosphorylation at Ser296 augments catalytic efficiency (gorecki2021clinicalcandidatestargeting pages 21-22, matthews2013structurebaseddesigndiscovery pages 19-20).  
AKT targets Ser280, modulating subcellular localization (gorecki2021clinicalcandidatestargeting pages 21-22).  
ATM together with NBS1 also phosphorylates Ser317 in response to ionizing radiation (gatei2003ataxiatelangiectasiamutated(atm)and pages 6-7).  
Activated CHK1 is ubiquitinated by SCF^β-TrCP, promoting proteasomal degradation, whereas PP2A removes activating phosphates to restore basal activity (gorecki2021clinicalcandidatestargeting pages 21-22, dent2019investigationalchk1inhibitors pages 1-3).  
Phosphorylation-induced conformational changes and interaction between the kinase domain and C-terminal KA1 domain mediate allosteric control (matthews2013structurebaseddesigndiscovery pages 19-20, chen2000implicationsforchk1 pages 8-9).

## Function

CHK1 is expressed ubiquitously with highest levels in proliferative tissues and exhibits cell cycle-dependent abundance (gorecki2021clinicalcandidatestargeting pages 21-22).  
ATR recruits CHK1 to RPA-coated single-stranded DNA via the adaptor CLASPIN during replication stress (gorecki2021clinicalcandidatestargeting pages 2-5).  
CHK1 phosphorylates CDC25A/B/C, leading to their degradation or sequestration and consequent inhibition of CDK/cyclin complexes to enforce intra-S and G2/M checkpoints (dent2019investigationalchk1inhibitors pages 1-3, gorecki2021clinicalcandidatestargeting pages 2-5).  
Additional substrates include WEE1, MYT1, treslin, DBF4, MCM complex components and FANCD2, coordinating origin firing, fork stability and homologous recombination repair (gorecki2021clinicalcandidatestargeting pages 2-5, matthews2013structurebaseddesigndiscovery pages 19-20).  
CHK1 also contributes to chromatin assembly and mitotic spindle regulation through phosphorylation of Aurora B kinase (gorecki2021clinicalcandidatestargeting pages 2-5).  
These activities collectively maintain genome integrity under both stressed and unperturbed conditions (dent2019investigationalchk1inhibitors pages 1-3).

## Inhibitors

ATP-competitive inhibitors with low-nanomolar potency include Prexasertib, Rabusertib, AZD7762, SRA737, UCN-01 and CCT241533 (gorecki2021clinicalcandidatestargeting pages 21-22, matthews2013structurebaseddesigndiscovery pages 17-19).  
These molecules anchor to Glu85 and Cys87 in the hinge, extend into the ribose pocket and exploit the Asn59-centred water network for selectivity (matthews2013structurebaseddesigndiscovery pages 2-4, matthews2013structurebaseddesigndiscovery pages 14-16).  
First-generation UCN-01 displayed suboptimal pharmacokinetics, whereas later oral series such as imidazo[1,2-a]pyrazines and thioquinazolinones achieved improved selectivity and bioavailability (dent2019investigationalchk1inhibitors pages 1-3, matthews2013structurebaseddesigndiscovery pages 14-16).  
No CHK1 inhibitor has yet received regulatory approval (dent2019investigationalchk1inhibitors pages 1-3).

## Other Comments

CHK1 is frequently over-expressed in cancers and is essential for survival of tumors with high replication stress or TP53 deficiency, supporting synthetic-lethality approaches (matthews2013structurebaseddesigndiscovery pages 19-20, gorecki2021clinicalcandidatestargeting pages 21-22).  
Germline CHEK1 variants such as E85K and C87R are linked to developmental disorders including microcephaly, underscoring the kinase’s role in genome maintenance during development (gorecki2021clinicalcandidatestargeting pages 21-22, zhang2019understandingtheactivation pages 32-36).  
CHEK1 amplification or overexpression contributes to chemoresistance across several tumor types (dent2019investigationalchk1inhibitors pages 1-3).

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