## Phylogeny

Serine/threonine-protein kinase Chk2 is a member of the CMGC kinase group and is the mammalian ortholog of the yeast checkpoint kinases Rad53 (S. cerevisiae) and Cds1 (S. pombe). Orthologs are found in all mammals and functionally analogous proteins exist in more distant eukaryotes, indicating an early evolutionary origin and strong conservation of the DNA-damage checkpoint from yeast to humans (Buscemi et al., 2014; Nevanlinna & Bartek, 2006).

## Reaction Catalyzed

ATP + [protein]-OH ⇌ ADP + [protein]-O-phosphate + H⁺ (Li & Stern, 2005)

## Cofactor Requirements

Mg²⁺ is required for catalytic activity (Seo et al., 2003; Li & Stern, 2005).

## Substrate Specificity

Chk2 preferentially phosphorylates serine or threonine residues within the motif L-X-R-X-X-S/T (Leu at –5, Arg at –3). Hydrophobic and basic residues upstream of the phospho-acceptor enhance recognition. Confirmed cellular substrates include CDC25A/B/C, p53, BRCA1/2 and other DNA-damage response factors (Buscemi et al., 2014; Kim et al., 2007; Seo et al., 2003).

## Structure

The 543-residue protein contains:  
• N-terminal SQ/TQ cluster domain (SCD) phosphorylated by ATM  
• Central forkhead-associated (FHA) phosphopeptide-binding domain that mediates dimerization and substrate docking  
• C-terminal bilobal kinase domain with an activation loop carrying T383 and T387.  
Crystallographic and AlphaFold models confirm the canonical kinase fold and show the FHA domain positioned to facilitate activation-dependent dimerization and autophosphorylation (Buscemi et al., 2014; Stolarova et al., 2020; Wu et al., 2006).

## Regulation

DNA double-strand breaks trigger ATM-dependent phosphorylation of Chk2 at T68, inducing FHA/SCD-mediated homodimerization. Dimerization enables autophosphorylation at T383, T387 and S516, producing full kinase activity. Activity is attenuated by ubiquitination-driven proteolysis and by dephosphorylation through PP2A or WIP1. Nuclear import can be modulated by karyopherin-α interactions (Li & Stern, 2005; Wu et al., 2006; Stolarova et al., 2020).

## Function

Activated Chk2 enforces cell-cycle arrest at the G1/S and G2/M checkpoints and promotes DNA repair and apoptosis. Key actions include:  
• Phosphorylation-dependent inhibition of CDC25 phosphatases, preventing CDK activation.  
• Phosphorylation of p53 (Ser20) leading to p53 stabilization and transcriptional activation of repair and apoptotic genes.  
• Modulation of homologous recombination via BRCA2 phosphorylation and RAD51 recruitment.  
Non-canonical roles include regulation of mitotic spindle assembly and autophagy under oxidative stress. Collectively, Chk2 acts as a tumor suppressor safeguarding genomic integrity (Bell et al., 2007; Buscemi et al., 2014; Smith et al., 2020).

## Inhibitors

Several small-molecule inhibitors (e.g., PV1019, VRX0466617, bis-guanylhydrazone derivatives) bind the ATP site of Chk2, but many also inhibit the related kinase Chk1 (Lountos et al., 2011; Stolz et al., 2011).

## Other Comments

Germ-line and somatic CHEK2 variants such as 1100delC and I157T are linked to elevated risks of breast, prostate and colorectal cancers. Loss or attenuation of Chk2 function contributes to chromosomal instability and therapy resistance, making the kinase a potential target for sensitizing tumors to DNA-damaging agents (Stolarova et al., 2020; Bell et al., 2007; Chrisanthar et al., 2008).

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