## Phylogeny

Human WEE1 belongs to the Wee1 family within the “Other” group of the kinome according to Manning et al. (Fu et al., 2018; Hamer et al., 2011; Esposito et al., 2021). Other reports place it in either the tyrosine-kinase-like (TKL) or CMGC groups (Esposito et al., 2021; Geenen & Schellens, 2017; Moiseeva et al., 2019). Although WEE1 phosphorylates tyrosine, its catalytic domain is serine/threonine-kinase-like, implying evolutionary divergence from that lineage (Esposito et al., 2021; Hamer et al., 2011). Orthologs are conserved across eukaryotes, including *Schizosaccharomyces pombe* (wee1) and *Saccharomyces cerevisiae* (SWE1) (Fu et al., 2018; Hamer et al., 2011). Human paralogs include PKMYT1 and WEE1B; the related kinase MIK1 is found in fission yeast (Fu et al., 2018; Rorà et al., 2020; Esposito et al., 2021).

## Reaction Catalyzed

ATP + Cyclin B1–CDK1 → ADP + Cyclin B1–CDK1-[pTyr15] (Esposito et al., 2021; Moiseeva et al., 2019).  
Overall: ATP-dependent transfer of the γ-phosphate to a substrate tyrosine residue (Esposito et al., 2021; Moiseeva et al., 2019).

## Cofactor Requirements

Mg²⁺ is essential for catalysis, coordinating ATP in the active site (Elbæk et al., 2020; Esposito et al., 2021; Moiseeva et al., 2019).

## Substrate Specificity

• Principal targets: CDK1 and CDK2 on Tyr15; some evidence for CDK1 Thr14 (Do et al., 2013; Fu et al., 2018).  
• Additional substrate: histone H2B Tyr37 (Fu et al., 2018; Esposito et al., 2021).  
• Peptide-array profiling groups WEE1 into a “Non-canonical (WEE)” cluster with atypical sequence preferences; a position-specific scoring matrix spanning −5 to +5 around the phosphotyrosine was reported (Yaron-Barir et al., 2024).

## Structure

WEE1 comprises an N-terminal regulatory region (phospho-degrons Ser53, Ser123; nuclear-localization signal; three PEST motifs), a central kinase domain with canonical C-helix and activation loop, and a short C-terminal region containing Ser642 (14-3-3 docking site) (Esposito et al., 2021; Elbæk et al., 2020; Moiseeva et al., 2019). Crystal structures of the kinase domain include PDB IDs 1JXD, 1X8B, 3BI6, 4FX3-5, and 6O6E (Elbæk et al., 2020; Moiseeva et al., 2019; Geenen & Schellens, 2017). High-confidence AlphaFold models supplement experimental data (Elbæk et al., 2020; Esposito et al., 2021).

## Regulation

Transcriptional control: repressed by KLF2 and miR-195 (Elbæk et al., 2020).  
Post-translational modifications  
• Phosphorylation:  
 – Ser53/Ser123 by CDK1 and PLK1 triggers degradation (Esposito et al., 2021; Hamer et al., 2011; Rorà et al., 2020).  
 – DNA-damage-induced CHK1 phosphorylation enhances activity (Fu et al., 2018).  
 – AKT phosphorylates Ser642, promoting 14-3-3 binding and cytoplasmic sequestration (Fu et al., 2018).  
• Ubiquitination: SCFβ-TrCP1/2 and SCF-Tome-1 ligases target phosphorylated WEE1 for proteasomal degradation (Elbæk et al., 2020; Rorà et al., 2020; Esposito et al., 2021).  
Protein interactions: HSP90 and MIG6 stabilize WEE1; 14-3-3 binding to pSer642 increases stability and interphase activity (Elbæk et al., 2020; Esposito et al., 2021; Fu et al., 2018).

## Function

Key nuclear kinase that enforces G2/M and S-phase checkpoints (Esposito et al., 2021; Elbæk et al., 2020).  
Upstream signaling: ATM/ATR activate CHK1, which in turn activates WEE1 (Geenen & Schellens, 2017).  
Downstream targets: CDK1 and CDK2 are inhibited via Tyr15 phosphorylation; CDC25 phosphatases reverse this inhibition (Do et al., 2013; Rorà et al., 2020; Fu et al., 2018).  
Pathway roles:  
• G2 DNA-damage checkpoint—suppresses Cyclin B1–CDK1 to allow repair (Esposito et al., 2021; Hamer et al., 2011).  
• S-phase—modulates CDK2 to stabilize replication forks (Esposito et al., 2021; Rorà et al., 2020).  
• Epigenetic—phosphorylation of histone H2B Tyr37 represses histone gene transcription (Fu et al., 2018; Esposito et al., 2021).

## Inhibitors

• Adavosertib (AZD1775/MK-1775): potent, selective ATP-competitive inhibitor; abrogates G2 checkpoint and induces mitotic catastrophe, especially in p53-deficient cancers (Geenen & Schellens, 2017; Hamer et al., 2011).  
• PROTACs: under investigation for targeted WEE1 degradation (Elbæk et al., 2020).

## Other Comments

WEE1 dysregulation is frequent in glioblastoma, ovarian, colorectal, and breast cancers and generally associates with poor prognosis; some studies also link down-regulation to adverse outcomes (Fu et al., 2018; Geenen & Schellens, 2017; Esposito et al., 2021). p53-mutant tumors rely heavily on the WEE1-mediated G2 checkpoint, creating a therapeutic vulnerability (Elbæk et al., 2020; Geenen & Schellens, 2017). Mouse WEE1 knockout causes embryonic lethality, underscoring its essential role (Esposito et al., 2021).

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