## Phylogeny

Member of the AGC protein-kinase group, S6K sub-family. It is the vertebrate paralogue of RPS6KB1/S6K1, sharing ~83 % amino-acid identity in the catalytic core (Khalil et al., 2024, pp. 7–9). Orthologues occur from yeast to mammals (Magnuson et al., 2012, pp. 1–2; Malanchuk et al., 2024, pp. 1–2). Like other AGC kinases (PKC, Akt, RSK, SGK), activity depends on a C-terminal hydrophobic-motif phosphorylation mechanism (Unknown authors, 2016, pp. 36–41).

## Reaction Catalyzed

ATP + [protein]-Ser/Thr ⇌ ADP + [protein]-O-phospho-Ser/Thr (Magnuson et al., 2012, pp. 1–2).

## Cofactor Requirements

Requires Mg²⁺ (Mn²⁺ can substitute) coordinated with ATP in the active site (Fenton & Gout, 2011, pp. 1–2).

## Substrate Specificity

• Prefers the basic consensus sequence R/K-X-R/K-X-X-S/T (Unknown authors, 2016, pp. 25–30).  
• Major target: ribosomal protein S6 at Ser235/236 and Ser240/244 (Pende & Treins, 2014, pp. 5–7).  
• Additional validated substrates: eIF4B, eEF2K and PDCD4 (Pende & Treins, 2014, pp. 5–7).  
• Phosphorylates TRBP to influence miRNA biogenesis and augments YY1 expression downstream of its kinase activity (Khalil et al., 2024, pp. 7–9).

## Structure

N-terminal nuclear-localisation signal, bilobal kinase domain, short kinase extension, and C-terminal regulatory tail harbouring the hydrophobic-motif Thr388 plus an autoinhibitory pseudosubstrate (Unknown authors, 2016, pp. 25–30). The AlphaFold model AF-Q9UBS0-F1 adopts the canonical AGC fold with catalytic Lys100, DFG motif (Asp215-Phe216-Gly217) and activation-loop Thr228 (Khalil et al., 2024, pp. 9–11). Phosphorylation of Thr388 stabilises the αC helix and creates a PDK1-docking site, mirroring the S6K1 crystal structure 3A60 (Sunami et al., 2010, pp. 1–2). Activation-loop phosphorylation on Thr228 aligns the catalytic and regulatory spines for full activity (Khalil et al., 2024, pp. 7–9). A unique hinge Cys150 (Tyr in S6K1) enables covalent inhibitor design (Gerstenecker et al., 2021, pp. 7–10).

## Regulation

• Sequential phosphorylation cascade: Ser370 priming → mTORC1 on Thr388 → PDK1 on Thr228 (Khalil et al., 2024, pp. 7–9).  
• ERK pathway targets Ser410/417/423 to relieve autoinhibition (Khalil et al., 2024, pp. 7–9).  
• PKC phosphorylates Ser486, masking the NLS and retaining the kinase in the cytoplasm (Khalil et al., 2024, pp. 9–11).  
• Fyn phosphorylates Tyr45, linking receptor tyrosine kinase signals (Khalil et al., 2024, pp. 9–11).  
• PRMT-mediated arginine methylation of C-terminal RXR motifs promotes nuclear localisation and survival signalling (Khalil et al., 2024, pp. 9–11).  
• Lysine acetylation by p300/PCAF stabilises the protein; HDACs and sirtuins reverse this mark (Khalil et al., 2024, pp. 9–11).  
• Polyubiquitination directs degradation (Fenton & Gout, 2011, p. 13).  
• PP2A removes activating phosphates, while PTEN and TSC1/2 restrain upstream PI3K–mTOR signalling (Unknown authors, 2016, pp. 36–41).

## Function

Predominantly nuclear owing to an intrinsic NLS; p54 and p56 isoforms accumulate in the nucleus (Unknown authors, 2016, pp. 25–30). Acts downstream of mTORC1 to drive ribosome biogenesis and protein synthesis via rpS6 phosphorylation, integrating PI3K/AKT, ERK and PKC inputs (Magnuson et al., 2012, pp. 1–2). Enhances translation initiation (eIF4B phosphorylation, PDCD4 degradation) and elongation (inhibitory phosphorylation of eEF2K) (Pende & Treins, 2014, pp. 5–7). Regulates miRNA processing through TRBP and increases YY1 levels, linking activity to transcriptional programmes (Khalil et al., 2024, pp. 7–9). Provides negative feedback to PI3K signalling (Fenton & Gout, 2011, pp. 1–2). Gene amplification or high activity supports survival, migration and chemoresistance in several cancers, including small-cell lung cancer and NRAS-mutant melanoma (Khalil et al., 2024, pp. 24–25). Combined S6K1/S6K2 knockout causes perinatal lethality, underscoring developmental importance (Khalil et al., 2024, pp. 11–12).

## Inhibitors

• LY2584702 – ATP-competitive pan-p70S6K inhibitor with limited clinical benefit (Khalil et al., 2024, pp. 11–12).  
• PF-4708671 – S6K1-biased tool compound exhibiting partial S6K2 inhibition (Khalil et al., 2024, pp. 11–12).  
• “Compound 2” – covalent inhibitor targeting Cys150, highly potent and S6K2-selective (Gerstenecker et al., 2021, pp. 7–10).  
• Rapamycin/rapalogues – block mTORC1, preventing Thr388 phosphorylation and indirectly suppressing S6K2 activity (Magnuson et al., 2012, pp. 1–2).

## Other Comments

Over-expression or hyperactivation correlates with poor prognosis and therapy resistance in multiple tumour types (Khalil et al., 2024, pp. 24–25). PRMT-dependent methylation-driven nuclear localisation underlies chemoresistance in small-cell lung cancer (Khalil et al., 2024, pp. 9–11). Enhanced rpS6 phosphorylation downstream of S6Ks contributes to neurodevelopmental phenotypes in PPP2R5D variant disorders (Unknown authors, 2024, pp. 127–130).

## 9. References

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