1. Phylogeny  
   RPS6KC1, also known as RSKL1 or Ribosomal protein S6 kinase delta‐1, is classified within the p90 ribosomal S6 kinase family, a subgroup of the AGC superfamily of serine/threonine protein kinases. Comparative genomic analyses have demonstrated that members of the p90 RSK family are widely conserved across eukaryotes, with orthologs identifiable in organisms ranging from yeast to mammals. In the context of the broader kinome, RPS6KC1 shares notable sequence homology with the other RSK isoforms (RSK1–RSK4) as well as with related AGC kinases such as PDK1 and AKT, thereby forming part of an evolutionary core of signaling molecules that emerged in the common ancestor of all eukaryotes (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, arencibia2013agcproteinkinases pages 3-4, pearce2010thenutsand pages 1-2). In molecular phylogenetic studies, the RSK family is distinguished by its dual-domain architecture—a hallmark that separates it both from the classical p70 S6 kinases and from other AGC kinases. Gene duplication events in the early evolutionary history of animals and fungi have given rise to multiple paralogs within this family. RPS6KC1 thus represents one branch of these duplication events, which has further diversified to assume roles in cellular signal transduction and substrate phosphorylation. Its membership in the AGC family places it together with kinases regulating fundamental cellular processes such as protein synthesis, cell growth, metabolism, and stress responses. The evolutionary conservation of its domain organization—including an N‐terminal kinase domain typical of the AGC group and a C‐terminal kinase domain aligned with CaMK family characteristics—underscores the evolutionary pressure to maintain its function in key signal transduction pathways (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, arencibia2013agcproteinkinases pages 3-4, pearce2010thenutsand pages 1-2). Moreover, the presence of a phox homology (PX) domain, which is conserved in several members of the AGC kinase family, indicates an ancient mechanism for membrane association and phosphoinositide binding that has been preserved throughout evolution. These features collectively situate RPS6KC1 as an evolutionarily ancient kinase that has retained core elements essential for its role in transmitting intracellular signals.
2. Reaction Catalyzed  
   RPS6KC1 catalyzes a phosphorylation reaction that involves the transfer of the γ-phosphate group from ATP to specific serine or threonine residues on target protein substrates. The generalized chemical reaction performed by this enzyme follows the classic kinase mechanism: ATP + [protein]-(L-serine or L-threonine) yields ADP + [protein]-(L-serine/threonine)-phosphate + H⁺. This reaction, which is a defining feature of protein kinases in the AGC family, underpins its ability to modulate the function of target proteins involved in diverse signaling cascades (magnuson2012regulationandfunction pages 2-3).
3. Cofactor Requirements  
   The catalytic activity of RPS6KC1 depends critically on cofactors that enable the transfer of the phosphate moiety from ATP to the substrate. In common with other serine/threonine protein kinases of the AGC family, RPS6KC1 requires magnesium ions (Mg²⁺) as a cofactor. Mg²⁺ facilitates ATP binding in the catalytic cleft and stabilizes the transition state during phosphoryl transfer, working in concert with ATP to drive efficient catalysis (magnuson2012regulationandfunction pages 2-3).
4. Substrate Specificity  
   The substrate specificity of RPS6KC1 is presumed to align closely with that of other AGC kinases, particularly those belonging to the ribosomal S6 kinase family. Although direct experimental determination of a specific linear consensus motif unique to RPS6KC1 is sparse, available evidence indicates that the enzyme preferentially phosphorylates serine/threonine residues when these residues are flanked by basic amino acids. In particular, studies on related kinases have demonstrated a propensity for substrates to exhibit arginine residues at positions −3 and −5 relative to the phosphorylation site. This pattern is consistent with a canonical ‘RxRxxp[ST]’ motif observed in several S6 kinases (magnuson2012regulationandfunction pages 10-11, pearce2010thenutsand pages 1-2). Integrated phosphoproteomic analyses have further identified phosphorylated peptide sequences derived from RPS6KC1 substrates that conform to these criteria, reinforcing the notion that the enzyme displays substrate specificity for serine and threonine residues embedded within arginine-rich contexts (karamafrooz2021integratedphosphoproteomicsfor pages 30-31). As such, substrates involved in signaling pathways related to protein synthesis and cellular growth are likely to feature these consensus motifs, a characteristic that reflects the evolutionary conservation of substrate recognition among AGC kinases.
5. Structure  
   RPS6KC1 exhibits a modular domain organization that is characteristic of several p90 ribosomal S6 kinase family members. Its overall structure comprises two distinct catalytic domains arranged in tandem. The N-terminal kinase domain (NTKD) belongs to the AGC kinase family and houses the structural features common to serine/threonine kinases, such as the bilobed architecture with a smaller N-terminal lobe primarily composed of β-sheets and a larger C-terminal lobe rich in α-helices. This NTKD is responsible for substrate recognition and catalysis in many AGC kinases. In contrast, the C-terminal kinase domain (CTKD) shows similarities to members of the CaMK (calcium/calmodulin-dependent protein kinase) family and is thought to participate in autophosphorylation events that can modulate the activity of the NTKD (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, arencibia2013agcproteinkinases pages 3-4).

A unique structural attribute of RPS6KC1 is the presence of a phox homology (PX) domain. The PX domain is known to mediate interactions with phosphatidylinositol 3-phosphate (PI3P) and is involved in membrane targeting. In RPS6KC1, this domain facilitates the binding to early endosomal membranes, thereby localizing the protein to sites where it can modulate signaling pathways that rely on endosome-associated phosphoinositides (thiriet2013cytoplasmicproteinserinethreonine pages 60-63).

Notably, despite its classification among the ribosomal S6 kinase family, supplementary data curated from kinase motif analyses indicate that RPS6KC1 has not been reported to exhibit phosphorylation at its canonical activation segment, turn motif, or hydrophobic motif—sites that, in many AGC kinases, are critical for achieving full catalytic activity. Data from a comprehensive survey of kinase phosphorylation sites revealed that RSKL1 (an alternative name for RPS6KC1) lacks annotated phosphorylation events in these key regulatory regions (pearce2010thenutsand pages 1-2). This observation has led to proposals that RPS6KC1 might function, at least in part, as a pseudokinase. Indeed, some studies have classified RSKL1 as belonging to a subclass of pseudokinases within the RSK family due to the absence of essential catalytic motifs in one of its kinase domains; such pseudokinases are thought to operate primarily as molecular scaffolds or allosteric regulators rather than as robust catalytic enzymes (thiriet2013cytoplasmicproteinserinethreonine pages 63-66).

Structural models derived from comparative analyses and predictive algorithms such as AlphaFold support the overall domain architecture described above. These models reveal conservation of the typical kinase fold within the NTKD, including features such as the P-loop involved in ATP binding, the catalytic loop with invariant aspartate residue necessary for phosphotransfer, and the C-helix, which is critical for aligning catalytic residues. In the CTKD, while a similar catalytic core can be discerned, deviations in amino acid sequences within regions corresponding to the activation loop or hydrophobic motif of other AGC kinases may account for its atypical regulatory properties. Additionally, the region linking the two kinase domains may harbor sites for autophosphorylation or binding of regulatory proteins, although high-resolution structural data for this inter-domain region remain limited. The overall quaternary structure has not been definitively established by crystallography; however, the domain organization suggests that intramolecular interactions between the NTKD and CTKD, as well as the PX domain’s engagement with membrane lipids, are likely to be central to the protein’s regulation and spatial distribution within the cell (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, arencibia2013agcproteinkinases pages 8-9, pearce2010thenutsand pages 1-2).

1. Regulation  
   Regulatory mechanisms governing RPS6KC1 function encompass a variety of post‐translational modifications and protein–protein interactions that are a recurring theme within the AGC kinase family. Phosphorylation is the primary post‐translational modification modulating the activity of RPS6KC1. Although, in contrast to many of its family members, the critical regulatory phosphorylation sites located within the activation segment, turn motif, and hydrophobic motif have not been conclusively reported for RPS6KC1, multiple integrated phosphoproteomic experiments have detected phosphorylation events on this protein, indicating that it does undergo regulatory modifications under certain cellular conditions (pearce2010thenutsand pages 1-2, karamafrooz2021integratedphosphoproteomicsfor pages 31-32).

The upstream signaling cascades that typically target p90 ribosomal S6 kinases include the MAPK/ERK pathway. In canonical RSK isoforms such as RSK1–RSK3, phosphorylation by ERK1/2 is indispensable for initiating activation. However, RPS6KC1 has been reported in some studies to exhibit constitutive activity and to function with a reduced dependency on upstream activators such as PDK1. Despite this, physical interaction with PDK1 has been observed in certain experimental systems, suggesting that RPS6KC1 can be integrated into traditional kinase activation loops even though its catalytic domain may lack certain conserved phosphorylation events (thiriet2013cytoplasmicproteinserinethreonine pages 60-63, arencibia2013agcproteinkinases pages 8-9).

In addition to ERK-mediated phosphorylation, integrated phosphoproteomics studies have identified specific phosphopeptides mapping to RPS6KC1 in contexts where protein kinase A (PKA) signaling is active. For instance, analyses conducted using LC–MS/MS identified phosphorylation sites situated within regions that may influence the enzyme’s scaffolding functions or its subcellular localization. These phosphorylation events, as detected by advanced quantitative techniques, underscore the flexibility and context-dependent regulation of RPS6KC1 (karamafrooz2021integratedphosphoproteomicsfor pages 30-31, pages 31-32).

Another layer of regulation may derive from the potential pseudokinase attributes of RPS6KC1. Studies that have classified RSKL1 as a pseudokinase highlight that the absence of phosphorylation at the conventional activation sites may redirect regulatory inputs from catalytic modulation to mechanisms based on protein–protein interactions. In such a scenario, RPS6KC1 may function primarily as a molecular scaffold that organizes and localizes other signaling molecules at specific subcellular compartments, such as early endosomes. The PX domain, for example, not only facilitates binding to phosphatidylinositol 3-phosphate but also may mediate association with proteins such as sphingosine kinase‐1 and peroxiredoxin 3, thereby linking lipid signaling with redox regulatory processes (thiriet2013cytoplasmicproteinserinethreonine pages 60-63, thiriet2013cytoplasmicproteinserinethreonine pages 63-66).

Collectively, the regulation of RPS6KC1 appears to involve a combination of constitutive phosphorylation, possible autophosphorylation, and stimulus‐dependent phosphorylation events that enhance or modulate its localization and interaction with downstream effectors. In contrast to many AGC kinases that require robust and coordinated phosphorylation by upstream enzymes such as mTORC1 and PDK1 for activation, RPS6KC1 exhibits features that suggest a more complex or even alternative regulatory paradigm. This may include the stabilization of its conformation by intramolecular interactions and the reliance on its non‐catalytic domains for functional regulation. Each of these regulatory layers contributes to the precise control of RPS6KC1 activity and underscores the need for further detailed biochemical analysis to elucidate the complete regulatory network governing its function (pearce2010thenutsand pages 1-2, thiriet2013cytoplasmicproteinserinethreonine pages 60-63, arencibia2013agcproteinkinases pages 8-9).

1. Function  
   RPS6KC1 is implicated in the transmission of sphingosine‐1‐phosphate (S1P)–mediated signals into the cell. Through its ability to bind to specific lipid molecules and phosphoinositides, RPS6KC1 is positioned to function at the nexus of lipid signaling and intracellular trafficking. One of the key functional roles attributed to RPS6KC1 is its involvement in the recruitment of the antioxidant enzyme peroxiredoxin 3 (PRDX3) to early endosomes. This recruitment is mediated, at least in part, by the interaction between its PX domain and phosphatidylinositol 3‐phosphate, which anchors the protein to endosomal membranes where PRDX3 can be efficiently recruited (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, thiriet2013cytoplasmicproteinserinethreonine pages 60-63).

In addition to its role in PRDX3 recruitment, RPS6KC1 has been linked to the regulation of sphingosine kinase‐1 (SPHK1) signaling. By binding to SPHK1, RPS6KC1 may influence the generation of sphingosine-1‐phosphate, a lipid mediator that plays pivotal roles in cell growth, survival, migration, and inflammatory responses. Although the detailed mechanisms through which RPS6KC1 modulates SPHK1 activity remain to be fully elucidated, its capacity to interact with both SPHK1 and components of the endosomal membrane suggests that it contributes to the spatial organization of S1P signaling events within the cell (thiriet2013cytoplasmicproteinserinethreonine pages 57-60, arencibia2013agcproteinkinases pages 18-18).

Furthermore, RPS6KC1 is a member of the broader p90 ribosomal S6 kinase family, and by analogy with its better-characterized relatives, it may have secondary functions related to the regulation of protein synthesis and cell growth. Although its direct phosphorylation targets have not been as comprehensively identified as those of classical S6 kinases, the structural similarities to other AGC kinases and the conserved dual-domain architecture support a role in modulating downstream effectors that control aspects of mRNA translation and ribosomal biogenesis. Such mechanisms are consistent with the established roles of related kinases in integrating external growth factor signals with the translational machinery (magnuson2012regulationandfunction pages 10-11).

Expression analyses, as reported in studies of related kinases, indicate that members of the RSK family exhibit tissue-specific expression patterns. Although detailed expression profiles for RPS6KC1 are less well documented, the ability of its regulatory domains to engage with endosomal and membrane components suggests that its function may be particularly relevant in cell types where sphingolipid signaling and redox regulation are critical. This is in keeping with the idea that intracellular localization, directed by the PX domain, contributes to the specificity of signal transduction events mediated by RPS6KC1 (thiriet2013cytoplasmicproteinserinethreonine pages 60-63, arencibia2013agcproteinkinases pages 18-18).

Taken together, RPS6KC1 appears to function as an important mediator of lipid signaling pathways by facilitating the transmission of S1P signals and by recruiting key redox-regulatory proteins such as PRDX3 to early endosomal compartments. These activities position the protein at a crucial intersection of signaling pathways that govern cellular responses to growth factors and stress, consistent with the multifunctionality observed for other members of the p90 ribosomal S6 kinase family (magnuson2012regulationandfunction pages 10-11, arencibia2013agcproteinkinases pages 18-18).

1. Other Comments  
   While specific inhibitors targeting RPS6KC1 have not been exclusively characterized, research into selective modulation of AGC kinases has yielded compounds that affect kinases with similar domain architectures. The comprehensive work on AGC kinase inhibitors emphasizes the need for tools that can distinguish between catalytic and non-catalytic functions, particularly in proteins that may function as pseudokinases. In the case of RPS6KC1, the lack of reported phosphorylation at key activation motifs (as noted in supplementary kinase phosphorylation analyses) suggests that any pharmacological intervention may need to target protein–protein interactions or the PX domain rather than conventional ATP-binding sites (pearce2010thenutsand pages 1-2, karamafrooz2021integratedphosphoproteomicsfor pages 31-32).

Additionally, alterations in sphingosine‐1‐phosphate signaling have been associated with a range of pathological conditions, including inflammatory diseases and cancer. Although direct disease associations specific to RPS6KC1 remain to be conclusively established, its role—by virtue of influencing S1P signaling and endosomal dynamics—positions it as a potential contributor to disease states in which dysregulated lipid signaling and oxidative stress are central features. In particular, its ability to recruit PRDX3 to early endosomes may have implications for the management of cellular redox balance in disease contexts (arencibia2013agcproteinkinases pages 18-18, pearce2010thenutsand pages 1-2).

Notably, some literature has classified RPS6KC1 as a pseudokinase. In such cases, the protein’s function may derive predominantly from its capacity to serve as a molecular scaffold or to mediate allosteric interactions within signaling complexes rather than from robust catalytic activity. This characteristic may influence both its regulatory properties and its potential utility as a therapeutic target, as modulating scaffold functions can be as critical as inhibiting enzymatic activity in controlling cellular signaling networks (thiriet2013cytoplasmicproteinserinethreonine pages 63-66).

Finally, integrated phosphoproteomic studies have provided preliminary maps of phosphorylation sites on RPS6KC1, raising the possibility that dynamic regulatory modifications may exist under specific signaling conditions. Such data, while not yet fully detailed in the literature, offer a foundation for future investigations aimed at defining the physiological relevance of these modifications and identifying candidate sites for pharmacological targeting (karamafrooz2021integratedphosphoproteomicsfor pages 30-31, pages 31-32). The cumulative evidence underscores the importance of continuing to refine our understanding of RPS6KC1, both from the perspective of basic kinase biology and for its potential role in mediating critical cellular signaling events.

References

1. (magnuson2012regulationandfunction pages 10-11): Brian Magnuson, Bilgen Ekim, and Diane C. Fingar. Regulation and function of ribosomal protein s6 kinase (s6k) within mtor signalling networks. Biochemical Journal, 441:1-21, Dec 2012. URL: https://doi.org/10.1042/bj20110892, doi:10.1042/bj20110892. This article has 1235 citations and is from a domain leading peer-reviewed journal.
2. (thiriet2013cytoplasmicproteinserinethreonine pages 57-60): M Thiriet M Thiriet. Cytoplasmic protein serine/threonine kinases. Biomathematical and Biomechanical Modeling of the Circulatory and Ventilatory Systems, pages 175-310, Jul 2013. URL: https://doi.org/10.1007/978-1-4614-4370-4\_5, doi:10.1007/978-1-4614-4370-4\_5. This article has 11 citations.
3. (thiriet2013cytoplasmicproteinserinethreonine pages 60-63): M Thiriet M Thiriet. Cytoplasmic protein serine/threonine kinases. Biomathematical and Biomechanical Modeling of the Circulatory and Ventilatory Systems, pages 175-310, Jul 2013. URL: https://doi.org/10.1007/978-1-4614-4370-4\_5, doi:10.1007/978-1-4614-4370-4\_5. This article has 11 citations.
4. (thiriet2013cytoplasmicproteinserinethreonine pages 63-66): M Thiriet M Thiriet. Cytoplasmic protein serine/threonine kinases. Biomathematical and Biomechanical Modeling of the Circulatory and Ventilatory Systems, pages 175-310, Jul 2013. URL: https://doi.org/10.1007/978-1-4614-4370-4\_5, doi:10.1007/978-1-4614-4370-4\_5. This article has 11 citations.
5. (arencibia2013agcproteinkinases pages 18-18): José M. Arencibia, Daniel Pastor-Flores, Angelika F. Bauer, Jörg O. Schulze, and Ricardo M. Biondi. Agc protein kinases: from structural mechanism of regulation to allosteric drug development for the treatment of human diseases. Biochimica et Biophysica Acta (BBA) - Proteins and Proteomics, 1834:1302-1321, Jul 2013. URL: https://doi.org/10.1016/j.bbapap.2013.03.010, doi:10.1016/j.bbapap.2013.03.010. This article has 238 citations.
6. (arencibia2013agcproteinkinases pages 3-4): José M. Arencibia, Daniel Pastor-Flores, Angelika F. Bauer, Jörg O. Schulze, and Ricardo M. Biondi. Agc protein kinases: from structural mechanism of regulation to allosteric drug development for the treatment of human diseases. Biochimica et Biophysica Acta (BBA) - Proteins and Proteomics, 1834:1302-1321, Jul 2013. URL: https://doi.org/10.1016/j.bbapap.2013.03.010, doi:10.1016/j.bbapap.2013.03.010. This article has 238 citations.
7. (arencibia2013agcproteinkinases pages 8-9): José M. Arencibia, Daniel Pastor-Flores, Angelika F. Bauer, Jörg O. Schulze, and Ricardo M. Biondi. Agc protein kinases: from structural mechanism of regulation to allosteric drug development for the treatment of human diseases. Biochimica et Biophysica Acta (BBA) - Proteins and Proteomics, 1834:1302-1321, Jul 2013. URL: https://doi.org/10.1016/j.bbapap.2013.03.010, doi:10.1016/j.bbapap.2013.03.010. This article has 238 citations.
8. (karamafrooz2021integratedphosphoproteomicsfor pages 30-31): Adak Karamafrooz, Jack Brennan, David D. Thomas, and Laurie L. Parker. Integrated phosphoproteomics for identifying substrates of human protein kinase a (prkaca) and its oncogenic mutant dnajb1–prkaca. Journal of Proteome Research, 20:4815-4830, Aug 2021. URL: https://doi.org/10.1021/acs.jproteome.1c00500, doi:10.1021/acs.jproteome.1c00500. This article has 11 citations and is from a peer-reviewed journal.
9. (magnuson2012regulationandfunction pages 2-3): Brian Magnuson, Bilgen Ekim, and Diane C. Fingar. Regulation and function of ribosomal protein s6 kinase (s6k) within mtor signalling networks. Biochemical Journal, 441:1-21, Dec 2012. URL: https://doi.org/10.1042/bj20110892, doi:10.1042/bj20110892. This article has 1235 citations and is from a domain leading peer-reviewed journal.
10. (pearce2010thenutsand pages 1-2): Laura R. Pearce, David Komander, and Dario R. Alessi. The nuts and bolts of agc protein kinases. Nature Reviews Molecular Cell Biology, 11:9-22, Jan 2010. URL: https://doi.org/10.1038/nrm2822, doi:10.1038/nrm2822. This article has 1655 citations and is from a domain leading peer-reviewed journal.
11. (karamafrooz2021integratedphosphoproteomicsfor pages 31-32): Adak Karamafrooz, Jack Brennan, David D. Thomas, and Laurie L. Parker. Integrated phosphoproteomics for identifying substrates of human protein kinase a (prkaca) and its oncogenic mutant dnajb1–prkaca. Journal of Proteome Research, 20:4815-4830, Aug 2021. URL: https://doi.org/10.1021/acs.jproteome.1c00500, doi:10.1021/acs.jproteome.1c00500. This article has 11 citations and is from a peer-reviewed journal.