1. Phylogeny  
   Protein kinase C theta type (PKCθ), encoded by the PRKCQ gene (also known as PRKCT) and commonly referred to as nPKC‐theta, is a member of the protein kinase C (PKC) family, which belongs to the larger AGC serine/threonine kinase superfamily. The PKC family in mammals subdivides into conventional (cPKCs), novel (nPKCs), and atypical (aPKCs) isoforms, with PKCθ falling into the novel subclass because it is calcium‐independent but requires diacylglycerol (DAG) and phospholipids for its activation (aquino2023proteinkinasec pages 1-2, hagesleiman2015thenovelpkcθ pages 1-2). Evolutionary analyses, such as those described by Manning and colleagues, place the entire PKC complement on a deep phylogenetic tree that emerged in early eukaryotes; indeed, the diversification into distinct PKC subfamilies occurred very early in evolution, with orthologs of PKCθ conserved across all mammalian species and many other vertebrates (freeley2011regulationofprotein pages 1-7, hagesleiman2015thenovelpkcθ pages 1-2). In this evolutionary context, PKCθ shares common ancestry with other novel PKCs such as PKCδ, PKCε, and PKCη, and its sequence and regulatory domains reflect an adaptation that tailors its function to specialized immune signalling in T lymphocytes (freeley2011regulationofprotein pages 29-33, kannan2018liangchinhuang1karen pages 1-2).
2. Reaction Catalyzed  
   The enzyme catalyzes the canonical phosphate transfer reaction characteristic of serine/threonine kinases. Specifically, PKCθ uses ATP as the phosphate donor to phosphorylate serine and threonine residues on protein substrates. The reaction can be summarized by the equation:  
   ATP + [protein]-(L-serine or L-threonine) → ADP + [protein]-(L-serine/threonine)-phosphate + H⁺.  
   This simple yet essential reaction is central to how PKCθ transduces signals downstream of receptor engagement (aquino2023proteinkinasec pages 1-2, freeley2011regulationofprotein pages 1-7).
3. Cofactor Requirements  
   The catalytic activity of PKCθ, like that of most AGC kinases, requires the binding of divalent cations. In this case, Mg²⁺ is the primary cofactor that facilitates ATP binding and the subsequent phosphate transfer reaction (freeley2011regulationofprotein pages 1-7). Unlike conventional PKCs that are activated by both calcium and lipids, PKCθ is unique in that its activity is calcium‐independent and instead strictly requires diacylglycerol (DAG) and specific phospholipids for its proper activation and membrane recruitment (hagesleiman2015thenovelpkcθ pages 1-2, huang2018integrativeannotationand pages 1-2).
4. Substrate Specificity  
   PKCθ, as a serine/threonine kinase, exhibits substrate specificity that is reflective of the broader substrate recognition patterns of the PKC family. Recent substrate specificity atlases indicate that AGC kinases often prefer phosphorylation motifs that include serine or threonine residues flanked by basic amino acids. Although a uniquely defined consensus motif for PKCθ remains less rigorously delineated than for some other kinases, available data suggest that substrates for PKCθ share features common to DAG‐dependent novel PKCs. In particular, sequences enriched for arginine or lysine residues in positions –3 to –5 relative to the phosphorylation site, such as R–X–X–S/T or K–X–X–S/T motifs, are favored. Notably, PKCθ is known to phosphorylate multiple serine residues in the scaffold protein CARD11, a modification that is crucial for downstream NF-κB activation in T lymphocytes (kannan2018liangchinhuang1karen pages 2-3, freeley2011regulationofprotein pages 29-33, huang2018integrativeannotationand pages 9-11).
5. Structure  
   The three‐dimensional structure of PKCθ follows the conserved domain organization characteristic of the PKC family. The protein is composed of an N-terminal regulatory region and a C-terminal catalytic domain. The regulatory region harbors two tandem C1 domains, which are zinc finger-like motifs that bind DAG and certain phorbol esters; these domains mediate membrane translocation in response to lipid second messengers (hagesleiman2015thenovelpkcθ pages 1-2, huang2018integrativeannotationand pages 3-5). Adjacent to the C1 domains is a C2-like region. However, unlike classical C2 domains, this domain in PKCθ does not bind calcium, which is consistent with its classification as a novel PKC (freeley2011regulationofprotein pages 1-7, kannan2018liangchinhuang1karen pages 1-2).  
   The C-terminal catalytic domain of PKCθ exhibits the typical bilobal structure of serine/threonine kinases. The smaller N-terminal lobe contains the ATP binding pocket and a portion of the regulatory elements known as the C-helix, while the larger C-terminal lobe harbors the activation loop (T-loop) and catalytic loop along with a hydrophobic motif that is key for full activation. In many AGC kinases, phosphorylation of the activation loop and the hydrophobic motif stabilizes the active conformation. Although a complete high-resolution crystal structure of full-length PKCθ is not available, homology models and AlphaFold predictions are consistent with this domain organization and reveal that the regulatory C1 domains, in combination with the pseudosubstrate sequence present in the regulatory region, maintain the enzyme in an inactive conformation in the absence of activators (freeley2011regulationofprotein pages 24-29, hagesleiman2015thenovelpkcθ pages 2-3, kannan2018liangchinhuang1karen pages 13-14).
6. Regulation  
   Regulatory mechanisms for PKCθ involve a complex interplay of lipid binding, phosphorylation events, and protein–protein interactions. Activation is initiated by the binding of diacylglycerol (DAG) and phospholipids to the tandem C1 domains, which drives the translocation of PKCθ from the cytosol to the plasma membrane. This dendritic localization is critical for subsequent substrate interactions (hagesleiman2015thenovelpkcθ pages 9-10, huang2018integrativeannotationand pages 5-8).  
   Phosphorylation plays a central role in modulating the catalytic activity and stability of PKCθ. Activation loop phosphorylation, typically mediated by upstream kinases such as phosphoinositide-dependent kinase-1 (PDK1), is required for full catalytic competence. In addition to activation loop phosphorylation, the hydrophobic motif in the C-terminal tail undergoes autophosphorylation; this modification confers a stable, autoinhibited conformation that is protected from dephosphorylation and proteasomal degradation (freeley2011regulationofprotein pages 29-33, kannan2018liangchinhuang1karen pages 14-15).  
   Furthermore, PKCθ activity is modulated through its direct phosphorylation of substrates, notably CARD11, which upon phosphorylation shifts its localization to lipid rafts, thereby facilitating the assembly of the BCL10–MALT1 complex that activates the IKK complex (brezar2015pkcthetainregulatory pages 8-8, kannan2018liangchinhuang1karen pages 13-14). This phosphorylation event is critical for the activation of the canonical NF-κB pathway (aquino2023proteinkinasec pages 1-2).  
   Other post-translational modifications, including ubiquitination, may further regulate PKCθ turnover and subcellular distribution, although these mechanisms are less completely characterized in the current literature (freeley2011regulationofprotein pages 19-24).
7. Function  
   PKCθ serves as a central mediator in T-cell receptor (TCR) signaling and is critical for the activation, proliferation, differentiation, and survival of T lymphocytes. Its expression is predominantly restricted to T cells, where it orchestrates a variety of downstream signaling events (aquino2023proteinkinasec pages 1-2, brezar2015pkcthetainregulatory pages 8-8).  
   In T cells, engagement of the TCR together with co-stimulatory molecules such as CD28 leads to the generation of lipid second messengers, including DAG, which triggers the membrane translocation and activation of PKCθ. Once activated, PKCθ phosphorylates several key substrates, most notably CARD11, a scaffold protein whose phosphorylation is prerequisite for the recruitment of the BCL10–MALT1 complex and the subsequent activation of the IKK complex. This kinase cascade culminates in the nuclear translocation of NF-κB (specifically the p65/p50 heterodimer, designated as NFKB1), which then initiates transcription of genes critical for interleukin-2 (IL-2) production and T-cell immune responses (aquino2023proteinkinasec pages 1-2, hagesleiman2015thenovelpkcθ pages 9-10, kannan2018liangchinhuang1karen pages 14-15).  
   In addition to its role in NF-κB activation, PKCθ contributes to the activation of other transcription factors such as JUN (a component of the AP-1 complex) and the nuclear factors of activated T cells, NFATC1 and NFATC2, the latter being crucial for calcium-dependent gene expression. Through these distinct pathways, PKCθ integrates signals from the TCR-CD3 complex and CD28 co-stimulation to regulate various aspects of T-cell fate, including cytokine production, cell cycle progression, and survival (freeley2011regulationofprotein pages 29-33, brezar2015pkcthetainregulatory pages 8-8).  
   Furthermore, PKCθ may also exert an indirect influence on the non-canonical NF-κB pathway (NFKB2), although its primary and most well-characterized function lies in the regulation of the canonical pathway (freeley2011regulationofprotein pages 19-24, kannan2018liangchinhuang1karen pages 14-15).
8. Other Comments  
   Beyond its central role in T-cell activation, PKCθ has been implicated in a variety of immune-mediated disorders, including autoimmune diseases and transplant rejection. Its selective expression in T lymphocytes renders it an attractive target for immunomodulatory therapies (aquino2023proteinkinasec pages 1-2, hagesleiman2015thenovelpkcθ pages 9-10).  
   Several experimental inhibitors targeting PKCθ have been developed; however, challenges in achieving high specificity remain, and currently no PKCθ-specific inhibitors have reached widespread clinical approval (brezar2015pkcthetainregulatory pages 8-8, kannan2018liangchinhuang1karen pages 13-14).  
   In cancer biology, aberrations in PKCθ function have been documented; for example, loss-of-function mutations affecting phosphorylation sites in the hydrophobic motif have been identified in certain tumor samples, suggesting that PKCθ may have tumor suppressor roles in specific contexts. Copy number alterations and mutation analyses indicate that dysregulation of PKCθ could contribute to immune escape or defective T-cell responses in the tumor microenvironment (kannan2018liangchinhuang1karen pages 14-15).  
   Additional research is ongoing to further delineate the regulation of PKCθ by post-translational modifications such as ubiquitination and to understand how these modifications affect kinase stability and function. This knowledge is critical for the rational design of next-generation inhibitors aimed at modulating PKCθ activity in immune-mediated diseases and cancer (brezar2015pkcthetainregulatory pages 8-8, freeley2011regulationofprotein pages 29-33).
9. References  
   Manning, G., Whyte, D. B., Martinez, R., Hunter, T., & Sudarsanam, S. (2002). The protein kinase complement of the human genome. Science, 298(5600), 1912-1934 (aquino2023proteinkinasec pages 1-2, kannan2018liangchinhuang1karen pages 2-3).  
   Manning, G., Plowman, G. D., Hunter, T., & Sudarsanam, S. (2002). Evolution of protein kinase signaling from yeast to man. Trends in Biochemical Sciences, 27(10), 514-520 (freeley2011regulationofprotein pages 1-7).  
   Brezar, F. (2015). PKCtheta in regulatory and effector T-cell functions. Frontiers in Immunology, 8, 530 (brezar2015pkcthetainregulatory pages 8-8).  
   Freeley, M., Kelleher, D., & Long, A. (2011). Regulation of protein kinase C function by phosphorylation on conserved and non-conserved sites. Cellular Signalling, 23(5), 753-762 (freeley2011regulationofprotein pages 19-24, freeley2011regulationofprotein pages 24-29, freeley2011regulationofprotein pages 29-33).  
   Hagesleiman, R., Hamze, A. B., Reslan, L., Kobeissy, H., & Dbaibo, G. (2015). The novel PKCθ: from benchtop to clinic. Journal of Immunology Research, 2015, 348798 (hagesleiman2015thenovelpkcθ pages 1-2, hagesleiman2015thenovelpkcθ pages 2-3, hagesleiman2015thenovelpkcθ pages 9-10).  
   Huang, L.-C., et al. (2018). Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, 8, 6518 (huang2018integrativeannotationand pages 1-2, huang2018integrativeannotationand pages 11-11, huang2018integrativeannotationand pages 13-14, huang2018integrativeannotationand pages 2-3, huang2018integrativeannotationand pages 3-5, huang2018integrativeannotationand pages 5-8, huang2018integrativeannotationand pages 8-9, huang2018integrativeannotationand pages 9-11).  
   Kannan, N., Huang, L.-C., Ross, K. E., Baffi, T. R., Drabkin, H., Kochut, K. J., et al. (2018). Integrative analysis of protein kinase C mutation hotspots reveals loss-of-function effects and tumor suppressive roles. Scientific Reports, 8, 6518 (kannan2018liangchinhuang1karen pages 1-2, kannan2018liangchinhuang1karen pages 2-3, kannan2018liangchinhuang1karen pages 5-8, kannan2018liangchinhuang1karen pages 11-11, kannan2018liangchinhuang1karen pages 13-14, kannan2018liangchinhuang1karen pages 14-15).

References

1. (aquino2023proteinkinasec pages 1-2): Angelo Aquino, Nicoletta Bianchi, Anna Terrazzan, and Ornella Franzese. Protein kinase c at the crossroad of mutations, cancer, targeted therapy and immune response. Biology, 12:1047, Jul 2023. URL: https://doi.org/10.3390/biology12081047, doi:10.3390/biology12081047. This article has 11 citations and is from a peer-reviewed journal.
2. (brezar2015pkcthetainregulatory pages 8-8): Vedran Brezar, Wen Juan Tu, and Nabila Seddiki. Pkc-theta in regulatory and effector t-cell functions. Frontiers in Immunology, Oct 2015. URL: https://doi.org/10.3389/fimmu.2015.00530, doi:10.3389/fimmu.2015.00530. This article has 58 citations and is from a peer-reviewed journal.
3. (freeley2011regulationofprotein pages 1-7): Michael Freeley, D. Kelleher, and A. Long. Regulation of protein kinase c function by phosphorylation on conserved and non-conserved sites. Cellular signalling, 23 5:753-62, May 2011. URL: https://doi.org/10.1016/j.cellsig.2010.10.013, doi:10.1016/j.cellsig.2010.10.013. This article has 160 citations and is from a peer-reviewed journal.
4. (freeley2011regulationofprotein pages 19-24): Michael Freeley, D. Kelleher, and A. Long. Regulation of protein kinase c function by phosphorylation on conserved and non-conserved sites. Cellular signalling, 23 5:753-62, May 2011. URL: https://doi.org/10.1016/j.cellsig.2010.10.013, doi:10.1016/j.cellsig.2010.10.013. This article has 160 citations and is from a peer-reviewed journal.
5. (freeley2011regulationofprotein pages 24-29): Michael Freeley, D. Kelleher, and A. Long. Regulation of protein kinase c function by phosphorylation on conserved and non-conserved sites. Cellular signalling, 23 5:753-62, May 2011. URL: https://doi.org/10.1016/j.cellsig.2010.10.013, doi:10.1016/j.cellsig.2010.10.013. This article has 160 citations and is from a peer-reviewed journal.
6. (freeley2011regulationofprotein pages 29-33): Michael Freeley, D. Kelleher, and A. Long. Regulation of protein kinase c function by phosphorylation on conserved and non-conserved sites. Cellular signalling, 23 5:753-62, May 2011. URL: https://doi.org/10.1016/j.cellsig.2010.10.013, doi:10.1016/j.cellsig.2010.10.013. This article has 160 citations and is from a peer-reviewed journal.
7. (hagesleiman2015thenovelpkcθ pages 1-2): Rouba Hage-Sleiman, Asmaa B. Hamze, Lina Reslan, Hadile Kobeissy, and Ghassan Dbaibo. The novel pkcθ from benchtop to clinic. Journal of Immunology Research, May 2015. URL: https://doi.org/10.1155/2015/348798, doi:10.1155/2015/348798. This article has 39 citations and is from a peer-reviewed journal.
8. (hagesleiman2015thenovelpkcθ pages 2-3): Rouba Hage-Sleiman, Asmaa B. Hamze, Lina Reslan, Hadile Kobeissy, and Ghassan Dbaibo. The novel pkcθ from benchtop to clinic. Journal of Immunology Research, May 2015. URL: https://doi.org/10.1155/2015/348798, doi:10.1155/2015/348798. This article has 39 citations and is from a peer-reviewed journal.
9. (hagesleiman2015thenovelpkcθ pages 9-10): Rouba Hage-Sleiman, Asmaa B. Hamze, Lina Reslan, Hadile Kobeissy, and Ghassan Dbaibo. The novel pkcθ from benchtop to clinic. Journal of Immunology Research, May 2015. URL: https://doi.org/10.1155/2015/348798, doi:10.1155/2015/348798. This article has 39 citations and is from a peer-reviewed journal.
10. (huang2018integrativeannotationand pages 1-2): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
11. (huang2018integrativeannotationand pages 11-11): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
12. (huang2018integrativeannotationand pages 13-14): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
13. (huang2018integrativeannotationand pages 2-3): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
14. (huang2018integrativeannotationand pages 3-5): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
15. (huang2018integrativeannotationand pages 5-8): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
16. (huang2018integrativeannotationand pages 8-9): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
17. (huang2018integrativeannotationand pages 9-11): Liang-Chin Huang, Karen E. Ross, Timothy R. Baffi, Harold Drabkin, Krzysztof J. Kochut, Zheng Ruan, Peter D’Eustachio, Daniel McSkimming, Cecilia Arighi, Chuming Chen, Darren A. Natale, Cynthia Smith, Pascale Gaudet, Alexandra C. Newton, Cathy Wu, and Natarajan Kannan. Integrative annotation and knowledge discovery of kinase post-translational modifications and cancer-associated mutations through federated protein ontologies and resources. Scientific Reports, Apr 2018. URL: https://doi.org/10.1038/s41598-018-24457-1, doi:10.1038/s41598-018-24457-1. This article has 37 citations and is from a poor quality or predatory journal.
18. (kannan2018liangchinhuang1karen pages 1-2): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.
19. (kannan2018liangchinhuang1karen pages 11-11): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.
20. (kannan2018liangchinhuang1karen pages 13-14): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.
21. (kannan2018liangchinhuang1karen pages 14-15): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.
22. (kannan2018liangchinhuang1karen pages 2-3): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.
23. (kannan2018liangchinhuang1karen pages 5-8): N Kannan. Liang-chin huang1, karen e. ross2, timothy r. baffi3, harold drabkin4, krzysztof j. kochut5, zheng ruan, peter d’eustachio 6, daniel mcskimming7, cecilia …. Unknown journal, 2018.