1. Phylogeny  
   Calcium/calmodulin-dependent protein kinase II subunit delta (CAMK2D, also termed CaMKIIδ) belongs to the CaMKII family, a group of serine/threonine protein kinases that are evolutionarily conserved across species. Phylogenetic analyses indicate that the CaMKII family is ancient, traceable to early metazoans, and the four isoforms (α, β, γ, and δ) likely arose by gene duplication prior to the radiation of modern vertebrates (beghi2022calciumsignallingin pages 7-9, bhattacharyya2020structuralinsightsinto pages 3-5). CAMK2D is the predominant isoform expressed in cardiac tissue and can be found in mammalian species with well‐conserved kinase and association domains. Its evolutionary conservation is underscored by the maintenance of key regulatory modules, such as the catalytic core and the Ca²⁺/calmodulin (CaM)–binding regulatory region, among orthologs from diverse organisms (beghi2022calciumsignallingin pages 7-9, bhattacharyya2020structuralinsightsinto pages 3-5).
2. Reaction Catalyzed  
   CAMK2D catalyzes the transfer of the γ-phosphate group from ATP to serine/threonine residues on target proteins. In biochemical terms, the reaction can be summarized as:  
     ATP + [protein] – (L-serine or L-threonine) → ADP + [protein] – (L-serine/threonine)-phosphate + H⁺  
   This phosphorylation reaction is central to its role in regulating diverse substrates involved in calcium homeostasis and excitation–contraction coupling (beghi2022calciumsignallingin pages 16-17, swulius2008ca2+calmodulindependentproteinkinases pages 10-13).
3. Cofactor Requirements  
   The enzymatic activity of CAMK2D is dependent on Ca²⁺ binding to calmodulin, which serves as a calcium sensor to activate the kinase. In the presence of elevated intracellular Ca²⁺, calmodulin binds four Ca²⁺ ions and undergoes a conformational change that allows it to interact with CAMK2D, relieving autoinhibition. Additionally, typical serine/threonine kinases require a divalent metal ion, most commonly Mg²⁺, as a cofactor that facilitates ATP binding and proper phosphoryl transfer (beghi2022calciumsignallingin pages 6-7, zhang2021calciumcalmodulin–dependentproteinkinase pages 2-4).
4. Substrate Specificity  
   CAMK2D exhibits substrate specificity for serine/threonine residues within target proteins. Its activity is directed toward several key substrates critical in cardiac physiology. Among these, CAMK2D phosphorylates proteins involved in the regulation of calcium cycling and excitation–contraction coupling, including:  
    • Phospholamban, which modulates sarcoplasmic reticulum Ca²⁺ uptake.  
    • Ryanodine receptor 2 (RYR2), where phosphorylation (notably at Ser-2808) modulates Ca²⁺ release from the sarcoplasmic reticulum.  
    • Myosin-binding protein-C and titin, which ultimately impact contractile function and passive diastolic stiffness.  
    • Voltage-gated sodium and potassium channels, contributing to the regulation of cardiac excitability.  
   Furthermore, in the nucleus CAMK2D phosphorylates histone deacetylase 4 (HDAC4), facilitating its export and thereby modulating transcriptional programs associated with hypertrophy (beghi2022calciumsignallingin pages 16-17, gray2014camkiideltasubtypeslocalization pages 7-8).
5. Structure  
   CAMK2D consists of a highly conserved multidomain architecture typical for CaMKII isoforms. Its domain organization includes:  
    • An N-terminal catalytic domain responsible for ATP binding and phosphate transfer.  
    • A central regulatory domain that harbors an autoinhibitory region overlapping with a Ca²⁺/calmodulin–binding site; binding of Ca²⁺/CaM to this region relieves autoinhibition.  
    • A C-terminal association (or hub) domain that mediates oligomerization into a multimeric holoenzyme, usually a dodecamer arranged as two stacked hexameric rings.  
   Structural studies using crystallography and electron microscopy have revealed that the holoenzyme architecture facilitates intersubunit autophosphorylation—critical for its transition to a Ca²⁺-independent, or “autonomous”, active state. Key catalytic features include the activation loop (often phosphorylated at a residue analogous to Thr286 in other isoforms, with Thr287 being the corresponding site in CAMK2D), the conserved C-helix necessary for aligning the active site, and hydrophobic spines that stabilize the active conformation (bhattacharyya2020structuralinsightsinto pages 3-5, rostas2023calciumcalmodulinstimulatedproteinkinase pages 15-17, swulius2008ca2+calmodulindependentproteinkinases pages 9-10).
6. Regulation  
   CAMK2D is tightly regulated by several distinct mechanisms:  
    • Ca²⁺/calmodulin Binding: Elevated intracellular Ca²⁺ leads to CaM binding, which displaces the autoinhibitory segment from the catalytic site and activates the kinase.  
    • Autophosphorylation: Once active, CAMK2D undergoes intersubunit autophosphorylation at key threonine residues (e.g., Thr287), which sustains its activity independently of Ca²⁺/CaM (termed “autonomy”). Subsequent phosphorylation events at sites such as Thr305/306 can inhibit further Ca²⁺/CaM rebinding, thereby modulating sensitivity to calcium signals (bhattacharyya2020structuralinsightsinto pages 15-16, rostas2023calciumcalmodulinstimulatedproteinkinase pages 14-15).  
    • Oxidative Modifications: Under conditions of oxidative stress, specific methionine residues within CAMK2D may become oxidized, resulting in Ca²⁺/CaM-independent activation.  
    • Alternative Splicing: Multiple splice variants of CAMK2D exist, with differences in regulatory or targeting sequences that can influence subcellular localization and functional responses (gray2014camkiideltasubtypeslocalization pages 7-8).  
   Together, these regulatory mechanisms enable CAMK2D to integrate transient Ca²⁺ signals into longer-lasting phosphorylation events, crucial for its roles in cardiac signal transduction (bhattacharyya2020structuralinsightsinto pages 5-6, hidalgo2013themultifunctionalca2+calmodulindependent pages 1-2).
7. Function  
   CAMK2D plays a central role in the regulation of cardiac calcium homeostasis and excitation–contraction coupling (ECC). Key functional roles include:  
    • Modulating Ca²⁺ Cycling: By phosphorylating proteins such as the ryanodine receptor (RYR2) and phospholamban, CAMK2D regulates calcium release, reuptake, and overall sarcoplasmic reticulum function, which are critical for proper cardiac contractility (beghi2022calciumsignallingin pages 16-17, beghi2022calciumsignallingin pages 6-7).  
    • Control of Electrical Activity: Through phosphorylation of voltage-gated sodium and potassium channels, CAMK2D contributes to the regulation of membrane excitability in cardiomyocytes.  
    • Transcriptional Regulation: In the nucleus, CAMK2D phosphorylates HDAC4. This modification promotes HDAC4 nuclear export and subsequent activation of transcription factors such as myocyte enhancer factor 2 (MEF2), leading to the expression of genes involved in the hypertrophic response of the heart.  
    • Pathological Roles: Upon chronic activation, CAMK2D is involved in the pathogenesis of dilated cardiomyopathy and heart failure, contributing to maladaptive cardiac remodeling and decompensation following myocardial infarction (beghi2022calciumsignallingin pages 16-17, rostas2023calciumcalmodulinstimulatedproteinkinase pages 11-12).
8. Other Comments  
   Experimental inhibitors of CaMKII, such as KN-93, have been used to elucidate the functional role of CAMK2D in cardiac physiology; these inhibitors reduce phosphorylation of downstream targets and attenuate pathological signaling in preclinical models (hidalgo2013themultifunctionalca2+calmodulindependent pages 1-2, rokita2012newtherapeutictargets pages 2-3). CAMK2D’s activity profile and its central role in both normal cardiac function and pathological cardiac remodeling make it an attractive therapeutic target. Clinically, dysregulation of CAMK2D has been linked to heart failure, arrhythmias, and ventricular remodeling after myocardial infarction, underscoring its potential as a biomarker and as a candidate for targeted intervention. In addition, alternative splicing producing variants with distinct subcellular localizations—such as nuclear-targeted versus cytoplasmic isoforms—adds another layer of functional complexity and specificity (gray2014camkiideltasubtypeslocalization pages 7-8, mohanan2022roleofca2+calmodulindependent pages 29-30).
9. References  
   [1] Sofia Beghi, Malgorzata Furmanik, Armand Jaminon, Rogier Veltrop, Nikolas Rapp, Kanin Wichapong, Elham Bidar, Annamaria Buschini, and Leon J. Schurgers, “Calcium signalling in heart and vessels: role of calmodulin and downstream calmodulin‐dependent protein kinases,” International Journal of Molecular Sciences, vol. 23, article 16139, Dec. 2022 (pages 6-7, 7-9, 16-17, 12-13).  
   [2] Moitrayee Bhattacharyya, Deepti Karandur, and John Kuriyan, “Structural insights into the regulation of ca²⁺/calmodulin-dependent protein kinase ii (camkii),” Cold Spring Harbor Perspectives in Biology, vol. 12, article a035147, Oct. 2020 (pages 1-3, 3-5, 5-6, 15-16, 19-20).  
   [3] Charles B. B. Gray and Joan Heller Brown, “Camkiidelta subtypes: localization and function,” Frontiers in Pharmacology, Feb. 2014 (pages 7-8).  
   [4] Carlos G. Hidalgo, Charles S. Chung, Chandra Saripalli, Mei Methawasin, Kirk R. Hutchinson, George Tsaprailis, Siegfried Labeit, Alicia Mattiazzi, and Henk L. Granzier, “The multifunctional ca²⁺/calmodulin-dependent protein kinase ii delta (camkiiδ) phosphorylates cardiac titin’s spring elements,” Journal of Molecular and Cellular Cardiology, vol. 54, pp. 90–97, Jan. 2013 (pages 1-2, 5-7).  
   [5] Xuejing Zhang, Jaclyn Connelly, Edwin S. Levitan, Dandan Sun, and Jane Q. Wang, “Calcium/calmodulin–dependent protein kinase ii in cerebrovascular diseases,” Translational Stroke Research, vol. 12, pp. 513–529, Mar. 2021 (pages 1-2, 2-4).  
   [6] Pamela Gaitán-González, Rommel Sánchez-Hernández, José-Antonio Arias-Montaño, and Angélica Rueda, “Tale of two kinases: protein kinase A and ca²⁺/calmodulin-dependent protein kinase ii in pre-diabetic cardiomyopathy,” World Journal of Diabetes, vol. 12, pp. 1704–1718, Oct. 2021 (pages 11-12).  
   [7] Archana G. Mohanan, Sowmya Gunasekaran, Reena Sarah Jacob, and R. V. Omkumar, “Role of ca²⁺/calmodulin-dependent protein kinase type ii in mediating function and dysfunction at glutamatergic synapses,” Frontiers in Molecular Neuroscience, Jun. 2022 (pages 1-2, 15-16, 20-21, 22-22, 29-30).  
   [8] Adam G. Rokita and Mark E. Anderson, “New therapeutic targets in cardiology,” Circulation, vol. 126, pp. 2125–2139, Oct. 2012 (pages 2-3).  
   [9] John A. P. Rostas and Kathryn A. Skelding, “Calcium/calmodulin-stimulated protein kinase ii (camkii): different functional outcomes from activation, depending on the cellular microenvironment,” Cells, vol. 12, article 401, Jan. 2023 (pages 1-2, 2-4, 4-5, 5-7, 9-11, 11-12, 12-14, 14-15, 15-17).  
   [10] F. Z. Saddouk, Li-yan Sun, Y. F. Liu, M. Jiang, D. Singer, J. Backs, Dee Van Riper, R. Ginnan, J. Schwarz, and Harold A. Singer, “Ca²⁺/calmodulin‐dependent protein kinase ii‐γ (camkiiγ) negatively regulates vascular smooth muscle cell proliferation and vascular remodeling,” The FASEB Journal, vol. 30, pp. 1051–1064, Mar. 2016 (pages 13-14).  
   [11] M. T. Swulius and M. N. Waxham, “Ca²⁺/calmodulin-dependent protein kinases,” Cellular and Molecular Life Sciences, vol. 65, pp. 2637–2657, May 2008 (pages 1-2, 2-4, 4-6, 6-8, 8-9, 9-10, 10-13, 14-16, 17-18).  
   [12] Hiroshi Tokumitsu and Hiroyuki Sakagami, “Molecular mechanisms underlying ca²⁺/calmodulin-dependent protein kinase kinase signal transduction,” International Journal of Molecular Sciences, vol. 23, article 11025, Sep. 2022 (pages 15-16).  
   [13] Anshua Ghosh and Karl Peter Giese, “Calcium/calmodulin-dependent kinase ii and Alzheimer’s disease,” Molecular Brain, Nov. 2015 (pages 1-2).

Each citation was selected from peer-reviewed publications which provide comprehensive details on the enzyme’s evolutionary relationships, catalytic reaction, cofactor usage, substrate recognition, three-dimensional structure, modes of regulation, and biological function in cardiac physiology and pathology.

References

1. (beghi2022calciumsignallingin pages 16-17): Sofia Beghi, Malgorzata Furmanik, Armand Jaminon, Rogier Veltrop, Nikolas Rapp, Kanin Wichapong, Elham Bidar, Annamaria Buschini, and Leon J. Schurgers. Calcium signalling in heart and vessels: role of calmodulin and downstream calmodulin-dependent protein kinases. International Journal of Molecular Sciences, 23:16139, Dec 2022. URL: https://doi.org/10.3390/ijms232416139, doi:10.3390/ijms232416139. This article has 34 citations and is from a peer-reviewed journal.
2. (beghi2022calciumsignallingin pages 7-9): Sofia Beghi, Malgorzata Furmanik, Armand Jaminon, Rogier Veltrop, Nikolas Rapp, Kanin Wichapong, Elham Bidar, Annamaria Buschini, and Leon J. Schurgers. Calcium signalling in heart and vessels: role of calmodulin and downstream calmodulin-dependent protein kinases. International Journal of Molecular Sciences, 23:16139, Dec 2022. URL: https://doi.org/10.3390/ijms232416139, doi:10.3390/ijms232416139. This article has 34 citations and is from a peer-reviewed journal.
3. (bhattacharyya2020structuralinsightsinto pages 3-5): Moitrayee Bhattacharyya, Deepti Karandur, and John Kuriyan. Structural insights into the regulation of ca2+/calmodulin-dependent protein kinase ii (camkii). Cold Spring Harbor Perspectives in Biology, 12:a035147, Oct 2020. URL: https://doi.org/10.1101/cshperspect.a035147, doi:10.1101/cshperspect.a035147. This article has 90 citations and is from a peer-reviewed journal.
4. (gray2014camkiideltasubtypeslocalization pages 7-8): Charles B. B. Gray and Joan Heller Brown. Camkiidelta subtypes: localization and function. Frontiers in Pharmacology, Feb 2014. URL: https://doi.org/10.3389/fphar.2014.00015, doi:10.3389/fphar.2014.00015. This article has 102 citations and is from a peer-reviewed journal.
5. (hidalgo2013themultifunctionalca2+calmodulindependent pages 1-2): Carlos G. Hidalgo, Charles S. Chung, Chandra Saripalli, Mei Methawasin, Kirk R. Hutchinson, George Tsaprailis, Siegfried Labeit, Alicia Mattiazzi, and Henk L. Granzier. The multifunctional ca2+/calmodulin-dependent protein kinase ii delta (camkiiδ) phosphorylates cardiac titin’s spring elements. Journal of Molecular and Cellular Cardiology, 54:90-97, Jan 2013. URL: https://doi.org/10.1016/j.yjmcc.2012.11.012, doi:10.1016/j.yjmcc.2012.11.012. This article has 117 citations and is from a domain leading peer-reviewed journal.
6. (zhang2021calciumcalmodulin–dependentproteinkinase pages 2-4): Xuejing Zhang, Jaclyn Connelly, Edwin S. Levitan, Dandan Sun, and Jane Q. Wang. Calcium/calmodulin–dependent protein kinase ii in cerebrovascular diseases. Translational Stroke Research, 12:513-529, Mar 2021. URL: https://doi.org/10.1007/s12975-021-00901-9, doi:10.1007/s12975-021-00901-9. This article has 53 citations and is from a peer-reviewed journal.
7. (beghi2022calciumsignallingin pages 6-7): Sofia Beghi, Malgorzata Furmanik, Armand Jaminon, Rogier Veltrop, Nikolas Rapp, Kanin Wichapong, Elham Bidar, Annamaria Buschini, and Leon J. Schurgers. Calcium signalling in heart and vessels: role of calmodulin and downstream calmodulin-dependent protein kinases. International Journal of Molecular Sciences, 23:16139, Dec 2022. URL: https://doi.org/10.3390/ijms232416139, doi:10.3390/ijms232416139. This article has 34 citations and is from a peer-reviewed journal.
8. (bhattacharyya2020structuralinsightsinto pages 15-16): Moitrayee Bhattacharyya, Deepti Karandur, and John Kuriyan. Structural insights into the regulation of ca2+/calmodulin-dependent protein kinase ii (camkii). Cold Spring Harbor Perspectives in Biology, 12:a035147, Oct 2020. URL: https://doi.org/10.1101/cshperspect.a035147, doi:10.1101/cshperspect.a035147. This article has 90 citations and is from a peer-reviewed journal.
9. (bhattacharyya2020structuralinsightsinto pages 5-6): Moitrayee Bhattacharyya, Deepti Karandur, and John Kuriyan. Structural insights into the regulation of ca2+/calmodulin-dependent protein kinase ii (camkii). Cold Spring Harbor Perspectives in Biology, 12:a035147, Oct 2020. URL: https://doi.org/10.1101/cshperspect.a035147, doi:10.1101/cshperspect.a035147. This article has 90 citations and is from a peer-reviewed journal.
10. (mohanan2022roleofca2+calmodulindependent pages 29-30): Archana G. Mohanan, Sowmya Gunasekaran, Reena Sarah Jacob, and R. V. Omkumar. Role of ca2+/calmodulin-dependent protein kinase type ii in mediating function and dysfunction at glutamatergic synapses. Frontiers in Molecular Neuroscience, Jun 2022. URL: https://doi.org/10.3389/fnmol.2022.855752, doi:10.3389/fnmol.2022.855752. This article has 36 citations and is from a peer-reviewed journal.
11. (rokita2012newtherapeutictargets pages 2-3): Adam G. Rokita and Mark E. Anderson. New therapeutic targets in cardiology. Circulation, 126:2125-2139, Oct 2012. URL: https://doi.org/10.1161/circulationaha.112.124990, doi:10.1161/circulationaha.112.124990. This article has 151 citations and is from a highest quality peer-reviewed journal.
12. (rostas2023calciumcalmodulinstimulatedproteinkinase pages 14-15): John A. P. Rostas and Kathryn A. Skelding. Calcium/calmodulin-stimulated protein kinase ii (camkii): different functional outcomes from activation, depending on the cellular microenvironment. Cells, 12:401, Jan 2023. URL: https://doi.org/10.3390/cells12030401, doi:10.3390/cells12030401. This article has 46 citations and is from a peer-reviewed journal.
13. (rostas2023calciumcalmodulinstimulatedproteinkinase pages 15-17): John A. P. Rostas and Kathryn A. Skelding. Calcium/calmodulin-stimulated protein kinase ii (camkii): different functional outcomes from activation, depending on the cellular microenvironment. Cells, 12:401, Jan 2023. URL: https://doi.org/10.3390/cells12030401, doi:10.3390/cells12030401. This article has 46 citations and is from a peer-reviewed journal.
14. (swulius2008ca2+calmodulindependentproteinkinases pages 10-13): M. T. Swulius and M. N. Waxham. Ca2+/calmodulin-dependent protein kinases. Cellular and Molecular Life Sciences, 65:2637-2657, May 2008. URL: https://doi.org/10.1007/s00018-008-8086-2, doi:10.1007/s00018-008-8086-2. This article has 466 citations and is from a domain leading peer-reviewed journal.
15. (swulius2008ca2+calmodulindependentproteinkinases pages 9-10): M. T. Swulius and M. N. Waxham. Ca2+/calmodulin-dependent protein kinases. Cellular and Molecular Life Sciences, 65:2637-2657, May 2008. URL: https://doi.org/10.1007/s00018-008-8086-2, doi:10.1007/s00018-008-8086-2. This article has 466 citations and is from a domain leading peer-reviewed journal.
16. (rostas2023calciumcalmodulinstimulatedproteinkinase pages 11-12): John A. P. Rostas and Kathryn A. Skelding. Calcium/calmodulin-stimulated protein kinase ii (camkii): different functional outcomes from activation, depending on the cellular microenvironment. Cells, 12:401, Jan 2023. URL: https://doi.org/10.3390/cells12030401, doi:10.3390/cells12030401. This article has 46 citations and is from a peer-reviewed journal.