Phylogeny  
• Classified within the Ca²⁺/calmodulin-dependent protein kinase (CaMK) group, CAMK1/2 branch of the human kinome (Byrne et al., 2023).  
• Closest paralogue is PSKH2, a catalytically impaired pseudokinase that diverged from PSKH1 but retains the kinase fold (Byrne et al., 2023).  
• Shares conserved catalytic and regulatory motifs with CaMK1γ and CaMK2 (“Illuminating the Regulation…”, 2024, pp. 9–11).  
• PSKH1 orthologues are present throughout vertebrates, whereas PSKH2 is lost from mouse and rat genomes, indicating differential evolutionary retention (Shrestha et al., 2020).

Reaction Catalyzed  
ATP + protein-Ser/Thr ⇌ ADP + protein-Ser/Thr-P (“Illuminating the Regulation…”, 2024, pp. 78–84, 131–137).

Cofactor Requirements  
• Mg²⁺ is obligatory for phosphotransfer (“Illuminating the Regulation…”, 2024, pp. 78–84).  
• Ca²⁺/calmodulin enhances, but is not essential for, basal activity (“Illuminating the Regulation…”, 2024, pp. 101–108, 131–137).

Substrate Specificity  
• Positional-scanning peptide arrays defined a basophilic consensus L-x-R-T-x-S\*-F-x-x-x with a strict Arg at –3 and a strong preference for serine over threonine as the phospho-acceptor (“Illuminating the Regulation…”, 2024, pp. 175–180).  
• Independent profiling confirmed the Arg(–3) bias and overall serine selectivity (Horne et al., 2025).

Structure  
• N-terminus (res. 1–30): dual lipidation sites Gly2 (N-myristoylation) and Cys3 (palmitoylation) plus a PxxP SH3-binding motif (“Illuminating the Regulation…”, 2024, pp. 201–206).  
• Two calmodulin-binding domains (CBDs): an N-terminal CBD (~80–100; inverted 1-5-8 motif, Phe90 anchor) and a C-terminal CBD containing autophospho-Ser372 that modulates CaM affinity (“Illuminating the Regulation…”, 2024, pp. 140–144).  
• Catalytic core (≈86–424) retains canonical VAIK (Lys104), HRD (Asp218) and DFG (Asp254) motifs; deletions beyond residue 98 abolish activity (“Illuminating the Regulation…”, 2024, pp. 140–144).  
• Activation segment sites: Thr256 (major autophosphorylation), Thr260, Ser363 and Ser372 (“Illuminating the Regulation…”, 2024, pp. 101–108, 140–144).  
• AlphaFold2 predicts a head-to-toe homodimer that can engage CaM at both termini; no crystal or NMR structure is available (model AF-P11801-F1) (“Illuminating the Regulation…”, 2024, pp. 140–144).

Regulation  
Post-translational modifications  
– Cis-autophosphorylation at Thr256, Thr260, Ser363 and Ser372 increases catalytic turnover (“Illuminating the Regulation…”, 2024, pp. 101–108, 140–144).  
– N-myristoylation (Gly2) and palmitoylation (Cys3) drive membrane and Golgi localisation (“Illuminating the Regulation…”, 2024, pp. 201–206).

Allosteric control  
– Ca²⁺/calmodulin modestly stimulates activity at low Ca²⁺ (“Illuminating the Regulation…”, 2024, pp. 131–137).  
– Ca²⁺ sensors Reticulocalbin-1/-3 suppress autophosphorylation and catalytic output, whereas calumenin selectively reduces autophosphorylation (“Illuminating the Regulation…”, 2024, pp. 131–137).  
– UNC119B binds the kinase domain and enhances activity independently of N-terminal acylation (Horne et al., 2025).  
– Glucose withdrawal up-regulates kinase activity, linking PSKH1 to metabolic stress (“Illuminating the Regulation…”, 2024, pp. 16–21).

Function  
Expression & localisation  
• Broadly expressed with highest levels in testis; over-expressed in prostate, lung and kidney cancers (Horne et al., 2025; “Illuminating the Regulation…”, 2024, pp. 21–24).  
• Localises to Golgi, centrosome, nucleus and, when dually acylated, the plasma membrane (“Illuminating the Regulation…”, 2024, pp. 189–193, 201–206).

Signalling roles  
• Phosphorylates SR-rich splice factors, modulating pre-mRNA splicing (“Illuminating the Regulation…”, 2024, pp. 16–21).  
• Acts as a metabolic sensor that promotes fatty-acid utilisation and prostate cancer cell proliferation under glucose scarcity (“Illuminating the Regulation…”, 2024, pp. 175–180, 189–193).  
• Regulates vesicle trafficking and cell migration via interactions with SORBS1, PAK4 and other actin-related proteins (“Illuminating the Regulation…”, 2024, pp. 210–214).  
• Direct substrate: phosphorylates RSK1 at Ser380, facilitating PDK1-dependent RSK1 activation and downstream inhibition of eEF2K (“Illuminating the Regulation…”, 2024, pp. 180–185).  
• Upstream kinase: PAK1 can phosphorylate PSKH1 Thr256 in vitro (“Illuminating the Regulation…”, 2024, pp. 157–163).  
• Interactome includes Golgi resident GOLGA8R and Ca²⁺ sensor Reticulocalbin-3 (Horne et al., 2025).

Inhibitors  
• ATP-competitive compound “C2” inhibits PSKH1 and blocks proliferation of PSKH1-positive prostate cancer cells under glucose stress (“Illuminating the Regulation…”, 2024, pp. 175–180).  
• Chemoproteomic screens identified afatinib and neratinib as covalent binders, although potency was not reported (Shrestha et al., 2020).

Other Comments  
• Disease links: driver of metastatic prostate cancer, loss-of-function mutations associated with hepatorenal ciliopathy, and P66L/R79L variants linked to Crohn’s disease (“Illuminating the Regulation…”, 2024, pp. 175–180, 201–206; Horne et al., 2025).  
• Experimental gap: lack of a phospho-Thr256–specific antibody hampers direct monitoring of activation-loop status (“Illuminating the Regulation…”, 2024, pp. 157–163).

References  
Byrne, D. P., Shrestha, S., Daly, L. A., Marensi, V., Ramakrishnan, K., Eyers, C. E., Kannan, N., & Eyers, P. A. (2023). Evolutionary and cellular analysis of the ‘dark’ pseudokinase PSKH2. Biochemical Journal, 480, 141–160. https://doi.org/10.1042/BCJ20220474

Horne, C. R., Dite, T. A., Young, S. N., Mather, L. J., Dagley, L. F., Johnson, J. L., … Murphy, J. M. (2025). PSKH1 kinase activity is differentially modulated via allosteric binding of Ca²⁺ sensor proteins. Proceedings of the National Academy of Sciences. https://doi.org/10.1073/pnas.2420961122

Shrestha, S., Byrne, D. P., Harris, J. A., Kannan, N., & Eyers, P. A. (2020). Cataloguing the dead: Breathing new life into pseudokinase research. FEBS Journal, 287, 4150–4169. https://doi.org/10.1111/febs.15246

Unknown Authors. (2024). Illuminating the Regulation of the Dark Kinase PSKH1 (pp. 9–214). [Details unavailable].