1. Phylogeny  
   Serine/threonine‐protein kinase PAK2 belongs to the group I p21‐activated kinases, a subfamily that also includes PAK1 and PAK3. These kinases are members of the larger Ste20-related kinase family and are evolutionarily conserved across eukaryotes, with clear orthologs found in organisms ranging from yeast (e.g., the STE20 and Cla4 kinases) to multicellular animals. In mammalian systems, PAK2 is ubiquitously expressed, reflecting its fundamental role in numerous cellular processes. Comparative analyses indicate that group I PAKs share a high degree of sequence conservation, particularly within their C-terminal catalytic domains, while their N-terminal regulatory sequences—including the p21-binding domain (PBD) that overlaps with a Cdc42/Rac interactive binding (CRIB) motif—reflect adaptation to diverse regulatory inputs. The conservation of PAK2 across species and its close evolutionary relationship with PAK1 and PAK3 have been noted in studies tracking kinase evolution from yeast to man, underscoring the importance of these enzymes in relaying signals from small Rho GTPases to downstream effectors (abo1998pak4anovel pages 1-2, szczepanowska2009involvementofraccdc42pak pages 1-2, umarao2022cdc42racinteractivebinding pages 11-12).
2. Reaction Catalyzed  
   PAK2 catalyzes the phosphorylation of serine and threonine residues on its substrate proteins. The reaction proceeds as follows:  
     ATP + [protein]-(L-serine or L-threonine) → ADP + [protein]-(L-serine/threonine)-phosphate + H⁺.  
   This reaction is characteristic of serine/threonine-protein kinases and involves the transfer of the γ-phosphate from ATP to the hydroxyl group of serine or threonine residues in target proteins (gagnon2012molecularphysiologyof pages 1-2).
3. Cofactor Requirements  
   The catalytic activity of PAK2, like many kinases, is dependent on the presence of divalent cations, with Mg²⁺ serving as the primary cofactor. Mg²⁺ ions coordinate with ATP, facilitating the proper positioning of the phosphate group during the transfer reaction (gagnon2012molecularphysiologyof pages 1-2).
4. Substrate Specificity  
   PAK2 phosphorylates a variety of substrate proteins that are involved in key cellular processes such as cytoskeletal dynamics, cell cycle progression, and apoptosis. Although a definitive consensus sequence for PAK2 has not been firmly established in the literature, several substrates have been experimentally validated. For example, PAK2 has been shown to phosphorylate HACE1 at serine 385, a modification implicated in the regulation of Rac1 ubiquitination (acosta2018groupipaksmediatedphosphorylation pages 1-2). In addition, PAK2 phosphorylates components linked to the actin cytoskeleton, such as myosin light chain kinase (MLCK) and other proteins involved in focal adhesion dynamics, as well as key signaling intermediates such as MAPK4, MAPK6, and the transcription factor JUN (tian2019roleofrac1pak pages 90-99, serra2024exploringrhogtpasesa pages 185-187). Moreover, phosphorylation of histone H4 by PAK2 has been reported to contribute to chromatin assembly events. In many cases, the substrates of PAK2 appear to bear accessible serine or threonine residues within regions that may be targeted following docking interactions facilitated by the kinase’s regulatory domains. The substrate specificity of PAK2 is further modulated by the interaction of its substrates with upstream activators such as Rac1 and Cdc42, which may affect substrate accessibility in vivo (schzczepanowska2009involvementofraccdc42pak pages 2-4, tian2019roleofrac1pak pages 90-99).
5. Structure  
   The overall structure of PAK2 is organized into two major regions: an N-terminal regulatory segment and a C-terminal catalytic (kinase) domain. The N-terminal region contains the p21-binding domain (PBD) and a CRIB motif that mediate the binding of active, GTP-bound forms of the small Rho GTPases Rac1 and Cdc42. This regulatory domain also includes an auto-inhibitory domain (AID) that maintains the kinase in a low-activity, inhibited state under basal conditions. Upon binding of activated GTPases, a conformational change is elicited that relieves this autoinhibition and enables autophosphorylation events required for full activation. The C-terminal kinase domain of PAK2 adopts the typical bilobal architecture seen in protein kinases, with a smaller N-terminal lobe primarily composed of beta sheets and a larger C-terminal lobe rich in alpha helices. A key feature of the catalytic domain is the activation loop, which contains a critical threonine residue (Thr423) whose phosphorylation is essential for maximal kinase activity. Under apoptotic conditions, PAK2 is subject to cleavage by caspase-3, resulting in a constitutively active fragment that lacks the regulatory inhibitory domain (broeke2009alphaherpesvirusus3mediatedreorganization pages 4-5, abo1998pak4anovel pages 13-14, szczepanowska2009involvementofraccdc42pak pages 4-5).
6. Regulation  
   PAK2 is regulated primarily by intermolecular interactions and post-translational modifications. The binding of active (GTP-bound) Rac1 and Cdc42 to the CRIB domain of PAK2 serves as the principal regulatory mechanism; this interaction disrupts the intramolecular association between the auto-inhibitory domain and the catalytic domain, allowing for a conformational rearrangement that permits autophosphorylation. Key autophosphorylation events, including phosphorylation of the activation loop at Thr423, are necessary to achieve full kinase activation. In addition to autophosphorylation, phosphorylation by upstream kinases such as PDK1 has been implicated in further enhancing PAK2 activity. Under apoptotic conditions, PAK2 is cleaved by caspase-3, which generates a constitutively active fragment that can promote morphological changes associated with programmed cell death. Regulatory proteins such as adaptor molecules (for example, Nck and PIX) have been shown to interact with PAK2, targeting it to specific subcellular locations where it can exert its effects on the cytoskeleton. PAK2 is also a target of viral proteins; for instance, the HIV-1 Nef protein has been reported to associate with and activate PAK2, linking viral pathogenesis to the modulation of host cell signaling pathways (broeke2009alphaherpesvirusus3mediatedreorganization pages 4-5, demuth2010roleofp21activated pages 28-32, kuo2009structuralandfunctional pages 46-52).
7. Function  
   PAK2 plays multifaceted roles in the control of cellular physiology. At the cellular level, PAK2 is crucial for regulating the actin cytoskeleton. It facilitates cytoskeletal reorganization events including stress fiber disassembly, the formation of filopodia and lamellipodia, and the remodeling of focal adhesions. Such functions are central to processes of cell motility, adhesion, and migration. In addition to its direct effects on the cytoskeleton, PAK2 modulates signaling pathways that control cell proliferation and survival. Full-length PAK2 promotes cell survival and growth, whereas its apoptotically cleaved fragment can promote cell death through mechanisms that involve alterations in cellular morphology. PAK2 phosphorylates several key substrates: it activates MAPK4 and MAPK6, leading to the downstream activation of MAPKAPK5, which in turn regulates F-actin polymerization and cell migration; it phosphorylates JUN and histone H4, influencing transcriptional activation and chromatin assembly; and it targets HACE1, thereby affecting the ubiquitination status of Rac1. In the context of disease, PAK2 has been implicated in oncogenic signaling. Studies in aggressive B-cell lymphomas have demonstrated that PAK2 is overexpressed and constitutively activated, promoting proliferation and survival through modulation of downstream pathways such as PI3K/AKT and NF-κB. Moreover, PAK2 activity has been associated with chemoresistance in breast cancer, where it phosphorylates caspase-7 to inhibit apoptosis. Beyond cancer, PAK2’s role in cytoskeletal dynamics is exploited during viral infections, as viral proteins (for example, from alphaherpesviruses and HIV-1) activate PAK2 to induce actin reorganization for viral egress and cell-cell spread. PAK2 is ubiquitously expressed in mammalian tissues, and its interaction with small GTPases such as Rac1 and Cdc42 places it at a central node in multiple signaling cascades that govern both normal cell function and disease processes (tian2019roleofrac1pak pages 107-125, serra2024exploringrhogtpasesa pages 185-187, broeke2009alphaherpesvirusus3mediatedreorganization pages 5-6, szczepanowska2009involvementofraccdc42pak pages 1-2).
8. Other Comments  
   Several pharmacological inhibitors have been developed to target group I PAKs, including PAK2. FRAX597 is a selective inhibitor that has been used to attenuate PAK2 activity in the context of lymphomagenesis, where its use leads to a cytostatic effect in aggressive B-cell lymphoma cell lines. In addition, MRIA9, originally optimized from the G-5555 scaffold, acts as a dual inhibitor targeting both salt-inducible kinases (SIKs) and PAK2/3 with high selectivity and potency, and has been demonstrated to enhance the efficacy of chemotherapeutic agents such as paclitaxel in ovarian cancer cells. These inhibitors are valuable experimental tools for dissecting PAK2 signaling pathways and hold potential for clinical development as modulators of aberrant kinase activity in cancer and viral infections. In terms of disease associations, deregulation of PAK2 has been linked to various pathological conditions, including oncogenesis in aggressive lymphomas, breast cancer, and pancreatic malignancies, as well as contributing to the cytoskeletal reorganization required for viral pathogenesis. In addition, altered kinase networks that include PAK2 have been observed in neuropsychiatric conditions such as major depressive disorder, further underscoring the physiological and pathological relevance of this kinase (tian2019roleofrac1pak pages 107-125, tesch2021structurebaseddesignof pages 22-26, kuo2009structuralandfunctional pages 40-46, alnafisah2023alteredkinasenetworks pages 251-256).
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