

Phosphoprotein Phosphatases

Key References

- Barford, D. "Molecular mechanisms of the protein serine threonine phosphatases." *Trends Biochem. Sci.* **21**, 407-412 (1996).
- Cohen, P. "Classification of protein-serine/threonine phosphatases: Identification and quantitation in cell extracts." *Methods Enzymol.* **201**, 389-398 (1991).
- Cohen, P.T.W. "Novel protein serine/threonine phosphatases: Variety is the spice of life." *Trends Biochem. Sci.* **22**, 245-251 (1997).
- Fauman, E.B., Saper, M.A. "Structure and function of the protein tyrosine phosphatases." *Trends Biochem. Sci.* **21**, 413-417 (1996).
- Greengard, P. et al. "Beyond the dopamine receptor: the DARPP-32/protein phosphatase-1 cascade." *Neuron* **23**, 435-447 (1999).
- Keyse, S.M. "Protein phosphatases and the regulation of mitogen-activated protein kinase signalling." *Curr. Opin. Cell Biol.* **12**, 186-192 (2000).
- Matozaki, T., Kasuga, M. "Roles of protein-tyrosine phosphatases in growth factor signaling." *Cell Signal.* **8**, 13-19 (1996).
- Millward, T.A. et al. "Regulation of protein kinase cascades by protein phosphatase 2A." *Trends Biochem. Sci.* **24**, 186-191 (1999).
- Tonks, N.K., Neel, B.G. "From form to function: Signaling by protein tyrosine phosphatases." *Cell* **87**, 365-368 (1996).
- Unkeless, J.C., Jin, J. "Inhibitory receptors, ITIM sequences and phosphatases." *Curr. Opin. Immunol.* **9**, 338-343 (1997).
- Villafranca, J.E. et al. "Protein serine/threonine phosphatases." *Curr. Opin. Biotechnol.* **7**, 397-402 (1996).
- Wera, S., Hemmings, B.A. "Serine threonine protein phosphatases." *Biochem. J.* **311**, 17-29 (1995).

Overview

Phosphoprotein phosphatases comprise a large family of enzymes that hydrolyze the phosphoester bonds of phosphoserines, phosphothreonines or phosphotyrosines. While many phosphatases inhibit the activities of phosphorylation cascades, some activate them. The tyrosine phosphatase CD45, for example, activates Src family tyrosine kinases by dephosphorylating an inhibitory phosphotyrosine residue. Phosphoserine/threonine phosphatases comprise two gene families of metallo-enzymes: phosphoprotein phosphatase P (PPP) family and phosphoprotein phosphatase M family (PPM). Phosphotyrosine phosphatases (PTPs) and dual specificity phosphatases belong to several gene families which possess a conserved cysteine for catalysis and some conserved features of 3-dimensional structure.

The serine/threonine phosphatases of the PPP family are mostly regulated by protein-protein interactions. For example, in basal ganglia neurons, the PP1 catalytic subunit is inhibited by the phosphorylated form of DARPP-32 (32 kDa dopamine and cAMP-regulated protein), that is regulated by dopamine and several other neurotransmitters. The PP2A catalytic subunit forms heterotrimeric complexes with two other subunits and interacts with several inhibitory proteins. In contrast, the serine/threonine phosphatase calcineurin (PP2B) is directly activated by calcium and calmodulin.

A number of natural toxins are powerful inhibitors of PP1 and PP2A. Calcineurin is the target of two immunosuppressant drugs, cyclosporin A and FK506, complexed with their intracellular receptors (immunophilins), cyclophilin and FK506-binding protein (FKBP), respectively. In

contrast to the PPP family, the regulation and function of the PPM family are still poorly understood.

The "classical" PTPs form a large group of enzymes, many of which have a single transmembrane domain and a large extracellular domain. Although these structural features suggest that they may function as receptors, no soluble activating ligands for PTPs have been identified to date. However, it is known that transmembrane PTPs are involved in cell-cell or cell-matrix interactions and have properties in common with adhesion molecules. As such, they are implicated in neurite outgrowth, focal adhesions and adherens junctions. In addition to their physiological role, PTPs are used as weapons by pathogenic bacteria. For example, one of the virulence genes of the genus *Yersinia* (to which belongs the agent of bubonic plague) codes for a PTP.

Many non-receptor PTPs have targeting domains, including SH2 domains. In lymphocytes, for example, SH2-containing PTPs are recruited to specific tyrosine phosphorylated motifs (ITIMs - immunoreceptor tyrosine-based inhibition motifs) and participate in the modulation of the immune response.

The specific activities of PTPs are very high and these enzymes are tightly regulated within cells. Mechanisms of control may include inactivating dimerization and restriction to specific locations by precise targeting. Most of the pharmacological inhibitors of PTPs are nonspecific and include agents which mimic phosphorylated residues (e.g. orthovanadate) or which oxidize the catalytic cysteine.

Dual specificity phosphatases belong to two different groups. One comprises an enzyme of the vaccinia virus and its relatives in mammals. Members of this group are induced by the MAP kinase pathway and dephosphorylate MAP kinases, providing a negative feedback mechanism. Another group consists of Cdc25, a dual specificity phosphatase which plays a critical role in the control of cell cycle by removing the inhibitory phosphates on amino-terminal threonine and tyrosine residues of cyclin-dependent kinases (Cdks). At the present time, however, there are no specific pharmacological inhibitors of these phosphatases.

Phosphoprotein Phosphatases - Serine/Threonine Phosphatases

GENE FAMILY	PPP							PPM
TYPE	PP1 (P 7937)	PP2A	PP2B (C 1907) = calcineurin	PP4	PP5	PP6	PP7	PP2C
MOL. MASS CATAL. SUBUNIT (kDa)	~37	~36	~60	~35	~60	35	75	42-44
QUAT. STRUCTURE	Associated with targeting subunits	Heterotrimer: 1 catalytic sub. 2 regulatory sub.	Heterodimer: A: catalytic + CaM binding B: Ca ²⁺ binding	—	—	—	—	Monomer
EFFECT OF CATIONS	None	—	Activated by Ca ²⁺	—	—	—	Mg ²⁺ , Ca ²⁺	Activated by high Mg ²⁺
ENDOGENOUS INHIBITORS	Phospho-Inhibitor-1 Phospho-DARPP-32 Inhibitor-2 NIPP RIPP CPI17 Phosphatidic acid (P 9511)	I1PP2A I2PP2A	Cain AKAP-79 ^a DSCR1 ZAKI-4	—	—	—	—	—
PHARMACOLOGICAL INHIBITORS ^b	Calyculin A (C 5552) >Microcystin LR (M 2912) >Nodularin >Tautomycin >Okadaic acid (O 9381 , O 7760) >Cantharidin (C 7632)	Nodularin >Microcystin LR (M 2912) >Calyculin A (C 5552) ~Okadaic acid (O 9381 , O 7760) >Fostriecin (F 4425) >Tautomycin >Cantharidin (C 7632)	Cyclosporin A (C 3662) ^c FK506 ^d	Okadaic acid (O 9381 , O 7760) >Microcystin LR (M 2912) >Tautomycin >Calyculin A (C 5552) >Fostriecin (F 4425) >Cantharidin (C 7632)	Okadaic acid (O 9381 , O 7760) Microcystin LR (M 2912)	—	—	—

ABBREVIATIONS

AKAP-79: A kinase anchor protein 79
Cain: Calcineurin inhibitor
CaM: Calmodulin
CPI17: 17 kDa PKC-potentiated inhibitory protein of PPI
DARPP-32: Dopamine and cAMP-regulated phosphoprotein
DSCR1: Down syndrome critical region 1
FKBP: FK506 binding protein
I1PP2A: Protein inhibitor 1 of protein phosphatase 2A
I2PP2A: Protein inhibitor 2 of protein phosphatase 2A
NIPP: Nuclear inhibitor of protein phosphatase 1
PP: Phosphoprotein phosphatase
PP1: Phosphoprotein phosphatase 1
PP2A: Phosphoprotein phosphatase 2A
PP2B: Phosphoprotein phosphatase 2B or calcineurin
PP2C: Phosphoprotein phosphatase 2C
PPM: Phosphoprotein phosphatase M family
PPP: Phosphoprotein phosphatase P family
RIPP: Ribosomal inhibitor of protein phosphatase 1
ZAKI-4: Down syndrome critical region 1-like 1

FOOTNOTES

- a** Binding protein, some inhibitory effect.
b Inhibitors are listed in order of decreasing potency.
c Combined with cyclophilin.
d Combined with FKBP.

Phosphoprotein Phosphatases - Protein Tyrosine Phosphatases

GENE FAMILY	Classical PTPs Receptor-like	Classical PTPs Non-receptor	Low MW PTPs	VH-1 group	cdc-25 group
MOLECULAR WEIGHT (kDa)	Variable	Variable	~18	~20	30-40
SUBSTRATE SPECIFICITY	P-Tyr, broad	P-Tyr, broad	P-Tyr, broad	Dual specificities, MAP-kinases	Dual specificities, CDKs
INHIBITORS	Orthovanadate (S 6508) Pervanadate bpV(phen) Phenylarsine oxide (P 3075) Dephostatin (D 8065) PTP inhibitors I, II, III	Orthovanadate (S 6508) Pervanadate bpV(phen) Phenylarsine oxide (P 3075) Dephostatin (D 8065) PTP inhibitors I, II, III	Vanadate (S 6508) NO H ₂ O ₂ (H 1009)	Vanadate (S 6508)	Vanadate (S 6508)

ABBREVIATIONS

BpV(phen): Bisperoxo(1,10-phenanthroline)oxovanadate (V)

CDK: Cyclin-dependent kinase

PTP: Protein tyrosine phosphatase