Protein tyrosine phosphatases: from genes, to function, to disease

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Abstract | The protein tyrosine phosphatase (PTP) superfamily of enzymes functions in a coordinated manner with protein tyrosine kinases to control signalling pathways that underlie a broad spectrum of fundamental physiological processes. In this review, I describe recent breakthroughs in our understanding of the role of the PTPs in the regulation of signal transduction and the aetiology of human disease.

In this era of 'omics' research that has followed the sequencing of the human genome, families of signalling molecules are being defined and the complexities of the regulation of signal transduction are now becoming apparent. In contrast to initial views of signalling pathways as simple linear arrangements of phosphorylation cascades that function in isolation, it is now clear that there are multiple interacting signalling networks, the coordination of which in response to a stimulus determines physiological outcome. The importance of phosphorylation and the regulatory role of protein kinases is established; however, it has recently become apparent that protein phosphatases can no longer be viewed as passive housekeeping enzymes in these processes. Instead, the kinases and phosphatases are partners, and their activities are coordinated in the regulation of signalling responses. The distinct but complementary function of these enzymes is emphasized by recent studies in which the kinases have been implicated in controlling the amplitude of a signalling response, whereas phosphatases are thought to have an important role in controlling the rate and duration of the response[1,2].

Unlike the protein kinases, which are derived from a common ancestor, the protein phosphatases have evolved in separate families that are structurally and mechanistically distinct. Among these families, the serine/threonine phosphatases exist *in vivo* as a range of holoenzyme complexes, which consist of multiple combinations of catalytic and regulatory subunits that control a broad spectrum of signalling pathways[3]. These enzymes catalyse the direct hydrolysis of phosphosubstrate, a process that is facilitated by two metal ions at the active centre of the enzyme. The importance of the serine/threonine phosphatases was highlighted by their sensitivity to several inhibitors, such as the tumour promoter okadaic acid, which have become valuable tools in the characterization of phosphorylation-dependent signal transduction. Attention has been drawn recently to haloacid dehalogenases, which include phosphatases such as Eyes absent (Eya), which also functions as a transcription factor[4], and chronophin, which is a regulator of cofilin phosphorylation and, thereby, affects the dynamics of the actin cytoskeleton[5]. These enzymes catalyse dephosphorylation through an unusual mechanism that involves essential aspartic acid residues at the active site. Various phosphatases also regulate signal transduction through effects on non-protein substrates, including Src homology-2 (SH2)-domain-containing inositide phosphatases (SHIPs) and synaptojanins, which catalyse the dephosphorylation of inositol phospholipids[6].

In this review, I focus on the protein tyrosine phosphatases (PTPs), which are encoded by the largest family of phosphatase genes. These enzymes are defined by the active-site signature motif HCX_5R, in which the cysteine residue functions as a nucleophile and is essential for catalysis. Having described the composition of the PTP family in humans, and other organisms, attention has now turned to the analysis of their function. I highlight recent developments in our understanding of the signalling function of PTPs, including the regulation of receptor PTPs by ligands and the role of reversible oxidation in the control of PTP activity and, therefore, tyrosine phosphorylation *in vivo*. The crucial role of these enzymes as regulators of signal transduction has been further emphasized by recent examples of the disruption of PTP function as an underlying cause of human disease, including the identification of the first PTP oncogene. These developments reveal the potential for PTPs to represent a new class of therapeutic targets.

The composition and diversity of the PTP family

Since the completion of the sequence of the human genome, it has become possible to catalogue the genes that comprise the PTP superfamily (FIGS 1,2).

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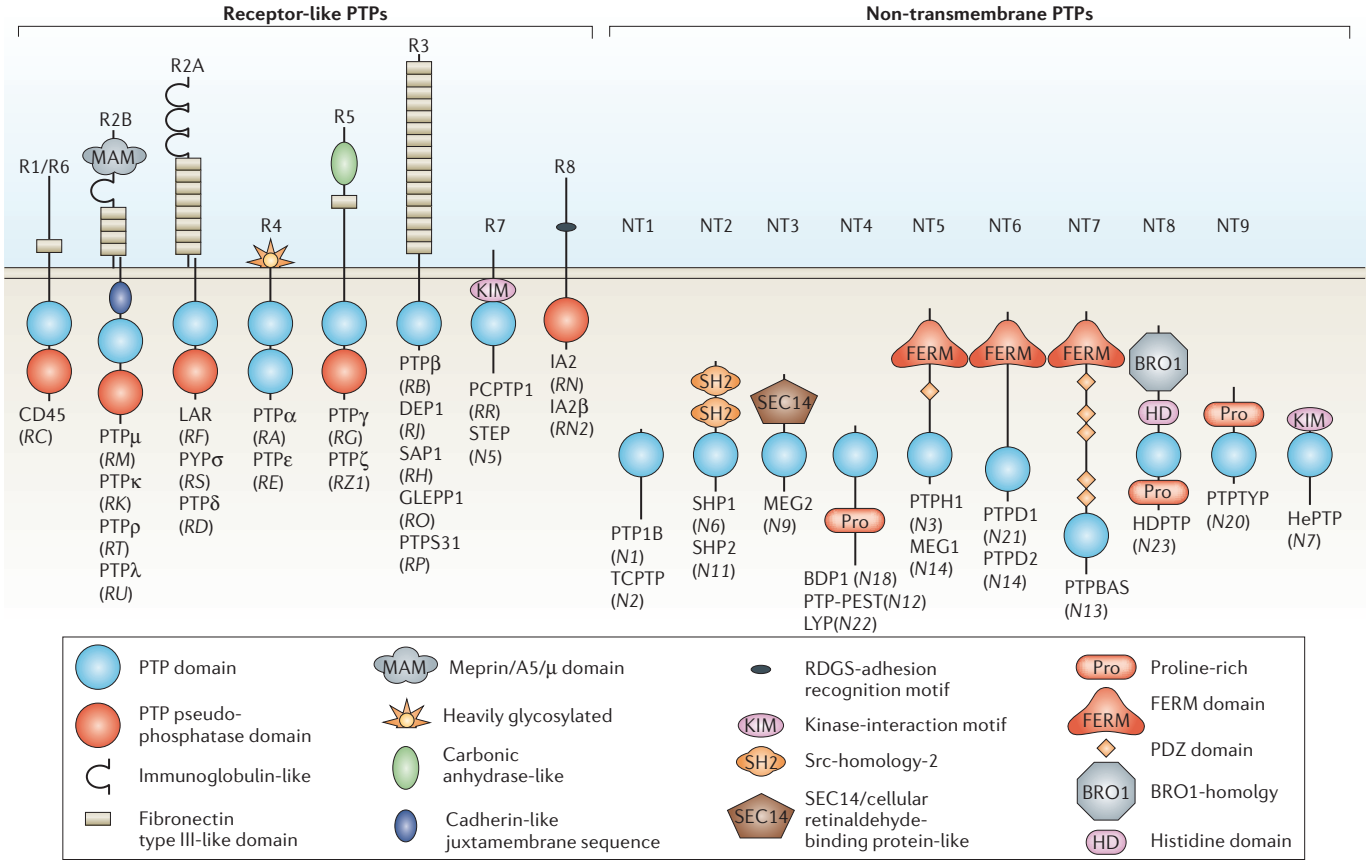


Figure 1 | **The classical PTPs.** The classical protein tyrosine phosphatases (PTPs) can be categorized as receptor-like (R) or non-transmembrane (NT) proteins. However, this is not an absolute distinction. For example, use of alternative promoters (as in the case of PTPε (*PTPRE*)) or alternative splicing (as in the case of GLEPP1 (*PTPRO*)) leads to the production of transmembrane and cytoplasmic forms of some PTPs from a single gene. In the PTPR7 subgroup, receptor-like and non-transmembrane variants of PCPTP1 (*PTPRR*) and STEP (*PTPN5*) have been described; however, as no receptor-like isoforms of HePTP (*PTPN7*) have been identified, this is included with the non-transmembrane enzymes. For those receptor-like PTPs (RPTP) with two intracellular PTP domains, the membrane-proximal D1 domain is catalytically active. In receptor subtype R4, PTPα is unique among the RPTPs in that the membrane-distal D2 domain also displays a low residual activity[117]. For the remaining RPTPs, including PTPε, which is the other enzyme in receptor subtype R4, the D2 domain maintains a PTP fold but lacks activity and can be classified as a pseudophosphatase domain. In each case, the PTPs have been designated by a name that is commonly used in the literature. Where this differs from the gene symbol, the latter is included in parentheses for clarification. In each case, the various subdivisions are based upon sequence similarity.

These encode enzymes that are divided into the classical, phosphotyrosine (pTyr)-specific phosphatases (FIG. 1) and the dual specificity phosphatases (DSPs)[7,8] (FIG. 2). Overall, there are approximately 100 human PTP-superfamily genes, compared to 90 human protein tyrosine kinase (*PTK*) genes, suggesting similar levels of complexity between the two families. However, the number of genes only illustrates the minimal level of complexity in the family, as additional diversity is introduced through the use of alternative promoters, alternative mRNA splicing and post-translational modifications. This structural diversity is indicative of the functional importance of the PTPs in the control of cell signalling. It is now apparent that the PTPs have the capacity to function both positively and negatively in the regulation of signal transduction. Furthermore, PTPs have the potential to display exquisite substrate, and functional, specificity *in vivo*. Therefore, the definition of the 'PTP-ome' provides a foundation for detailed analyses of the structure, regulation and physiological function of this important family of signal-transducing enzymes.

Classical pTyr-specific PTPs. The catalytic domain of classical PTPs comprises approximately 280 residues and is defined by several short sequence motifs, in particular the signature sequence that functions as a phosphate-binding loop at the active site[9]. The mouse and rat genomes contain 38 classical PTP genes, including osteotesticular (*OST*)-*PTP*, which are predicted to encode functional proteins. In humans, it seems that there are 37 classical *PTP* genes, with the status of *OST-PTP* being unclear[8]. The human *OST-PTP* sequence has greater than expected divergence from that of the functional rat and mouse gene and it has been suggested that in humans it is a pseudogene[10]. In addition, there are 12 *PTP* pseudogenes that are unique to humans, some of which are transcribed, but their function is unclear[8].

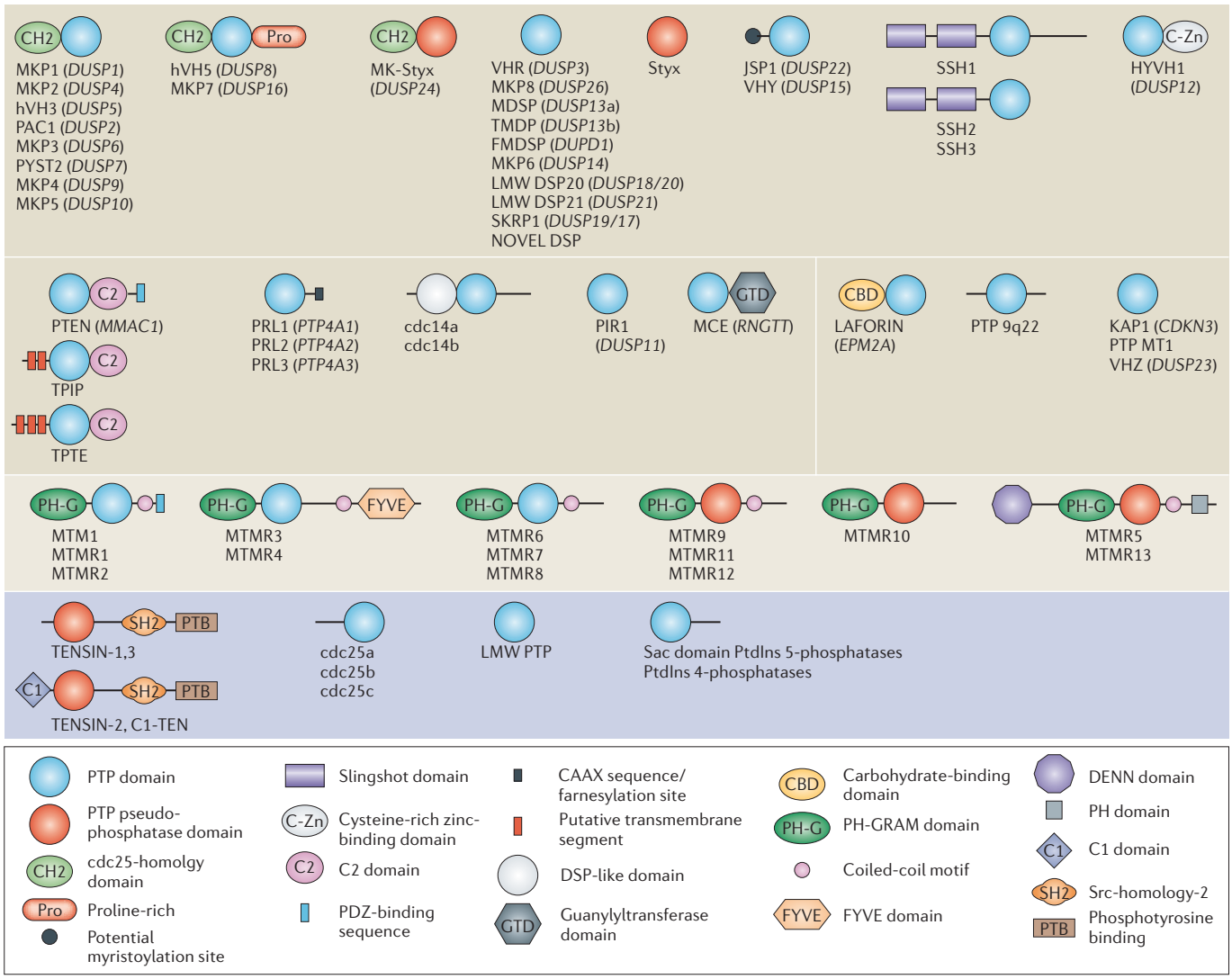
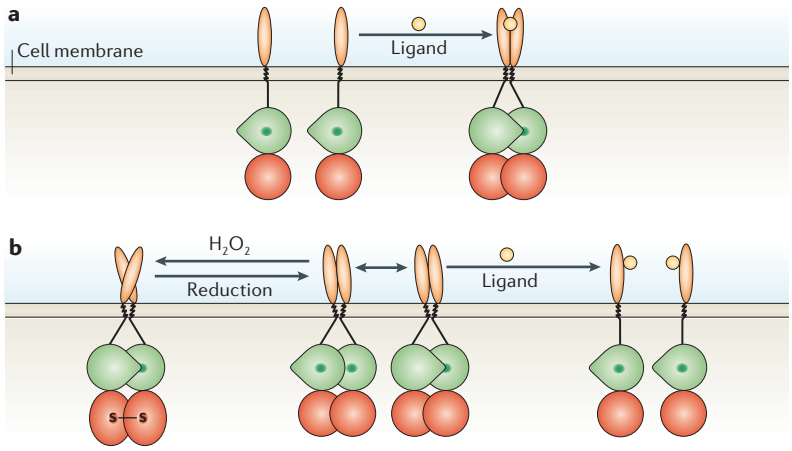


Figure 2 | **The dual specificity phosphatases.** The second category of protein tyrosine phosphatases (PTPs) are broadly defined as dual specificity phosphatases (DSPs). Although they are characterized by the presence of the $HC(X)_5R$ active-site signature motif, the DSPs are more structurally diverse than the classical PTPs and possess a smaller conserved catalytic domain. Like the classical PTPs, both catalytically-competent and inactive, pseudophosphatase DSP domains have been identified. Various non-catalytic motifs that serve a regulatory function are highlighted. In each case, the DSPs have been designated by a name that is commonly used in the literature. Where this differs from the gene symbol, the latter is included in parentheses for clarification. In each case, the various subdivisions are based upon sequence similarity. MTM, myotubularin; MTMR, myotubularin-related; PtdIns, phosphatidylinositol.

The classical PTPs include transmembrane receptor-like proteins (RPTPs) that have the potential to regulate signalling through ligand-controlled protein tyrosine dephosphorylation (FIG. 1). Many of the RPTPs display features of cell-adhesion molecules in their extracellular segment and have been implicated in processes that involve cell–cell and cell–matrix contact. Of the 21 RPTPs, 12 possess a tandem arrangement of PTP domains in their intracellular segment. So far, studies of these RPTPs indicate that all of the activity resides in the membrane-proximal (named D1) PTP domain. With the exception of PTPα[11], the membrane-distal (named D2) domain is inactive. Nevertheless, the structural integrity of the D2 domain is important for the activity, specificity and stability of the RPTP as a whole[12,13]. Furthermore, the D2 domain is important for protein–protein interactions that regulate RPTP dimerization[14,15].

There are also non-transmembrane, cytoplasmic PTPs (FIG. 1). These enzymes are characterized by regulatory sequences that flank the catalytic domain and control activity either directly, by interactions at the active site that modulate activity (as described below for SH2 domain-containing protein tyrosine phosphatase-2 (SHP2)), or by controlling substrate specificity (such as the interaction of PTP-PEST with $p130^{cas}$ (REF. 16) or STEP with the mitogen-activated protein kinase (MAPK) ERK[17]). These non-catalytic sequences also control subcellular distribution, thereby indirectly regulating activity by restricting access to particular substrates at defined subcellular locations.

Box 1 | **Regulation of receptor PTP function by dimerization**



In a similar manner to receptor protein tyrosine kinases (RPTKs), the activity of receptor protein tyrosine phosphatases (RPTPs) may be controlled by ligand-regulated dimerization (panel **a**). Ligand binding is shown as inhibiting RPTP activity by inducing dimerization. In the dimeric state, there is reciprocal inhibition of the catalytically-competent D1 PTP domains (green); the wedge motif of one D1 domain occludes the active site of the apposing D1 domain in the dimer. This is the converse of the situation for RPTKs, in which ligand-induced dimerization stimulates autophosphorylation and activation.

Recent data from studies of PTPα suggest another model of regulation of RPTP activity (panel **b**). PTPα was shown to exist predominantly as a homodimer on the cell surface[15,90]. A 'zipper model' was proposed in which interactions between multiple components of the PTP contribute to stabilization of the dimeric state, including the extracellular and transmembrane segments, as well as the D1 wedge and D2 (red) domains[15]. In this context, ligand binding would have the potential to disrupt dimer formation, thereby promoting PTP activation. The rotational coupling within the dimeric forms of PTPα determines the relative orientation of each monomer and, thereby, the activation status of the enzyme[91]. In some conformations, presumably those in which the orientation of the monomers precludes D1 wedge-motif-mediated inhibition, the enzyme may retain activity in the dimeric state[91].

A further level of regulation may be conferred by oxidative stress. Treatment of cells with H_2O_2 resulted in the stabilization of inactive PTPα dimers[92]. The cysteine residue at the active site of D2 (Cys723) is more sensitive to reactive oxygen species than the active site cysteine of D1, suggesting that D2 may function as a redox sensor[63]. Oxidation of Cys723 induces a conformational change in D2 (REF. 92) that then triggers the formation of an S–S bond between the active site cysteine residues of the D2 domains, thereby stabilizing the inactive dimer[64]. Oxidation changes the rotational coupling of the PTPα molecules, which results also in a change in the orientation of the extracellular segments within the dimer, consistent with the potential for 'inside-out' signalling[93]. Such redox regulation might also introduce a new tier of control through triggering the formation of heterodimers between distinct RPTPs[14]. Ultimately, these are reversible modifications. Following the reduction of the S–S bond that links the D2 domains in the dimer, the original conformations of the PTPα protein are restored.

Dual specificity phosphatases. There are approximately 65 genes that encode a heterogeneous group of phosphatases that are broadly described as DSPs (FIG. 2). These enzymes are less well conserved, have little sequence similarity beyond the cysteine-containing signature motif and possess smaller catalytic domains than the classical PTPs. In general, they share the same catalytic mechanism as the classical PTPs, but the construction of the DSP active site allows them to accommodate phosphoserine (pSer)/phosphothreonine (pThr) residues as well as pTyr residues in proteins. The DSPs include the VH1-like enzymes, which are related to the prototypic VH1 DSP, a 20 kDa protein that is a virulence factor of vaccinia virus[18].

One of the best characterized sub-divisions of these enzymes catalyses the inactivation of MAPKs by dephosphorylation of both tyrosine and threonine phosphorylation sites in the kinase activation loop. These MAPK phosphatases (MKPs) display distinct patterns of induction, subcellular location and specificity for individual MAPKs, thereby representing a response network of phosphatases that functions in attenuating MAPK-dependent signalling pathways. Nonetheless, in terms of physiological function, several of the DSPs may actually show preference for either tyrosine or serine/threonine residues such as KAP (cyclin-dependent kinase (CDK)-associated phosphatase), which dephosphorylates the threonine residue in the activation loop of CDKs (Thr160 in CDK1)[19], or the VH1-related DSP (VHR), which preferentially dephosphorylates tyrosine residues in MAPKs[20]. Some DSPs, such as the RNA-capping enzymes[21] and various phosphatidylinositol phosphatases (including the myotubularins (MTMs)), even target non-protein substrates[22].

Pseudophosphatase PTPs. Several members of the PTP superfamily possess conserved domains with core features of a PTP, but which lack residues that are critical for catalysis[23]. Recently there have been important developments in our understanding of the function of these proteins. The prototypic example, Styx, is catalytically inactive because it contains a glycine residue at the position expected for the active site cysteine. A single point mutation, to convert the glycine to cysteine, yields a mutant protein with activity that is similar to a *bona fide* DSP[23]. Styx interacts with the spermatid phosphoprotein CRHSP24 (calcium-regulated heat-stable protein of apparent molecular mass 24k) and mice that carry a germline mutation in Styx are defective for sperm production[24]. However, the molecular details of the function of this catalytically-impaired pseudophosphatase remain to be defined.

Several D2 domains of RPTPs maintain a PTP fold but lack residues that are critical for activity. For example, in LAR only two point mutations are required to convert its D2 domain from an inactive to an active PTP[25]. Similarly, two point mutations in the D2 of PTPα were sufficient to enhance its activity to the level of the D1 domain[11]. This situation is again reminiscent of examples encountered among the protein kinases, including STRAD, which is a Ste20-like pseudokinase that regulates the function of the tumour suppressor kinase LKB1, a member of the AGC family of kinases[26]. Like the RPTPs, the Janus PTKs (JAKs), which are critical regulators of cytokine and growth-factor signalling, contain both an active and a pseudokinase domain[27]; the latter suppresses the activity of the catalytic domain and is mutated in the myeloproliferative disease polycythemia vera, leading to enhanced JAK activity[27].

Within the PTP superfamily, pseudophosphatases are most prevalent among the MTMs[22,28] (FIG. 2). Of the 14 *MTM* genes in the human genome, 6 encode pseudophosphatases. Recently, it has been demonstrated that inactive MTMs form complexes with the active enzymes. For example, active MTM-related-2

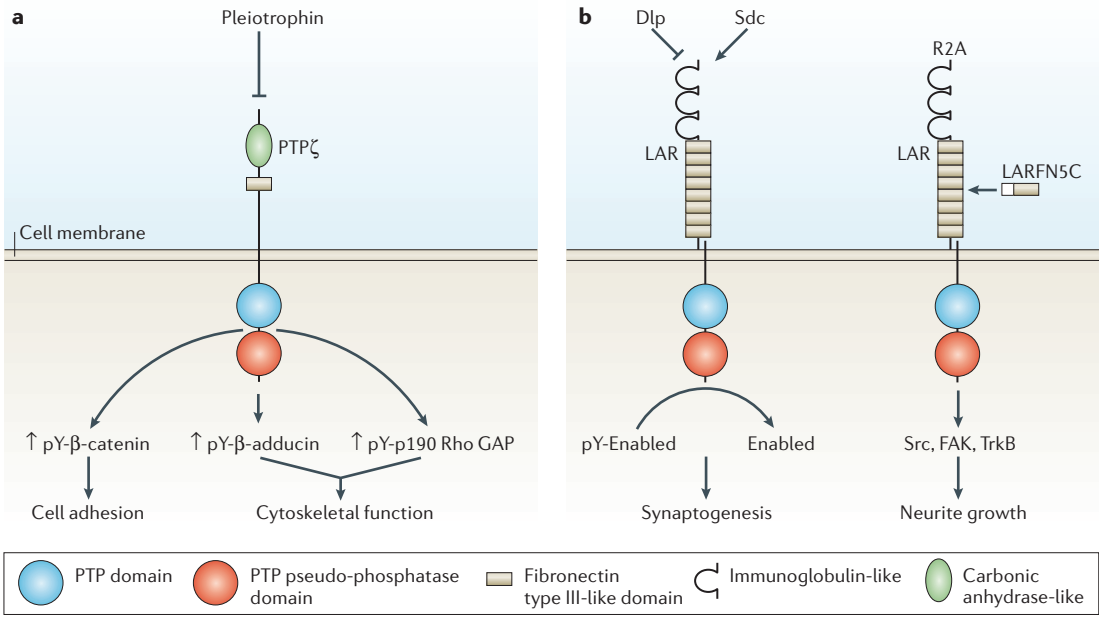


Figure 3 | **Regulation of receptor PTP function by ligands.** Recent studies have defined signalling pathways that are triggered by ligand-induced changes in the activity of receptor protein tyrosine phosphatases (RPTPs). **a** | The soluble cytokine pleiotrophin functions as a ligand for RPTPζ. Binding of pleiotrophin inhibits RPTPζ activity and promotes the tyrosine phosphorylation of proteins that regulate cell adhesion and cytoskeletal function. **b** | LAR, a cell-adhesion-molecule-like RPTP, can respond to surface bound or soluble ligands. A high-affinity interaction between the immunoglobulin (Ig) domains of LAR and the transmembrane protein syndecan (Sdc) promotes PTP function, whereas the glycosylphosphatidylinositol-anchored protein Dallylike (Dlp) also binds with high affinity to the Ig domains of LAR, but suppresses PTP function. These ligands modulate a common pathway — they antagonize the effects of the protein tyrosine kinase Abl and regulate the tyrosine phosphorylation of the LAR-associated protein Enabled, which in turn regulates the actin cytoskeleton and controls synapse morphogenesis and function. The soluble ligand LARFN5C binds homophilically to the fibronectin domains in the extracellular segment of LAR, activating various signalling pathways to promote neurite outgrowth. Whether or not these effects involve changes in the dimerization of LAR remains to be established.

(MTMR2) protein binds to the pseudophosphatases MTMR5 (REF. 29) and MTMR13 (REF. 30), whereas MTMR7 binds to the pseudophosphatase MTMR9 (REF. 31). These interactions regulate both the enzymatic activity and the subcellular location of the active phosphatase. Inactivating mutations in MTMR2 have been associated with type 4B Charcot–Marie–Tooth (CMT) disease, a neuropathy characterized by abnormal nerve myelination[22,28]. Now, in a particularly exciting development, it has been shown that mutations in the pseudophosphatase MTMR13, which probably result in the loss of the protein, also give rise to type 4B CMT disease[30,32], consistent with the functional importance of the phosphatase–pseudophosphatase interaction.

Recent structural studies have defined the molecular basis for the recognition of specific phosphoinositide substrates by the active MTMs[33]. Modelling analyses have revealed that although the fold is conserved, the residues in MTMR2 that coordinate substrate binding are not conserved in the pseudophosphatases[33]. Therefore, it is most likely that the MTM pseudophosphatases are inactive, and also, contrary to original suggestions, do not function in docking substrates. The active site of the MTMs is located on a positively charged face of the protein, whereas the area that surrounds the 'active site' of the pseudophosphatase MTMs is primarily negatively charged. Perhaps interaction between these charged faces may aid in the pseudophosphatase scaffolding function.

Regulation of RPTP function

As might be anticipated for a family of enzymes that play critical roles in the regulation of cell signalling, the activity of PTPs is tightly controlled *in vivo* by a variety of mechanisms. An important aspect of this regulation was revealed by the existence of receptor-linked PTPs, through which signal transduction may be regulated by ligand-controlled dephosphorylation of tyrosyl residues in proteins. Recently, we have witnessed great progress in defining how RPTPs may be controlled by ligand binding, the identity of ligands for RPTPs and some of the signalling events that these phosphatases may regulate.

The role of RPTP dimerization. The solution of the crystal structure of the membrane-proximal PTP domain of RPTPα represented an important development for the field. Within the crystal, the PTP domains were organized in symmetrical dimers, in which an inhibitory helix–turn–helix wedge motif from one domain occluded the active site of the partner domain[34]. It was proposed that

Table 1 | **Reversible oxidation of PTPs in response to various stimuli**

PTP	Stimulus-inducing oxidation	Citation
PTP1B	EGF	119
PTP1B	Insulin	120,121
TCPTP	Insulin	121
SHP2	PDGF	122
SHP2	T-cell receptor stimulation	123
SHP2	Endothelin-1	124
SHP1	B-cell receptor stimulation	125
CD45	B-cell receptor stimulation	125
PTPκ	Ultraviolet irradiation	126
PTP-PEST	Endothelial cell migration	127
MKPs	TNFα	68
MKPs	Neuronal oxidative stress	128
PTEN	PDGF, EGF, insulin	129
PTEN	Insulin	129, 130
Low-molecular-mass PTP	PDGF	131
Low-molecular-mass PTP	Integrin engagement	132
Low-molecular-mass PTP	Activated Rac mutants	133

EGF, epidermal growth factor; MKP, mitogen-activated-protein-kinase phosphatase; PDGF, platelet-derived growth factor; PTP, protein tyrosine phosphatase; SHP, Src-homology-2-domain-containing protein tyrosine phosphatase; TNFα, tumour-necrosis factor-α.

in a dimeric state the catalytic activity of RPTPs might be attenuated by reciprocal occlusion of the active sites. Such a model presents an appealing contrast to RPTKs, which are activated by ligand-induced dimerization[35]. Therefore, regulation of RPTP dimerization by ligand binding may directly modulate phosphatase activity and the phosphorylation of important downstream signalling molecules (BOX 1).

Many RPTPs contain two intracellular PTP domains, D1 and D2, which raises the question of whether constructs that contain both domains would dimerize in the same manner as the isolated D1 domain of PTPα. LAR is a RPTP that displays features of a cell-adhesion molecule and has been implicated in control of cell–cell and cell–matrix interactions[36]. The crystal structure of the tandem D1 and D2 domains of LAR revealed that although the wedge motif was present in this structure, there was no evidence of dimerization in the crystal[25]. In addition, the D1 and D2 domains were orientated such that both active sites were accessible, and steric hindrance caused by the presence of D2 would prevent wedge-mediated dimerization of D1. The two domains were tightly packed against each other, constrained by a short linker sequence and an array of non-covalent interactions, which led the authors to propose that this orientation would also be favoured in solution.

The structure of the tandem D1 and D2 domains of the prototypic RPTP CD45 has now also been solved[37]. The orientation of the D1 and D2 domains is very similar to that of LAR and was observed in crystals in two different space groups with distinct crystallographic contacts between neighbouring molecules. Again, the two domains are joined by a short linker and the domain interface is stabilized by extensive non-covalent interactions, suggesting that this reflects a favoured orientation. Nevertheless, it is important to remember that in RPTPs *in vivo*, the PTP domains are connected through a transmembrane segment to an extracellular segment, which may influence the orientation of D1 and D2 and the accessibility of the active site of D1 to a wedge motif in a dimer. In fact, studies of PTPα showed that multiple segments of the protein, including the D2 domain as well as the extracellular and transmembrane segments, contribute to the dimeric state[15].

What is the function of the wedge? Using a chimeric molecule comprising the extracellular segment of the epidermal growth factor (EGF) receptor fused to the intracellular segment of CD45, it was shown that ligand-induced dimerization inhibited the function of the RPTP in the regulation of T-cell signalling and that this inhibitory effect was attenuated by the mutation of residues in the wedge motif[38]. To investigate further the importance of the inhibitory wedge in the regulation of CD45 function, a knock-in mouse was generated in which Glu613, which is predicted to be at the tip of the wedge, was mutated to arginine. This mutation would be expected to prevent the wedge-mediated inhibition of CD45 in a dimer. Mice that express the CD45-Glu613Arg mutant seemed normal early in life, but with age display lymphocyte activation, developing a lymphoproliferative syndrome and ultimately a lupus-like autoimmune disease[39].

More recent studies have shown that genetic elimination of B cells in these knock-in mice ablated the lymphoproliferative disorder[40]. The consequences of expressing CD45-Glu613Arg in the knock-in mice are essentially the opposite of those observed following knockout of the CD45 gene, consistent with the notion that the wedge mutation removed an inhibitory constraint on CD45 function. In CD45-knockout mice, B cells show diminished signalling responses to stimulation of the B-cell receptor (BCR). By contrast, CD45-Glu613Arg B cells are hyperproliferative and display activated Ca^{2+} and MAPK signalling, which manifests itself in a disruption of B-cell development.

The physiological functions of CD45 have been linked to the control of the phosphorylation status of tyrosyl residues in the Src family of PTKs[41]. The autoimmune phenotype of CD45-Glu613Arg mice resembles the effect of mutating negative regulators of BCR signalling, such as the Src-family PTK Lyn, and can be explained in terms of the phosphorylation status of this PTK. Src-family PTKs are regulated by two sites of tyrosine phosphorylation: an autophosphorylation site in the activation loop, which promotes activity, and an inhibitory site at the C terminus. In CD45-knockout mice, in which BCR signalling is attenuated, the autophosphorylation site of Lyn is hyperphosphorylated, whereas there is decreased phosphorylation of the C-terminal inhibitory site. Therefore, the function of Lyn as an inhibitor of BCR signalling is potentiated. By contrast, the CD45-Glu613Arg mice display the opposite phenotype; the autophosphorylation site

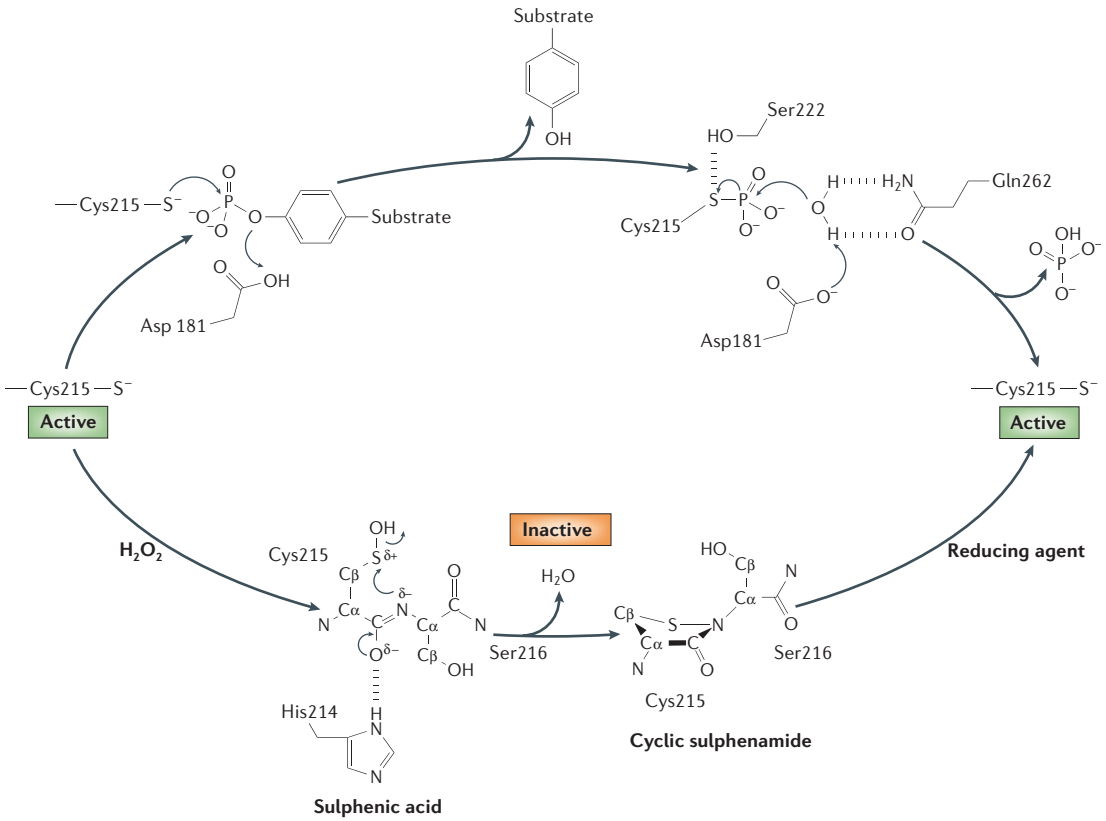


Figure 4 | **Reversible oxidation of PTPs.** All members of the protein tyrosine phosphatase (PTP) family use the same basic catalytic mechanism. The $HC(X)_5R$ signature motif contains the essential nucleophilic cysteinyl residue, forms the base of the active-site cleft and recognizes the phosphate of the target substrate. Catalysis proceeds through a two-step mechanism that involves the production of a cysteinyl-phosphate intermediate. In the first step, there is nucleophilic attack on the phosphate by the sulphur atom of the thiolate ion of the essential cysteine residue (Cys215 in PTP1B). This is coupled with protonation of the tyrosyl leaving group of the substrate by the conserved aspartic acid residue (Asp181 in PTP1B). The second step involves the hydrolysis of the phosphoenzyme intermediate, mediated by a glutamine residue (Gln262 in PTP1B), which coordinates a water molecule, and Asp181, which now functions as a general base, culminating in the release of phosphate. Oxidation of the active site cysteine residue abrogates its nucleophilic function, thereby inhibiting PTP activity. As shown for PTP1B, and presumably reflective of the classical PTPs in general, oxidation is reversible due to the rapid conversion of the sulphenic acid form of the oxidised cysteine to a novel 5-atom-ring structure, a cyclic sulphenamide, which is promoted by the environment of the catalytic site. In particular, the juxtaposition of His214 with Cys215 polarizes the amide bond, promoting nucleophilic attack by the amide nitrogen of Ser216 on the sulphur atom of the Cys215 sulphenic acid, leading to condensation and formation of a covalent bond between the sulphur and nitrogen atoms. The cyclic sulphenamide can be readily reduced to the active, thiolate form of the active-site cysteine residue.

is hypophosphorylated, whereas the C-terminal site is hyperphosphorylated, consistent with Lyn being functionally inactive in the presence of the wedge mutant. These observations suggest that the primary function of CD45 in B cells is to regulate Lyn and may explain why the CD45-Glu613Arg phenotype mirrors that of Lyn-deficient mice. The authors of the study propose that CD45 functions as a rheostat to set signalling thresholds during B-cell development. They conclude that the wedge motif negatively regulates CD45 activity, enabling maximal activation of Lyn, which subsequently functions to attenuate BCR signalling. These data clearly show the regulatory importance of the wedge. However, whether the mechanism involves dimer-induced inhibition or some other effect, such as controlling the interaction with substrates, remains to be determined.

Ligands for RPTPs. If dimerization is considered as a mechanism for regulation of RPTP activity, this raises the question of the identity of potential ligands. In the case of CD45, which is highly glycosylated and comprises up to 10% of the surface of haematopoietic cells, lectins, such as CD22, may bind to the extracellular segment. Nevertheless, these interactions do not appear to modulate PTP activity[41]. The glycosylation of the extracellular segment of CD45, and therefore its size and shape, varies according to the alternative splicing of primarily three exons (designated A, B and C). The larger, more highly glycosylated and sialylated forms, such as RABC, which express all 3 exons, are less efficient at forming dimers than the smallest form, termed RO[42]. When the CD45-deficient T-cell line H45 was reconstituted with physiological levels of the RO

Lectins
Lectins are carbohydrate-binding proteins. Individual lectins show specificity for particular sugar structures.

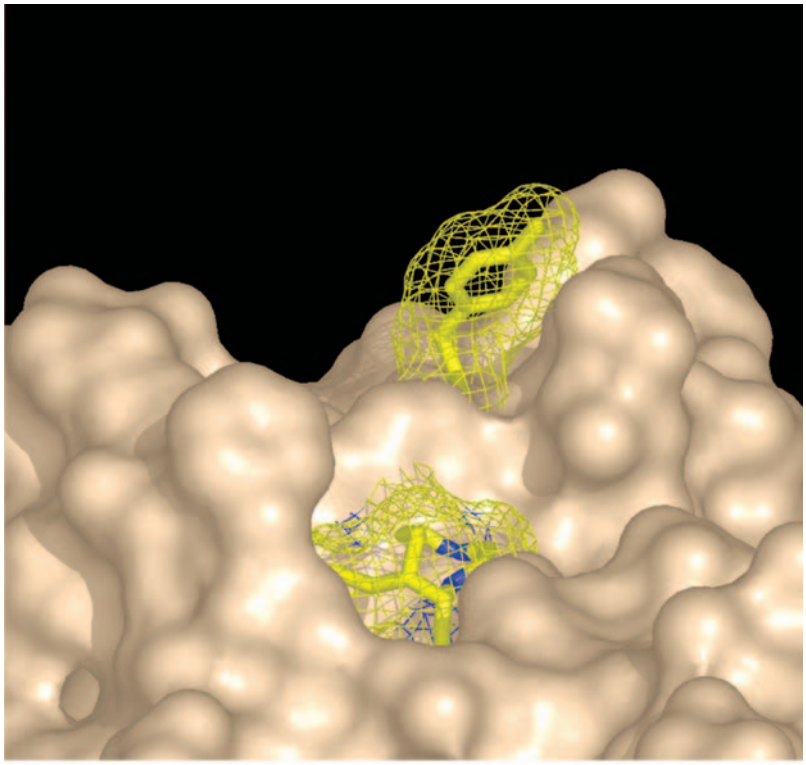


Figure 5 | **The structural consequences of oxidation at the PTP active site.** Incubation of protein tyrosine phosphatase-1B (PTP1B) crystals with stoichiometric quantities of H_2O_2 induced formation of the cyclic sulphenamide, which was accompanied by profound conformational changes at the active site[62,118]. The elements of the active site that change conformation are highlighted in yellow over the background of the structure of the reduced enzyme. The PTP loop, containing the signature motif, and Tyr46 from the phosphotyrosine binding loop, which are normally buried in the structure, flip out of the active site to adopt solvent exposed positions. These conformational changes are readily reversible, consistent with a mechanism for reversible regulation of PTP function. However, it will be important for future studies to establish whether this modification occurs *in vivo* and whether it is of regulatory significance.

and RABC isoforms, RO was found to dimerize more efficiently than RABC, but it was less effective than RABC at reconstituting signalling through the T-cell receptor[42]. This led to the proposal that there is an equilibrium between monomers and dimers of CD45 on the cell surface, with the activity of CD45 set by the differential dimerization of specific isoforms. However, as isoform exchange on the cell surface occurs on a timescale of days, this would reflect a mechanism for regulation in the long term[42]. Mutants that retain only the extracellular and transmembrane segments of CD45 maintained the capability to dimerize. Furthermore, the Glu613Arg-wedge mutation did not alter the extent of dimerization of the RO isoform of CD45. Therefore, the role of the wedge in modulating activity in this context, and potential mechanisms for acute regulation of CD45 activity, remain unclear.

The best characterized example of acute regulation of a RPTP by ligand binding is the inhibition of RPTPζ activity following binding of pleiotrophin (PTN)[43] (FIG. 3A), but whether this effect is mediated by PTP dimerization is unknown. Potential downstream substrates of RPTPζ that show increased phosphorylation in response to PTN include β-catenin[43], which is a component of the cadherin–catenin cell–adhesion complexes, and β-adducin[44], which is a cytoskeletal protein that regulates actin filaments. These findings suggest that some of the effects on cytoskeletal architecture triggered by PTN may be mediated by RPTPζ-induced changes in tyrosine phosphorylation. More recently, p190 Rho GAP, a GTPase-activating protein that is a potent inhibitor of Rho, was identified as a PTN-modulated substrate of RPTPζ[45]. Aberrant phosphorylation of p190 Rho GAP disrupts the Rho–ROCK (Rho-associated kinase) signalling pathway, which regulates cell morphogenesis through the remodelling of the actin cytoskeleton and may underlie the impairment of spatial learning and the enhancement of long-term potentiation in RPTPζ-deficient mice[46].

Further insights from neuronal systems. In an intriguing symmetry with the regulation of RPTKs[47], heparan sulphate proteoglycans (HSPGs) have now been recognized as playing an important role in regulating the function of RPTPs. For example, the HSPGs agrin and collagen XVIII were identified as ligands for RPTPσ in neurons[48]. More recently, the extracellular segment of LAR, a close relative of PTPσ, was used as a probe to compare the staining of wild-type and deletion mutant *Drosophila melanogaster* embryo slices, which identified the HSPG syndecan (Sdc) as a ligand for LAR[49]. This is a high-affinity interaction, with LAR displaying subnanomolar affinity for Sdc *in vitro*. *In vivo*, Sdc, which is a transmembrane cell surface protein on muscles, interacts with LAR in *trans* to regulate positively the function of this RPTP in motor-neuron growth cones, thereby mediating signalling events that are associated with the axon-guidance decisions that lead to muscle innervation[49] (FIG. 3B). In a separate study, Sdc was also identified as a high-affinity ligand that promoted LAR function during neuromuscular-junction formation in *D. melanogaster* larvae[50]. In the same study, a distinct HSPG, Dallylike (Dlp), which is a glycosylphosphatidylinositol-anchored glypican, was also identified as a high-affinity LAR ligand[50] (FIG. 3B). The tyrosine phosphorylated protein Enabled (Ena), which binds to the cytoplasmic segment of LAR, was identified as a substrate. RNA interference (RNAi)-mediated depletion of LAR enhanced Ena phosphorylation in KC167 *D. melanogaster* cells, whereas RNAi-mediated depletion of Dlp decreased Ena phosphorylation. Therefore, in contrast to Sdc, Dlp acts to inhibit LAR function at synapses. It appears that Sdc and Dlp influence a common signalling pathway in regulating synaptic morphogenesis and function by competing for binding to LAR and modulating PTP activity, with Dlp being the dominant effector *in vivo*[50] (FIG. 3B).

LAR displays features of an immunoglobulin (Ig)-superfamily cell-adhesion molecule, possessing an extracellular segment that comprises Ig and fibronectin type III (FNIII) domains[36]. The heterotypic ligand Sdc binds to the Ig domains, but not the FNIII domains of LAR[50]. Additional ligands have been identified that

Immunoglobulin (Ig) domain
This is a homology unit of ~110 amino acids. Although there is variation in primary sequence, the structure of this domain is conserved and organized into two β-sheets, one of three and the other of four strands, which are stabilized by disulphide bonds in most of the cases. Ig domains are found in several proteins, including cell-adhesion molecules and signalling receptors, and have been implicated in protein–protein interactions.

Box 2 | **PTPs as tumour suppressors**

In light of the prevalence of activated protein tyrosine kinases (PTKs) as oncoproteins, it was initially anticipated that many protein tyrosine phosphatases (PTPs) would be identified as tumour suppressors — nevertheless, such a discovery was slow in coming. The first example was PTEN, however its tumour suppressor function is associated with the dephosphorylation of the 3-position in the sugar head group of inositol phospholipids, thereby regulating phosphatidylinositol 3-kinase-dependent signalling pathways that are associated with cell survival[72]. More recently, several potential tumour suppressors have been identified among the classical PTPs, including receptor PTPs such as DEP1 (density-enhanced phosphatase-1, encoded by *PTPRJ*[94]) and PTPκ (encoded by *PTPRK*)[95] (TABLE 2). An analysis of mutations in *PTP* genes in colorectal cancers also identified several candidate tumour suppressors[96] — the most frequently mutated was the receptor PTPρ (encoded by *PTPRT*). In addition to inactivating mutations in the intracellular PTP domains, a large number of missense mutations were detected in the extracellular segment of this receptor-like PTP (RPTP), which highlights the importance of this segment, and its potential role in ligand binding, for the function of the enzyme. Similar observations were made for two other RPTPs, LAR (encoded by *PTPRF*) and PTPγ (encoded by *PTPRG*). The spectrum of potential tumour suppressor PTPs has now broadened with the observation of epigenetic regulation by hypermethylation of CpG sites in the promoters of several members of the PTP family[97]. This includes the receptor PTPRO (also known as GLEPP1) in hepatocellular carcinoma[98] and lung cancer[99], Src-homology-2-domain-containing protein tyrosine phosphatase-1 (SHP1; encoded by *PTPN6*) in leukaemia and lymphoma[100,101], FAP1 (encoded by *PTPN13*) in hepatocellular carcinoma[102] as well as the dual specificity phosphatases PTEN in breast cancer[103] and mitogen-activated-protein-kinase phosphatase-3 (MKP3; encoded by *DUSP6*) in pancreatic cancer[104]. It will be interesting to see whether it will be possible to target such gene-silencing events as a therapeutic strategy for cancer.

interact with the FNIII motifs. For example, the extracellular matrix laminin–nidogen complex binds to a specific spliced variant of LAR that includes a nine residue sequence in FNIII domain-5 (REF. 51). This interaction regulates morphology in HeLa cells[51]. Several other RPTPs also display features of Ig-superfamily cell-adhesion molecules and, as first shown for PTPμ[52,53], participate in homophilic binding interactions (that is, the ligand for the RPTP on the surface of one cell is a molecule of the same enzyme on an adjacent cell).

Further attention has been focused on LAR with the identification of a novel isoform of its extracellular segment, which contains a unique N-terminal sequence that is fused to the C-terminal portion of FNIII domain-5. This protein, designated LARFN5C, displays the capacity for homophilic binding and associates with full length LAR in neurons[54], promoting neurite outgrowth by activating multiple signalling pathways[55] (FIG. 3B). Characterization of the mechanisms that coordinate these various ligand-binding interactions will be the subject of future study.

Regulation of RPTP by reversible oxidation

Recently, the production of reactive oxygen species (ROS), such as hydrogen peroxide, and the resulting post-translational modification of proteins by reversible oxidation have been implicated in the regulation of tyrosine phosphorylation-dependent signalling pathways that are initiated by a wide variety of stimuli, including growth factors, hormones, cytokines and cellular stresses[56–58]. Attention was drawn to the PTPs as targets of ROS because the signature motif of this family, (I/V)HCXXGXXR(S/T), contains an invariant cysteine residue which, due to the unique environment of the PTP active site, is characterized by an extremely low pKα[59–61]. The low pKα means that this cysteine residue is present as the thiolate ion at neutral pH, which promotes its function as a nucleophile in catalysis but renders it highly susceptible to oxidation with concomitant abrogation of nucleophilic function and inhibition of PTP activity. This represents a new tier of control of tyrosine-phosphorylation-dependent signalling that is exerted at the level of the PTPs.

Mechanistic insights. Many PTPs have been shown to be oxidized transiently in response to various cellular stimuli (TABLE 1). Depending upon the extent of oxidation, the active-site cysteine in PTPs can be converted to either sulphenic (SOH), sulphinic (SO_2H) or sulphonic (SO_3H) acid. For oxidation to represent a mechanism for the reversible regulation of PTP function, it is essential that the active-site cysteine is not oxidized further than sulphenic acid, because higher oxidation is usually an irreversible modification. In the classical PTPs, such as PTP1B, we now have a molecular explanation of how this is achieved. Oxidation of the nucleophilic cysteine to sulphenic acid is accompanied by its conversion to a cyclic sulphenamide species (FIG. 4), which induces profound conformational changes at the PTP active site that both disrupt the interaction with substrate and expose the oxidized cysteine to the environment of the cell[62] (FIG. 5). This has the potential to serve the dual purpose of preventing irreversible oxidation to higher oxidized forms and facilitating the reduction of the sulphenamide to restore the active form of the PTP.

The dual specificity phosphatases cdc25c and PTEN, as well as the low-M_r PTP, are also sensitive to oxidation. Unlike the classical PTPs, these enzymes contain a second cysteine residue within the active site. Following oxidation of the nucleophilic cysteine within the signature motif, a disulfide bond is formed with the neighbouring cysteine protecting the enzymes from the irreversible inactivation that would result from higher oxidized species[60]. The S–S bond can be readily reduced, which ensures the transient nature of the modification and returns the enzymes to their active form.

Fibronectin type III (FNIII) domain
FNIII motifs, originally described in the extracellular matrix protein fibronectin, comprise ~90 amino acids. Their three-dimensional structure is similar to that of the immunoglobulin domain. In fibronectin itself, a RGD sequence in the loop that connects the β-sheets in FNIII motif 10 has been implicated in promoting adhesion by binding to integrins. This motif is also found in a wide variety of signalling proteins.

pKα
The pKα (also known as pKa) is a measure of the uptake/release of protons by amino acids. It is the negative log to the base 10 of the acid-dissociation constant, which reflects the equilibrium between protonation and deprotonation and indicates the extent of proton dissociation. The log scale is used because this constant differs over orders of magnitude between individual acids; the smaller the pKα value, the stronger the acid.

Table 2 | **PTPs and cancer**

PTP (encoding gene)	Tumour suppressing functions
PTEN (*MMAC1*)	Tumour suppressor mutated in various human cancers. Cowden disease
DEP1 (*PTPRJ*)	Colon cancer susceptibility locus SCC1. Deletions and mutations in human colon, lung and breast cancer
PTPκ (*PTPRK*)	Potential tumour suppressor in primary central nervous system lymphomas
PTPρ (*PTPRT*)	Potential tumour suppressor in colorectal cancers
LAR (*PTPRF*)	
PTPγ (*PTPRG*)	
PTPH1 (*PTPN3*)	
PTPBAS (*PTPN13*)	
PTPD2 (*PTPN14*)	
GLEPP1 (*PTPRO*)	Promoter methylation in lung tumours and hepatocellular carcinoma — potential tumour suppressor
SHP1 (*PTPN6*)	Promoter methylation in leukaemia and/or lymphoma — potential tumour suppressor
FAP1 (*PTPN13*)	Promoter methylation in hepatocellular carcinoma — potential tumour suppressor
SHP2 (*PTPN11*)	Oncogene in leukaemia. Target of *Helicobacter pylori* CagA protein in gastric carcinoma
MKP3 (*DUSP6*)	Candidate pancreatic tumour suppressor at locus 12q22. Promoter methylation
cdc25	Cell-cycle control. Target of Myc and overexpressed in primary breast cancer
PRL3 (*PTP4A3*)	Upregulated in metastases of colon cancer
(*PTPRR*)	*TEL* and *PTPRR* chimeric gene. It fuses exon 4 of the *TEL* gene with exon 7 of the *PTPRR* gene in acute myelogenous leukaemia

For the designation of protein tyrosine phosphatases (PTPs), the most common protein names were chosen and were used in the tables and throughout this article. Here they are accompanied by the systematic gene names in parentheses. DEP1, density-enhanced phosphatase-1; LAR, leukocyte common antigen related; MKP3, Mitogen-activated-protein-kinase phosphatase-3; PRL3, phosphatase of regenerating liver-3; SHP, Src-homology-2-domain-containing protein tyrosine phosphatase.

Oxidation of RPTPs. An interesting further aspect of control by oxidation is manifested in the RPTPs. For those RPTPs with a tandem arrangement of PTP domains in their intracellular segment, there are structural distinctions between the two domains that suggest differences in function. For example, all D2 domains are phylogenetically distinct from D1 domains, defining a separate subfamily of PTP domains[9]. Within the LAR RPTP subtype, the sequence similarity between the D2 domains of LAR, PTPσ and PTPδ is even higher than between the corresponding, catalytically-functional D1 domains[9]. The indication from phylogenetic analyses that the duplication that engendered the RPTPs that bear two PTP domains occurred early in evolution, together with this high level of conservation of the D2 domains, suggests a separate function from D1 domains. Recently, based on studies of RPTPα, it has been shown that the D2 domain displays greater sensitivity to oxidation than the D1 domain[63] and can form intermolecular (S–S) bonds following treatment with H_2O_2 that contribute to stabilization of PTP dimers[64]. Furthermore, oxidation induces a conformational change in D2 that can be transmitted to the extracellular segment of the receptor PTP, consistent with the potential for 'inside-out' signalling[59] (BOX 1). These observations suggest that D2 domains may function as redox sensors, and it will be interesting to explore whether this applies to other RPTPs. In addition, dissecting the mechanisms by which oxidation of D2 domains may affect the catalytic activity of D1 domains will help to define the signalling function of RPTPs as a whole.

Important unresolved issues. The current model is that physiological stimuli, such as growth factors or engagement of antigen receptors, trigger localized production of ROS. This leads to oxidation and inactivation of those PTPs that normally function to attenuate the signalling response, thereby promoting tyrosine phosphorylation and enhancing signalling. Oxidation is transient, with the PTPs being reduced back to their active state by the action of cellular reducing agents such as thioredoxin or glutathione, leading to signal termination.

Although particularly exciting as a new tier of control of pTyr-dependent signalling, there are several issues that must be resolved before the physiological importance of redox regulation of PTPs can be established. In particular, a prevalent view of ROS as agents of indiscriminate damage rather than as second messengers in cell signalling will have to be addressed. Tight control is exerted over the production of ROS by Nox (NADPH oxidase) enzymes, including requirement for assembly into a multiprotein complex with scaffold and activator proteins, phosphorylation of regulatory subunits, binding of inositol phospholipids and the small GTPase Rac[57]. Regulation of the Duox (dual oxidase) NADPH oxidases by Ca^{2+} adds further control[57]. Nevertheless, it will be essential to explain how ROS once produced display specificity for the oxidation of particular PTPs. Although the active site cysteine is particularly sensitive, it is likely that there will be intrinsic differences in sensitivity to oxidation between individual PTPs[65]. It is also likely that those PTPs that are colocalized with sites of ROS production will be preferentially oxidized. For example, it has been shown that Nox4 regulates the oxidation of PTP1B in response to insulin[66] and colocalizes with PTP1B on intracellular membranes[67]. Therefore, it will be important to define further where in the cell oxidation occurs, the precise source of ROS, such as Nox enzymes[66] or mitochondria[68], and how the localization of the PTPs is regulated. Furthermore, the studies of PTP oxidation reported to date have been conducted under O_2-rich conditions, rather than at the lower physiological O_2 concentrations that would be encountered in tissues. Interestingly, it has been reported that in cardiomyocytes, hypoxia induces an increase in ROS relative to normoxic conditions, as measured by DCF (2′,7′-dichlorodihydrofluorescein) fluorescence, due to an increased generation of ROS by mitochondria[69]. Nevertheless, it will be important to assess the effects of hypoxia on PTP oxidation.

Box 3 | **PTPN11, the first PTP oncogene**

Src-homology-2 (SH2) domain-containing protein tyrosine phosphatase-2 (SHP2) comprises two SH2 domains in its N-terminal segment, a protein tyrosine phosphatase (PTP) catalytic domain and a C-terminal tail, which contains two tyrosine phosphorylation sites and a proline-rich sequence and may participate in protein–protein interactions. Under basal conditions, SHP2 exists in a low-activity state in which the active site is occluded by an intramolecular interaction with the N-terminal SH2 domain. Engagement of the SH2 domains by phosphotyrosine (pTyr) residues on proteins that target the PTP to signalling complexes induces a conformational change that releases the autoinhibitory interaction and 'opens up' the structure to create an active form of the phosphatase that can now dephosphorylate substrates[105]. Gain-of-function mutations in SHP2, initially identified in patients with Noonan syndrome[106], facilitate activation of the PTP. Mutations have been noted in residues in and around the N-terminal SH2 domain, which may facilitate activation by pTyr ligands and in key residues at the interface between the N-terminal SH2 and catalytic domains, which disrupt the autoinhibitory interaction and may induce the active conformation in the absence of a stimulus[107].

SHP2 normally facilitates Ras activation, for example by dephosphorylating docking sites for Ras GTPase activating proteins (GAPs) in growth factor receptor protein tyrosine kinases (PTKs), by regulating the activity of Src-family PTKs, or at the level of Sprouty proteins[105,108]. Activating somatic mutations in *PTPN11* (which encodes SHP2) have been associated with increased risk of certain sporadic childhood malignancies, such as juvenile myelomonocytic leukaemia and acute myeloid leukaemia [109–111]. These gain-of-function mutations induce hypersensitivity to granulocyte-macrophage colony-stimulating factor (GM-CSF) in haematopoietic progenitor cells[112–114]. Furthermore, upon transplantation into lethally-irradiated mice, bone marrow expressing leukaemia-associated SHP2 mutants gives rise to a fatal invasive myeloproliferative disease that is associated with hyperactivation of the mitogen-activated protein kinase (MAPK) ERK and other signalling pathways[113]. Therefore, activating mutations in SHP2 directly cause leukaemia. LEOPARD syndrome, which shares clinical features with Noonan syndrome, is also associated with mutations in *PTPN11* (REF. 115); however, in this case the mutations cause loss of function in SHP2, generating mutant proteins that act as dominant negatives and interfere with ERK MAPK activation[116]. Future efforts will have to define in which cell populations, and at what stage of development, SHP2 exerts its effects, and will need to identify the critical substrates and signalling pathways that mediate these effects.

PTPs and human disease

It has long been appreciated that PTPs not only have the capacity to function as inhibitors of pTyr-dependent signalling, but also to act as positive regulators in promoting signalling, as in the case of CD45 (REF. 41). Several PTPs have been identified as the products of tumour suppressor genes[70] (BOX 2; TABLE 2). Now there are examples of aberrant upregulation of PTPs in human cancer; overexpression of the cell-cycle regulatory phosphatase cdc25 has been detected in multiple cancers, often correlating with poor prognosis[71]. An exciting new development has been the identification of the first oncogene that encodes a PTP. Just as several components of the phosphatidylinositol 3-kinase signalling pathway are targeted in cancer, or cancer-predisposition syndromes, including the tumour-suppressor phosphatase PTEN[72], it is now apparent that a variety of syndromes are explained in terms of aberrant regulation of the Ras–MAPK signalling pathway[73]. One aspect of this is gain-of-function mutations in the *PTPN11* gene, which encodes the SH2-domain-containing PTP SHP2 (BOX 3).

PTPN22/PTP Lyp and autoimmunity. It is important to note that aberrant regulation of PTP-superfamily members is also associated with diseases other than cancer[8]. In an exciting series of studies of the *PTPN22* gene, which encodes the PTP Lyp, a single nucleotide polymorphism (SNP) that generated an Arg620 to tryptophan point mutant was identified. This mutation has been determined to be a common risk factor for autoimmune diseases, including type I diabetes[74], Graves' disease[75], rheumatoid arthritis[76,77] and systemic lupus erythematosus[78]. *PTPN22* Lyp is expressed in haematopoietic cells and acts as an inhibitor of T-cell activation. Consistent with this, the autophosphorylation site of the PTKs Lck and ZAP70 and the T-cell receptor-ζ (TCRζ) chain have been identified as substrates of this phosphatase[79]. The initial characterization of the *PTPN22*-Arg620Trp SNP focused on the fact that this mutation occurs in a proline-rich stretch of the non-catalytic segment of the enzyme and disrupts an association with the SH3 domain of Csk, the Src C-terminal kinase[74]. Now it has been shown that this is actually a gain-of-function mutation that generates a more active PTP that is a more effective inhibitor of T-cell signalling than the wild-type enzyme[80]. However, the mechanism by which this mutation in the non-catalytic segment leads to activation of the phosphatase remains to be defined. It has been proposed that this activating mutation in *PTPN22* may cause a predisposition to autoimmune disease either by failure to delete autoreactive T cells or due to insufficient activity of regulatory T cells[80]. It will be interesting to pursue the possibility suggested by this analysis that small molecule inhibitors of PTP-Lyp may be of therapeutic benefit in autoimmune disease.

Future directions

Recently, we have witnessed important breakthroughs in the functional characterization of members of the PTP superfamily that have provided new perspectives on the regulation of signal transduction. Ligands for some RPTPs, and the signalling pathways that they regulate, have been identified and characterized. Now, there are specific examples of receptor PTPs that regulate signal transduction through ligand-controlled dephosphorylation of tyrosyl residues in proteins. Reversible oxidation is becoming established as a novel

mechanism for the control of PTP function. Future studies must define the links between oxidation of specific PTPs and the regulation of pTyr-dependent signalling in a physiological context. Of particular significance has been the definition of links between the disruption of PTP function and the aetiology of human disease.

As our understanding of the physiological importance of PTPs has matured, so has enthusiasm for the development of PTP-based therapeutics, fuelled in particular by compelling data that define PTP1B as an outstanding target for the treatment of diabetes and obesity[81]. However, the PTPs are challenging targets for the development of active-site-directed inhibitors. Their susceptibility to oxidation can cause problems in high-throughput screens and the tendency for potent inhibitors to be highly charged, such as non-hydrolysable pTyr mimetics, presents challenges with respect to bioavailability. Antisense-based therapeutics that target PTP1B have shown efficacy in type II diabetes and are now in phase 2 clinical trials[82,83], providing further validation of this target in humans. The future will no doubt see alternative approaches for therapeutic development, such as those involving secondary allosteric sites recently described for PTP1B[84]. The development of 'wedge domain' peptides as inhibitors of RPTPs, based upon the dimerization model of the regulation of PTP function, suggests a new strategy for PTP inhibition[85]. Recent data highlighting the importance of RPTPσ in regulating post-injury axon regrowth in the peripheral and central nervous system[86] suggest that in some circumstances there may also be utility for therapeutic agents that target the extracellular segment of RPTPs.

Nonetheless, the PTP field is still in its infancy and many of these enzymes remain largely uncharacterized. The description of the 'PTP-ome' will now facilitate functional analyses of the family as a whole, for example by RNAi[87]. The application of substrate-trapping mutant technology[88] will help to define the physiological substrate specificity of members of the PTP superfamily, as illustrated by the recent elucidation of the role of PTPRO/GLEPP1 in the dephosphorylation and inactivation of Eph receptor PTKs, and thereby in the regulation of retinal axon guidance in the developing nervous system[89]. As further progress is made in defining the signalling function of PTPs and elucidating novel links to human disease, it is anticipated that new insights into therapeutic development will be revealed, either at the level of the PTPs themselves or from targets within the pathways they regulate.

1. Heinrich, R., Neel, B. G. & Rapoport, T. A. Mathematical models of protein kinase signal transduction. *Mol. Cell* **9**, 957–970 (2002).
2. Hornberg, J. J. *et al.* Principles behind the multifarious control of signal transduction. ERK phosphorylation and kinase/phosphatase control. *FEBS J.* **272**, 244–258 (2005).
3. Cohen, P. T. W. in *Overview of Protein Serine/Threonine Phosphatases* (eds Arino, J. & Alexander D. R.) (Spring-Verlag, Berlin, 2004).
4. Epstein, J. A. & Neel, B. G. Signal transduction: an eye on organ development. *Nature* **426**, 238–239 (2003).
5. Wiggan, O., Bernstein, B. W. & Bamburg, J. R. A phosphatase for cofilin to be HAD. *Nature Cell Biol.* **7**, 8–9 (2005).
6. Hughes, W. E., Cooke, F. T. & Parker, P. J. Sac phosphatase domain proteins. *Biochem. J.* **350**, 337–352 (2000).
7. Alonso, A. *et al.* Protein tyrosine phosphatases in the human genome. *Cell* **117**, 699–711 (2004).
8. Andersen, J. N. *et al.* A genomic perspective on protein tyrosine phosphatases: gene structure, pseudogenes, and genetic disease linkage. *FASEB J.* **18**, 8–30 (2004).
9. Andersen, J. N. *et al.* Structural and evolutionary relationships among protein tyrosine phosphatase domains. *Mol. Cell. Biol.* **21**, 7117–7136 (2001). **References 7, 8 and 9 provide comprehensive overviews of the structure, regulation and function of the PTP superfamily.**
10. Cousin, W., Courseaux, A., Ladoux, A., Dani, C. & Peraldi, P. Cloning of hOST-PTP: the only example of a protein-tyrosine-phosphatase the function of which has been lost between rodent and human. *Biochem. Biophys. Res. Commun.* **321**, 259–265 (2004).
11. Buist, A. *et al.* Restoration of potent protein-tyrosine phosphatase activity into the membrane-distal domain of receptor protein-tyrosine phosphatase α. *Biochemistry* **38**, 914–922 (1999).
12. Felberg, J. & Johnson, P. Characterization of recombinant CD45 cytoplasmic domain proteins. Evidence for intramolecular and intermolecular interactions. *J. Biol. Chem.* **273**, 17839–17845 (1998).
13. Streuli, M., Krueger, N. X., Thai, T., Tang, M. & Saito, H. Distinct functional roles of the two intracellular phosphatase like domains of the receptor-linked protein tyrosine phosphatases LCA and LAR. *EMBO J.* **9**, 2399–2407 (1990).
14. Blanchetot, C., Tertoolen, L. G., Overvoorde, J. & den Hertog, J. Intra- and intermolecular interactions between intracellular domains of receptor protein-tyrosine phosphatases. *J. Biol. Chem.* **277**, 47263–47269 (2002).
15. Jiang, G., den Hertog, J. & Hunter, T. Receptor-like protein tyrosine phosphatase α homodimerizes on the cell surface. *Mol. Cell. Biol.* **20**, 5917–5929 (2000).
16. Garton, A. J., Burnham, M. R., Bouton, A. H. & Tonks, N. K. Association of PTP-PEST with the SH3 domain of p130cas; a novel mechanism of protein tyrosine phosphatase substrate recognition. *Oncogene* **15**, 877–885 (1997).
17. Pulido, R., Zuniga, A. & Ullrich, A. PTP-SL and STEP protein tyrosine phosphatases regulate the activation of the extracellular signal-regulated kinases ERK1 and ERK2 by association through a kinase interaction motif. *EMBO J.* **17**, 7337–7350 (1998).
18. Guan, K. L., Broyles, S. S. & Dixon, J. E. A Tyr/Ser protein phosphatase encoded by vaccinia virus. *Nature* **350**, 359–362 (1991).
19. Poon, R. Y. & Hunter, T. Dephosphorylation of Cdk2 Thr160 by the cyclin-dependent kinase-interacting phosphatase KAP in the absence of cyclin. *Science* **270**, 90–93 (1995).
20. Schumacher, M. A., Todd, J. L., Rice, A. E., Tanner, K. G. & Denu, J. M. Structural basis for the recognition of a bisphosphorylated MAP kinase peptide by human VHR protein phosphatase. *Biochemistry* **41**, 3009–3017 (2002).
21. Wen, Y., Yue, Z. & Shatkin, A. J. Mammalian capping enzyme binds RNA and uses protein tyrosine phosphatase mechanism. *Proc. Natl Acad. Sci. USA* **95**, 12226–12231 (1998).
22. Begley, M. J. & Dixon, J. E. The structure and regulation of myotubularin phosphatases. *Curr. Opin. Struct. Biol.* **15**, 614–620 (2005). **Excellent review of the myotubularin subfamily of phosphatases.**
23. Wishart, M. J. & Dixon, J. E. Gathering STYX: phosphatase-like form predicts functions for unique protein-interaction domains. *Trends Biochem. Sci.* **23**, 301–306 (1998).
24. Wishart, M. J. & Dixon, J. E. The archetype STYX/dead-phosphatase complexes with a spermatid mRNA-binding protein and is essential for normal sperm production. *Proc. Natl Acad. Sci. USA* **99**, 2112–2117 (2002).
25. Nam, H. J., Poy, F., Krueger, N. X., Saito, H. & Frederick, C. A. Crystal structure of the tandem phosphatase domains of RPTP LAR. *Cell* **97**, 449–457 (1999).
26. Alessi, D. R., Sakamoto, K. & Bayascas, J. R. LKB1-dependent signaling pathways. *Annu. Rev. Biochem.* **75**, 137–163 (2006).
27. Shannon, K. & Van Etten, R. A. JAKing up hematopoietic proliferation. *Cancer Cell* **7**, 291–293 (2005).
28. Laporte, J., Bedez, F., Bolino, A. & Mandel, J. L. Myotubularins, a large disease-associated family of cooperating catalytically active and inactive phosphoinositides phosphatases. *Hum. Mol. Genet.* **12**, R285–R292 (2003).
29. Kim, S. A., Vacratsis, P. O., Firestein, R., Cleary, M. L. & Dixon, J. E. Regulation of myotubularin-related (MTMR)2 phosphatidylinositol phosphatase by MTMR5, a catalytically inactive phosphatase. *Proc. Natl Acad. Sci. USA* **100**, 4492–4497 (2003).
30. Robinson, F. L. & Dixon, J. E. The phosphoinositide-3-phosphatase MTMR2 associates with MTMR13, a membrane-associated pseudophosphatase also mutated in type 4B Charcot–Marie–Tooth disease. *J. Biol. Chem.* **280**, 31699–31707 (2005).
31. Mochizuki, Y. & Majerus, P. W. Characterization of myotubularin-related protein 7 and its binding partner, myotubularin-related protein 9. *Proc. Natl Acad. Sci. USA* **100**, 9768–9773 (2003).
32. Azzedine, H. *et al.* Mutations in MTMR13, a new pseudophosphatase homologue of MTMR2 and Sbf1, in two families with an autosomal recessive demyelinating form of Charcot–Marie–Tooth disease associated with early-onset glaucoma. *Am. J. Hum. Genet.* **72**, 1141–1153 (2003).
33. Begley, M. J. *et al.* Molecular basis for substrate recognition by MTMR2, a myotubularin family phosphoinositide phosphatase. *Proc. Natl Acad. Sci. USA* **103**, 927–932 (2006).
34. Bilwes, A. M., den Hertog, J., Hunter, T. & Noel, J. P. Structural basis for inhibition of receptor protein-tyrosine phosphatase-α by dimerization. *Nature* **382**, 555–559 (1996).
35. Weiss, A. & Schlessinger, J. Switching signals on or off by receptor dimerization. *Cell* **94**, 277–280 (1998).
36. Chagnon, M. J., Uetani, N. & Tremblay, M. L. Functional significance of the LAR receptor protein tyrosine phosphatase family in development and diseases. *Biochem. Cell Biol.* **82**, 664–675 (2004).
37. Nam, H. J., Poy, F., Saito, H. & Frederick, C. A. Structural basis for the function and regulation of the receptor protein tyrosine phosphatase CD45. *J. Exp. Med.* **201**, 441–452 (2005).

38. Majeti, R., Bilwes, A. M., Noel, J. P., Hunter, T. & Weiss, A. Dimerization-induced inhibition of receptor protein tyrosine phosphatase function through an inhibitory wedge. *Science* **279**, 88–91 (1998).
This is a highly significant study, which, together with reference 91, provides important insights into dimerization as a potential mechanism for the regulation of receptor PTP function.
39. Majeti, R. *et al.* An inactivating point mutation in the inhibitory wedge of CD45 causes lymphoproliferation and autoimmunity. *Cell* **103**, 1059–1070 (2000).
40. Hermiston, M. L., Tan, A. L., Gupta, V. A., Majeti, R. & Weiss, A. The juxtamembrane wedge negatively regulates CD45 function in B cells. *Immunity* **23**, 635–647 (2005).
41. Hermiston, M. L., Xu, Z. & Weiss, A. CD45: a critical regulator of signaling thresholds in immune cells. *Annu. Rev. Immunol.* **21**, 107–137 (2003).
42. Xu, Z. & Weiss, A. Negative regulation of CD45 by differential homodimerization of the alternatively spliced isoforms. *Nature Immunol.* **3**, 764–771 (2002).
43. Meng, K. *et al.* Pleiotrophin signals increased tyrosine phosphorylation of β-catenin through inactivation of the intrinsic catalytic activity of the receptor-type protein tyrosine phosphatase β/ζ. *Proc. Natl Acad. Sci. USA* **97**, 2603–2608 (2000).
An important study, characterizing the regulation of a receptor PTP by ligand binding.
44. Pariser, H., Perez-Pinera, P., Ezquerra, L., Herradon, G. & Deuel, T. F. Pleiotrophin stimulates tyrosine phosphorylation of β-adducin through inactivation of the transmembrane receptor protein tyrosine phosphatase β/ζ. *Biochem. Biophys. Res. Commun.* **335**, 232–239 (2005).
45. Tamura, H., Fukada, M., Fujikawa, A. & Noda, M. Protein tyrosine phosphatase receptor type Z is involved in hippocampus-dependent memory formation through dephosphorylation at Y1105 on p190 RhoGAP. *Neurosci. Lett.* **399**, 33–38 (2006).
46. Niisato, K. *et al.* Age-dependent enhancement of hippocampal long-term potentiation and impairment of spatial learning through the Rho-associated kinase pathway in protein tyrosine phosphatase receptor type Z-deficient mice. *J. Neurosci.* **25**, 1081–1088 (2005).
47. Eswarakumar, V. P., Lax, I. & Schlessinger, J. Cellular signaling by fibroblast growth factor receptors. *Cytokine Growth Factor Rev.* **16**, 139–149 (2005).
48. Aricescu, A. R., McKinnell, I. W., Halfter, W. & Stoker, A. W. Heparan sulfate proteoglycans are ligands for receptor protein tyrosine phosphatase σ. *Mol. Cell. Biol.* **22**, 1881–1892 (2002).
49. Fox, A. N. & Zinn, K. The heparan sulfate proteoglycan syndecan is an *in vivo* ligand for the *Drosophila* LAR receptor tyrosine phosphatase. *Curr. Biol.* **15**, 1701–1711 (2005).
50. Johnson, K. G. *et al.* The HSPGs Syndecan and Dallylike bind the receptor phosphatase LAR and exert distinct effects on synaptic development. *Neuron* **49**, 517–531 (2006).
An elegant study, together with reference 49, characterizing the regulation of the receptor PTP LAR by ligand binding at neuromuscular junctions.
51. O'Grady, P., Thai, T. C. & Saito, H. The laminin–nidogen complex is a ligand for a specific splice isoform of the transmembrane protein tyrosine phosphatase LAR. *J. Cell Biol.* **141**, 1675–1684 (1998).
52. Brady-Kalnay, S. M., Flint, A. J. & Tonks, N. K. Homophilic binding of PTPmu, a receptor-type protein tyrosine phosphatase, can mediate cell–cell aggregation. *J. Cell Biol.* **122**, 961–972 (1993).
53. Zondag, G. C. M. *et al.* Homophilic interactions mediated by receptor tyrosine phosphatases μ and κ. *J. Biol. Chem.* **270**, 14247–14250 (1995).
54. Yang, T. *et al.* Leukocyte antigen-related protein tyrosine phosphatase receptor: a small ectodomain isoform functions as a homophilic ligand and promotes neurite outgrowth. *J. Neurosci.* **23**, 3353–3363 (2003).
55. Yang, T., Yin, W., Derevyanny, V. D., Moore, L. A. & Longo, F. M. Identification of an ectodomain within the LAR protein tyrosine phosphatase receptor that binds homophilically and activates signalling pathways promoting neurite outgrowth. *Eur. J. Neurosci.* **22**, 2159–2170 (2005).
56. Finkel, T. Oxidant signals and oxidative stress. *Curr. Opin. Cell Biol.* **15**, 247–254 (2003).
An excellent overview of the targets and functions of ROS.
57. Lambeth, J. D. NOX enzymes and the biology of reactive oxygen. *Nature Rev. Immunol.* **4**, 181–189 (2004).
A comprehensive review of the NOX family of enzymes.
58. Rhee, S. G. *et al.* Intracellular messenger function of hydrogen peroxide and its regulation by peroxiredoxins. *Curr. Opin. Cell Biol.* **17**, 183–189 (2005).
An excellent review of the role of hydrogen peroxide as a second messenger.
59. den Hertog, J., Groen, A. & van der Wijk, T. Redox regulation of protein-tyrosine phosphatases. *Arch. Biochem. Biophys.* **434**, 11–15 (2005).
60. Salmeen, A. & Barford, D. Functions and mechanisms of redox regulation of cysteine-based phosphatases. *Antioxid. Redox Signal* **7**, 560–577 (2005).
61. Tonks, N. K. Redox redux: revisiting PTPs and the control of cell signaling. *Cell* **121**, 667–670 (2005).
This review, together with references 59 and 60, provides a comprehensive overview of the regulation of PTP function by reversible oxidation.
62. Salmeen, A. *et al.* Redox regulation of protein tyrosine phosphatase 1B involves a sulphenyl-amide intermediate. *Nature* **423**, 769–773 (2003).
An important paper that, together with reference 118, provides structural insights into the effects of ROS on the active site of PTP1B. The data suggest an explanation, at the molecular level, of how oxidation can be harnessed as a mechanism for reversible regulation of PTP function.
63. Persson, C. *et al.* Preferential oxidation of the second phosphatase domain of receptor-like PTP-α revealed by an antibody against oxidized protein tyrosine phosphatases. *Proc. Natl Acad. Sci. USA* **101**, 1886–1891 (2004).
64. van der Wijk, T., Overvoorde, J. & den Hertog, J. H_2O_2-induced intermolecular disulfide bond formation between receptor protein-tyrosine phosphatases. *J. Biol. Chem.* **279**, 44355–44361 (2004).
65. Groen, A. *et al.* Differential oxidation of protein-tyrosine phosphatases. *J. Biol. Chem.* **280**, 10298–10304 (2005).
66. Goldstein, B. J., Mahadev, K., Wu, X., Zhu, L. & Motoshima, H. Role of insulin-induced reactive oxygen species in the insulin signaling pathway. *Antioxid. Redox Signal* **7**, 1021–1031 (2005).
67. Martyn, K. D., Frederick, L. M., von Loehneysen, K., Dinauer, M. C. & Knaus, U. G. Functional analysis of Nox4 reveals unique characteristics compared to other NADPH oxidases. *Cell Signal.* **18**, 69–82 (2006).
68. Kamata, H. *et al.* Reactive oxygen species promote TNFα-induced death and sustained JNK activation by inhibiting MAP kinase phosphatases. *Cell* **120**, 649–661 (2005).
69. Duranteau, J., Chandel, N. S., Kulisz, A., Shao, Z. & Schumacker, P. T. Intracellular signaling by reactive oxygen species during hypoxia in cardiomyocytes. *J. Biol. Chem.* **273**, 11619–11624 (1998).
70. Ostman, A., Hellberg, C. & Bohmer, F. D. Protein-tyrosine phosphatases and cancer. *Nature Rev. Cancer* **6**, 307–320 (2006).
A comprehensive review of the function of PTPs in tumorigenesis.
71. Kristjansdottir, K. & Rudolph, J. Cdc25 phosphatases and cancer. *Chem. Biol.* **11**, 1043–1051 (2004).
72. Cully, M., You, H., Levine, A. J. & Mak, T. W. Beyond PTEN mutations: the PI3K pathway as an integrator of multiple inputs during tumorigenesis. *Nature Rev. Cancer* **6**, 184–192 (2006).
73. Bentires-Alj, M., Kontaridis, M. I. & Neel, B. G. Stops along the RAS pathway in human genetic disease. *Nature Med.* **12**, 283–285 (2006).
74. Bottini, N. *et al.* A functional variant of lymphoid tyrosine phosphatase is associated with type I diabetes. *Nature Genet.* **36**, 337–338 (2004).
75. Smyth, D. *et al.* Replication of an association between the lymphoid tyrosine phosphatase locus (*LYP/PTPN22*) with type 1 diabetes, and evidence for its role as a general autoimmunity locus. *Diabetes* **53**, 3020–3023 (2004).
76. Begovich, A. B. *et al.* A missense single-nucleotide polymorphism in a gene encoding a protein tyrosine phosphatase (*PTPN22*) is associated with rheumatoid arthritis. *Am. J. Hum. Genet.* **75**, 330–337 (2004).
77. Carlton, V. E. *et al. PTPN22* genetic variation: evidence for multiple variants associated with rheumatoid arthritis. *Am. J. Hum. Genet.* **77**, 567–581 (2005).
78. Kyogoku, C. *et al.* Genetic association of the *R620W* polymorphism of protein tyrosine phosphatase *PTPN22* with human SLE. *Am. J. Hum. Genet.* **75**, 504–507 (2004).
79. Wu, J. *et al.* Identification of substrates of human protein tyrosine phosphatase PTPN22. *J. Biol. Chem.* **281**, 11002–11010 (2006).
80. Vang, T. *et al.* Autoimmune-associated lymphoid tyrosine phosphatase is a gain-of-function variant. *Nature Genet.* **37**, 1317–1319 (2005).
81. Andersen, J. N. & Tonks, N. K. in *Protein Tyrosine Phosphatase-Based Therapeutics: Lessons from PTP1B* (eds Arino, J. & Alexander, D. R.) (Spring-Verlag, Berlin, 2004).
An overview of drug-discovery efforts targetting members of the PTP superfamily, in particular PTP1B.
82. Liu, G. Technology evaluation: ISIS-113715, Isis. *Curr. Opin. Mol. Ther.* **6**, 331–336 (2004).
83. Zinker, B. A. *et al.* PTP1B antisense oligonucleotide lowers PTP1B protein, normalizes blood glucose, and improves insulin sensitivity in diabetic mice. *Proc. Natl Acad. Sci. USA* **99**, 11357–11362 (2002).
84. Wiesmann, C. *et al.* Allosteric inhibition of protein tyrosine phosphatase 1B. *Nature Struct. Mol. Biol.* **11**, 730–737 (2004).
85. Xie, Y. *et al.* Protein-tyrosine phosphatase (PTP) wedge domain peptides: a novel approach for inhibition of PTP function and augmentation of protein-tyrosine kinase function. *J. Biol. Chem.* **281**, 16482–16492 (2006).
86. Sapieha, P. S. *et al.* Receptor protein tyrosine phosphatase σ inhibits axon regrowth in the adult injured CNS. *Mol. Cell Neurosci.* **28**, 625–635 (2005).
87. MacKeigan, J. P., Murphy, L. O. & Blenis, J. Sensitized RNAi screen of human kinases and phosphatases identifies new regulators of apoptosis and chemoresistance. *Nature Cell Biol.* **7**, 591–600 (2005).
88. Flint, A. J., Tiganis, T., Barford, D. & Tonks, N. K. Development of "substrate trapping" mutants to identify physiological substrates of protein tyrosine phosphatases. *Proc. Natl. Acad. Sci. USA* **94**, 1680–1685 (1997).
The first description of the use of 'substrate trapping' mutants, an approach now applied widely in the field to define the substrate specificity of members of the PTP superfamily.
89. Shintani, T. *et al.* Eph receptors are negatively controlled by protein tyrosine phosphatase receptor type O. *Nature Neurosci.* **9**, 761–769 (2006).
90. Tertoolen, L. G. *et al.* Dimerization of receptor protein-tyrosine phosphatase α in living cells. *BMC Cell Biol.* **2**, 8 (2001).
91. Jiang, G. *et al.* Dimerization inhibits the activity of receptor-like protein-tyrosine phosphatase-α. *Nature* **401**, 606–610 (1999).
This is a highly significant study, which, together with reference 38, provides important insights into dimerization as a potential mechanism for the regulation of receptor PTP function.
92. Blanchetot, C., Tertoolen, L. G. & den Hertog, J. Regulation of receptor protein-tyrosine phosphatase α by oxidative stress. *EMBO J.* **21**, 493–503 (2002).
93. van der Wijk, T., Blanchetot, C., Overvoorde, J. & den Hertog, J. Redox-regulated rotational coupling of receptor protein-tyrosine phosphatase α dimers. *J. Biol. Chem.* **278**, 13968–13974 (2003).
94. Ruivenkamp, C. A. *et al. Ptprj* is a candidate for the mouse colon-cancer susceptibility locus *Scc1* and is frequently deleted in human cancers. *Nature Genet.* **31**, 295–300 (2002).
95. Nakamura, M. *et al.* Novel tumor suppressor loci on 6q22–23 in primary central nervous system lymphomas. *Cancer Res.* **63**, 737–741 (2003).
96. Wang, Z. *et al.* Mutational analysis of the tyrosine phosphatome in colorectal cancers. *Science* **304**, 1164–1166 (2004).
97. Jacob, S. T. & Motiwala, T. Epigenetic regulation of protein tyrosine phosphatases: potential molecular targets for cancer therapy. *Cancer Gene Ther.* **12**, 665–672 (2005).
98. Motiwala, T. *et al.* Suppression of the protein tyrosine phosphatase receptor type O gene (*PTPRO*) by methylation in hepatocellular carcinomas. *Oncogene* **22**, 6319–6331 (2003).
99. Motiwala, T. *et al.* Protein tyrosine phosphatase receptor-type O (PTPRO) exhibits characteristics of a candidate tumor suppressor in human lung cancer. *Proc. Natl Acad. Sci. USA* **101**, 13844–13849 (2004).

100. Oka, T. *et al.* Gene silencing of the tyrosine phosphatase *SHP1* gene by aberrant methylation in leukemias/lymphomas. *Cancer Res.* **62**, 6390–6394 (2002).
101. Zhang, Q. *et al.* STAT3- and DNA methyltransferase 1-mediated epigenetic silencing of SHP-1 tyrosine phosphatase tumor suppressor gene in malignant T lymphocytes. *Proc. Natl Acad. Sci. USA* **102**, 6948–6953 (2005).
102. Yeh, S. H. *et al.* Genetic characterization of Fas-associated phosphatase-1 as a putative tumor suppressor gene on chromosome 4q21.3 in hepatocellular carcinoma. *Clin. Cancer Res.* **12**, 1097–1108 (2006).
103. Garcia, J. M. *et al.* Promoter methylation of the *PTEN* gene is a common molecular change in breast cancer. *Genes Chromosomes Cancer* **41**, 117–124 (2004).
104. Xu, S., Furukawa, T., Kanai, N., Sunamura, M. & Horii, A. Abrogation of DUSP6 by hypermethylation in human pancreatic cancer. *J. Hum. Genet.* **50**, 159–167 (2005).
105. Neel, B. G., Gu, H. & Pao, L. The 'Shp'ing news: SH2 domain-containing tyrosine phosphatases in cell signaling. *Trends Biochem. Sci.* **28**, 284–293 (2003).
106. Tartaglia, M. & Gelb, B. D. Noonan syndrome and related disorders: genetics and pathogenesis. *Annu. Rev. Genomics Hum. Genet.* **6**, 45–68 (2005).
107. Keilhack, H., David, F. S., McGregor, M., Cantley, L. C. & Neel, B. G. Diverse biochemical properties of Shp2 mutants. Implications for disease phenotypes. *J. Biol. Chem.* **280**, 30984–30993 (2005).
108. Salmond, R. J. & Alexander, D. R. SHP2 forecast for the immune system: fog gradually clearing. *Trends Immunol.* **27**, 154–160 (2006).
This review, together with references 105 and 106, provides a comprehensive overview of the structure, regulation and function of SHP2 — the first PTP oncoprotein.
109. Niihori, T. *et al.* Functional analysis of PTPN11/SHP-2 mutants identified in Noonan syndrome and childhood leukemia. *J. Hum. Genet.* **50**, 192–202 (2005).
110. Tartaglia, M. *et al.* Somatic mutations in *PTPN11* in juvenile myelomonocytic leukemia, myelodysplastic syndromes and acute myeloid leukemia. *Nature Genet.* **34**, 148–150 (2003).
111. Bentires-Alj, M. *et al.* Activating mutations of the Noonan syndrome-associated *SHP2/PTPN11* gene in human solid tumors and adult acute myelogenous leukemia. *Cancer Res.* **64**, 8816–8820 (2004).
112. Chan, R. J. *et al.* Human somatic *PTPN11* mutations induce hematopoietic-cell hypersensitivity to granulocyte-macrophage colony-stimulating factor. *Blood* **105**, 3737–3742 (2005).
113. Mohi, M. G. *et al.* Prognostic, therapeutic, and mechanistic implications of a mouse model of leukemia evoked by Shp2 (*PTPN11*) mutations. *Cancer Cell* **7**, 179–191 (2005).
114. Schubbert, S. *et al.* Functional analysis of leukemia-associated *PTPN11* mutations in primary hematopoietic cells. *Blood* **106**, 311–317 (2005).
115. Digilio, M. C. *et al.* Grouping of multiple-lentigines/LEOPARD and Noonan syndromes on the *PTPN11* gene. *Am. J. Hum. Genet.* **71**, 389–394 (2002).
116. Kontaridis, M. I., Swanson, K. D., David, F. S., Barford, D. & Neel, B. G. *PTPN11* (Shp2) mutations in LEOPARD syndrome have dominant negative, not activating, effects. *J. Biol. Chem.* **281**, 6785–6792 (2006).
117. Lim, K. L., Kolatkar, P. R., Ng, K. P., Ng, C. H. & Pallen, C. J. Interconversion of the kinetic identities of the tandem catalytic domains of receptor-like protein-tyrosine phosphatase PTPα by two point mutations is synergistic and substrate-dependent. *J. Biol. Chem.* **273**, 28986–28993 (1998).
118. van Montfort, R. L., Congreve, M., Tisi, D., Carr, R. & Jhoti, H. Oxidation state of the active-site cysteine in protein tyrosine phosphatase 1B. *Nature* **423**, 773–777 (2003).
An important paper that, together with reference 62, provides structural insights into the effects of ROS on the active site of PTP1B. The data suggest an explanation, at the molecular level, of how oxidation can be harnessed as a mechanism for reversible regulation of PTP function.
119. Lee, S. R., Kwon, K. S., Kim, S. R. & Rhee, S. G. Reversible inactivation of protein-tyrosine phosphatase 1B in A431 cells stimulated with epidermal growth factor. *J. Biol. Chem.* **273**, 15366–15372 (1998).
An important paper that provided the first demonstration of reversible oxidation and inactivation of a defined PTP in response to a physiological stimulus.
120. Mahadev, K., Zilbering, A., Zhu, L. & Goldstein, B. J. Insulin-stimulated hydrogen peroxide reversibly inhibits protein-tyrosine phosphatase 1B *in vivo* and enhances the early insulin action cascade. *J. Biol. Chem.* **276**, 21938–21942 (2001).
121. Meng, T. C., Buckley, D. A., Galic, S., Tiganis, T. & Tonks, N. K. Regulation of insulin signaling through reversible oxidation of the protein-tyrosine phosphatases TC45 and PTP1B. *J. Biol. Chem.* **279**, 37716–37725 (2004).
122. Meng, T. C., Fukada, T. & Tonks, N. K. Reversible oxidation and inactivation of protein tyrosine phosphatases *in vivo*. *Mol. Cell* **9**, 387–399 (2002).
An analysis of the regulation of PTP function by reversible oxidation, which introduces the use of a modified in-gel phosphatase assay to measure PTP oxidation. The study highlights the role of oxidation of SHP2 in regulating platelet-derived growth factor (PDGF) receptor signalling.
123. Kwon, J. *et al.* Receptor-stimulated oxidation of SHP-2 promotes T-cell adhesion through SLP-76–ADAP. *EMBO J.* **24**, 2331–2341 (2005).
124. Chen, C. H. *et al.* Reactive oxygen species generation is involved in epidermal growth factor receptor transactivation through the transient oxidization of Src homology 2-containing tyrosine phosphatase in endothelin-1 signaling pathway in rat cardiac fibroblasts. *Mol. Pharmacol.* **69**, 1347–1355 (2006).
125. Singh, D. K. *et al.* The strength of receptor signaling is centrally controlled through a cooperative loop between Ca^{2+} and an oxidant signal. *Cell* **121**, 281–293 (2005).
126. Xu, Y., Shao, Y., Voorhees, J. J. & Fisher, G. J. Oxidative inhibition of receptor type protein tyrosine phosphatase κ by ultraviolet irradiation activates EGFR in human keratinocytes. *J. Biol. Chem.* 18 July 2006 (doi:10.1074/jbc.M602355200).
127. Wu, R. F. *et al.* Subcellular targeting of oxidants during endothelial cell migration. *J. Cell Biol.* **171**, 893–904 (2005).
128. Levinthal, D. J. & Defranco, D. B. Reversible oxidation of ERK-directed protein phosphatases drives oxidative toxicity in neurons. *J. Biol. Chem.* **280**, 5875–5883 (2005).
129. Kwon, J. *et al.* Reversible oxidation and inactivation of the tumor suppressor PTEN in cells stimulated with peptide growth factors. *Proc. Natl Acad. Sci. USA* **101**, 16419–16424 (2004).
130. Seo, J. H., Ahn, Y., Lee, S. R., Yeol Yeo, C. & Chung Hur, K. The major target of the endogenously generated reactive oxygen species in response to insulin stimulation is phosphatase and tensin homolog and not phosphoinositide-3 kinase (PI-3 kinase) in the PI-3 kinase/Akt pathway. *Mol. Biol. Cell* **16**, 348–357 (2005).
131. Chiarugi, P. *et al.* Two vicinal cysteines confer a peculiar redox regulation to low molecular weight protein tyrosine phosphatase in response to platelet-derived growth factor receptor stimulation. *J. Biol. Chem.* **276**, 33478–33487 (2001).
132. Chiarugi, P. *et al.* Reactive oxygen species as essential mediators of cell adhesion: the oxidative inhibition of a FAK tyrosine phosphatase is required for cell adhesion. *J. Cell Biol.* **161**, 933–944 (2003).
133. Nimnual, A. S., Taylor, L. J. & Bar-Sagi, D. Redox-dependent downregulation of Rho by Rac. *Nature Cell Biol.* **5**, 236–241 (2003).

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Competing interests statement
The author declares no competing financial interests.

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FURTHER INFORMATION
Nicholas Tonks's homepage: http://www.cshl.edu/public/SCIENCE/tonks.html
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