Biochimica et Biophysica Acta 1795 (2009) 1–15

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbacan

Review

Protein phosphatase 2A regulatory subunits and cancer

Pieter J.A. Eichhorn [1], Menno P. Creyghton [2], René Bernards *

Division of Molecular Carcinogenesis, Center for Cancer Genomics and Center for Biomedical Genetics, The Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX Amsterdam, The Netherlands

ARTICLE INFO

Article history:
Received 7 April 2008
Received in revised form 20 May 2008
Accepted 21 May 2008
Available online 3 June 2008

Keywords:
Protein phosphatase 2A
Signal transduction
Cancer

ABSTRACT

The serine/threonine protein phosphatase (PP2A) is a trimeric holoenzyme that plays an integral role in the regulation of a number of major signaling pathways whose deregulation can contribute to cancer. The specificity and activity of PP2A are highly regulated through the interaction of a family of regulatory B subunits with the substrates. Accumulating evidence indicates that PP2A acts as a tumor suppressor. In this review we summarize the known effects of specific PP2A holoenzymes and their roles in cancer relevant pathways. In particular we highlight PP2A function in the regulation of MAPK and Wnt signaling.

© 2008 Elsevier B.V. All rights reserved.

Contents

1. Introduction . . . 1
2. PP2A structure and function . . . 2
   2.1. The catalytic subunit (PP2Ac) . . . 2
   2.2. The structural subunit (PR65) . . . 3
   2.3. The regulatory B subunits . . . 3
      2.3.1. The B/PR55 family of B subunits . . . 3
      2.3.2. The B′/PR61 family of β subunits . . . 4
      2.3.3. The B″/PR72 family of β subunits . . . 4
      2.3.4. The PTP/PR53 family of β subunits . . . 4
3. PP2A subunits and cancer . . . 4
   3.1. PP2A and transformation . . . 4
   3.2. PP2A and MAP kinase signaling . . . 7
   3.3. PP2A and WNT signaling . . . 9
4. Conclusions . . . 9
References . . . 11

1. Introduction

The ability of a cell to rapidly adapt to external changes is a vital requirement for evolutionary selection in a dynamic environment. Post-translational modifications of proteins can influence the structure, charge and thus the enzymatic activity of proteins thereby conferring onto a relatively static pool of proteins the ability to rapidly adapt to external cues.

The ever increasing pattern of modifications includes changes in ubiquitination, phosphorylation, glycosylation, methylation, acetylation, farnecylation etc., thus giving rise to a vast amount of differentially modified proteins harboring different activities. Reversible phosphorylation was the first described modification able to alter the enzymatic activity of a protein [1]. Inappropriate regulation of the reversible phosphorylation of proteins can have a profound effect on how the cell responds to its environment. It is therefore not surprising that genetic or chemical disturbance of the action of these kinases and phosphatases can lead to aberrant cellular behavior in an organism, giving rise to a wide array of disease phenotypes including many

* Corresponding author. Tel.: +31 20 512 1952; fax: +31 20 512 1954.
E-mail address: R.Bernards@nki.nl (R. Bernards).

[1] Present address: Institut de Recerca, Hospital Vall d Hebron, Passeig Vall d Hebron 119-129, 08035 Barcelona, Spain.

[2] Present address: Whitehead Institute for Biomedical Research, 9 Cambridge Center, 02142 MA Cambridge, USA.

0304-419X/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.bbcan.2008.05.005

forms of cancer [2]. Specific dependence of tumor cells on their aberrantly regulated kinases or phosphatases for growth and survival, in addition to the drug-able enzymatic activities of these proteins, makes them attractive drug targets [3–6]. Although many kinases are deregulated in cancer, the role of phosphatases in cellular transformation has remained underexplored. The ubiquitously expressed protein phosphatase type 2A (PP2A) is a serine/threonine phosphatase that makes up 1% of all cellular proteins and along with protein phosphatase 1 (PP1) accounts for over 90% of all Ser/Thr phosphatase activity in the cell. A number of studies have highlighted the role of PP2A as a tumor suppressor. This was first suggested in experiments showing that okadaic acid, a selective but not specific inhibitor of PP2A, promotes tumor growth in mice [7–11]. Similarly, it was established that the SV40 small tumor antigen, which is required for cellular transformation in human cells, can alter PP2A activity by displacing the regulatory B subunit from the holoenzyme complex [12,13]. Finally, mutations have been identified in different components of the PP2A holoenzyme complex, which have been linked to a variety of primary human tumors. In this review we will discuss the role of PP2A in cancer with special emphasis on MAPK and Wnt signaling.

2. PP2A structure and function

PP2A is a phosphatase that functions to reverse the action of kinases in most major signaling cascades [11,14,15]. Although once perceived as a single broad specificity phosphatase, this notion now seems oversimplified. PP2A represents a family of holoenzyme complexes (Fig. 1 and Table 1) with different activities and diverse substrate specificities [15]. Three distinct functional components give rise to the holoenzyme complex: Typically the catalytic subunit (PP2Ac) interacts with the structural core subunit (PP2Aa/PR65) making up the core of the enzyme. The association with a wide variety of B regulatory subunits to the core enzyme results in the formation of heterotrimeric PP2A holoenzyme complexes with diverse specificities (Fig. 1).

2.1. The catalytic subunit (PP2Ac)

The catalytic subunit has a large conserved domain that forms a bimetallic active site for phosphor-ester hydrolysis. It targets phosphate groups on either serine or threonine residues and under some conditions harbors activity towards phosphorylated tyrosine [16]. PP2A catalytic activity is encoded by two distinct ubiquitously expressed genes [17], the Cα and the Cβ subunits, the latter being expressed approximately 10 fold lower due to a weaker promoter [18]. Both are 35 kDa in size and share 97% sequence identity. The levels of PP2Ac are tightly regulated in the cell at the translational or post-translational level making ectopic expression of this protein extremely difficult [19]. Loss of Cα in yeast and in mice is lethal [20] underscoring the importance of this phosphatase for proper maintenance of cellular homeostasis.

The recent elucidation of the crystal structure of the PP2A holoenzyme determined that the highly conserved C-terminal tail (^{304}TPDYFL^{309}) of the PP2Ac subunit resides at a critical interface between the PR65 structural subunit and the B subunit PR61γ [21,22]. As such, the recruitment of the B subunit to the core enzyme is tightly regulated by the methylation and phosphorylation patterns of the C-terminal tail. Methylation on the carboxyl group Leu^{309} by *S*-adenosylmethionine-dependent leucine carboxyl transferase 1 (LCMT1) was shown to be required for the binding of the PR55B family members but not for other B subunits [23]. The methylation of the C-terminal tail can be reversed by the specific phosphatase methylesterase (PME-1) thus adding another dimension to holoenzyme regulation [24].

Phosphorylation appears to occur at either Tyr^{307} or the newly identified Thr^{304} [23]. Phosphorylation of Tyr^{307} inhibits the recruitment of the PR55B and PR61B αβε family members to the core

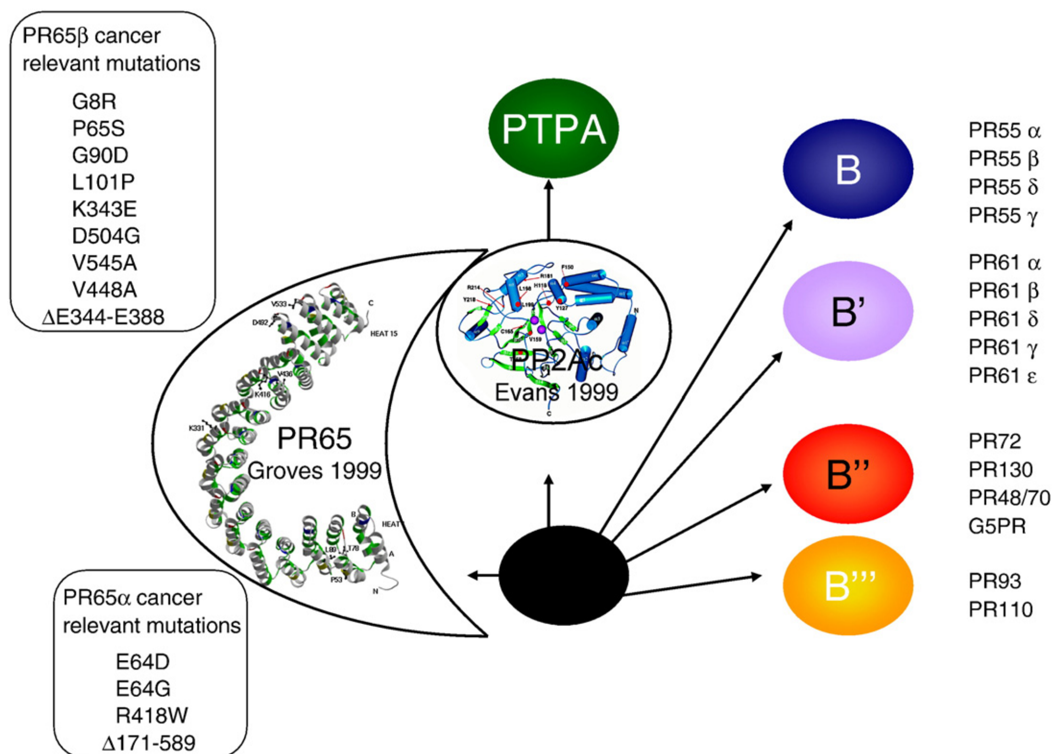

Fig. 1. Schematic overview of PP2A holoenzyme composition showing interaction of the PP2A catalytic subunit [264] to the PR65 scaffold protein at five C-terminal Huntington/elongation/A-subunit/TOR (HEAT) repeats [28]. The variable B subunit interacts with the N-terminal HEAT domains of the PR65 scaffold subunit or directly to the PP2A catalytic subunit. The boxes highlight the cancer relevant mutations described to date in the two isoforms of PR65.

Table 1
Different subunits of the PP2A holoenzyme shown with gene name (first column), the two forms of nomenclature used in the literature (columns 2 and 3), Ensembl transcript number (column 4) and reference (column 5)

Gene name	Alternate name		Ensembl nr.	Reference
Catalytic subunit				
PPP2CA	Ca	PP2Aca	ENST00000231504	[17]
PPP2CB	Cβ	PP2Acβ	ENST00000221138	[17]
Structural subunit				
PPP2R1A	Aα	PR65α	ENST00000322088	[29]
			ENST00000361107	
PPP2R1B	Aβ	PR65β	ENST00000341980	[29]
			ENST00000311129	
Regulatory subunit				
PPP2R2A	Bα	PR55α	ENST00000315985	[38,145]
PPP2R2B	Bβ	PR55β	ENST00000287031	[38,145]
			ENST00000336640	
			ENST00000355212	
PPP2R2C	Bγ	PR55γ	ENST00000264959	[39]
			ENST00000335585	
PPP2R2D	Bδ	PR55δ	ENST00000314348	[37]
PPP2R3A	B″α	PR130	ENST00000264977	[47]
	B″β	PR72	ENST00000334546	[47]
PPP2R3B		PR70/48	ENST00000300846	[51,146]
			ENST00000361450	
C14orf10		G5PR	OTTHUMG00000028723	[147]
PPP2R4	PTPA	PR53	ENST00000337738	[53,55]
			ENST00000266101	[55]
			ENST00000347048	[55]
			ENST00000357197	[55]
PPP2R5A	B′α	PR61α	ENST00000261461	[45,148]
PPP2R5B	B′β	PR61β	ENST00000164133	[45,148]
PPP2R5C	B′γ1	PR61γ1	ENST00000334743	[148,149]
	B′γ2	PR61γ2	ENST00000334756	[148,149]
	B′γ3	PR61γ3	ENST00000350249	[148,149]
PPP2R5D	B′δ	PR61δ	ENST00000230402	[148,150]
			ENST00000344268	
PPP2R5E	B′ε	PR61ε	ENST00000337537	[148]
STRN	Striatin	PR110	ENSG00000115808	[151]
STRN3	SG2NA	PR93	OTTHUMT00000073367	[151]

enzyme [25]. Interestingly this site is regulated by the oncogene c-SRC raising the possibility that inhibition of one or all of these subunits may be one of the requirements for c-SRC induced transformation. However, mutational analysis of Tyr^{307} limited the methylation status at Leu^{309} possibly explaining the deficiency in PR55B binding [23]. Similarly the C-terminal tail can be phosphorylated at Thr^{304}, which was also shown to inhibit the binding of PR55B subunits but not the other B subunits [23]. The methylation and phosphorylation status of the C-terminus had no bearing on the binding of PR70, PR72 or PR61δ B subunits.

The formation of the holoenzyme complex and the activation of PP2Ac are also kept in check by a series of interlocking steps involving the dual action of PME-1 and the peptidyl prolyl isomerase PTPA. PTPA binds to structural subunit of PP2A and is an essential and specific activator of the inactive form of PP2A (PP2Ai). [26]. The precise mechanism of how PTPA induces C subunit activity remains to be determined, but it appears that the PR65/PTPA complex may function partly by inhibiting the methylesterase activity of PME-1. PME-1 on the other hand appears to have a multifaceted role. In normal cells PME-1 acts as a negative regulator of PR55 B subunit binding by demethylating Leu^{309} of the PP2Ac C-terminal tail; however, in the absence of PR65, PME-1 can also inhibit the generation of the active C subunit. [27].

2.2. The structural subunit (PR65)

The catalytic subunit interacts with the PR65 scaffold protein at four C-terminal Huntington/elongation/A-subunit/TOR (HEAT) repeats (Fig. 1) [28]. Upon the formation of the core enzyme the scaffold protein folds in on itself forming a more horseshoe shape-like structure [21,22]. The bending of the structural subunit allows the catalytic subunit unimpeded access to the PP2A substrate, which is recruited to the holoenzyme by the B regulatory subunit. Two alternative genes, PR65α and PR65β encode the two flavors of the PR65 scaffold protein [29]. Most PP2A holoenzymes contain the PR65α isoform, while only a small fraction (10%) contain the PR65β isoform. Although the 2 isoforms share an 87% sequence identity, PR65β has unique biochemical properties and is unable to substitute for the loss of PR65α in mice. Moreover PR65β is expressed at much lower levels than PR65α in adult tissues. However, in oocytes and during the early stages of vertebrate development, PR65β mRNA is more abundant than PR65α [30,31]. As such these two alternative transcripts differ in their ability to interact with the various regulatory B subunits [32]. Interestingly several tumor-specific mutations have been identified in PR65β which disrupt the ability of PR65β to form holoenzymes with specific regulatory subunits in vitro (discussed in detail below).

2.3. The regulatory B subunits

To date, 15 genes have been identified in the human genome that encode at least 26 different alternative transcripts and splice forms representing the B subunits of the PP2A holoenzyme (Table 1). These B subunits can be expressed in a tissue specific manner and are proposed to mediate substrate specificity of the PP2A holoenzyme complex [15,33,34]. While the PR65α scaffold can interact with all regulatory B subunits, the PR65β scaffold is unable to interact with the B/PR55 family of B subunits and shows a preference for binding to PR72 [32]. Assuming that there are no significant differences between PP2Acα and PP2Acβ, around 30 PP2A holoenzyme combinations are possible, taking into account the inability of PR65β to interact with the PR55 family of B subunits [35] and not including predicted splice variants. Functionally the number of holoenzymes might even be lower since different subunits within a family might be functionally redundant or expressed in a tissue specific manner under different promoters. Conversely it could be higher since not all regulatory subunits are necessarily identified. The different holoenzyme complexes likely account for the growing list of phosphoproteins and signaling pathways known to be regulated by PP2A. The B subunits have been subdivided into five distinct families (Table 1), which share no sequence similarity, apart from a few conserved amino acids that allow the interaction with the N-terminal HEAT domains of the PR65 scaffold subunit [36].

2.3.1. The B/PR55 family of B subunits

The B/PR55 family consists of at least six members, transcribed from four different genes (Table 1) with more variants likely to exist [38]. PR55 family members exhibit both temporal and spatial expression patterns [37–39] with both PR55α and PR55δ being expressed almost ubiquitously while PR55β and PR55γ being highly enriched in the brain. Furthermore, the expression of PR55β and PR55γ is also developmentally regulated with PR55β levels decreasing and PR55γ levels increasing sharply after birth [37]. The different PR55 family member proteins also show distinct spatial distribution in the cell [40]. However, whether this variable localization is a regulatory function of these subunits or it simply represents the location of their binding partners, is unclear.

Substrate regulation by PR55 family members appears to be dependent on a number of factors. First, substrate binding by the PR55 family members appears to be dependent on a stretch of five degenerate WD40 repeats. WD40 repeats are conserved 40 amino acid sequences that end with a characteristic tryptophan-aspartate (WD) that appears to directly mediate protein–protein interactions. Secondly, the PP2Ac subunit of the core enzyme needs to be methylated

on Leu^{309} and dephosphorylated at Thr^{304} for its interaction with the PR55 regulatory B subunits [23,41,42]. However, these PP2Ac modifications are more likely to be a consequence of the spatial distribution of the core enzyme rather than anything else. Surprisingly, it was found that the PR65 subunit is not strictly required for PR55 to interact with PP2Ac subunit [42,43].

2.3.2. The B′/PR61 family of β subunits

The PR61 family of regulatory subunits contains eight members represented by five different genes (Table 1) [44] showing diverse tissue distribution [45,46]. PR61 family members show distinct spatial distribution inside the cell with PR61α, PR61β, and PR61ε being expressed in the cytoplasm, while PR61γ1, PR61γ2 and PR61γ3 are expressed in the nucleus. PR61δ appears to be expressed in both the nucleus and cytoplasm [15]. Interestingly, differentiation induced by retinoic acid treatment in IMR-32 cells results in the specific increase in the expression of PR61β and PR61δ suggesting that these two B isoforms are developmentally regulated [44]. These subunits display strong conservation within their central region (80%) but differ in their N- and C-terminal regions. Furthermore, PR61 family proteins show very little to no similarity with any of the other β subunit families. The recent elucidation of the crystal structure of PR61γ1 reveals that PR61 family members consist exclusively of α helices [21,22]. Unexpectedly, the structural composition of PR61γ resembles that of the PR65 structural subunit even though they exhibit little sequence homology. The formation of the holoenzyme structure results in an arrangement whereby the highly acidic, concave side of the PR61 family members remains unoccupied. It is believed that this highly acidic surface is used to recruit substrate proteins [21,22]. Interestingly, Shugoshin, a target of PR61B is a basic protein [22].

2.3.3. The B″/PR72 family of β subunits

PR72 and PR130 were the first members of this family discovered and are transcribed from the same gene albeit from different promoters [47]. PR130 and PR72 differ in their N-terminal regions with PR130 having a specific stretch of 665 amino acids which is functionally replaced by 44 amino acids in PR72. This results in a tissue specific distribution of these subunits [47]. PR72 contains two EFX hand domains that bind calcium, resulting in a conformational change in protein structure [48]. The strongest Ca^{2+} binding EFX domain (EFX1) is required for binding to the PR65 core subunit and nuclear localization of PR72 when overexpressed.

Notably G5PR, the newest proposed member of this subunit family, also contains an EFX hand domain and was also found to be able to associate with both PP5 and PP2Ac/PR65 [49] although the in vivo relevance of these interactions remains unexplored. A recently published functional knockout mouse of G5PR suggests a role for this protein in promoting B-cell survival [49].

A third member of the B″/PR72 family, PR70 has recently been demonstrated to bind to the tumor suppressor RB [50]. While PR48, a splice variant of PR70, was initially found to be able to interact with cdc6 [51]. Notably, similar to PR72, G5PR and PR130, both PR48 and PR70 also harbor an EFX domain which is dependent upon Ca^{2+} levels to regulate its binding to the core enzyme and subsequent phosphatase activity [50].

Finally a mouse specific subunit, PR59 was identified as a p107-interacting protein, which preferentially bound to p107 and not pRB when overexpressed [52]. No obvious human orthologue of mouse PR59 has been identified to date.

2.3.4. The PTP/PR53 family of β subunits

The PTPase activator (PTPA/PR53) was initially described as a B regulatory subunit due to its ability to interact with PR65. However, unlike other B regulatory subunits, which recruit the core enzyme to targeted PP2A substrates, PTPA appears to act like a chaperone directly affecting the activity of PP2Ac. PTPA transiently induces phosphotyrosyl phosphatase (PTP) activity of the PP2Ac and PP2Ac/PR65 complexes in vitro [53,54]. Similarly ATP and Mg^{2+} could activate PTP activity of PP2A on their own. There are 8 splice variants of this gene although only 4 were found to be efficiently translated [55]. Interestingly recent reports have described that PTPA loss of function mimics some of the effects seen with suppression of regulatory B subunits in yeast and apoptosis in mammalian cells [16]. A list of known PP2A interacting proteins is presented in Table 2.

3. PP2A subunits and cancer

3.1. PP2A and transformation

PP2A was first suggested to act as a tumor suppressor based on the tumor-promoting actions of okadaic acid, a more or less selective inhibitor of PP2A [10]. Treatment of mice with okadaic acid gave rise to tumors on the skin [7-9] which was later demonstrated to be caused by the activation of several cancer-promoting pathways [34]. Similarly, it was found that several tumor-promoting viruses are capable of displacing the B regulatory subunits from the core enzyme [12,56] resulting in altered enzymatic activity towards PP2A substrates [57–59]. Alternatively these could function as B subunits themselves. The alteration of PP2A by viral proteins leads to the deregulation of similar pathways found disturbed by okadaic acid [15,34,60,61]. Both the polyoma small T (pyST) antigen and polyoma middle T (pyMT) antigens but also the simian virus SV40 small T (ST) and E4orf4 antigens are able to form complexes with the PP2A/PR65 dimer [12,62]. Furthermore, the ability of the pyMT antigen to induce transformation in 3T3 cells is directly dependent on its ability to interact with PP2A [13]. Interestingly, while both the viral proteins pyMT/pyST and ST contain the ability to displace the B subunit from the core enzyme, their role in PP2A inhibition leads to activation of discrete cellular pathways [63]. PyMT appears to preferentially activate the MAP kinase pathway while ST stimulates AKT (Ser^{473}) phosphorylation in a PP2A dependent manner [63,64]. Curiously, like PyMT, PyST can activate the MAP kinase pathway; however, its ability to regulate AKT phosphorylation and its downstream effector pathways is highly dependent upon serum concentration levels. In 10% serum PyST dramatically stimulates Thr^{308} phosphorylation inciting the pro-proliferative aspects of AKT function but sensitizing cells to apoptosis. No clear effect is observed on Ser^{473} phosphorlyation. Conversely, during serum starvation PyST enhances phosphorylation of both Thr^{308} and Ser^{473} inducing the antiapoptotic functions of AKT through downstream phosphorylation of FOXO3 and BAD, thus promoting cell survival [64].

Since both PyMT and ST are able to induce transformation this suggests that cellular transformation resulting from PP2A inhibition can occur through distinct mechanisms. Being that global inhibition of PP2A is generally lethal to cells, it is therefore likely that these proteins target specific PP2A complexes required to block cellular transformation. In agreement with this notion, the overexpression of ST appears to correlate with the loss of PR55 activity, resulting in enhanced phosphorylation of proteins involved in controlling cell growth (see below) [35]. It was later found that expression of SV40 ST antigen is required for full transformation of both primary human embryonic kidney (HEK) cells and human foreskin fibroblasts expressing RAS^{V12}/hTERT/SV40 LT (hereby referred to as HEK-TER or BJ-TER cells respectively), suggesting that disruption of PP2A function is one of the requirements to generate engineered transformed human cells [65]. Following on this, a number of groups have used this transformation model to identify other components that could substitute for ST in this system. Surprisingly it was shown that targeted inhibition of only the B subunit PR61γ could enhance low level spontaneous transformation in RAS^{V12}/hTERT/SV40 LT expressing human fibroblasts [66], thus indicating that ST selectively targets PR61γ. However, PR61γ knockdown only weakly phenocopies

Table 2
Proteins that have been shown to directly interact with PP2A either by co-immunoprecipitation, mass-spectrometry, yeast two-hybrid, or functional assays

PP2A interacting protein	B subunit	Substrate	Regulatory effect	Reference
AC8	–	–	–	[152]
AKT	PR55α	Thr^{308}	Inhibit	[85]
Alpha 4	–	–	–	[153]
Androgen receptor	–	–	Inhibit	[154]
AP-1/AP-2	PR55α	Thr^{156}	Inhibit	[155]
APC	PR61	–	Inhibit	[131]
ARL2	PR55α/PR61ε	–	–	[156]
ATM	–	Ser^{1981}	Inhibit	[157]
Aurora A	–	Ser^{51}	Inhibit	[158]
AXIN	PR61	–	Inhibit	[133]
B-Arrestin II	–	–	–	[159]
B-CATENIN	–	–	Activate	[139]
BAD	–	Ser^{112}	Activate	[75]
BAX	–	Ser^{184}	Activate	[160]
BCL-2	PR61α	–	Inhibit	[161]
B2-ADRENERGIC FACTOR	–	–	–	[162]
Bestrophin	–	–	–	[163]
E-CADHERIN	–	–	Activate	[139]
Cav1.2	PR61	Ser^{1928}	Inhibit	[164]
Calpain	–	–	Inhibit	[165]
CaM kinase II	–	–	–	[166]
CaM kinase IV	–	–	Inhibit	[167]
Carboxypeptidase D	–	–	Inhibit	[168]
CAS	–	–	Inhibit	[169]
Cdc2	–	–	–	[89]
Chk1	–	Ser^{317}/ Ser^{345}	Inhibit	[170]
Chk2	–	Thr^{68}	Inhibit	[171]
CK2	–	–	–	[172]
Caspase-3	–	–	Inhibit	[173]
Cdc6	PR48	–	Activate	[51]
Cdc25c	PR61δ	–	Inhibit	[174]
Cdk4	–	–	–	[89]
Cdk9	–	–	–	[175]
CFTR	–	–	Inhibit	[176]
CG-NAP	PR130	–	–	[177]
CIP2A	–	–	–	[76]
Cofilin	–	–	–	[178]
Connexin43	–	–	–	[179]
CXCR2	–	–	Inhibit	[180]
Cyclin G1	PR61α/PR61β	–	–	[79]
Cyclin G2	PR61γ/PR61β	–	–	[181]
DARPP-32	PR72	Thr^{75}	Inhibit	[182]
DNA polymerase A	–	–	–	[183]
E4orf4	–	–	–	[56]
EMI2	–	–	–	[184]
eRF1	–	–	–	[185]
ERK	PR61γ1/PR61β	–	Inhibit	[108]
Estrogen receptor A	–	Ser^{118}	Inhibit	[186]
FMRP	–	–	–	[187]
Galpha12	–	–	–	[188]
Glutamate receptor	–	–	–	[189]
Gp130	–	Ser^{782}	Activate	[190]
H2AX	–	–	Inhibit	[191]
HAND-1	PR61δ	–	Inhibit	[192]
HAND-2	PR61δ	–	Inhibit	[192]
HCP-6	PR55	–	–	[193]
HDAC4	PR55α	Ser^{298}	Inhibit	[194]
HSF2	–	–	–	[195]
HIV Vpr	PR55	–	–	[196]
HOX11	–	–	–	[197]
HRX	–	–	–	[198]
IEX-1	–	–	–	[108]
IKKB	–	Ser^{177}/ Ser^{181}	Activate, inhibit	[199,200]
IKKG	–	–	Inhibit	[201]
IQGAP1	–	–	–	[202]
JAK2	–	–	–	[203]
JNK	–	–	Inhibit	[204]
KCNQ2	PR61γ	–	–	[205]
Keratin 8	–	–	–	[206]

Table 2 *(continued)*

PP2A interacting protein	B subunit	Substrate	Regulatory effect	Reference
Keratin 18	–	–	–	[206]
KSR	PR55α	–	Activate	[97]
LCMT	–	–	–	[207]
Mdm2		Thr^{216}	Activate	[77]
MEK3	–	Thr^{193}	Inhibit	[208]
MEKK3	–	Ser^{526}	Inhibit	[209]
c-MET	–	Ser^{985}	Inhibit	[210]
Mid-1	–	–	–	[211]
MKK4	–	–	Inhibit	[212]
MPM-2	–	–	–	[213]
c-MYC	PR61α	Ser^{62}	Inhibit	[72]
Myosin	–	–	–	[214]
Naked	PR72/PR130	–	–	[134,140]
Nemo	–	Ser^{68}	Inhibit	[215]
Neuroephrine transporter	–	–	–	[216]
Neurofilament proteins	PR55	–	–	[217]
NKCC1	–	–	–	[218]
NM23H2	–	–	–	[219]
NMDA receptor	–	–	–	[220]
NR3A	–	–	–	[221]
Nucleoredoxin	PR55β	–	–	[222]
Occludin	–	–	Inhibit	[223]
OSBP	–	–	–	[224]
P38	–	–	–	[173]
P53	PR55α PR61γ1/ PR61γ3	Ser^{37} Thr^{55}	Activate	[80,81]
P63	PR61α	–	–	[132]
P70-S6K	–	–	–	[225]
P107	PR59	–	Inhibit	[52]
PACS-1	–	Ser^{278}	Inhibit	[226]
PAK1	–	–	–	[225]
PAK3	–	–	–	[225]
Paxillin	PR61γ1	–	Inhibit	[69]
Period	–	–	Activate	[227]
P70 S6 kinase	–	–	–	[225]
PIM	PR61β	–	Inhibit	[228]
PME-1	–	–	–	[24]
Pin1	–	–	–	[229,230]
PKA	–	–	–	[231]
PKCα	–	–	–	[232]
PKCβ II	–	–	Inhibit	[233]
PKCδ	–	–	Inhibit	[234]
PKR	PR61α	–	–	[235]
PLK1	–	–	–	[236]
PyMT	–	–	–	[62,237]
PyST	–	–	–	[100]
Raf1	PR55α/PR55δ	Ser^{259}	Activate	[98]
RalA	–	Ser^{183}/ Ser^{194}	Inhibit	[89]
RACK1	–	–	–	[238]
R-RAS	–	–	–	[219]
RB	PR70	Thr^{826}	Inhibit	[50]
RelA	–	Ser^{536}	Inhibit	[200–203,239]
RHEB	–	–	–	[219]
Rho-B	–	–	–	[219]
RSA1-2	–	–	–	[240]
Runx2	–	–	–	[241]
SCR	PR61	–	Inhibit	[242]
Securin	–	–	Activate	[243]
Seratonin	–	–	Activate	[244]
Separase	PR61	–	–	[245]
SET	–	–	–	[112]
Src	PR55γ	Ser^{12}	Inhibit	[104]
SG2NA	–	–	–	[151]
Shc	–	–	Inhibit	[246]
Shugosin	Pr61	–	–	[247–249]
SMG-2	–	–	–	[250]
Sp1	–	Ser^{59}/ Thr^{681}	Activate	[251]
Sprouty	–	Ser^{112}/ Ser^{115}	Activate	[252]

(continued on next page)

Table 2 *(continued)*

PP2A interacting protein	B subunit	Substrate	Regulatory effect	Reference
STAT5	–	–	–	[253]
STE20	–	–	–	[218]
SV40 ST	–	–	–	[100]
Tap42/alpha4	–	–	–	[254]
TAU	PR55	Ser^{202}/ Thr^{205}	Activate	[255–257]
TAX	–	–	–	[201]
TIP	–	–	–	[258]
TGFBR1	PR55α	–	–	[259]
TOM22	PR55β	–	–	[260]
TRAF2	PR61γ	Thr^{117}	Inhibit	[200]
TSC2	–	–	–	[219]
TTP	–	–	–	[261]
UPF1	–	–	–	[262]
Vimentin	PR55	–	Activate	[263]

expression of ST antigen [67] suggesting that other B subunits might also be involved in tumor suppression. Recent clinical evidence has shown that the expression of PR61γ is suppressed in melanomas compared with non-transformed melanocytic naevi [68] supporting a role for this subunit in tumor suppression. Another interesting finding came from studying the metastatic capability of B16 mouse melanoma subclones that have different metastatic potential. The authors found that a truncated form of PR61γ was partially responsible for the metastatic potential of one of the subclones [69]. They found the protein co-localizes and interacts with paxillin at focal adhesions similar to wild type PR61γ. However, in the presence of mutant PR61γ, paxillin was hyperphosphorylated. Notably the PR61γ mutant did not lead to a loss of PP2A associated with paxillin, suggesting that the mutant PR61γ functions in a dominant negative fashion regulating PP2A activity rather than targeting. Furthermore, a number of reports have described that ectopic expression of various B subunits can lead to competition with other regulatory subunits for binding to the core enzyme [70,71]. As such the truncated form of PR61γ may inhibit the ability of other B subunits to recruit the core enzyme therefore limiting the tumor suppressor capabilities of these other holoenzyme complexes.

Recent evidence has indicated that c-MYC is also a direct target of PP2A regulation [72]. Levels of c-MYC are tightly regulated by opposing phosphorylation events at SER^{62}, which stabilizes c-MYC, and the subsequent phosphorylation of MYC^{T58} which is required for its degradation. Holoenzymes containing the B subunit PR61α dephosphorylate c-MYC at SER^{62} targeting c-MYC for ubiquitination and subsequent degradation. Conversely, inhibition of PR61α increased the levels of Ser^{62} and led to c-MYC stabilization and an increase in c-MYC activity. Interestingly stable expression of a c-MYC mutant MYC^{T58A}, which is no longer able to be degraded, functionally replaced ST in the transformation assays of primary human cells described above [73].

PR61α also interacts and co-localizes with the anti-apoptotic protein BCL2 at the mitochondrial membranes. This leads to increased dephosphorylation and inactivation of BCL2 [74]. Interestingly, PR61α was stabilized by the pro-apoptotic agent ceramide and the authors speculate on this as a possible mechanism for ceramide-induced apoptosis. Similarly, PP2A can also activate the pro-apoptotic function of BAD although the specific holoenzyme complex has of yet to be determined [75].

Junttila et al. have shown that the dephosphorylation of Ser^{62} of c-MYC by PR61α is regulated by the c-MYC specific PP2A inhibitor CIP2A (Cancerous Inhibitor of PP2A) [76]. Expression of CIP2A stabilizes c-MYC expression by inhibiting the catalytic activity of the PP2A holoenzyme towards Ser^{62} without affecting PP2A binding potential. Conversely inhibition of CIP2A in Hela cells resulted in reduced phosphorylation of Ser^{62} and decreased c-MYC stabilization. Importantly, ectopic expression CIP2A can also replace ST in the HEK-TER transformation model. However, similar to the results observed with knockdown of PR61γ, CIP2A only weakly phenocopied the expression of the ST antigen in the HEK-TER system. Considering the functional role of c-MYC in oncogenesis and the inhibition of c-MYC activity by PR61α it would be surprising if knockdown of PR61α could not also partly recapitulate the phenotype of ST in the HEK-TER system or if the dual inhibition PR61α and PR61γ would not result in a synergist effect on tumor formation closer to that seen with ST.

On top of its role as a regulator of cMyc, PR61α also appears to play an indirect role in the regulation of the tumor suppressor p53. It was shown that cyclin G, a p53 target, recruits PP2A holoenzyme complexes containing either PR61α or PR61β to form a quaternary complex with the E3 ligase MDM2 [77]. This leads to dephosphorylation of MDM2 at Thr^{216} and the subsequent activation of the protein. MDM2 binds to p53 and ubiquitinates p53 leading to its degradation [78]. In agreement with this, cyclin G null cells exhibit increased MDM2 phosphorylation and stabilized p53 protein levels. Interestingly, the interaction between PR61 and cyclin G is dependent on p53 [79]. These results suggest that PP2A can act as a negative regulator of p53 signaling. Although this piece of data would not support a role for PP2A as a tumor suppressor, recent studies indicate that PP2A can also act as a positive regulator of p53 signaling. PP2A has been shown to directly dephosphorylate p53 at either Ser^{37} or Thr^{55} resulting in increased p53 stabilization and increased apoptosis following DNA damage [80,81]. In agreement with this, targeted inhibition of either PR61γ1 or PR61γ3 abolished Thr^{55} dephosphorylation and reduced overall p53 levels [81].

Finally, recent data have indicated that activation of the oncogene AKT is inhibited by the actions of PP2A. AKT is a serine/threonine kinase that has been shown to be the central node in a number of tumor-promoting pathways (reviewed in Manning et al. [82]). Furthermore a number of studies have demonstrated that AKT is deregulated in human cancers [83,84]. Activation of AKT is tightly regulated by phosphorylation events at either Thr^{308} or Ser^{473}. Holoenzymes containing PR55α directly dephosphorylate AKT at Thr^{308} resulting in the downstream inhibition of the pro-survival effector pathways and overall growth retardation [85].

In addition to the negative regulatory function of specific B subunits in cancer-associated pathways several cancer-associated mutations have been found in the two PR65 scaffold subunits, indicating a causal role for loss of specific PP2A holoenzyme complexes in cancer. Extensive sequencing of human tumor samples revealed that the PR65β scaffold subunit was mutated in 15% (5 out of 33) of primary lung tumors, 6% (4 out of 70) of lung tumor-derived cell lines, and 15% (2 out of 13) of primary colon tumors [86]. The PR65β gene locus (11q23) was found to display LOH in 30%–50% of breast lung ovary cervical carcinomas and melanomas and in 15% of non-Hodgkin's lymphomas and chronic lymphocytic leukemias [86] although ATM kinase, which is frequently mutated in cancer [87], is also present at this locus. Notably the B isoform of PR65 gene was also found to be specifically reduced in approximately 50% of 34 tumor cell lines tested [32]. Most of the PR65β cancer-associated mutants are composed of missense mutations including G8R, P65S, G90D, L101P, K343E, D504G, V545A, V448A, one contains the double mutant L101P/V448A, and one an in frame deletion ΔE344–E388 [88,89]. Mutational analysis of these cancer-associated mutations determined that individual B subunit binding varies greatly between each PR65β mutation with only the ΔE344–E388 mutant incapable of binding to any of the B subunits tested [88,89]. A further five of these mutants compromised catalytic subunit binding leading to an overall loss in phosphatase activity. Interestingly, none of the PR65β mutants tested exhibited a decrease in one specific B subunit. However, only the holoezymes containing PR61γ showed complete loss of integrity in all of the mutants tested [90,88,89]. This suggests that while PR61γ appears to be the critical determinant in the tumor suppressor function of PP2A the loss of other subunits may indeed play a role in cellular transformation.

When addressing the integrity of the PR65α isoform, no reduction in expression was found in 34 tumor cell lines tested which was in contrast to the β isoform [32]. In a different study, PR65α levels were found to be reduced in 43% of primary human gliomas [91], while established glioma cell lines showed uniformly high levels of PR65α [32]. This could suggest that for the establishment of primary tumors in tissue culture, strong selective pressure in favor of high PR65α levels exists. Mutations have also been described in PR65α but at a lot lower frequency [92]. Of these four cancer-associated mutations that were identified, two (R418W and Δ171–589) displayed reduced binding to the catalytic subunit and all regulatory subunits tested. A further two (E64D and E64G) specifically lost efficient binding to the PR61 subunits [93]. Interestingly residue Glu64 of PR65α resides at the interface between PP2Ac and the B subunit forming a critical hydrogen bond between PR65α and residue 309 of PP2Ac [22], therefore providing a logical link between PR65 cancer-associated mutants and loss of holoenzyme composition. Since loss of PR65α is lethal in rats it would be highly surprising to observe a null-mutant in tumors as it appears that at least a minimal level of PR65α is likely to be required to maintain cell viability [94,95].

To assess the individual roles of each of the structural subunits and cancer-derived mutants in tumorigenesis, Hahn and colleagues substituted ST with either knockdown vectors targeting either PR65α or PR65β in HEK-TER cells [89]. As expected, near complete suppression of PR65α induced cell death by apoptosis, while partial inhibition imparts a tumorigenic phenotype. Interestingly, loss of PR65β had full tumor-forming potential, which could not be rescued by ectopic expression of PR65α. Moreover, it was later confirmed that ST exclusively interacts with PR65α and not PR65β suggesting that the tumor-forming capabilities of PR65α and PR65β occur through unique and independent mechanisms. Further studies indicated that the GTPase RalA bound specifically to PR65β containing complexes [89]. This interaction is then lost when PR65β is replaced by the cancer-derived PR65β mutants, leading to increased RalA activity and increased tumor incidence. Moreover it was shown in a lung carcinoma cell line harboring non-functional PR65β alleles that there is a concomitant increase in RalA phosphorylation. As these studies suggest that there appears to be a clear importance in the regulation of RalA activity by PP2A:PR65β complexes. Furthermore, suppression of RalA severely diminished the ability of PR65β mutants to form tumors in the HEK-TER system. Together, these observations indicate that inhibition of PP2A complexes containing either PR65α or PR65β can lead to cellular transformation. However, their mechanistic actions are distinct and unique.

Collectively these data provide compelling evidence for the tumor suppressor function of PP2A. However, the use of artificially engineered transformed fibroblasts, although elegant, probably does not reflect the actual role of PP2A deregulation during in vivo tumor progression. Furthermore, the fact that ST itself can also function as a regulatory B subunit [96] adds another level of complexity to the system. If this ability contributes to the transformation phenotype in engineered tumor cells, loss of endogenous subunits might never fully complement the effects of ST expression. It is also possible that different sets of genetic aberrations during tumor formation require the loss of different PP2A holoenzyme complexes for the tumor to survive. Nevertheless, the data on specific holoenzyme complexes clearly indicate that all B subunit families regulate PP2A in pathways that are deregulated in cancer. However, it appears that the functional loss of PR61γ is the most relevant event for tumorigenesis. As such, the role of PP2A as a tumor suppressor is likely to be more diverse than initially suggested and to be largely context dependent.

3.2. PP2A and MAP kinase signaling

A major function of PP2A, which seems to be conserved between yeast, flies and vertebrates, is the regulation of the RAS-RAF-mitogen-activated protein (MAP) kinase signaling pathways. PP2A can have both activating and inhibitory effects on these pathways, depending on the cell type. Interestingly PP2A selectively targets a number of kinases in this seemingly linear pathway, suggesting that PP2A regulation of the RAF-MEK-ERK pathway might be context dependent. In response to growth factors, PR55α is recruited to a complex at the plasma membrane containing Kinase Suppressor of RAS (KSR1), RAF1 and the PR65/PP2Ac core dimer [97]. This correlates with dephosphorylation of critical 14-3-3 binding sites on KSR1 and RAF-1 resulting in downstream activation of the MAPK pathway. Furthermore, both PR55α and PR55δ can positively regulate MAPK signaling by directly dephosphorylating the inhibitory phosphorylation site on Raf-1 (Ser^{259}) [98]. Similar results were obtained in *C. elegans* although here it was shown that the action of PR55 (cdc55) does not involve RAF1 dephosphorylation [99]. Furthermore it was found that PR55 translocates to the plasma membrane following treatment with platelet-derived growth factor (PDGF) [97] thus activating the RAF1 pathway. These studies support a positive role for PP2A as activators of the RAF1-MAPK pathway. However, both inhibition of PR55 by ST [100] or treatment of cells with okadaic acid [101] also result in the phosphorylation and activation of Extracellular Regulated Kinase (ERK) which is downstream of RAF1. In support of these data, it was found in *Drosophila* cells that ablation of either the PP2A catalytic subunit, the PR65 core subunit or the B/PR55 subunit caused enhanced MAPK signaling in response to insulin [102]. This would suggest that while PP2A positively regulates MAPK signaling at the level of KSR and RAF1, there is also a negative regulatory role for PR55 and PP2A in this pathway. This notion was supported by studies in *Drosophila* where it was shown with activated RAS and RAF mutants that PP2A stimulates MAPK signaling downstream of RAF1, while negatively influencing MAPK signaling upstream of RAF1 [103]. It was subsequently found in mammalian cells that PR55γ could interact with and function as a negative modulator of c-SRC activity [104]. Interestingly it was found by others that c-SRC could indeed activate RAF1 independently of RAS [105,106] and thereby activate the MAPK pathway [107]. These data suggest that PR55 mediates the activation of mitogenic signaling through KSR1 downstream of RAF1 and force the inhibition of mitogenic signaling through c-SRC inhibition upstream of RAF1. Interestingly it also appears that PP2A targets ERK. Recent studies have shown that ablation of PR61β or PR61γ leads to increased ERK activation with no concomitant increase in pMEK levels, the kinase directly upstream of ERK, suggesting that PP2A acts on ERK directly [108].

In agreement with the antagonistic role of PP2A in the MAPK pathway, overexpression of the endogenous inhibitor of PP2A, I2PP2A, increases both c-JUN and ERK activation [109,110]. I1PP2A (PHAP-I) and I2PP2A (SET/PHAP-II) are both potent inhibitors of PP2A [111,112]. I2PP2A was initially identified as a truncated from of the myeloid leukemia associated protein SET which is fused to nucleoporin Nup214 in acute non-lymphocytic myeloid leukemia [112]. Furthermore, high levels of I2PP2A have been observed in a number of different human malignancies, including Wilms' tumor and leukemias [113,114], thus implying a plausible role of PP2A regulation by I2PP2A in cancer. Recent evidence has demonstrated that BCR/ABL requires the induction of I2PP2A, and the resulting PP2A inhibition, to fulfill its tumorigenic potential in chronic myelogenous leukemia (CML) [115]. Suppression of I2PP2A by short hairpin RNAs in BCR/ABL positive cell lines resulted in increased PP2A activity and a reduction in BCR/ABL leukemogenesis in vivo. This corresponded with the dephosphorylation of a number of PP2A target genes including ERK, AKT, MYC, BAD, JAK2, and RB, which have previously been shown to be essential downstream signaling components in BCR/ABL induced leukemogenesis [115]. Moreover, inhibition of BCR-ABL activity by the tyrosine kinase inhibitor imatinib methylate (Gleevac, STI571) inhibits the expression of I2PP2A resulting in the reactivation of PP2A and impairing BCR/ABL function [115].

Although targeting of the BCR/ABL oncogene by imatinib has transformed the treatment of CML, patients who initially respond to this treatment invariably develop resistance. However, a number of PP2A activators have been described capable of potently inhibiting BCR/ABL function including forskolin and FTY720 [115,116]. FTY720, a synthetic myriocin analog structurally similar to sphingosine, was originally developed as a immunosuppressive agent and has shown to induce responses in an number of solid tumors [116-121]. Addition of FTY720 to BCR/ABL overexpressing cell lines results in the dephosphorylation of the PP2A target genes AKT, STAT5 and ERK culminating in the induction of apoptosis in both imatinib sensitive and resistant cells while exhibiting minimal effects on wild type cells. Moreover, the addition of ST or okadaic acid to FTY720 treated cells prevents FTY720 induced BCR/ABL degradation restoring BCR/ABL activity and rescuing the oncogenic potential of these leukemias. Interestingly, reduced PR65β transcript levels have been observed in a subset of leukemia cases [122]. Together these data highlight the importance of PP2A deregulation in BCR/ABL induced leukemogenesis. Furthermore, it supports the use of PP2A activators as a clinically relevant approach to the treatment of a variety of cancers.

Collectively, these data present convincing evidence that PP2A can play both a protagonistic and antagonistic role in regulating the RAF-MEK-ERK pathway, determination of which appears to be largely dependent on stimulus and cell type. A schematic overview of the role of PP2A holoenzymes in MAPK regulation is depicted in Fig. 2.

It was demonstrated both in yeast [123] and in mammalian cells [124] that the adenoviral protein E4orf4 binds to PR55α and in transformed mammalian cells this interaction is required for E4orf4 to induce apoptosis. Furthermore, in yeast ectopic expression of E4orf4 phenocopies loss of PR55 [123]. The interaction between PR55 and E4orf4 was later confirmed by others [125]. However, it was found that an E4orf4 mutant that can no longer interact with PR55 still induces apoptosis, which is dependent on its interaction with c-SRC [125]. Recent data indicate that following cell stress suppression of PR55γ or PR55δ prolongs and enhances the activation of the JNK stress kinase pathway leading to apoptosis [104]. Interestingly, the enhanced JNK activation following PR55γ suppression is dependent on c-SRC [104].This phenotype is similar to that seen with the two c-SRC interacting viral proteins, E4orf4 and pyMT [125]. Expression of pyMT is able to activate JNK by virtue of its interaction with PP2A [126]. Furthermore, pyMT was also found to interact with c-SRC leading to its activation [127,128] and the subsequent activation of the JNK stress kinase pathway. Therefore it is tempting to speculate that both these viral proteins function by displacing PR55γ from the holoenzyme complex, leading to enhanced c-SRC activity and increased apoptosis.

While a number of early studies have shown that loss of holoenzyme integrity is a prerequisite for tumorigenesis by tumor-promoting viral antigens, a number of recent studies have also shown that a number of B subunits can be bumped off following cell stress. PR55α is targeted by an ATM dependent pathway following ionizing radiation, which results in the transient dissociation of PR55α from complexes containing the PR65/PP2Ac core dimer [129]. Similar results have been observed between PR55γ and c-SRC in which the interaction between c-SRC and PR55γ is lost following exposure to UV [104]. Interestingly, the loss of PR55γ binding to c-SRC following cell stress is not dependent on the phosphorylation status of c-SRC.

Holoenzyme formation can also be directly regulated by the substrates themselves. For example, recent studies have indicated that the ERK target gene IEX-1 can form a ternary structure with PP2A and ERK which decreases the phosphatase activity of PP2A towards ERK [108]. Interestingly IEX-1 inhibition of PP2A appears to be ERK dependent. When complexed with IEX-1 ERK transphosphorylates PR61 at a Ser-Pro site, a site conserved in all PR61 family members, triggering the dissociation of PR61 from the core enzyme. This suggests that while the interaction between PP2A subunits and their targets is primarily dependent on the phosphorylation status of the substrate it insinuates that holoenzyme formation is dynamic and can be negatively regulated by various mechanisms.

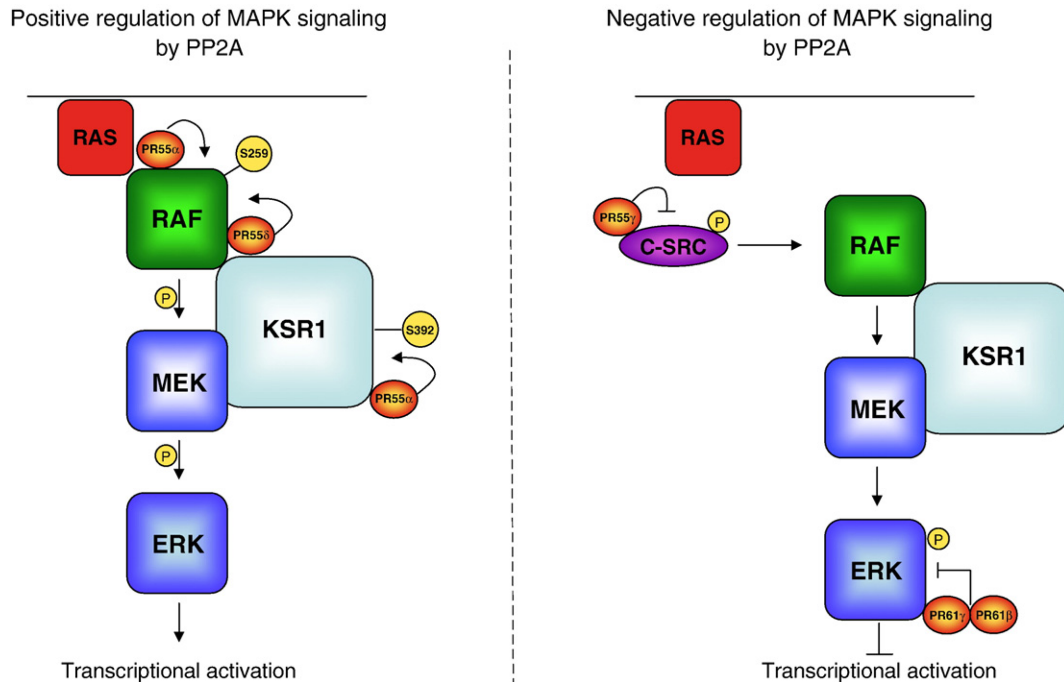

Fig. 2. Schematic overview of MAPK signaling by PP2A. PP2A can play both a positive and negative role in MAPK signaling. Upon stimulation RAS promotes binding of PR55α to KSR1 and RAF leading to the dephosphorylation of Ser^{392} and Ser^{295}, respectively. This results in the loss of 14-3-3 inhibition and the subsequent recruitment of RAF to the plasma membrane facilitating the RAS/RAF interaction. Similarly, PR55α can dephosphorylate key inhibitory 14-3-3 sites on KSR1 allowing KSR1 to act as a scaffold protein permitting the activation the signaling cascade. RAF can also be dephosphorylated at Ser^{295} by PR55δ leading to downstream ERK activation. In contrast, PP2A can negatively regulate MAPK signaling by inhibiting the kinase activity of c-SRC. c-SRC activates RAF independently of RAS leading to ERK activation. Furthermore, ERK can be directly dephosphorylated by PR61β and PR61γ resulting in decreased ERK activation.

Since it was previously shown that expression of ST results in the activation of a number of these signaling pathways, which appear to be primarily but not exclusively regulated by PR55B subunits, it would be surprising if the loss of these subunits did not play some role in cellular transformation.

3.3. PP2A and WNT signaling

The WNT signaling cascade is required for several crucial steps during early embryogenesis. Deregulation of the WNT signaling cascade in adult tissues can also lead to uncontrolled cellular proliferation and tumorigenesis [130]. One of the targets of the WNT pathway is β-catenin which, when stabilized, accumulates in the nucleus leading to evolution of many cancers. The function of PP2A in WNT regulation is similar to its role in the RAF-MEK-ERK signaling cascade with individual PP2A family members exerting both dominant and negative effects on this pathway. However, the net effect of PP2A inhibition through the use of okadaic acid does lead to stabilization of β-catenin [131], suggesting that the dominate role of PP2A in this pathway is that of a negative regulator. It is however important to note that the stabilization of β-catenin with okadaic acid does not correlate with its phosphorylation status, suggesting that its stabilization may be unrelated to WNT signaling [132].

The first evidence that PP2A controls WNT signaling was obtained using the adenomatous polyposis coli (APC) protein as bait in a yeast two-hybrid assay. This led to the isolation of the PP2A B subunits PR61α and PR61δ. Further experiments indicated that overexpression of PR61α decreased the levels of β-catenin leading to an overall reduction in WNT signaling both in tissue culture and in *Xenopus* cell extracts [131]. However, the effects presented were not specific for PR61α only but conserved among all the PR61 family members tested [131]. The same group later reported that the PP2A subunits PR61α, PR61β, and PR61γ could interact with axin as well and when overexpressed could inhibit WNT signaling during vertebrate development [133]. It should be noted that as of yet no direct substrate has been identified for the phosphatase activity of PP2A in the WNT signaling pathway. However this should not diminish the clear role that PP2A plays in WNT regulation.

One of the downstream target genes of the WNT signaling cascade is the protein Naked cuticle (Naked). Naked is required to restrict WNT signaling during *Drosophila* embryonic segmentation, thus acting as negative feedback regulator of WNT signaling. Interestingly, recent data have shown that Naked requires the presence of the PP2A subunit PR72 for its negative regulatory function [134]. As previously described, ectopic expression of Naked inhibited WNT signaling [135]. However, co-expression of hairpins targeting PR72 completely limited the restrictive capacity of Naked on the WNT pathway [134]. Interestingly, PR72 is required to tether the PP2A/PR65 dimer to Naked and it was shown that PR72 itself is a target of PP2A, suggesting that PR72 might have different functions, which are regulated by direct PP2A-mediated dephosphorylation. Notably, Naked, like PR72, also harbors an EFX hand domain, suggesting that both these proteins are regulated by calcium to alter their conformation [136]. Taken together these reports show that PP2A plays a negative role in WNT signaling and clearly demonstrates that PP2A can associate with a number of WNT signaling components such as APC [131], axin [133,137] and Naked [134].

Initial experiments using PR61ε also described this subunit as a negative regulator of the WNT pathway. PR61ε binds to both disheveled and APC inhibiting WNT/β-catenin signaling [131]. However in direct contrast to these data, knockdown experiments of PR61ε later convincingly established that PR61ε, rather than being a negative regulator of WNT signaling, acts as a positive modulator of WNT signaling during vertebrate development upstream of disheveled [70]. Earlier studies on PR55 (named "twins" in *Drosophila*) also report differing phenotypes comparing overexpression and knockdown of the same subunit [71]. It has been proposed in both these reports that regulatory subunits when overexpressed can exert a dominant negative effect due to sequestration of essential components of the PP2A complex or endogenous targets [70,71]. The majority of experiments discussed in the reports above are based on overexpression of B subunits. This approach has the potential artifact that overexpression of one type of B subunit may displace endogenous B subunits. We therefore urge the reader to interpret these data with caution until confirmed by knockdown or knockout experiments.

Be that as it may, during development PR55/twins is required for WNT signaling downstream of disheveled and upstream of the GSK3β, establishing a positive role for PR55 during *Drosophila* development [71]. Analyses of twins (−/−) mitotic clones suggest that twins is required for stabilization of β-catenin in response to WNT signaling. The authors speculate on a role for Vn/EGFR signaling in the deregulation of WNT signaling by PR55 since Vn/EGFR is a known antagonist of wing development. Since c-SRC is a regulator of EGFR [138] it would be interesting to explore whether this developmental role for PR55 is also mediated through c-SRC deregulation. In addition, a role for PR61α in the regulation of WNT signaling was found when studying another member of the p53 family: p63 [132]. A yeast two-hybrid screen revealed that PR61α was able to interact with p63 and overexpression of p63 led to β-catenin stabilization through the negative regulation of GSK3β. Notably the interaction of PR61α with p63 did not lead to recruitment of the PP2A holoenzyme to p63 [132]. Strangely however the authors do find a drop in general PP2A activity when both these proteins are overexpressed although the relevance of this remains undetermined.

PP2A can also stabilize β-catenin directly. PP2A has been observed to form a complex with E-cadherin and β-catenin at the plasma membrane resulting in the stabilization of both proteins. In contrast, ablation of PP2Acα expression results in β-catenin being redistriduted to the cytoplasm where it is rapidly degraded by the proteasome [139]. Although the specific B subunit has not been described loss of PP2A clearly plays a positive role in Wnt signaling.

Finally, PR130, encoded by the larger alternative transcript of PR72, was also found to interact with Naked [140]. However, unlike its sibling, PR130 acts as a positive regulator of WNT signaling inhibiting the effects of Naked both in tissue culture and during embryonic development where it is required for the correct alignment of somite boundaries [140]. Notably, both loss of PR72 and loss of PR130 respectively phenocopy gain and loss of WNT signaling during vertebrate somitogenesis [141]. Similarly, the role of PP2A catalytic activity during this regulation remains elusive, since no dephosphorylation of naked cuticle or other possible targets was observed. A schematic overview of the role of PP2A holoenzymes in Wnt signaling is depicted in Fig. 3.

Collectively these reports provide convincing evidence that PP2A, through a variety of mechanisms, plays both a positive and negative role in the regulation of WNT signaling, thus emphasizing the critical role in PP2A mediated regulation of both embryogenesis and the regulation of cellular proliferation in adult tissues.

4. Conclusions

Our initial understanding of PP2A as a putative tumor suppressor was primarily based on loss of function analysis using the general PP2A inhibitors okadaic acid or expression of tumor antigens from several tumor-promoting viruses. As such, the precise role of the individual PP2A B regulatory subunits remained undisclosed. Not until recent advances in gene knockdown technology have we been able to dissect the individual roles of the PP2A holoenzymes and their specific roles in several signaling pathways. Considering the positive and negative roles PP2A appears to play in a number of different cancer-promoting pathways, the description of PP2A as a *bona fide* tumor suppressor is a bit misleading since not all holoenzyme complexes can

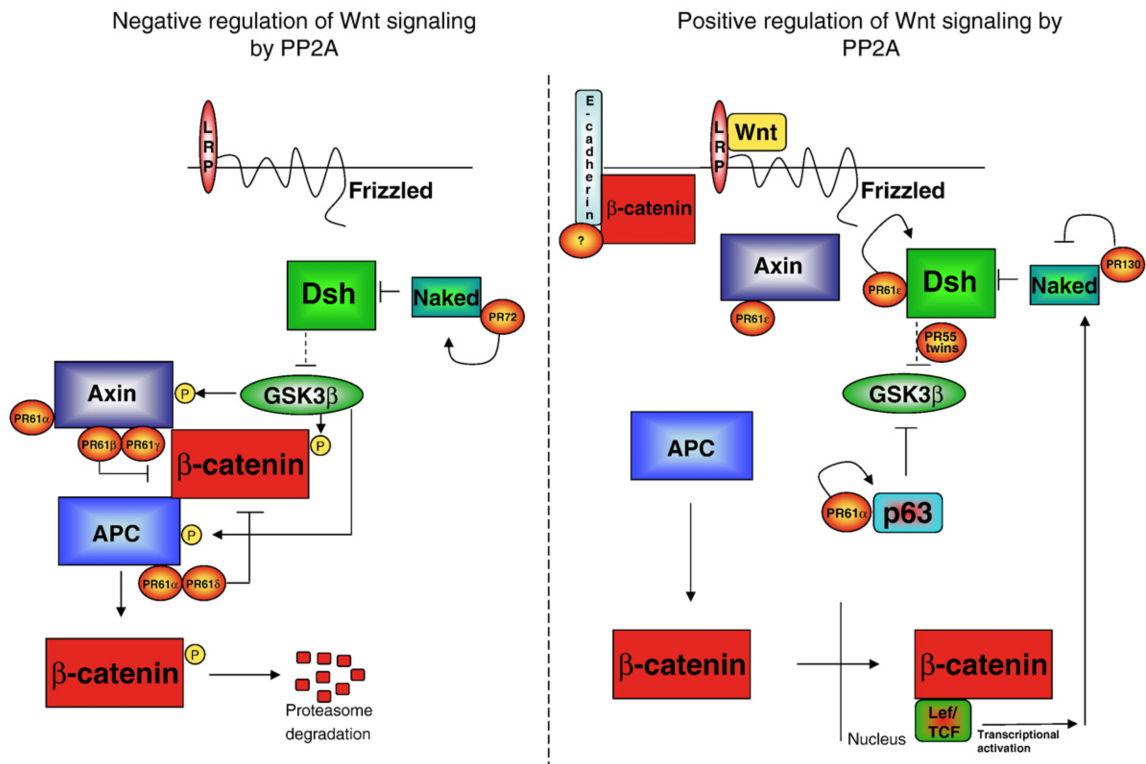

Fig. 3. Schematic overview of Wnt regulation by PP2A. PP2A exerts both dominant and negative effects on the Wnt pathway. In the absence of Wnt signal β-catenin is complexed with APC, AXIN, and GSK-3β in the cytosol. GSK-3β negatively regulates β-catenin by phosphorylation rendering it unstable. Phosphorylated β-catenin is subsequently ubiquitinated and targeted by the proteasome for degradation. The PP2A B subunits PR61α and PR61δ bind to APC leading to decreased levels of β-catenin. Similarly, the PP2A B subunits PR61α, PR61β, and PR61γ bind to Axin also leading to decreased levels of β-catenin. The precise mechanism of how PP2A negatively regulates the Wnt signaling pathway through the destabilization of β-catenin remains unclear. Upon Wnt activation the APC, AXIN, and GSK-3β complex is degraded by Dsh. Unphosphorylated β-catenin localizes in the nucleus permitting transactivation of specific Wnt target genes. Naked, one of the downstream target genes of the Wnt signaling cascade, inhibits Dsh expression through a negative feedback mechanism. Naked appears to require PR72 for its negative regulatory function. In contrast, the ability of Naked to bind to Dsh is repressed by PR130, resulting in increased Dsh expression. Dsh activity itself is positively regulated by PR61ε. Furthermore, PR55 (twins) acts downstream of Dsh. Both PR61ε and PR55 positively regulate Wnt signaling by destabilizing the inhibitory properties of GSK-3β. Similarly p63 stabilizes β-catenin through the negative regulation of GSK-3β. β-catenin may also reside at the plasma membrane complexed with E-cadherin and PP2A. Here, PP2A appears to play a role in the stabilization of E-cadherin and β-catenin, however, its precise role in Wnt signaling remains to be determined.

subscribe to this function. Rather it appears that only a few individual β subunits can be classified as true tumor suppressors and their function as such can be context dependent. Surprisingly however no mutations have been found in any B subunits, suggesting that deregulation of a single holoenzyme complex may not be sufficient to promote tumorigenesis. Recent reports have described that specific inhibition of only the B subunit PR61γ could enhance the transformation in RAS^{V12}/hTERT/SV40 LT transformed human fibroblasts [66]. However, PR61γ knockdown only weakly phenocopied expression of ST antigen in these cells, suggesting that other B subunits are involved in tumor suppression [67]. Arguments for this can be taken from the fact that the cancer-associated mutations observed in the structural subunits of PP2A Aα and PP2A Aβ are not specific to the loss of PR61γ binding, but lead to the deregulation of distinct PP2A holoenzymes [86,88]. Even so the loss of PR61γ binding was observed in all PR65β mutations, suggesting that the loss of PR61γ may be a key factor in cellular transformation, but probably requires the loss of other B subunits for complete tumorigenesis.

One of the most interesting recent observations is that partial inhibition of PR65α or complete suppression of PR65β can induce transformation in the HEK-TER system, albeit through the deregulation of distinct downstream signaling pathways. Similarly ectopic expression of either polyoma and SV40 proteins appears to differentially regulate PP2A holonzymes activating distinct signaling pathways leading to cellular transformation. ST, which binds only to PR65α, specifically regulates the AKT pathway, while PyMT specifically activates the MAPK pathway. This suggests that the deregulation of PP2A in a number of distinct but unique pathways is sufficient to promote tumorigenesis.

Another interesting finding has been the demonstration that the expression of PR61γ is suppressed in melanomas compared with non-transformed melanocytic naevi [68]. B-RAF mutations have frequently been observed in naevi leading to a senescent like phenotype [142]. Furthermore, overexpression of the oncogenic mutant of B-RAF (E600) induces cellular senescence in a number of cultured cell lines (reviewed in Michaloglou et al. [143]). As previously described, PP2A inhibits a number of proteins in the RAF-MEK-ERK pathway including RAF itself. Moreover, recent evidence has shown that PR61γ directly dephosphorylates ERK, thereby inhibiting ERK activity.

Similarly, a number of reports have suggested that another role of PP2A appears to be in the suppression of the downstream components of the RAS signaling pathway [89,144]. Indeed data clearly indicate that one of the major functions of PP2A is to antagonize the oncogenic ability of RAS by directly dephosphorylating and inhibiting the RAS downstream effectors c-MYC, RALA and AKT. Therefore it appears that deregulation of PP2A in either the RAS or B-RAF pathway is an essential requirement for oncogenic RAS and B-RAF to induce a fully transformed phenotype.

Collectively these observations highlight the role of PP2A deregulation in cancer. Through the upregulation of a number protein kinases involved in mitogenic and survival signaling (e.g. AKT and MAPK), the stabilization of protoncogenes (e.g. MYC), the destabilization of tumor suppressors (e.g. p53 and RB), or the loss of pro-apoptotic signaling pathways (e.g. BAD), the loss of specific PP2A

holoezymes is a critical determinant towards cellular transformation. No doubt further studies will lead to a better understanding of the dynamic interaction of PP2A regulatory β subunits within the diverse signaling cascades leading to a more complete understanding of which holoenzyme complexes function as tumor suppressors and in which genetic context(s).

References

[1] C. Nolan, W.B. Novoa, E.G. Krebs, E.H. Fischer, Further studies on the site phosphorylated in the phosphorylase B to a reaction, Biochemistry. 3 (1964) 542–551.

[2] Z. Wang, D. Shen, D.W. Parsons, A. Bardelli, J. Sager, S. Szabo, J. Ptak, N. Silliman, B.A. Peters, M.S. van der Heijden, G. Parmigiani, H. Yan, T.L. Wang, G. Riggins, S.M. Powell, J.K. Willson, S. Markowitz, K.W. Kinzler, B. Vogelstein, V.E. Velculescu, Mutational analysis of the tyrosine phosphatome in colorectal cancers, Science. 304 (2004) 1164–1166.

[3] J.S. Sebolt-Leopold, R. Herrera, Targeting the mitogen-activated protein kinase cascade to treat cancer, Nat. Rev. Cancer. 4 (2004) 937–947.

[4] C.L. Sawyers, Opportunities and challenges in the development of kinase inhibitor therapy for cancer, Genes Dev. 17 (2003) 2998–3010.

[5] J. Dancey, E.A. Sausville, Issues and progress with protein kinase inhibitors for cancer treatment, Nat. Rev. Drug Discov. 2 (2003) 296–313.

[6] J.P. MacKeigan, L.O. Murphy, J. Blenis, Sensitized RNAi screen of human kinases and phosphatases identifies new regulators of apoptosis and chemoresistance, Nat. Cell Biol. 7 (2005) 591–600.

[7] M. Suganuma, H. Fujiki, H. Suguri, S. Yoshizawa, M. Hirota, M. Nakayasu, M. Ojika, K. Wakamatsu, K. Yamada, T. Sugimura, Okadaic acid: an additional non-phorbol-12-tetradecanoate-13-acetate-type tumor promoter, Proc. Natl. Acad. Sci. U. S. A. 85 (1988) 1768–1771.

[8] M. Suganuma, H. Fujiki, H. Furuya-Suguri, S. Yoshizawa, S. Yasumoto, Y. Kato, N. Fusetani, T. Sugimura, Calyculin A, an inhibitor of protein phosphatases, a potent tumor promoter on CD-1 mouse skin, Cancer Res. 50 (1990) 3521–3525.

[9] H. Fujiki, M. Suganuma, Tumor promotion by inhibitors of protein phosphatases 1 and 2A: the okadaic acid class of compounds, Adv. Cancer Res. 61 (1993) 143–194.

[10] C. Bialojan, A. Takai, Inhibitory effect of a marine-sponge toxin, okadaic acid, on protein phosphatases, Specificity kinetics Biochem. J. 256 (1988) 283–290.

[11] A.H. Schonthal, Role of PP2A in intracellular signal transduction pathways, Front Biosci. 3 (1998) D1262–1273.

[12] D.C. Pallas, L.K. Shahrik, B.L. Martin, S. Jaspers, T.B. Miller, D.L. Brautigan, T.M. Roberts, Polyoma small and middle T antigens and SV40 small t antigen form stable complexes with protein phosphatase 2A, Cell. 60 (1990) 167–176.

[13] K.S. Campbell, K.R. Auger, B.A. Hemmings, T.M. Roberts, D.C. Pallas, Identification of regions in polyomavirus middle T and small t antigens important for association with protein phosphatase 2A, J. Virol. 69 (1995) 3721–3728.

[14] E. Sontag, Protein phosphatase 2A: the Trojan Horse of cellular signaling, Cell Signal. 13 (2001) 7–16.

[15] V. Janssens, J. Goris, Protein phosphatase 2A: a highly regulated family of serine/threonine phosphatases implicated in cell growth and signalling, Biochem. J. 353 (2001) 417–439.

[16] T. Fellner, D.H. Lackner, H. Hombauer, P. Piribauer, I. Mudrak, K. Zaragoza, C. Juno, E. Ogris, A novel and essential mechanism determining specificity and activity of protein phosphatase 2A (PP2A) in vivo, Genes. Dev. 17 (2003) 2138–2150.

[17] S.R. Stone, J. Hofsteenge, B.A. Hemmings, Molecular cloning of cDNAs encoding two isoforms of the catalytic subunit of protein phosphatase 2A, Biochemistry. 26 (1987) 7215–7220.

[18] Y. Khew-Goodall, B.A. Hemmings, Tissue-specific expression of mRNAs encoding alpha- and beta-catalytic subunits of protein phosphatase 2A, FEBS. Lett. 238 (1988) 265–268.

[19] Z. Baharians, A.H. Schonthal, Autoregulation of protein phosphatase type 2A expression, J. Biol. Chem. 273 (1998) 19019–19024.

[20] J. Gotz, A. Probst, E. Ehler, B. Hemmings, W. Kues, Delayed embryonic lethality in mice lacking protein phosphatase 2A catalytic subunit Calpha, Proc. Natl. Acad. Sci. U. S. A. 95 (1998) 12370–12375.

[21] U.S. Cho, W. Xu, Crystal structure of a protein phosphatase 2A heterotrimeric holoenzyme, Nature. 445 (2007) 53–57.

[22] Y. Xu, Y. Xing, Y. Chen, Y. Chao, Z. Lin, E. Fan, J.W. Yu, S. Strack, P.D. Jeffrey, Y. Shi, Structure of the protein phosphatase 2A holoenzyme, Cell. 127 (2006) 1239–1251.

[23] S. Longin, K. Zwaenepoel, J.V. Louis, S. Dilworth, J. Goris, V. Janssens, Selection of protein phosphatase 2A regulatory subunits is mediated by the C terminus of the catalytic Subunit, J. Biol. Chem. 282 (2007) 26971–26980.

[24] E. Ogris, X. Du, K.C. Nelson, E.K. Mak, X.X. Yu, W.S. Lane, D.C. Pallas, A protein phosphatase methylesterase (PME-1) is one of several novel proteins stably associating with two inactive mutants of protein phosphatase 2A, J. Biol. Chem. 274 (1999) 14382–14391.

[25] J. Chen, B.L. Martin, D.L. Brautigan, Regulation of protein serine–threonine phosphatase type-2A by tyrosine phosphorylation, Science. 257 (1992) 1261–1264.

[26] S. Longin, J. Jordens, E. Martens, I. Stevens, V. Janssens, E. Rondelez, I. De Baere, R. Derua, E. Waelkens, J. Goris, C. Van Hoof, An inactive protein phosphatase 2A population is associated with methylesterase and can be re-activated by the phosphotyrosyl phosphatase activator, Biochem. J. 380 (2004) 111–119.

[27] H. Hombauer, D. Weismann, I. Mudrak, C. Stanzel, T. Fellner, D.H. Lackner, E. Ogris, Generation of active protein phosphatase 2A is coupled to holoenzyme assembly, PLoS. Biol. 5 (2007) e155.

[28] M.R. Groves, N. Hanlon, P. Turowski, B.A. Hemmings, D. Barford, The structure of the protein phosphatase 2A PR65/A subunit reveals the conformation of its 15 tandemly repeated HEAT motifs, Cell. 96 (1999) 99–110.

[29] B.A. Hemmings, C. Adams-Pearson, F. Maurer, P. Muller, J. Goris, W. Merlevede, J. Hofsteenge, S.R. Stone, alpha- and beta-forms of the 65-kDa subunit of protein phosphatase 2A have a similar 39 amino acid repeating structure, Biochemistry. 29 (1990) 3166–3173.

[30] P. Hendrix, P. Turowski, R.E. Mayer-Jaekel, J. Goris, J. Hofsteenge, W. Merlevede, B.A. Hemmings, Analysis of subunit isoforms in protein phosphatase 2A holoenzymes from rabbit and *Xenopus*, J. Biol. Chem. 268 (1993) 7330–7337.

[31] M. Bosch, X. Cayla, C. Van Hoof, B.A. Hemmings, R. Ozon, W. Merlevede, J. Goris, The PR55 and PR65 subunits of protein phosphatase 2A from *Xenopus laevis*. Molecular cloning and developmental regulation of expression, Eur. J. Biochem. 230 (1995) 1037–1045.

[32] J. Zhou, H.T. Pham, R. Ruediger, G. Walter, Characterization of the Aalpha and Abeta subunit isoforms of protein phosphatase 2A: differences in expression, subunit interaction, and evolution, Biochem. J. 369 (2003) 387–398.

[33] C. Van Hoof, J. Goris, Phosphatases in apoptosis: to be or not to be, PP2A is in the heart of the question, Biochim. Biophys. Acta. 1640 (2003) 97–104.

[34] A.H. Schonthal, Role of serine/threonine protein phosphatase 2A in cancer, Cancer Lett. 170 (2001) 1–13.

[35] J. Zhou, H.T. Pham, G. Walter, The formation and activity of PP2A holoenzymes do not depend on the isoform of the catalytic subunit, J. Biol. Chem. 278 (2003) 8617–8622.

[36] X. Li, D.M. Virshup, Two conserved domains in regulatory B subunits mediate binding to the A subunit of protein phosphatase 2A, Eur. J. Biochem. 269 (2002) 546–552.

[37] S. Strack, D. Chang, J.A. Zaucha, R.J. Colbran, B.E. Wadzinski, Cloning and characterization of B delta, a novel regulatory subunit of protein phosphatase 2A, FEBS. Lett. 460 (1999) 462–466.

[38] R.E. Mayer, P. Hendrix, P. Cron, R. Matthies, S.R. Stone, J. Goris, W. Merlevede, J. Hofsteenge, B.A. Hemmings, Structure of the 55-kDa regulatory subunit of protein phosphatase 2A: evidence for a neuronal-specific isoform, Biochemistry. 30 (1991) 3589–3597.

[39] S. Zolnierowicz, C. Csortos, J. Bondor, A. Verin, M.C. Mumby, A.A. DePaoli-Roach, Diversity in the regulatory B-subunits of protein phosphatase 2A: identification of a novel isoform highly expressed in brain, Biochemistry. 33 (1994) 11858–11867.

[40] S. Strack, J.A. Zaucha, F.F. Ebner, R.J. Colbran, B.E. Wadzinski, Brain protein phosphatase 2A: developmental regulation and distinct cellular and subcellular localization by B subunits, J. Comp. Neurol. 392 (1998) 515–527.

[41] J.C. Bryant, R.S. Westphal, B.E. Wadzinski, Methylated C-terminal leucine residue of PP2A catalytic subunit is important for binding of regulatory Balpha subunit, Biochem. J. 339 (Pt 2) (1999) 241–246.

[42] R. Koren, L. Rainis, T. Kleinberger, The scaffolding A/Tpd3 subunit and high phosphatase activity are dispensable for Cdc55 function in the *Saccharomyces cerevisiae* spindle checkpoint and in cytokinesis, J. Biol. Chem. 279 (2004) 48598–48606.

[43] C. Kamibayashi, R.L. Lickteig, R. Estes, G. Walter, M.C. Mumby, Expression of the A subunit of protein phosphatase 2A and characterization of its interactions with the catalytic and regulatory subunits, J. Biol. Chem. 267 (1992) 21864–21872.

[44] B. McCright, A.M. Rivers, S. Audlin, D.M. Virshup, The B56 family of protein phosphatase 2A (PP2A) regulatory subunits encodes differentiation-induced phosphoproteins that target PP2A to both nucleus and cytoplasm, J. Biol. Chem. 271 (1996) 22081–22089.

[45] B. McCright, D.M. Virshup, Identification of a new family of protein phosphatase 2A regulatory subunits, J. Biol. Chem. 270 (1995) 26123–26128.

[46] C. Csortos, S. Zolnierowicz, E. Bako, S.D. Durbin, A.A. DePaoli-Roach, High complexity in the expression of the B¢ subunit of protein phosphatase 2A0. Evidence for the existence of at least seven novel isoforms, J. Biol. Chem. 271 (1996) 2578–2588.

[47] P. Hendrix, R.E. Mayer-Jackel, P. Cron, J. Goris, J. Hofsteenge, W. Merlevede, B.A. Hemmings, Structure and expression of a 72-kDa regulatory subunit of protein phosphatase 2A. Evidence for different size forms produced by alternative splicing, J. Biol. Chem. 268 (1993) 15267–15276.

[48] V. Janssens, J. Jordens, I. Stevens, C. Van Hoof, E. Martens, H. De Smedt, Y. Engelborghs, E. Waelkens, J. Goris, Identification and functional analysis of two Ca2+-binding EF-hand motifs in the B²/PR72 subunit of protein phosphatase 2A, J. Biol. Chem. 278 (2003) 10697–10706.

[49] Y. Xing, H. Igarashi, X. Wang, N. Sakaguchi, Protein phosphatase subunit G5PR is needed for inhibition of B cell receptor-induced apoptosis, J. Exp. Med. 202 (2005) 707–719.

[50] A. Magenta, P. Fasanaro, S. Romani, V. Di Stefano, M.C. Capogrossi, F. Martelli, PP2A-PR70 interacts with pRb and mediates its de-phosphorylation, Mol. Cell. Biol (2007).

[51] Z. Yan, S.A. Fedorov, M.C. Mumby, R.S. Williams, PR48, a novel regulatory subunit of protein phosphatase 2A, interacts with Cdc6 and modulates DNA replication in human cells, Mol. Cell Biol. 20 (2000) 1021–1029.

[52] P.M. Voorhoeve, E.M. Hijmans, R. Bernards, Functional interaction between a novel protein phosphatase 2A regulatory subunit, PR59, and the retinoblastoma-related p107 protein, Oncogene. 18 (1999) 515–524.

[53] X. Cayla, J. Goris, J. Hermann, P. Hendrix, R. Ozon, W. Merlevede, Isolation and characterization of a tyrosyl phosphatase activator from rabbit skeletal muscle and *Xenopus laevis* oocytes, Biochemistry. 29 (1990) 658–667.
[54] J. Goris, C.J. Pallen, P.J. Parker, J. Hermann, M.D. Waterfield, W. Merlevede, Conversion of a phosphoseryl/threonyl phosphatase into a phosphotyrosyl phosphatase, Biochem. J. 256 (1988) 1029–1034.
[55] V. Janssens, C. van Hoof, E. Martens, I. de Baere, W. Merlevede, J. Goris, Identification and characterization of alternative splice products encoded by the human phosphotyrosyl phosphatase activator gene, Eur. J. Biochem. 267 (2000) 4406–4413.
[56] R. Shtrichman, R. Sharf, H. Barr, T. Dobner, T. Kleinberger, Induction of apoptosis by adenovirus E4orf4 protein is specific to transformed cells and requires an interaction with protein phosphatase 2A, Proc. Natl. Acad. Sci. U. S A. 96 (1999) 10080–10085.
[57] X. Cayla, K. Ballmer-Hofer, W. Merlevede, J. Goris, Phosphatase 2A associated with polyomavirus small-T or middle-T antigen is an okadaic acid-sensitive tyrosyl phosphatase, Eur. J. Biochem. 214 (1993) 281–286.
[58] C. Kamibayashi, R. Estes, R.L. Lickteig, S.I. Yang, C. Craft, M.C. Mumby, Comparison of heterotrimeric protein phosphatase 2A containing different B subunits, J. Biol. Chem. 269 (1994) 20139–20148.
[59] S.I. Yang, R.L. Lickteig, R. Estes, K. Rundell, G. Walter, M.C. Mumby, Control of protein phosphatase 2A by simian virus 40 small-t antigen, Mol. Cell Biol. 11 (1991) 1988–1995.
[60] S.M. Dilworth, Polyoma virus middle T antigen and its role in identifying cancer-related molecules, Nat Rev Cancer. 2 (2002) 951–956.
[61] C. Skoczylas, K.M. Fahrbach, K. Rundell, Cellular targets of the SV40 small-t antigen in human cell transformation, Cell Cycle. 3 (2004) 606–610.
[62] G. Walter, R. Ruediger, C. Slaughter, M. Mumby, Association of protein phosphatase 2A with polyoma virus medium tumor antigen, Proc. Natl. Acad. Sci. U. S. A. 87 (1990) 2521–2525.
[63] P. Rodriguez-Viciana, C. Collins, M. Fried, Polyoma and SV40 proteins differentially regulate PP2A to activate distinct cellular signaling pathways involved in growth control, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 19290–19295.
[64] S. Andrabi, O.V. Gjoerup, J.A. Kean, T.M. Roberts, B. Schaffhausen, Protein phosphatase 2A regulates life and death decisions via Akt in a context-dependent manner, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 19011–19016.
[65] W.C. Hahn, C.M. Counter, A.S. Lundberg, R.L. Beijersbergen, M.W. Brooks, R.A. Weinberg, Creation of human tumour cells with defined genetic elements, Nature. 400 (1999) 464–468.
[66] W. Chen, R. Possemato, K.T. Campbell, C.A. Plattner, D.C. Pallas, W.C. Hahn, Identification of specific PP2A complexes involved in human cell transformation, Cancer Cell. 5 (2004) 127–136.
[67] C.S. Moreno, S. Ramachandran, D.G. Ashby, N. Laycock, C.A. Plattner, W. Chen, W.C. Hahn, D.C. Pallas, Signaling and transcriptional changes critical for transformation of human cells by simian virus 40 small tumor antigen or protein phosphatase 2A B56gamma knockdown, Cancer Res. 64 (2004) 6978–6988.
[68] M. Deichmann, M. Polychronidis, J. Wacker, M. Thome, H. Naher, The protein phosphatase 2A subunit Bgamma gene is identified to be differentially expressed in malignant melanomas by subtractive suppression hybridization, Melanoma Res. 11 (2001) 577–585.
[69] A. Ito, T.R. Kataoka, M. Watanabe, K. Nishiyama, Y. Mazaki, H. Sabe, Y. Kitamura, H. Nojima, A truncated isoform of the PP2A B56 subunit promotes cell motility through paxillin phosphorylation, EMBO. J. 19 (2000) 562–571.
[70] J. Yang, J. Wu, C. Tan, P.S. Klein, PP2A:B56epsilon is required for Wnt/beta-catenin signaling during embryonic development, Development. 130 (2003) 5569–5578.
[71] R. Bajpai, K. Makhijani, P.R. Rao, L.S. Shashidhara, *Drosophila* Twins regulates Armadillo levels in response to Wg/Wnt signal, Development. 131 (2004) 1007–1016.
[72] H.K. Arnold, R.C. Sears, Protein phosphatase 2A regulatory subunit B56alpha associates with c-myc and negatively regulates c-myc accumulation, Mol. Cell Biol. 26 (2006) 2832–2844.
[73] E. Yeh, M. Cunningham, H. Arnold, D. Chasse, T. Monteith, G. Ivaldi, W.C. Hahn, P.T. Stukenberg, S. Shenolikar, T. Uchida, C.M. Counter, J.R. Nevins, A.R. Means, R. Sears, A signalling pathway controlling c-Myc degradation that impacts oncogenic transformation of human cells, Nat. Cell Biol. 6 (2004) 308–318.
[74] P.P. Ruvolo, W. Clark, M. Mumby, F. Gao, W.S. May, A functional role for the B56 alpha-subunit of protein phosphatase 2A in ceramide-mediated regulation of Bcl2 phosphorylation status and function, J. Biol. Chem. 277 (2002) 22847–22852.
[75] C.W. Chiang, C. Kanies, K.W. Kim, W.B. Fang, C. Parkhurst, M. Xie, T. Henry, E. Yang, Protein phosphatase 2A dephosphorylation of phosphoserine 112 plays the gatekeeper role for BAD-mediated apoptosis, Mol. Cell Biol. 23 (2003) 6350–6362.
[76] M.R. Junttila, P. Puustinen, M. Niemela, R. Ahola, H. Arnold, T. Bottzauw, R. Ala-aho, C. Nielsen, J. Ivaska, Y. Taya, S.L. Lu, S. Lin, E.K. Chan, X.J. Wang, R. Grenman, J. Kast, T. Kallunki, R. Sears, V.M. Kahari, J. Westermarck, CIP2A inhibits PP2A in human malignancies, Cell. 130 (2007) 51–62.
[77] K. Okamoto, H. Li, M.R. Jensen, T. Zhang, Y. Taya, S.S. Thorgeirsson, C. Prives, Cyclin G recruits PP2A to dephosphorylate Mdm2, Mol. Cell. 9 (2002) 761–771.
[78] Y. Haupt, R. Maya, A. Kazaz, M. Oren, Mdm2 promotes the rapid degradation of p53, Nature. 387 (1997) 296–299.
[79] K. Okamoto, C. Kamibayashi, M. Serrano, C. Prives, M.C. Mumby, D. Beach, p53-dependent association between cyclin G and the B¢ subunit of protein phosphatase 2A, Mol. Cell Biol. 16 (1996) 6593–6602.
[80] K.M. Dohoney, C. Guillerm, C. Whiteford, C. Elbi, P.F. Lambert, G.L. Hager, J.N. Brady, Phosphorylation of p53 at serine 37 is important for transcriptional activity and regulation in response to DNA damage, Oncogene. 23 (2004) 49–57.
[81] H.H. Li, X. Cai, G.P. Shouse, L.G. Piluso, X. Liu, A specific PP2A regulatory subunit, B56gamma, mediates DNA damage-induced dephosphorylation of p53 at Thr55, EMBO. J. 26 (2007) 402–411.
[82] B.D. Manning, L.C. Cantley, AKT/PKB signaling: navigating downstream, Cell. 129 (2007) 1261–1274.
[83] A. Bellacosa, D. de Feo, A.K. Godwin, D.W. Bell, J.Q. Cheng, D.A. Altomare, M. Wan, L. Dubeau, G. Scambia, V. Masciullo, G. Ferrandina, P. Benedetti Panici, S. Mancuso, G. Neri, J.R. Testa, Molecular alterations of the AKT2 oncogene in ovarian and breast carcinomas, Int. J. Cancer. 64 (1995) 280–285.
[84] J.D. Carpten, A.L. Faber, C. Horn, G.P. Donoho, S.L. Briggs, C.M. Robbins, G. Hostetter, S. Boguslawski, T.Y. Moses, S. Savage, M. Uhlik, A. Lin, J. Du, Y.W. Qian, D.J. Zeckner, G. Tucker-Kellogg, J. Touchman, K. Patel, S. Mousses, M. Bittner, R. Schevitz, M.H. Lai, K.L. Blanchard, J.E. Thomas, A transforming mutation in the pleckstrin homology domain of AKT1 in cancer, Nature. 448 (2007) 439–444.
[85] Y.C. Kuo, K.Y. Huang, C.H. Yang, Y.S. Yang, W.Y. Lee, C.W. Chiang, Regulation of phosphorylation of Thr308 of Akt, cell proliferation and survival by the B55alpha regulatory subunit targeting of the protein phosphatase 2A holoenzyme to Akt, J. Biol. Chem (2007).
[86] S.S. Wang, E.D. Esplin, J.L. Li, L. Huang, A. Gazdar, J. Minna, G.A. Evans, Alterations of the PPP2R1B gene in human lung and colon cancer, Science. 282 (1998) 284–287.
[87] Y. Shiloh, ATM and related protein kinases: safeguarding genome integrity, Nat. Rev. Cancer. 3 (2003) 155–168.
[88] R. Ruediger, H.T. Pham, G. Walter, Alterations in protein phosphatase 2A subunit interaction in human carcinomas of the lung and colon with mutations in the A beta subunit gene, Oncogene. 20 (2001) 1892–1899.
[89] A.A. Sablina, W. Chen, J.D. Arroyo, L. Corral, M. Hector, S.E. Bulmer, J.A. DeCaprio, W.C. Hahn, The tumor suppressor PP2A Abeta regulates the RalA GTPase, Cell. 129 (2007) 969–982.
[90] E.D. Esplin, P. Ramos, B. Martinez, G.E. Tomlinson, M.C. Mumby, G.A. Evans, The glycine 90 to aspartate alteration in the Abeta subunit of PP2A (PPP2R1B) associates with breast cancer and causes a deficit in protein function, Genes Chromosomes Cancer. 45 (2006) 182–190.
[91] S. Colella, H. Ohgaki, R. Ruediger, F. Yang, M. Nakamura, H. Fujisawa, P. Kleihues, G. Walter, Reduced expression of the Aalpha subunit of protein phosphatase 2A in human gliomas in the absence of mutations in the Aalpha and Abeta subunit genes, Int. J. Cancer. 93 (2001) 798–804.
[92] G.A. Calin, M.G. di Iasio, E. Caprini, I. Vorechovsky, P.G. Natali, G. Sozzi, C.M. Croce, G. Barbanti-Brodano, G. Russo, M. Negrini, Low frequency of alterations of the alpha (PPP2R1A) and beta (PPP2R1B) isoforms of the subunit A of the serine–threonine phosphatase 2A in human neoplasms, Oncogene. 19 (2000) 1191–1195.
[93] R. Ruediger, H.T. Pham, G. Walter, Disruption of protein phosphatase 2A subunit interaction in human cancers with mutations in the A alpha subunit gene, Oncogene. 20 (2001) 10–15.
[94] S. Strack, J.T. Cribbs, L. Gomez, Critical role for protein phosphatase 2A heterotrimers in mammalian cell survival, J. Biol. Chem. 279 (2004) 47732–47739.
[95] J.D. Arroyo, W.C. Hahn, Involvement of PP2A in viral and cellular transformation, Oncogene. 24 (2005) 7746–7755.
[96] C.S. Yang, M.J. Vitto, S.A. Busby, B.A. Garcia, C.T. Kesler, D. Gioeli, J. Shabanowitz, D.F. Hunt, K. Rundell, D.L. Brautigan, B.M. Paschal, Simian virus 40 small t antigen mediates conformation-dependent transfer of protein phosphatase 2A onto the androgen receptor, Mol. Cell Biol. 25 (2005) 1298–1308.
[97] S. Ory, M. Zhou, T.P. Conrads, T.D. Veenstra, D.K. Morrison, Protein phosphatase 2A positively regulates Ras signaling by dephosphorylating KSR1 and Raf-1 on critical 14-3-3 binding sites, Curr. Biol. 13 (2003) 1356–1364.
[98] D.G. Adams, R.L. Coffee Jr., H. Zhang, S. Pelech, S. Strack, B.E. Wadzinski, Positive regulation of Raf1-MEK1/2-ERK1/2 signaling by protein serine/threonine phosphatase 2A holoenzymes, J. Biol. Chem. 280 (2005) 42644–42654.
[99] G. Kao, S. Tuck, D. Baillie, M.V. Sundaram, *C. elegans* SUR-6/PR55 cooperates with LET-92/protein phosphatase 2A and promotes Raf activity independently of inhibitory Akt phosphorylation sites. Development. 131 (2004) 755–765.
[100] E. Sontag, S. Fedorov, C. Kamibayashi, D. Robbins, M. Cobb, M. Mumby, The interaction of SV40 small tumor antigen with protein phosphatase 2A stimulates the map kinase pathway and induces cell proliferation, Cell. 75 (1993) 887–897.
[101] Y. Sonoda, T. Kasahara, Y. Yamaguchi, K. Kuno, K. Matsushima, N. Mukaida, Stimulation of interleukin-8 production by okadaic acid and vanadate in a human promyelocyte cell line, an HL-60 subline. Possible role of mitogen-activated protein kinase on the okadaic acid-induced NF-kappaB activation. J. Biol. Chem. 272 (1997) 15366–15372.
[102] A.M. Silverstein, C.A. Barrow, A.J. Davis, M.C. Mumby, Actions of PP2A on the MAP kinase pathway and apoptosis are mediated by distinct regulatory subunits, Proc. Natl. Acad. Sci. U. S. A. 99 (2002) 4221–4226.
[103] D.A. Wassarman, N.M. Solomon, H.C. Chang, F.D. Karim, M. Therrien, G.M. Rubin, Protein phosphatase 2A positively and negatively regulates Ras1-mediated photoreceptor development in *Drosophila*, Genes Dev. 10 (1996) 272–278.
[104] P.J. Eichhorn, M.P. Creyghton, K. Wilhelmsen, H. van Dam, R. Bernards, A RNA interference screen identifies the protein phosphatase 2A subunit PR55gamma as a stress-sensitive inhibitor of c-SRC, PLoS. Genet 3 (2007) 218.
[105] T.S. Chao, M. Abe, M.B. Hershenson, I. Gomes, M.R. Rosner, Src tyrosine kinase mediates stimulation of Raf-1 and mitogen-activated protein kinase by the tumor promoter thapsigargin, Cancer Res. 57 (1997) 3168–3173.

[106] D. Stokoe, F. McCormick, Activation of c-Raf-1 by Ras and Src through different mechanisms: activation in vivo and in vitro, EMBO J. 16 (1997) 2384–2396.
[107] A. Ziogas, K. Moelling, G. Radziwill, CNK1 is a scaffold protein that regulates Src-mediated Raf-1 activation, J. Biol. Chem. 280 (2005) 24205–24211.
[108] C. Letourneux, G. Rocher, F. Porteu, B56-containing PP2A dephosphorylate ERK and their activity is controlled by the early gene IEX-1 and ERK, EMBO. J. 25 (2006) 727–738.
[109] S.W. Al-Murrani, J.R. Woodgett, Z. Damuni, Expression of I2PP2A, an inhibitor of protein phosphatase 2A, induces c-Jun and AP-1 activity, Biochem. J. 341 (Pt 2) (1999) 293–298.
[110] A.S. Harmala-Brasken, A. Mikhailov, T.S. Soderstrom, A. Meinander, T.H. Holmstrom, Z. Damuni, J.E. Eriksson, Type-2A protein phosphatase activity is required to maintain death receptor responsiveness, Oncogene. 22 (2003) 7677–7686.
[111] M. Li, H. Guo, Z. Damuni, Purification and characterization of two potent heat-stable protein inhibitors of protein phosphatase 2A from bovine kidney, Biochemistry. 34 (1995) 1988–1996.
[112] M. Li, A. Makkinje, Z. Damuni, The myeloid leukemia-associated protein SET is a potent inhibitor of protein phosphatase 2A, J. Biol. Chem. 271 (1996) 11059–11062.
[113] S.G. Carlson, E. Eng, E.G. Kim, E.J. Perlman, T.D. Copeland, B.J. Ballermann, Expression of SET, an inhibitor of protein phosphatase 2A, in renal development and Wilms' tumor, J. Am. Soc. Nephrol. 9 (1998) 1873–1880.
[114] M. Fornerod, J. Boer, S. van Baal, M. Jaegle, M. von Lindern, K.G. Murti, D. Davis, J. Bonten, A. Buijs, G. Grosveld, Relocation of the carboxyterminal part of CAN from the nuclear envelope to the nucleus as a result of leukemia-specific chromosome rearrangements, Oncogene. 10 (1995) 1739–1748.
[115] P. Neviani, R. Santhanam, R. Trotta, M. Notari, B.W. Blaser, S. Liu, H. Mao, J.S. Chang, A. Galietta, A. Uttam, D.C. Roy, M. Valtieri, R. Bruner-Klisovic, M.A. Caligiuri, C.D. Bloomfield, G. Marcucci, D. Perrotti, The tumor suppressor PP2A is functionally inactivated in blast crisis CML through the inhibitory activity of the BCR/ABL-regulated SET protein, Cancer Cell. 8 (2005) 355–368.
[116] P. Neviani, R. Santhanam, J.J. Oaks, A.M. Eiring, M. Notari, B.W. Blaser, S. Liu, R. Trotta, N. Muthusamy, C. Gambacorti-Passerini, B.J. Druker, J. Cortes, G. Marcucci, C.S. Chen, N.M. Verrills, D.C. Roy, M.A. Caligiuri, C.D. Bloomfield, J.C. Byrd, D. Perrotti, FTY720, a new alternative for treating blast crisis chronic myelogenous leukemia and Philadelphia chromosome-positive acute lymphocytic leukemia, J. Clin. Invest. 117 (2007) 2408–2421.
[117] H. Azuma, S. Takahara, N. Ichimaru, J.D. Wang, Y. Itoh, Y. Otsuki, J. Morimoto, R. Fukui, M. Hoshiga, T. Ishihara, N. Nonomura, S. Suzuki, A. Okuyama, Y. Katsuoka, Marked prevention of tumor growth and metastasis by a novel immunosuppressive agent, FTY720, in mouse breast cancer models, Cancer Res. 62 (2002) 1410–1419.
[118] H. Azuma, S. Takahara, S. Horie, S. Muto, Y. Otsuki, Y. Katsuoka, Induction of apoptosis in human bladder cancer cells in vitro and in vivo caused by FTY720 treatment, J. Urol. 169 (2003) 2372–2377.
[119] T.K. Lee, K. Man, J.W. Ho, X.H. Wang, R.T. Poon, Y. Xu, K.T. Ng, A.C. Chu, C.K. Sun, I.O. Ng, H.C. Sun, Z.Y. Tang, R. Xu, S.T. Fan, FTY720: a promising agent for treatment of metastatic hepatocellular carcinoma, Clin. Cancer Res. 11 (2005) 8458–8466.
[120] Y. Sonoda, D. Yamamoto, S. Sakurai, M. Hasegawa, E. Aizu-Yokota, T. Momoi, T. Kasahara, FTY720, a novel immunosuppressive agent, induces apoptosis in human glioma cells, Biochem. Biophys. Res. Commun. 281 (2001) 282–288.
[121] H. Yasui, T. Hideshima, N. Raje, A.M. Roccaro, N. Shiraishi, S. Kumar, M. Hamasaki, K. Ishitsuka, Y.T. Tai, K. Podar, L. Catley, C.S. Mitsiades, P.G. Richardson, R. Albert, V. Brinkmann, D. Chauhan, K.C. Anderson, FTY720 induces apoptosis in multiple myeloma cells and overcomes drug resistance, Cancer Res. 65 (2005) 7478–7484.
[122] C. Kalla, M.O. Scheuermann, I. Kube, M. Schlotter, D. Mertens, H. Dohner, S. Stilgenbauer, P. Lichter, Analysis of 11q22–q23 deletion target genes in B-cell chronic lymphocytic leukaemia: evidence for a pathogenic role of NPAT, CUL5, and PPP2R1B, Eur. J. Cancer. 43 (2007) 1328–1335.
[123] D.E. Roopchand, J.M. Lee, S. Shahinian, D. Paquette, H. Bussey, P.E. Branton, Toxicity of human adenovirus E4orf4 protein in *Saccharomyces cerevisiae* results from interactions with the Cdc55 regulatory B subunit of PP2A, Oncogene. 20 (2001) 5279–5290.
[124] R. Shtrichman, R. Sharf, T. Kleinberger, Adenovirus E4orf4 protein interacts with both Balpha and B¢ subunits of protein phosphatase 2A, but E4orf4-induced apoptosis is mediated only by the interaction with Balpha, Oncogene. 19 (2000) 3757–3765.
[125] C. Champagne, M.C. Landry, M.C. Gingras, J.N. Lavoie, Activation of adenovirus type 2 early region 4 ORF4 cytoplasmic death function by direct binding to Src kinase domain, J. Biol. Chem. 279 (2004) 25905–25915.
[126] K.P. Mullane, M. Ratnofsky, X. Cullere, B. Schaffhausen, Signaling from polyomavirus middle T and small T defines different roles for protein phosphatase 2A, Mol. Cell Biol. 18 (1998) 7556–7564.
[127] S.A. Courtneidge, Activation of the pp60c-src kinase by middle T antigen binding or by dephosphorylation, EMBO. J. 4 (1985) 1471–1477.
[128] S.A. Courtneidge, A.E. Smith, Polyoma virus transforming protein associates with the product of the c-src cellular gene, Nature. 303 (1983) 435–439.
[129] C.Y. Guo, D.L. Brautigan, J.M. Larner, A-dependent dissociation of B55 regulatory subunit from nuclear PP2A in response to ionizing radiation, J. Biol. Chem. 277 (2002) 4839–4844.
[130] N. Barker, H. Clevers, Catenins, Wnt signaling and cancer, Bioessays. 22 (2000) 961–965.
[131] J.M. Seeling, J.R. Miller, R. Gil, R.T. Moon, R. White, D.M. Virshup, Regulation of beta-catenin signaling by the B56 subunit of protein phosphatase 2A, Science. 283 (1999) 2089–2091.
[132] M. Patturajan, S. Nomoto, M. Sommer, A. Fomenkov, K. Hibi, R. Zangen, N. Poliak, J. Califano, B. Trink, E. Ratovitski, D. Sidransky, DeltaNp63 induces beta-catenin nuclear accumulation and signaling, Cancer Cell. 1 (2002) 369–379.
[133] X. Li, H.J. Yost, D.M. Virshup, J.M. Seeling, Protein phosphatase 2A and its B56 regulatory subunit inhibit Wnt signaling in *Xenopus*, EMBO. J. 20 (2001) 4122–4131.
[134] M.P. Creyghton, G. Roel, P.J. Eichhorn, E.M. Hijmans, I. Maurer, O. Destree, R. Bernards, PR72, a novel regulator of Wnt signaling required for Naked cuticle function, Genes Dev. 19 (2005) 376–386.
[135] W. Zeng, K.A. Wharton Jr., J.A. Mack, K. Wang, M. Gadbaw, K. Suyama, P.S. Klein, M.P. Scott, Naked cuticle encodes an inducible antagonist of Wnt signalling, Nature. 403 (2000) 789–795.
[136] M. Ikura, Calcium binding and conformational response in EF-hand proteins, Trends Biochem Sci. 21 (1996) 14–17.
[137] H. Yamamoto, T. Hinoi, T. Michiue, A. Fukui, H. Usui, V. Janssens, C. Van Hoof, J. Goris, M. Asashima, A. Kikuchi, Inhibition of the Wnt signaling pathway by the PR61 subunit of protein phosphatase 2A, J. Biol. Chem.. 276 (2001) 26875–26882.
[138] R. Ishizawar, S.J. Parsons, c-Src and cooperating partners in human cancer, Cancer Cell. 6 (2004) 209–214.
[139] J. Gotz, A. Probst, C. Mistl, R.M. Nitsch, E. Ehler, Distinct role of protein phosphatase 2A subunit Calpha in the regulation of E-cadherin and beta-catenin during development, Mech Dev. 93 (2000) 83–93.
[140] M.P. Creyghton, G. Roel, P.J. Eichhorn, L.C. Vredeveld, O. Destree, R. Bernards, PR130 is a modulator of the Wnt-signaling cascade that counters repression of the antagonist Naked cuticle, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 5397–5402.
[141] A. Aulehla, B.G. Herrmann, Segmentation in vertebrates: clock and gradient finally joined, Genes Dev. 18 (2004) 2060–2067.
[142] C. Michaloglou, L.C. Vredeveld, M.S. Soengas, C. Denoyelle, T. Kuilman, C.M. van der Horst, D.M. Majoor, J.W. Shay, W.J. Mooi, D.S. Peeper, BRAFE600-associated senescence-like cell cycle arrest of human naevi, Nature. 436 (2005) 720–724.
[143] C. Michaloglou, L.C. Vredeveld, W.J. Mooi, D.S. Peeper, BRAF(E600) in benign and malignant human tumours, Oncogene (2007).
[144] M. Mumby, PP2A: unveiling a reluctant tumor suppressor, Cell. 130 (2007) 21–24.
[145] K. Schmidt, S. Kins, A. Schild, R.M. Nitsch, B.A. Hemmings, J. Gotz, Diversity, developmental regulation and distribution of murine PR55/B subunits of protein phosphatase 2A, Eur. J. Neurosci. 16 (2002) 2039–2048.
[146] I. Stevens, V. Janssens, E. Martens, S. Dilworth, J. Goris, C. Van Hoof, Identification and characterization of B²-subunits of protein phosphatase 2 A in *Xenopus laevis* oocytes and adult tissues, Eur. J. Biochem. 270 (2003) 376–387.
[147] Y. Kono, K. Maeda, K. Kuwahara, H. Yamamoto, E. Miyamoto, K. Yonezawa, K. Takagi, N. Sakaguchi, MCM3-binding GANP DNA-primase is associated with a novel phosphatase component G5PR, Genes Cells. 7 (2002) 821–834.
[148] B. McCright, A.R. Brothman, D.M. Virshup, Assignment of human protein phosphatase 2A regulatory subunit genes b56alpha, b56beta, b56gamma, b56delta, and b56epsilon (PPP2R5A–PPP2R5E), highly expressed in muscle and brain, to chromosome regions 1q41, 11q12, 3p21, 6p21.1, and 7p11.2®p12, Genomics. 36 (1996) 168–170.
[149] S. Muneer, V. Ramalingam, R. Wyatt, R.A. Schultz, J.D. Minna, C. Kamibayashi, Genomic organization and mapping of the gene encoding the PP2A B56gamma regulatory subunit, Genomics. 79 (2002) 344–348.
[150] O. Tanabe, T. Nagase, T. Murakami, H. Nozaki, H. Usui, Y. Nishito, H. Hayashi, H. Kagamiyama, M. Takeda, Molecular cloning of a 74-kDa regulatory subunit (B² or delta) of human protein phosphatase 2A, FEBS Lett. 379 (1996) 107–111.
[151] C.S. Moreno, S. Park, K. Nelson, D. Ashby, F. Hubalek, W.S. Lane, D.C. Pallas, WD40 repeat proteins striatin and S/G(2) nuclear autoantigen are members of a novel family of calmodulin-binding proteins that associate with protein phosphatase 2A, J. Biol. Chem. 275 (2000) 5257–5263.
[152] A.J. Crossthwaite, A. Ciruela, T.F. Rayner, D.M. Cooper, A direct interaction between the N terminus of adenylyl cyclase AC8 and the catalytic subunit of protein phosphatase 2A, Mol Pharmacol. 69 (2006) 608–617.
[153] M. Kong, C.J. Fox, J. Mu, L. Solt, A. Xu, R.M. Cinalli, M.J. Birnbaum, T. Lindsten, C.B. Thompson, The PP2A-associated protein alpha4 is an essential inhibitor of apoptosis, Science. 306 (2004) 695–698.
[154] C.S. Yang, H.W. Xin, J.B. Kelley, A. Spencer, D.L. Brautigan, B.M. Paschal, Ligand binding to the androgen receptor induces conformational changes that regulate phosphatase interactions, Mol. Cell Biol. 27 (2007) 3390–3404.
[155] D. Ricotta, J. Hansen, C. Preiss, D. Teichert, S. Honing, Characterization of a protein phosphatase 2A holoenzyme (PP2A) that dephosphorylates the clathrin adaptors AP-1 and AP-2. J. Biol. Chem. (2007).
[156] J.F. Shern, J.D. Sharer, D.C. Pallas, F. Bartolini, N.J. Cowan, M.S. Reed, J. Pohl, R.A. Kahn, Cytosolic Arl2 is complexed with cofactor D and protein phosphatase 2A, J. Biol. Chem. 278 (2003) 40829–40836.
[157] A.A. Goodarzi, J.C. Jonnalagadda, P. Douglas, D. Young, R. Ye, G.B. Moorhead, S.P. Lees-Miller, K.K. Khanna, Autophosphorylation of ataxia-telangiectasia mutated is regulated by protein phosphatase 2A, EMBO. J. 23 (2004) 4451–4461.
[158] V. Horn, J. Thelu, A. Garcia, C. Albiges-Rizo, M.R. Block, J. Viallet, Functional interaction of Aurora-A and PP2A during mitosis, Mol. Biol. Cell. 18 (2007) 1233–1241.
[159] J.M. Beaulieu, T.D. Sotnikova, S. Marion, R.J. Lefkowitz, R.R. Gainetdinov, M.G. Caron, An Akt/beta-arrestin 2/PP2A signaling complex mediates dopaminergic neurotransmission and behavior, Cell. 122 (2005) 261–273.
[160] M. Xin, X. Deng, Protein phosphatase 2A enhances the proapoptotic function of Bax through dephosphorylation, J. Biol. Chem. 281 (2006) 18859–18867.
[161] S.S. Lin, M.C. Bassik, H. Suh, M. Nishino, J.D. Arroyo, W.C. Hahn, S.J. Korsmeyer, T.M. Roberts, PP2A regulates BCL-2 phosphorylation and proteasome-mediated

degradation at the endoplasmic reticulum, J. Biol. Chem. 281 (2006) 23003–23012.
[162] K.M. Krueger, Y. Daaka, J.A. Pitcher, R.J. Lefkowitz, The role of sequestration in G protein-coupled receptor resensitization. Regulation of beta2-adrenergic receptor dephosphorylation by vesicular acidification, J. Biol. Chem. 272 (1997) 5–8.
[163] L.Y. Marmorstein, P.J. McLaughlin, J.B. Stanton, L. Yan, J.W. Crabb, A.D. Marmorstein, Bestrophin interacts physically and functionally with protein phosphatase 2A, J. Biol. Chem. 277 (2002) 30591–30597.
[164] D.D. Hall, J.A. Feekes, A.S. Arachchige Don, M. Shi, J. Hamid, L. Chen, S. Strack, G.W. Zamponi, M.C. Horne, J.W. Hell, Binding of protein phosphatase 2A to the L-type calcium channel Cav1.2 next to Ser1928, its main PKA site, is critical for Ser1928 dephosphorylation, Biochemistry. 45 (2006) 3448–3459.
[165] L. Xu, X. Deng, Suppression of cancer cell migration and invasion by protein phosphatase 2A through dephosphorylation of mu- and m-calpains, J. Biol. Chem. 281 (2006) 35567–35575.
[166] T. Yamashita, S. Inui, K. Maeda, D.R. Hua, K. Takagi, K. Fukunaga, N. Sakaguchi, Regulation of CaMKII by alpha4/PP2Ac contributes to learning and memory, Brain Res. 1082 (2006) 1–10.
[167] R.S. Westphal, K.A. Anderson, A.R. Means, B.E. Wadzinski, A signaling complex of Ca2+-calmodulin-dependent protein kinase IV and protein phosphatase 2A, Science. 280 (1998) 1258–1261.
[168] O. Varlamov, E. Kalinina, F.Y. Che, L.D. Fricker, Protein phosphatase 2A binds to the cytoplasmic tail of carboxypeptidase D and regulates post-*trans*-Golgi network trafficking, J Cell Sci. 114 (2001) 311–322.
[169] N. Yokoyama, W.T. Miller, Protein phosphatase 2A interacts with the Src kinase substrate p130(CAS), Oncogene. 20 (2001) 6057–6065.
[170] V. Leung-Pineda, C.E. Ryan, H. Piwnica-Worms, Phosphorylation of Chk1 by ATR is antagonized by a Chk1-regulated protein phosphatase 2A circuit, Mol. Cell Biol. 26 (2006) 7529–7538.
[171] X. Liang, E. Reed, J.J. Yu, Protein phosphatase 2A interacts with Chk2 and regulates phosphorylation at Thr-68 after cisplatin treatment of human ovarian cancer cells, Int J Mol Med. 17 (2006) 703–708.
[172] J.K. Heriche, F. Lebrin, T. Rabilloud, D. Leroy, E.M. Chambaz, Y. Goldberg, Regulation of protein phosphatase 2A by direct interaction with casein kinase 2alpha, Science. 276 (1997) 952–955.
[173] M. Alvarado-Kristensson, T. Andersson, Protein phosphatase 2A regulates apoptosis in neutrophils by dephosphorylating both p38 MAPK and its substrate caspase 3, J. Biol. Chem. 280 (2005) 6238–6244.
[174] S.S. Margolis, J.A. Perry, C.M. Forester, L.K. Nutt, Y. Guo, M.J. Jardim, M.J. Thomenius, C.D. Freel, R. Darbandi, J.H. Ahn, J.D. Arroyo, X.F. Wang, S. Shenolikar, A.C. Nairn, W.G. Dunphy, W.C. Hahn, D.M. Virshup, S. Kornbluth, Role for the PP2A/B56delta phosphatase in regulating 14-3-3 release from Cdc25 to control mitosis, Cell. 127 (2006) 759–773.
[175] T. Ammosova, K. Washington, Z. Debebe, J. Brady, S. Nekhai, Dephosphorylation of CDK9 by protein phosphatase 2A and protein phosphatase-1 in Tat-activated HIV-1 transcription, Retrovirology. 2 (2005) 47.
[176] A. Vastiau, L. Cao, M. Jaspers, G. Owsianik, V. Janssens, H. Cuppens, J. Goris, B. Nilius, J.J. Cassiman, Interaction of the protein phosphatase 2A with the regulatory domain of the cystic fibrosis transmembrane conductance regulator channel, FEBS Lett. 579 (2005) 3392–3396.
[177] M. Takahashi, H. Shibata, M. Shimakawa, M. Miyamoto, H. Mukai, Y. Ono, Characterization of a novel giant scaffolding protein, CG-NAP, that anchors multiple signaling enzymes to centrosome and the golgi apparatus, J. Biol. Chem. 274 (1999) 17267–17274.
[178] Y. Samstag, G. Nebl, Interaction of cofilin with the serine phosphatases PP1 and PP2A in normal and neoplastic human T lymphocytes, Adv Enzyme Regul. 43 (2003) 197–211.
[179] M.A. Meilleur, C.D. Akpovi, R.M. Pelletier, M.L. Vitale, Tumor necrosis factor-alpha-induced anterior pituitary folliculostellate TtT/GF cell uncoupling is mediated by connexin 43 dephosphorylation, Endocrinology. 148 (2007) 5913–5924.
[180] G.H. Fan, W. Yang, J. Sai, A. Richmond, Phosphorylation-independent association of CXCR2 with the protein phosphatase 2A core enzyme, J. Biol. Chem. 276 (2001) 16960–16968.
[181] D.A. Bennin, A.S. Don, T. Brake, J.L. McKenzie, H. Rosenbaum, L. Ortiz, A.A. DePaoli-Roach, M.C. Horne, Cyclin G2 associates with protein phosphatase 2A catalytic and regulatory B¢ subunits in active complexes and induces nuclear aberrations and a G1/S phase cell cycle arrest, J. Biol. Chem. 277 (2002) 27449–27467.
[182] J.H. Ahn, J.Y. Sung, T. McAvoy, A. Nishi, V. Janssens, J. Goris, P. Greengard, A.C. Nairn, The B²/PR72 subunit mediates Ca2+-dependent dephosphorylation of DARPP-32 by protein phosphatase 2A, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 9876–9881.
[183] S. Dehde, G. Rohaly, O. Schub, H.P. Nasheuer, W. Bohn, J. Chemnitz, W. Deppert, I. Dornreiter, Two immunologically distinct human DNA polymerase alpha-primase subpopulations are involved in cellular DNA replication, Mol. Cell Biol. 21 (2001) 2581–2593.
[184] J.Q. Wu, D.V. Hansen, Y. Guo, M.Z. Wang, W. Tang, C.D. Freel, J.J. Tung, P.K. Jackson, S. Kornbluth, Control of Emi2 activity and stability through Mos-mediated recruitment of PP2A, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 16564–16569.
[185] K. Lechward, S. Zolnierowicz, B.A. Hemmings, Eukaryotic translation termination factor 1 associates with protein phosphatase 2A and targets it to ribosomes, Biochemistry (Mosc). 64 (1999) 1373–1381.
[186] Q. Lu, H.K. Surks, H. Ebling, W.E. Baur, D. Brown, D.C. Pallas, R.H. Karas, Regulation of estrogen receptor alpha-mediated transcription by a direct interaction with protein phosphatase 2A, J. Biol. Chem. 278 (2003) 4639–4645.
[187] U. Narayanan, V. Nalavadi, M. Nakamoto, D.C. Pallas, S. Ceman, G.J. Bassell, S.T. Warren, FMRP phosphorylation reveals an immediate-early signaling pathway triggered by group I mGluR and mediated by PP2A, J. Neurosci. 27 (2007) 14349–14357.
[188] D. Zhu, K.S. Kosik, T.E. Meigs, V. Yanamadala, B.M. Denker, Galpha12 directly interacts with PP2A: evidence FOR Galpha12-stimulated PP2A phosphatase activity and dephosphorylation of microtubule-associated protein, tau, J. Biol. Chem. 279 (2004) 54983–54986.
[189] L. Mao, L. Yang, A. Arora, E.S. Choe, G. Zhang, Z. Liu, E.E. Fibuch, J.Q. Wang, Role of protein phosphatase 2A in mGluR5-regulated MEK/ERK phosphorylation in neurons, J. Biol. Chem. 280 (2005) 12602–12610.
[190] S. Mitsuhashi, H. Shima, N. Tanuma, S. Sasa, K. Onoe, M. Ubukata, K. Kikuchi, Protein phosphatase type 2A, PP2A, is involved in degradation of gp130, Mol. Cell Biochem. 269 (2005) 183–187.
[191] D. Chowdhury, M.C. Keogh, H. Ishii, C.L. Peterson, S. Buratowski, J. Lieberman, Gamma-H2AX dephosphorylation by protein phosphatase 2A facilitates DNA double-strand break repair, Mol Cell. 20 (2005) 801–809.
[192] B.A. Firulli, M.J. Howard, J.R. McDaid, L. McIlreavey, K.M. Dionne, V.E. Centonze, P. Cserjesi, D.M. Virshup, A.B. Firulli, PKA, PKC, and the protein phosphatase 2A influence HAND factor function: a mechanism for tissue-specific transcriptional regulation, Mol Cell. 12 (2003) 1225–1237.
[193] F.M. Yeong, H. Hombauer, K.S. Wendt, T. Hirota, I. Mudrak, K. Mechtler, T. Loregger, A. Marchler-Bauer, K. Tanaka, J.M. Peters, E. Ogris, Identification of a subunit of a novel Kleisin-beta/SMC complex as a potential substrate of protein phosphatase 2A, Curr. Biol. 13 (2003) 2058–2064.
[194] G. Paroni, N. Cernotta, C.D. Russo, P. Gallinari, M. Pallaoro, C. Foti, F. Talamo, L. Orsatti, C. Steinkuhler, C. Brancolini, PP2A regulates HDAC4 nuclear import, Mol. Biol. Cell (2007).
[195] H. Xing, Y. Hong, K.D. Sarge, Identification of the PP2A-interacting region of heat shock transcription factor 2, Cell Stress Chaperones. 12 (2007) 192–197.
[196] M. Hrimech, X.J. Yao, P.E. Branton, E.A. Cohen, Human immunodeficiency virus type 1 Vpr-mediated G(2) cell cycle arrest: Vpr interferes with cell cycle signaling cascades by interacting with the B subunit of serine/threonine protein phosphatase 2A, EMBO. J. 19 (2000) 3956–3967.
[197] T. Kawabe, A.J. Muslin, S.J. Korsmeyer, HOX11 interacts with protein phosphatases PP2A and PP1 and disrupts a G2/M cell-cycle checkpoint, Nature. 385 (1997) 454–458.
[198] H.T. Adler, F.S. Nallaseth, G. Walter, D.C. Tkachuk, HRX leukemic fusion proteins form a heterocomplex with the leukemia-associated protein SET and protein phosphatase 2A, J. Biol. Chem. 272 (1997) 28407–28414.
[199] A.E. Kray, R.S. Carter, K.N. Pennington, R.J. Gomez, L.E. Sanders, J.M. Llanes, W.N. Khan, D.W. Ballard, B.E. Wadzinski, Positive regulation of IkappaB kinase signaling by protein serine/threonine phosphatase 2A, J. Biol. Chem. 280 (2005) 35974–35982.
[200] S. Li, L. Wang, M.A. Berman, Y. Zhang, M.E. Dorf, RNAi screen in mouse astrocytes identifies phosphatases that regulate NF-kappaB signaling, Mol Cell. 24 (2006) 497–509.
[201] D.X. Fu, Y.L. Kuo, B.Y. Liu, K.T. Jeang, C.Z. Giam, Human T-lymphotropic virus type I tax activates I-kappa B kinase by inhibiting I-kappa B kinase-associated serine/threonine protein phosphatase 2A, J. Biol. Chem. 278 (2003) 1487–1493.
[202] K. Takahashi, K. Suzuki, Regulation of protein phosphatase 2A-mediated recruitment of IQGAP1 to beta1 integrin by EGF through activation of Ca2+/calmodulin-dependent protein kinase II, J. Cell Physiol. 208 (2006) 213–219.
[203] D.K. Fuhrer, Y.C. Yang, Complex formation of JAK2 with PP2A, P13K, and Yes in response to the hematopoietic cytokine interleukin-11, Biochem Biophys Res Commun. 224 (1996) 289–296.
[204] T.P. Shanley, N. Vasi, A. Denenberg, H.R. Wong, The serine/threonine phosphatase, PP2A: endogenous regulator of inflammatory cell signaling, J Immunol. 166 (2001) 966–972.
[205] M. Borsotto, L. Cavarec, M. Bouillot, G. Romey, F. Macciardi, A. Delaye, M. Nasroune, M. Bastucci, J.L. Sambucy, J.J. Luan, A. Charpagne, V. Jouet, R. Leger, M. Lazdunski, D. Cohen, I. Chumakov, PP2A-Bgamma subunit and KCNQ2 K+ channels in bipolar disorder, Pharmacogenomics J. 7 (2007) 123–132.
[206] G.Z. Tao, D.M. Toivola, Q. Zhou, P. Strnad, B. Xu, S.A. Michie, M.B. Omary, Protein phosphatase-2A associates with and dephosphorylates keratin 8 after hypos-motic stress in a site- and cell-specific manner, J. Cell Sci. 119 (2006) 1425–1432.
[207] J.A. Lee, D.C. Pallas, Leucine carboxyl methyltransferase-1 is necessary for normal progression through mitosis in mammalian cells, J. Biol. Chem. 282 (2007) 30974–30984.
[208] T.D. Prickett, D.L. Brautigan, Cytokine activation of p38 mitogen-activated protein kinase and apoptosis is opposed by alpha-4 targeting of protein phosphatase 2A for site-specific dephosphorylation of MEK3, Mol. Cell Biol. 27 (2007) 4217–4227.
[209] A. Fritz, K.J. Brayer, N. McCormick, D.G. Adams, B.E. Wadzinski, R.R. Vaillancourt, Phosphorylation of serine 526 is required for MEKK3 activity, and association with 14-3-3 blocks dephosphorylation, J. Biol. Chem. 281 (2006) 6236–6245.
[210] A. Hashigasako, M. Machide, T. Nakamura, K. Matsumoto, T. Nakamura, Bi-directional regulation of Ser-985 phosphorylation of c-met via protein kinase C and protein phosphatase 2A involves c-Met activation and cellular responsiveness to hepatocyte growth factor, J. Biol. Chem. 279 (2004) 26445–26452.
[211] K.M. Short, B. Hopwood, Z. Yi, T.C. Cox, MID1 and MID2 homo- and heterodimerise to tether the rapamycin-sensitive PP2A regulatory subunit, alpha 4, to microtubules: implications for the clinical variability of X-linked Opitz GBBB syndrome and other developmental disorders, BMC Cell Biol. 3 (2002) 1.
[212] N.J. Avdi, K.C. Malcolm, J.A. Nick, G.S. Worthen, A role for protein phosphatase-2A in p38 mitogen-activated protein kinase-mediated regulation of the c-Jun NH (2)-terminal kinase pathway in human neutrophils, J. Biol. Chem. 277 (2002) 40687–40696.

[213] A.E. Escargueil, A.K. Larsen, Mitosis-specific MPM-2 phosphorylation of DNA topoisomerase IIalpha is regulated directly by protein phosphatase 2A, Biochem J. 403 (2007) 235–242.

[214] J. Holst, A.T. Sim, R.I. Ludowyke, Protein phosphatases 1 and 2A transiently associate with myosin during the peak rate of secretion from mast cells, Mol. Biol. Cell. 13 (2002) 1083–1098.

[215] L. Palkowitsch, J. Leidner, S. Ghosh, R.B. Marienfeld, Phosphorylation of Serine 68 in the I{kappa}B kinase (IKK)-binding domain of NEMO interferes with the structure of the IKK complex and tumor necrosis factor-{alpha}-induced NF-{kappa}B activity, J. Biol. Chem. 283 (2008) 76–86.

[216] U. Sung, J.L. Jennings, A.J. Link, R.D. Blakely, Proteomic analysis of human norepinephrine transporter complexes reveals associations with protein phosphatase 2A anchoring subunit and 14-3-3 proteins, Biochem. Biophys. Res. Commun. 333 (2005) 671–678.

[217] T. Saito, H. Shima, Y. Osawa, M. Nagao, B.A. Hemmings, T. Kishimoto, S. Hisanaga, Neurofilament-associated protein phosphatase 2A: its possible role in preserving neurofilaments in filamentous states, Biochemistry. 34 (1995) 7376–7384.

[218] C.M. Liedtke, X. Wang, N.D. Smallwood, Role for protein phosphatase 2A in the regulation of Calu-3 epithelial Na+-K+-2Cl−, type 1 co-transport function, J. Biol. Chem. 280 (2005) 25491–25498.

[219] W.J. Lee, D.U. Kim, M.Y. Lee, K.Y. Choi, Identification of proteins interacting with the catalytic subunit of PP2A by proteomics, Proteomics. 7 (2007) 206–214.

[220] S.F. Chan, N.J. Sucher, An NMDA receptor signaling complex with protein phosphatase 2A, J. Neurosci. 21 (2001) 7985–7992.

[221] O.K. Ma, N.J. Sucher, Molecular interaction of NMDA receptor subunit NR3A with protein phosphatase 2A, Neuroreport. 15 (2004) 1447–1450.

[222] K. Lechward, E. Sugajska, I. de Baere, J. Goris, B.A. Hemmings, S. Zolnierowicz, Interaction of nucleoredoxin with protein phosphatase 2A, FEBS Lett. 580 (2006) 3631–3637.

[223] A. Seth, P. Sheth, B.C. Elias, R. Rao, Protein phosphatases 2A and 1 interact with occludin and negatively regulate the assembly of tight junctions in the CACO-2 cell monolayer, J. Biol. Chem. 282 (2007) 11487–11498.

[224] P.Y. Wang, J. Weng, R.G. Anderson, OSBP is a cholesterol-regulated scaffolding protein in control of ERK 1/2 activation, Science. 307 (2005) 1472–1476.

[225] R.S. Westphal, R.L. Coffee Jr., A. Marotta, S.L. Pelech, B.E. Wadzinski, Identification of kinase-phosphatase signaling modules composed of p70 S6 kinase-protein phosphatase 2A (PP2A) and p21-activated kinase-PP2A, J. Biol. Chem. 274 (1999) 687–692.

[226] G.K. Scott, F. Gu, C.M. Crump, L. Thomas, L. Wan, Y. Xiang, G. Thomas, The phosphorylation state of an autoregulatory domain controls PACS-1-directed protein traffic, EMBO. J. 22 (2003) 6234–6244.

[227] S. Sathyanarayanan, X. Zheng, R. Xiao, A. Sehgal, Posttranslational regulation of *Drosophila* PERIOD protein by protein phosphatase 2A, Cell. 116 (2004) 603–615.

[228] J. Ma, H.K. Arnold, M.B. Lilly, R.C. Sears, A.S. Kraft, Negative regulation of Pim-1 protein kinase levels by the B56beta subunit of PP2A, Oncogene. 26 (2007) 5145–5153.

[229] M. Michniewicz, M.K. Zago, L. Abas, D. Weijers, A. Schweighofer, I. Meskiene, M.G. Heisler, C. Ohno, J. Zhang, F. Huang, R. Schwab, D. Weigel, E.M. Meyerowitz, C. Luschnig, R. Offringa, J. Friml, Antagonistic regulation of PIN phosphorylation by PP2A and PINOID directs auxin flux, Cell. 130 (2007) 1044–1056.

[230] H.K. Huang, S.L. Forsburg, U.P. John, M.J. O'Connell, T. Hunter, Isolation and characterization of the Pin1/Ess1p homologue in *Schizosaccharomyces pombe*, J Cell Sci. 114 (2001) 3779–3788.

[231] J.H. Ahn, T. McAvoy, S.V. Rakhilin, A. Nishi, P. Greengard, A.C. Nairn, Protein kinase A activates protein phosphatase 2A by phosphorylation of the B56delta subunit, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 2979–2984.

[232] R.T. Boudreau, R. Garduno, T.J. Lin, Protein phosphatase 2A and protein kinase Calpha are physically associated and are involved in *Pseudomonas aeruginosa*-induced interleukin 6 production by mast cells, J. Biol. Chem. 277 (2002) 5322–5329.

[233] W. Huang, S. Batra, B.A. Atkins, V. Mishra, K.D. Mehta, Increases in intracellular calcium dephosphorylate histone H3 at serine 10 in human hepatoma cells: potential role of protein phosphatase 2A–protein kinase CbetaII complex, J. Cell Physiol. 205 (2005) 37–46.

[234] J. Srivastava, J. Goris, S.M. Dilworth, P.J. Parker, Dephosphorylation of PKCdelta by protein phosphatase 2Ac and its inhibition by nucleotides, FEBS Lett. 516 (2002) 265–269.

[235] Z. Xu, B.R. Williams, The B56alpha regulatory subunit of protein phosphatase 2A is a target for regulation by double-stranded RNA-dependent protein kinase PKR, Mol. Cell Biol. 20 (2000) 5285–5299.

[236] Y.J. Jang, J.H. Ji, Y.C. Choi, C.J. Ryu, S.Y. Ko, Regulation of Polo-like kinase 1 by DNA damage in mitosis. Inhibition of mitotic PLK-1 by protein phosphatase 2A, J. Biol. Chem. 282 (2007) 2473–2482.

[237] H.R. Glover, C.E. Brewster, S.M. Dilworth, Association between src-kinases and the polyoma virus oncogene middle T-antigen requires PP2A and a specific sequence motif, Oncogene. 18 (1999) 4364–4370.

[238] P.A. Kiely, D. O'Gorman, K. Luong, D. Ron, R. O'Connor, Insulin-like growth factor I controls a mutually exclusive association of RACK1 with protein phosphatase 2A and beta1 integrin to promote cell migration, Mol. Cell Biol. 26 (2006) 4041–4051.

[239] J. Yang, G.H. Fan, B.E. Wadzinski, H. Sakurai, A. Richmond, Protein phosphatase 2A interacts with and directly dephosphorylates RelA, J. Biol. Chem. 276 (2001) 47828–47833.

[240] A.L. Schlaitz, M. Srayko, A. Dammermann, S. Quintin, N. Wielsch, I. MacLeod, Q. de Robillard, A. Zinke, J.R. Yates 3rd, T. Muller-Reichert, A. Shevchenko, K. Oegema, A.A. Hyman, The *C. elegans* RSA complex localizes protein phosphatase 2A to centrosomes and regulates mitotic spindle assembly, Cell. 128 (2007) 115–127.

[241] A. Rajgopal, D.W. Young, K.A. Mujeeb, J.L. Stein, J.B. Lian, A.J. van Wijnen, G.S. Stein, Mitotic control of RUNX2 phosphorylation by both CDK1/cyclin B kinase and PP1/PP2A phosphatase in osteoblastic cells, J. Cell Biochem. 100 (2007) 1509–1517.

[242] M. Berry, W. Gehring, Phosphorylation status of the SCR homeodomain determines its functional activity: essential role for protein phosphatase 2A,B¢, EMBO. J. 19 (2000) 2946–2957.

[243] A.M. Gil-Bernabe, F. Romero, M.C. Limon-Mortes, M. Tortolero, Protein phosphatase 2A stabilizes human securin, whose phosphorylated forms are degraded via the SCF ubiquitin ligase, Mol. Cell Biol. 26 (2006) 4017–4027.

[244] A.L. Bauman, S. Apparsundaram, S. Ramamoorthy, B.E. Wadzinski, R.A. Vaughan, R.D. Blakely, Cocaine and antidepressant-sensitive biogenic amine transporters exist in regulated complexes with protein phosphatase 2A, J. Neurosci. 20 (2000) 7571–7578.

[245] A.J. Holland, F. Bottger, O. Stemmann, S.S. Taylor, Protein phosphatase 2A and separase form a complex regulated by separase autocleavage, J. Biol. Chem. 282 (2007) 24623–24632.

[246] S. Ugi, T. Imamura, W. Ricketts, J.M. Olefsky, Protein phosphatase 2A forms a molecular complex with Shc and regulates Shc tyrosine phosphorylation and downstream mitogenic signaling, Mol. Cell Biol. 22 (2002) 2375–2387.

[247] Z. Tang, H. Shu, W. Qi, N.A. Mahmood, M.C. Mumby, H. Yu, PP2A is required for centromeric localization of Sgo1 and proper chromosome segregation, Dev Cell. 10 (2006) 575–585.

[248] T.S. Kitajima, T. Sakuno, K. Ishiguro, S. Iemura, T. Natsume, S.A. Kawashima, Y. Watanabe, Shugoshin collaborates with protein phosphatase 2A to protect cohesin, Nature. 441 (2006) 46–52.

[249] C.G. Riedel, V.L. Katis, Y. Katou, S. Mori, T. Itoh, W. Helmhart, M. Galova, M. Petronczki, J. Gregan, B. Cetin, I. Mudrak, E. Ogris, K. Mechtler, L. Pelletier, F. Buchholz, K. Shirahige, K. Nasmyth, Protein phosphatase 2A protects centromeric sister chromatid cohesion during meiosis I, Nature. 441 (2006) 53–61.

[250] K.R. Anders, A. Grimson, P. Anderson, SMG-5, required for *C. elegans* nonsense-mediated mRNA decay, associates with SMG-2 and protein phosphatase 2A, EMBO. J. 22 (2003) 641–650.

[251] A. Vicart, T. Lefebvre, J. Imbert, A. Fernandez, B. Kahn-Perles, Increased chromatin association of Sp1 in interphase cells by PP2A-mediated dephosphorylations, J. Mol. Biol. 364 (2006) 897–908.

[252] D.H. Lao, P. Yusoff, S. Chandramouli, R.J. Philp, C.W. Fong, R.A. Jackson, T.Y. Saw, C.Y. Yu, G.R. Guy, Direct binding of PP2A to Sprouty2 and phosphorylation changes are a prerequisite for ERK inhibition downstream of fibroblast growth factor receptor stimulation, J. Biol. Chem. 282 (2007) 9117–9126.

[253] N. Yokoyama, N.C. Reich, W.T. Miller, Involvement of protein phosphatase 2A in the interleukin-3-stimulated Jak2-Stat5 signaling pathway, J. Interferon Cytokine Res. 21 (2001) 369–378.

[254] M. Nanahoshi, T. Nishiuma, Y. Tsujishita, K. Hara, S. Inui, N. Sakaguchi, K. Yonezawa, Regulation of protein phosphatase 2A catalytic activity by alpha4 protein and its yeast homolog Tap42, Biochem. Biophys. Res. Commun. 251 (1998) 520–526.

[255] E. Sontag, V. Nunbhakdi-Craig, G. Lee, G.S. Bloom, M.C. Mumby, Regulation of the phosphorylation state and microtubule-binding activity of Tau by protein phosphatase 2A, Neuron. 17 (1996) 1201–1207.

[256] E. Sontag, V. Nunbhakdi-Craig, G. Lee, R. Brandt, C. Kamibayashi, J. Kuret, C.L. White 3rd, M.C. Mumby, G.S. Bloom, Molecular interactions among protein phosphatase 2A, tau, and microtubules. Implications for the regulation of tau phosphorylation and the development of tauopathies, J. Biol. Chem. 274 (1999) 25490–25498.

[257] S.E. Merrick, D.C. Demoise, V.M. Lee, Site-specific dephosphorylation of tau protein at Ser202/Thr205 in response to microtubule depolymerization in cultured human neurons involves protein phosphatase 2A, J. Biol. Chem. 271 (1996) 5589–5594.

[258] J.L. McConnell, R.J. Gomez, L.R. McCorvey, B.K. Law, B.E. Wadzinski, Identification of a PP2A-interacting protein that functions as a negative regulator of phosphatase activity in the A/ATR signaling pathway, Oncogene. 26 (2007) 6021–6030.

[259] I. Griswold-Prenner, C. Kamibayashi, E.M. Maruoka, M.C. Mumby, R. Derynck, Physical and functional interactions between type I transforming growth factor beta receptors and Balpha, a WD-40 repeat subunit of phosphatase 2A, Mol. Cell Biol. 18 (1998) 6595–6604.

[260] R.K. Dagda, C.A. Barwacz, J.T. Cribbs, S. Strack, Unfolding-resistant translocase targeting: a novel mechanism for outer mitochondrial membrane localization exemplified by the Bbeta2 regulatory subunit of protein phosphatase 2A, J. Biol. Chem. 280 (2005) 27375–27382.

[261] L. Sun, G. Stoecklin, S. Van Way, V. Hinkovska-Galcheva, R.F. Guo, P. Anderson, T.P. Shanley, Tristetraprolin (TTP)-14-3-3 complex formation protects TTP from dephosphorylation by protein phosphatase 2a and stabilizes tumor necrosis factor-alpha mRNA, J. Biol. Chem. 282 (2007) 3766–3777.

[262] T. Ohnishi, A. Yamashita, I. Kashima, T. Schell, K.R. Anders, A. Grimson, T. Hachiya, M.W. Hentze, P. Anderson, S. Ohno, Phosphorylation of hUPF1 induces formation of mRNA surveillance complexes containing hSMG-5 and hSMG-7, Mol Cell. 12 (2003) 1187–1200.

[263] P. Turowski, T. Myles, B.A. Hemmings, A. Fernandez, N.J. Lamb, Vimentin dephosphorylation by protein phosphatase 2A is modulated by the targeting subunit B55, Mol. Biol. Cell. 10 (1999) 1997–2015.

[264] D.R. Evans, B.A. Hemmings, Mutation of the C-terminal leucine residue of PP2Ac inhibits PR55/B subunit binding and confers supersensitivity to microtubule destabilization in *Saccharomyces cerevisiae*, Mol. Gen. Genet. 264 (2000) 425–432.