Protein Phosphatase 2A as a Potential Target for Anticancer Therapy

Peter Kalev and Anna A. Sablina

1Department for Molecular and Developmental Genetics, Flanders Institute for Biotechnology (VIB); 2Center for Human Genetics, KULeuven, Belgium

Abstract: The kinase oncoproteins are well-characterized drivers of cancer development, and several targeted therapies focused on both specific and selectively nonselective kinase inhibitors have now been approved for clinical use. In contrast, much less is known about the role of protein phosphatases, although modulation of their activities might form the foundation for an effective anti-cancer approach. The serine-threonine protein phosphatase 2A (PP2A) is implicated in the regulation of numerous signaling pathways and may function as a tumor suppressor. Recently pharmacological modulation of PP2A activity has been showed to have a potent anti-tumor activity in both in vitro and in vivo cancer models. These studies implicate PP2A as a promising therapeutic target for the treatment of cancer.

Keywords: Protein phosphatase 2A, Drug Development, Cancer, CIP2A, FTY720, Forskolin, Norcantharidin derivatives, Cantharidin.

INTRODUCTION

The aberrant activation of cellular signalling cascades contributes directly to human diseases and plays a causal role in most if not all cancers. The kinase oncoproteins are well-characterized drivers of cancer development [1, 2], which makes them attractive targets for drug development. Several kinase inhibitors, including imatinib (Gleevec) for chronic myelogenous leukemia (CML) and gastrointestinal tumors, and gefitinib (Iressa) for non-small-cell lung cancer, have been recently approved for therapeutic use. Although it is clear that phosphatases antagonize the action of kinases, much less is known concerning the role of protein phosphatases in development and progression of human cancers.

The Protein Phosphatase 2A (PP2A), a large family of heterotrimeric serine-threonine phosphatases, regulates numerous cell signalling cascades mostly by opposing the activity of kinase oncoproteins [3]. Initial evidence for the role of PP2A in cell transformation and tumorigenesis came from studies with the sponge toxin okadaic acid (OA). OA is both a selective inhibitor of PP2A and a strong tumor promoter [4, 5]. The finding that PP2A is an important cellular target of several viral oncoproteins further supports the idea that PP2A contributes to tumor suppression. More recently, a decrease in PP2A activity have been found in different types of human cancers. The loss of PP2A function occurs either through inactivating mutations of PP2A structural subunits, or by upregulation of the cellular PP2A inhibitors such as Cancerous inhibitor of PP2A (CIP2A) and SET.

The promise of kinase inhibitors as anticancer agents raised the question whether protein phosphatases could also represent potential drug targets. Recently published reports have demonstrated that modulation of PP2A activity could be beneficial for the treatment of cancer. Furthermore, the involvement of PP2A in regulation of multiple physiological functions provides us an opportunity to employ several different strategies for PP2A targeting therapies. Here we will review recent progress in our understanding of PP2A as a potential drug target for cancer treatment.

THE PP2A FAMILY

PP2A accounts for the majority of serine/threonine phosphatase activity in mammalian cells [reviewed in [6]]. The term PP2A refers to a diverse family of phosphatases that contains a catalytic C subunit, whose activity is regulated by a diverse set of regulatory proteins. The most common forms of PP2A contain a highly active core dimer composed of a catalytic C subunit, also called PP2AC or PPP2C, and a scaffolding A subunit, also known as PR65 or PPP2R1. The AC dimer recruits an additional third regulatory B subunit, which is predicted to dictate the substrate specificity and localization of the PP2A heterotrimetric complex. Four unrelated families of B subunits have identified to date: B/B55/PR55/PPP2R2, B'/B56/PR61/PPP2R5, B''/PR72/PPP2R3, and Striatin [7] (Fig. 1).

![Fig. (1). The PP2A family of serine/threonine heterotrimeric phosphatases. The PP2A holoenzyme represents a heterotrimetric complex composed of a catalytic subunit C and a structural subunit A, and a single regulatory B subunit.](image)

The mechanism of assembly of heterotrimeric PP2A complexes is not completely understood. The dynamic exchange of PP2A regulatory subunits and PP2A activity could be modulated by reversible methylation and phosphorylation of the carboxy-terminal tail of the C subunit. Methylation at Lys309 by Leucine carboxyl methyltransferase-1 (LCMT1) enhances the affinity of the PP2A core dimer for PP2A regulatory subunits while phosphorylation at Tyr307 might indirectly affect the assembly of PP2A holoenzyme complexes by preventing methylation of the PP2AC subunit [8-11].
Multiple tyrosine kinases, including c-src, EGFR, Her-2, and JAK family members, have been shown to transiently inactivate PP2A activity by phosphorylation at Tyr307 in response to cytokine stimulation [12-16].

The assembly of PP2A complexes is also regulated by the PP2A activator (PTPA or PPP2R4) that was initially identified as a PP2A B regulatory subunit due to its ability to interact with the AC dimer. PTPA potentially serves as a metal chaperone by activating the phosphoserine/threonine activity of PP2AC from a poorly active state [8, 17]. PTPA has demonstrated peptidyl prolyl cis-trans-isomerase (PPIase) activity in vitro, which acts specifically on Pro190 of PP2AC [18]. This isomerase activity could potentially control the folding of PP2AC and its activity. However, the relative contribution of these PTPA activities to the assembly of specific PP2A complexes is unknown.

In the absence of these modifications, the C subunit preferentially associates with TAP42/α4 protein rather than with the PP2A structural A subunit. It has been suggested that TAP/α4 also acts as a general scaffold/chaperone protein in maintaining proper PP2A function and assembly [19].

The ability of the PP2A dimer to associate with numerous regulatory subunits suggests that the full complement of PP2A complexes may exceed more than 100 different multimers. The cellular localization, substrate specificity and physiological functions of specific PP2A complexes have been comprehensively reviewed in [55, 87]. This remarkable complexity in the assembly of the holoenzyme repertoire of different substrates and mediate a wide diversity of physiological functions.

**THE ROLE OF PP2A IN TUMOR SUPPRESSION**

The first evidence that PP2A acts as a tumor suppressor came from the longstanding observation that a PP2A inhibitor, OA, at low concentrations (50-500 pM) promotes tumor growth in mouse skin [5], stomach, and liver [20] and induces genomic instability [21, 22]. PP2A is also found to be one of the major cellular binding partners of several viral oncoproteins such as small and middle t antigens of polyoma virus, and adenoviral E4orI4 protein (reviewed in [23]), all of which contribute to cell transformation. More recently, activating alterations of PP2A subunits have been found to occur in human cancers, further supporting the idea that PP2A is involved in suppressing cancer development. Spontaneous mutations in both α and β isoforms of the A structural subunit have been found to occur in several types of human cancers. Specifically, somatic alterations of the gene encoding the PP2A Aα subunit have been identified in 8–15% of colon cancers, 15% of lung cancers, 13% of breast cancers, and 6% of tumor cell lines [24-29]. Mutations of the more abundant PP2A Aα subunit have also been observed in breast and lung tumors, although at a low frequency [24].

Most of the Aβ cancer-associated mutants are composed of missense mutations including G8R, P65S, L101P, K343E, D504G, V545A, V448A, one contains the double mutant L101P/V448A, and one an in frame deletion ΔE344–E388 [24-29]. All cancer-associated mutations of PP2A structural subunits interrupt interaction between A and C subunits by preventing the formation of the PP2A heterotrimeric holoenzymes [25, 26, 30]. Further studies demonstrated that loss of PP2A Aβ subunits contributes to cancer progression through dysregulation of the Ras-like GTPase RaLA activity [30]. On the other hand, haploinsufficiency of PP2A Aα seems to induce human cell transformation by activating the PI3K/AKT signaling pathway [31].

Recent studies identified several cellular inhibitors of PP2A (Fig. 1). Two endogenous inhibitors of PP2A, I1PP2A and I2PP2A, were purified from bovine kidney. I1PP2A is also called PHAP-I (putative class II human histocompatibility leucocyte-associated protein I) [32] while I2PP2A represents a truncated form of myeloid-leukemia associated protein SET (Suvar3-9, enhancer of zest, thithorax), also termed PHAP-II [33]. SET is a regulator of various physiological processes including histone acetylation, regulation of transcription [34], cell growth [35] and transformation [36]. SET upregulation has been observed in chronic myelogenous leukemia and Wilm’s tumors [37, 38]. Moreover, SET was found to be a fusion partner with the CAN/Nup214 gene in an acute undifferentiated leukemia fusion gene [33]. It is still unclear which specific cancer-related pathways are affected by SET dysregulation.

The recently identified CIP2A was found to inhibit PP2A activity towards c-Myc thereby preventing its proteolytic degradation [39]. Suppression of CIP2A inhibits anchorage independent cell growth and tumor growth in vivo while CIP2A overexpression cooperates with Ras and c-Myc oncoproteins to induce tumorigenic conversion of mouse primary embryo fibroblasts. Expression of CIP2A was found to be up-regulated in head and neck squamous cell carcinomas, and in colon cancer [39].

In addition to SET and CIP2A, PP2A activity in cancer cells could be upregulated due to overexpression of protein phosphatase methylesterase-1 (PME-1), which mediates PP2A methyl ester hydrolysis of the PP2AC C-terminal Leu307 and exerts negative regulation on the preassembled holoenzymes. PME-1-mediated inhibition of PP2A results in activation of the MAPK pathway. In malignant gliomas, PME-1 overexpression correlates with both increased Erk1/2 activity and the disease stage [40]. Another endogenous PP2A inhibitor with a potential link to human cancer is type 2A-interacting protein (TIP), which was also found to be overexpressed in human cancer cells. TIP directly inhibits PP2A activity in vitro while TIP depletion results in a significant decrease in the phosphorylation of a recently identified target protein substrate of ataxia-telangiectasia mutated (ATM) - and Rad3-related (ATR) kinases [41].

Taken together, these findings strongly suggest that PP2A plays a key role in tumor suppression. Moreover, inactivation of specific PP2A complexes by different mechanisms may alter multiple pathways, each of which may contribute independently to cancer development and progression.

**PP2A ACTIVATION AS AN ANTI-CANCER THERAPY**

Since PP2A has tumor suppressing activity by regulating cell signaling cascades in a way opposite to the activity of kinase oncoproteins [3, 87], it suggests that pharmacological activation of PP2A activity would restrain the activity of the kinase oncogenes and could be beneficial for cancer treatment (Fig. 2A). Consistently with this idea, the sphingolipid metabolite ceramide (Fig. 3A), which directly induces PP2A activity in vitro while TIP depletion results in a significant decrease in the phosphorylation of a recently identified target protein substrate of ataxia-telangiectasia mutated (ATM) - and Rad3-related (ATR) kinases [41].

PP2A Activators

The first example of PP2A activator is FTY720 (2-amino-2-[2-(4-octylphenyl) ethyl]-1,3-propanediol hydrochloride) (Fingolimod; Novartis), a synthetic myricin analogue structurally similar to sphingosine (Fig. 3A). It is a known immunomodulator, which is currently being evaluated in phase III clinical trials in patients with either multiple sclerosis or undergoing renal transplantation [70-73]. FTY720 is a relatively nontoxic drug with high oral bioavailability. Although the precise mechanism underlying induction by FTY720 of PP2A activation is unclear, evidence suggests that, like ceramide, FTY720 has a direct effect on the PP2A heterotrimERIC complexes [74]. However, a recent study demonstrates that ceramide directly binds to the PP2A inhibitor SET and induces PP2A activity by disturbing interaction between PP2A and the SET protein [75]. Increased SET levels were also observed in cells that undergo FTY720-induced apoptosis [76], suggesting that the effects of FTY720 might be as a result of restraining the inhibitory function of SET.
PP2A activity can be also indirectly triggered by forskolin (or its derivative, 1,9-dideoxyforskolin), a labdane diterpene that is produced by the Indian Coleus plant (*Coleus forskohlii*) (Fig. 3A). Forskolin is known to directly activate the adenylyl cyclase enzyme that results in an increase of cAMP intracellular levels and upregulation of protein kinase A (PKA). Although most of the forskolin-mediated effects depend on augmented PKA and adenylyl cyclase activities [77-79], the ability of forskolin and its derivatives to upregulate PP2A activity does not require PKA activation or increased cAMP levels [76]. The exact mechanism of PP2A activation by forskolin still has to be elucidated.

**PP2A Activators as Potential Drugs for Cancer Therapy**

PP2A activators have been recently suggested to use in treatment of different types of human leukemia. The potential efficacy of FTY720 in treatment of B-cell chronic lymphocytic leukemia (B-CLL) has been investigated by [80]. This study demonstrated that FTY720 mediates toxic effects in cell lines representing different B-cell malignancies and primary B cells from patients with CLL. Exposure with FTY720 of transformed B-cells results in decreased levels of phospho-ERK1/2 and Bcl-2 independent apoptosis. Notably, FTY720 treatment prolonged animal survival in a xenograft SCID mouse model of B cell lymphoma/leukemia [80].

Loss of PP2A enzymatic activity appears to contribute to the pathophysiology of p210- and p190-BCR/ABL-driven leukemias [76, 81]. Most of the chronic phase CML patients are effectively treated with BCR/ABL inhibitor imatinib. However, some of the patients develop resistance to imatinib and progress to the next stage of the disease, blast crisis. It was recently reported that PP2A activity is significantly decreased in blast crisis CML patients due to overexpression of the SET inhibitor [76, 81].

Restoration of PP2A phosphatase activity in BCR/ABL cell lines by forskolin or FTY720 promotes BCR/ABL tyrosine dephosphorylation leading to its proteasomal degradation. BCR/ABL proteolysis appears to depend on the PP2A-induced activation of the tumor suppressor SHP-1 tyrosine phosphatase [76]. Treatment with either forskolin or FTY720 inhibits cell growth and induces apoptosis by inhibiting dephosphorylation of both Akt and Erk1/2 [74, 76, 81]. FTY720-treated SCID mice injected with p210- and p190-BCR/ABL cells were alive and showed no signs of leukemia after 27 weeks, while all of the untreated animals died by the fifth week after injection [81]. Importantly, long-term *in vivo* administration of PP2A activators markedly suppresses the development of BCR/ABL-induced CML blast crisis without exerting significant adverse effects on normal hematopoiesis [76].

FTY720 has been proven effective in suppression of c-Kit-mediated tumourigenesis of acute myeloid leukemias (AML) [82]. Although the tyrosine kinase inhibitor imatinib has shown remarkable success in treating tumor patients that harbor juxtamembrane c-Kit mutations, mutations involving the kinase domain are resistant to imatinib inhibition. Clinical trials with the second generation tyrosine kinase inhibitor dasatinib (SPRYCEL, Bristol-Myers Squib) have also reported disappointing results [83]. Thus, there is a clinical need for additional therapies specifically for patients with c-Kit mutated tumors.

Cancer-associated c-Kit mutations result in inhibition of PP2A activity in myeloid cells. This effect is associated with the reduced expression of PP2A structural A subunit and several PP2A regulatory subunits, including B55α, B56α, B56γ, and B56δ. Pharmacologic reactivation of PP2A by FTY720 causes decreased phosphorylation of c-Kit receptor and its downstream signaling targets.
Fig. (3). Structures of PP2A modulators. A, PP2A activators. B, PP2A inhibitors.
including Akt, STAT5, and ERK1/2. As a result, addition of FTY720 blocks proliferation and induces apoptosis of cells harboring c-Kit mutants, while having no effect on cells with wild-type c-Kit. In addition, in vivo administration of FTY720 delays the growth of c-Kit harboring tumors, and results in inhibited splenic and bone marrow infiltration, and prolonged survival [82].

These findings indicate that incorporation of PP2A activating drugs in current therapeutic protocols for blast crisis CML, imatinib/dasatinib-resistant CML, and B-ALL patients may offer a novel attractive therapeutic strategy.

**PP2A Inhibition as a Strategy for Anti-Cancer Therapy**

PP2A is a negative regulator of the cell cycle, and inhibition of PP2A appears to accelerate cell cycle progression and abrogate cell cycle checkpoints [55]. This suggests a counterintuitive approach to treat cancer cells that have hyperactive oncoproteins with PP2A inhibitors that enhance their action further, in the hope of crossing a treatment barrier. In fact, treatment with the PP2A inhibitors such as OA (at concentration higher than 1 nM) or calyculin A results in uncontrolled-premature entry into mitosis and death of cancer cells [56]. Further studies supported a strong correlation between inhibition of PP2A activity and increased toxicity especially to actively dividing cells [57]. These observations validate inhibition of PP2A as a strategy for anticancer therapy (Fig. 2B).

**Natural Compounds that Inhibit PP2A**

The PP2A catalytic subunit shares 50% identity with the catalytic domains of other members of the serine/threonine phosphatase family. The structure of the catalytic subunit of PP2A has been recently solved, even though the protein was purified and cloned 20 years ago [7]. The structural analysis of the PP2A catalytic subunit reveals that it is similar to the catalytic subunits of PP1, PP2B/calcineurin, and PP5. As in the catalytic subunits of other serine-threonine phosphatases, the PP2A enzyme has the N- and C-terminal domains linked together by a central β-sandwich, which comprises the catalytic core. A narrow channel formed by the interface of the β-sheets creates the catalytic cleft, which contains two metal ions positioned by invariant amino acid residues within the loops of the β-sheets [42, 43].

The catalytic subunits of serine/threonine phosphatases are targets of several natural toxins produced by different microorganisms (Table 1, Fig. 3B). Consistently with the homology between catalytic subunits, all described compounds have overlapping substrate selectivity. Although some of these compounds such as microcystin-LR, nodularin, calyculin A, and cantharidin have similar IC₅₀ values for PP1, PP2A, PP4, and PP5, OA is a more potent inhibitor of PP2A (Table 1). The greater affinity of PP2A for OA is due to a “hydrophobic cage” within the binding pocket of PP2A that is not conserved in other serine-threonine phosphatases. This cage surrounds the hydrophobic portion of OA and is the structural feature responsible for the 100-fold higher affinity of OA for the PP2A enzyme [7]. Structure-activity relationship studies have facilitated the design and synthesis of inhibitor analogs with preferential selectivity for PP2A [44, 45].

The most successful example is the development of the norcantharidin analogue, LB1.2 [46]. Norcantharidin inhibits several serine/threonine phosphatases (Table 1) and demonstrates anticancer properties [47, 48]. It is currently unknown whether the anti-cancer effect of this compound is due to the inhibition of one or more phosphatases. Norcantharidin has several characteristics that make it ideal for development as an anticancer drug. It is a lipophilic compound that can easily traverse biological membranes and, importantly, is not a substrate for the drug transporter P-glycoprotein [49]. Significant effort has been devoted for development of norcantharidin analogues with high selectivity for PP2A [50-52]. Among numerous compounds that were generated during these studies, LB1.2 was selected for the future investigations as the most effective inhibitor of PP2A. LB1.2 inhibits PP2A (IC₅₀ = 0.4 μM) with more specificity than PP1 (IC₅₀ = 80 μM). It mimics the effect of OA, and has less pronounced toxic effect, which makes LB1.2 suitable for use in anti-cancer therapies [53, 54].

**PP2A Inhibitors as Potential Drugs for Cancer Therapy**

Over other natural PP2A inhibitors such as OA, microcystin-LR or calyculin A, fostriecin has a clear advantage for development as an anticancer drug because of its low toxicity and high specificity profiles [56]. Fostriecin was discovered based on its anti-tumor properties without any knowledge of its mechanism of action. At first, fostriecin's tumor cytotoxic activity was attributed to the inhibition of DNA topoisomerase II, but its effective concentration (IC₅₀ = 40 μM) and cell cycle effects were subsequently found to be inconsistent with classic topoisomerase II inhibitors [56, 58].

Preclinical studies revealed that fostriecin suppresses the growth of cancer cells by triggering premature mitotic entry and resulting in apoptotic cell death [56, 59, 60]. Fostriecin has completed a phase I clinical trial in 20 patients [61, 62]. Unfortunately, it was found to be highly susceptible to oxidation and have a short

### Table 1. Natural Inhibitors of Serine/Threonine Phosphatases

<table>
<thead>
<tr>
<th>Inhibitors</th>
<th>Origin</th>
<th>IC₅₀ (nM)†</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PP1</td>
<td>PP2A</td>
</tr>
<tr>
<td>Okadaic acid</td>
<td>Halichondria okadai</td>
<td>10-50</td>
</tr>
<tr>
<td></td>
<td>Halichondria malanodocia</td>
<td>Dinophysis acuminata</td>
</tr>
<tr>
<td>Fostriecin</td>
<td>Streptomyces pulveraceae</td>
<td>4.5×10⁴-5.8×10⁵</td>
</tr>
<tr>
<td>Cantharidin/Noncantharidin</td>
<td>Lytta vesicatoria</td>
<td>0.5 - 2.0</td>
</tr>
<tr>
<td>Nodularin-V</td>
<td>Nodularia spumigera</td>
<td>0.5 - 3.0</td>
</tr>
<tr>
<td>Microcystin-LR</td>
<td>Microcystis aeruginosa</td>
<td>0.3 – 1.0</td>
</tr>
<tr>
<td>Tautomycin</td>
<td>Streptomyces verticillus</td>
<td>0.23 – 22</td>
</tr>
<tr>
<td>Calyculin A/C</td>
<td>Discoderma calyx</td>
<td>0.6</td>
</tr>
</tbody>
</table>

*The IC₅₀ values are based on published studies conducted using purified catalytic subunits of the indicated phosphatases and phosphoproteins as substrates. Because IC₅₀ values are influenced by the mode of inhibition, and the amount of both enzyme and substrate employed in an assay the different values shown reflect the variations in the assay conditions used in different laboratories [90-93].
PP2A as a Drug Target

plasma half-life. No tumor response was observed, presumably due to drug instability and inadequate purity in the clinical supply [56]. To overcome these issues investigators used complementary strategies to generate analogues of fostriecin, including the development of several novel syntheses [56] and the manipulation of Streptomyces pulveraceus polyketide synthase gene clusters. The latter strategy has been successfully used to create bacterial strains that produce a single analog of phoslactomycin B, a drug structurally related to fostriecin [63]. Phoslactomycins are potent and selective inhibitors of PP2A, and phoslactomycin A has been shown to inhibit lung metastasis in mice through activation of natural killer cells [64]. The ability to produce a pure supply of phoslactomycins provides us a proof of principle for the production of pure supplies of fostriecin analogues that can be clinically tested for their anti-cancer activity [65].

Another natural product known for its antitumor properties is cantharidin secreted by many species of blister beetles, and most notably by the Spanish fly, Lytta vesicatoria. Despite its potential toxicities the Chinese have used cantharidin as a medicinal agent for over 2000 years and as an anti-cancer drug since 1264 for treatment of hepatomas and oesophageal carcinomas [66]. Western medicine has prohibited cantharidin from clinical use because of its high nephrotoxicity. However, the demethylated analogue of cantharidin, norcantharidin, not only displays less toxicity but induces haemopoiesis via bone marrow stimulation and retains antitumor activity. Norcantharidin was shown to suppress the growth of cancer cell lines both in vitro and in vivo [66].

Since 1984 norcantharidin has been used in cancer therapy of primary hepatomas and upper gastrointestinal carcinomas in China. It has been reported that norcantharidin increased the mean survival time of 285 patients with primary hepatoma from 4.7 to 11.1 months, and the 1 year survival rate from 17% to 30%, as compared to 102 patients treated with conventional chemotherapy (5-fluorouracil, hydroxycamptothecin, vincristine, thiophosphoramide, or mitomycin) [66]. Although further clinical trials are required to explore the anticancer properties of these compounds, the reported results look promising.

The Combination of Chemotherapy and PP2A Inhibitors for Cancer Treatment

A recent study [46] proposes to combine PP2A inhibitors with classical chemo- and radiotherapies to enhance the effectiveness of classical cytotoxic treatment (Fig. 2B). In this study, the authors used the described above synthetic derivative of norcantharidin, LB1.2. Treatment with LB1.2 in combination with temozolomide (TMZ), a DNA-methylating chemotherapeutic drug, causes complete regression of glioblastoma multiforme (GBM) xenografts without recurrence in 50% of animals (up to 28 weeks) and complete inhibition of growth of neuroblastoma xenografts. Treatment with either drug alone results in only short-term regression with all xenografts resuming rapid growth. Combined with another widely used anticancer drug, doxorubicin (DOX) LB1.2 also causes marked GBM xenograft regression, whereas DOX alone only slows down the tumor growth.

Exposure of cancer cells to LB1.2 induces increased phosphorylation of both Akt-1 and polo-like kinase 1 (Plk-1) that, in turn, abolishes G2/M checkpoint and promotes premature entry into mitosis. Moreover, LB-1.2 treatment leads to a marked decrease of transcriptionally controlled tumor protein (TCTP), which stabilizes microtubules before and after mitosis and exerts potent anti-apoptotic activity. As result, LB1.2 treatment induces mitotic catastrophe and cell death.

On the other hand, increased Akt-1 activity induced by LB1.2 treatment stabilizes the p53 ubiquitin ligase MDM2 that results in down-regulation of p53 expression and activity. Consistently, LB1.2 treatment impairs p53-mediated cell cycle arrest. Therefore, LB1.2 treatment triggers a chain of alterations in cancer cell signal-
tential therapeutic benefits in treating cancer. Accumulation of structural data for phosphatase holoenzymes and development of novel approaches for drug design will allow us to obtain more specific and less toxic modulators of PP2A activity.

Furthermore, several lines of evidence suggest that dysfunction of specific PP2A complexes is likely to be primarily responsible for the changes that lead to cell transformation. The discovery of small molecule selectively targeted specific PP2A complexes opens up new possibilities for disruption of particular phosphatase complexes to achieve a therapeutic effect. Systematic characterization of the regulation and function of specific PP2A complexes may lay the foundation for future studies aimed at developing specific antagonists for these complexes, which may ultimately prove to be valuable therapeutic agents for cancer treatment.

ACKNOWLEDGEMENTS

We thank Thomas Soin for helpful discussions on the manuscript. This work was supported in part by a VIB startup grant.

REFERENCES


