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REVIEW

TPP1 as a versatile player at the ends of chromosomes

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Abstract **Telomeres, the ends of linear eukaryotic chromosomes, are tandem DNA repeats and capped by various telomeric proteins. These nucleoprotein complexes protect telomeres from DNA damage response (DDR), recombination, and end-to-end fusions, ensuring genome stability. The human telosome/shelterin complex is one of the best-studied telomere-associated protein complexes, made up of six core telomeric proteins TRF1, TRF2, TIN2, RAP1, POT1, and TPP1. TPP1, also known as adrenocortical dysplasia protein homolog (ACD), is a putative mammalian homolog of TEBP-β and belongs to the oligonucleotide binding (OB)-fold-containing protein family. Three functional domains have been identified within TPP1, the N-terminal OB fold, the POT1 binding recruitment domain (RD), and the carboxyl-terminal TIN2-interacting domain (TID). TPP1 can interact with both POT1 and TIN2 to maintain telomere structure, and mediate telomerase recruitment for telomere elongation. These features have indicated TPP1 play an essential role in telomere maintenance. Here, we will review important findings that highlight the functional significance of TPP1, with a focus on its interaction with other telosome components and the telomerase. We will also discuss potential implications in disease therapies.**

Keywords telomere, TPP1, TIN2, telosome/shelterin, telomerase

Introduction

In human cells, telomeres consist of tandem DNA repeats of $(TTAGGG)_n$ with varying length (5–15 kb) (Moyzis et al., 1988; Holt et al., 1996; Zhao et al., 2009; Chow et al., 2012). Telomeres terminate in 3′ single-stranded DNA overhangs that invade into the double-stranded region to form the so-called T-loop (de Lange, 2004). A multitude of proteins maintain and protect chromosome ends. Telomeric proteins bind to telomeres and protect them from being recognized as DNA damage sites (de Lange, 2005; Songyang and Liu, 2006; Palm and de Lange, 2008; Nandakumar et al., 2012; Nandakumar and Cech, 2013). The structure formed by telomere DNA and its binding proteins ensures genome stability (Blackburn, 2001; de Lange, 2002).

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Telomeres are usually maintained by the telomerase, which consists of the catalytic reverse transcriptase and the RNA template, and adds repeat sequences to telomere ends during S phase of cell cycle (Greider and Blackburn, 1985; Kim et al., 1994; Zhao et al., 2009). Telomerase activity is tightly regulated. Progressive telomere shortening accelerates aging and apoptosis in somatic cells, where telomerase activity is low or undetectable (Harley et al., 1990; Levy et al., 1992). In cancer and stem cells, high telomerase activity supports cell growth and division (Colgin and Reddel, 1999). It should be noted that telomere maintenance can be achieved in telomerase-negative cells through a homologous recombination-based mechanism called alternative lengthening of telomeres (ALT) (Lundblad and Blackburn, 1993; Bryan et al., 1995; Le et al., 1999; Cesare and Reddel, 2010).

In mammalian cells, telosome/shelterin that consists of TRF1 (telomeric repeat binding factor 1), TRF2 (telomeric repeat binding factor 2), POT1 (protection of telomeres 1), TIN2 (TRF1-interacting nuclear protein 2), RAP1 (repressor and activator protein 1), and TPP1 (Liu et al., 2004a; de

Lange, 2005), functions as the core of the telomere interactome and plays an essential role in telomere maintenance (Songyang and Liu, 2006). In this complex, TRF1 and TRF2 directly bind to telomeric double-stranded DNA (dsDNA) (Zhong et al., 1992; Bilaud et al., 1997), while POT1 is a sequence-specific single-stranded telomeric DNA binding protein (Baumann and Cech, 2001). TIN2, RAP1, and TPP1 do not directly bind telomere DNA and exert their function through interacting with TRF1, TRF2, and POT1 (Kim et al., 1999; Li et al., 2000; Liu et al., 2004b).

Telomere dysfunction is associated with aging, cancer development, and other diseases (Collins and Mitchell, 2002; Granger et al., 2002; Hahn and Weinberg, 2002; Maser and DePinho, 2002; Blasco, 2005). For example, mutations of telomerase subunits and telosome/shelterin components have been linked to various diseases, including Duchenne muscular dystrophy, dyskeratosis congenital (DC), and pulmonary fibrosis (Sacco et al., 2010; Dokal, 2011; Yang et al., 2011; Zhong et al., 2011; Armanios and Blackburn, 2012; Zhong et al., 2012). Studies in the past decade have demonstrated pivotal roles of TPP1 in telomere regulation, e.g., telosome assembly, telomere protection, and telomerase recruitment. In the following sections, we will discuss TPP1 function and its interacting partners, particularly in light of recent findings.

Identification of TPP1 as a novel telomeric protein

Through large-scale affinity purification followed by mass spectrometry analysis, our laboratory found that a novel protein, which we named PTOP, could localize to telomeres and interact with TIN2 and POT1 (Liu et al., 2004b). Two other laboratories also reported similar findings around the same time and named the protein PIP1 (POT1-interacting protein) and TINT1 respectively (Houghtaling et al., 2004; Liu et al., 2004b; Ye et al., 2004). The protein was later renamed TPP1.

We now know that TPP1 is a putative mammalian homolog of TEBP-β and belongs to the oligonucleotide binding (OB)-fold-containing protein family (Wang et al., 2007; Xin et al., 2007). In addition to the N-terminal OB fold, TPP1 has a POT1 recruitment domain (RD), and a TIN2-interacting domain (TID) (Fig. 1). Human TPP1 mRNA contains two Kozak consensus sequences, leading to two isoforms – TPP1-L (with a predicated length of 544 amino acids) and TPP1-S (lacking the N-terminal 86 amino acids). TPP1-S is considered the predominant form in human cells (Chen et al., 2007). Both forms contain the essential domains for TPP1 function.

Co-immunoprecipitation and yeast two-hybrid assays revealed that TPP1 interaction with TIN2 and POT1 was independent of dsDNA and mediated through distinct regions on each protein (Liu et al., 2004b). For example, TPP1 can bind POT1 through the TPP1 RD domain and the POT1 PBR domain (Liu et al., 2004b), in an OB fold independent manner (Fig. 1). This explains the observation that the human POT1 mutant that did not have the OB fold could still be recruited to telomeres (Loayza and de Lange, 2003). Without the TIN2 binding domain (TID), TPP1 exhibited diffuse nucleoplasmic immunostaining, and it suggested TIN2 plays a critical role in TPP1 telomeric localization (Houghtaling et al., 2004). The RD domain is equally important, because RD domain is necessary for recruitment of POT1 to telomeres to protect telomere ssDNA (Liu et al., 2004b). Both knockdown of TPP1 and dominant expression of TPP1 mutants (e.g., TPP1ΔRD or TPP1 RD domain alone) resulted in telomere lengthening in HTC75 cells (Liu et al., 2004b; Ye et al., 2004; Chen et al., 2007), consistent with the model that the telosome complex can sequester telomere ends and negatively regulate telomerase access (Loayza and De Lange, 2003; Houghtaling et al., 2004; Liu et al., 2004b; Ye et al., 2004; Ye and de Lange, 2004).

More recent work on TPP1 structure and activity has yielded a more comprehensive picture of TPP1 (Wang et al., 2007; Xin et al., 2007; Tejera et al., 2010; Nandakumar et al., 2012; Zhong et al., 2012; Zhang et al., 2013). The multiple functional domains of TPP1, the N-terminal OB fold (OB-fold), POT1 recruitment domain (RD), and TIN2-interacting domain (TID) (Fig. 1), allow it to interact with members of the telosome as well as proteins outside of the telosome (e.g., OBFC1/Stn1 and UPF1) (Wan et al., 2009; Chawla et al., 2011; Lee et al., 2011; Chen et al., 2012b), to modulate telomerase recruitment and activity, control telomere length, and protect telomere ends, solidifying its role as a versatile player in all aspects of telomere maintenance.

The heterodimer of POT1 and TPP1 enhanced telomerase recruitment and activity regulation

In the ciliated protozoa *Oxytricha nova*, telomere end binding proteins (TEBP) TEBP-α and TEBP-β heterodimerize to cap the very ends of chromosomes (Gray et al., 1991). Human POT1 can specifically recognize single-stranded telomeric DNA (ssDNA) (Loayza and De Lange, 2003; Lei et al., 2004), and is a homolog of TEBP-α (Lei et al., 2004). Significant sequence variation exists between TPP1 and TEBP-β OB folds, especially in the connecting loop regions, which confounded initial bioinformatic analysis of the two proteins (Wang et al., 2007). However, computational threading and structural studies revealed extensive similarities between TPP1 and TEBP-β, and found TPP1 to be the putative mammalian homolog of TEBP-β (Wang et al., 2007; Xin et al., 2007). In fact, the OB-fold (90–250) of TPP1 resembles most the TEBP-β OB-fold, and much less the OB-fold in TEBP-α or other OB-fold containing proteins. These studies demonstrate an evolutionarily conserved mechanism

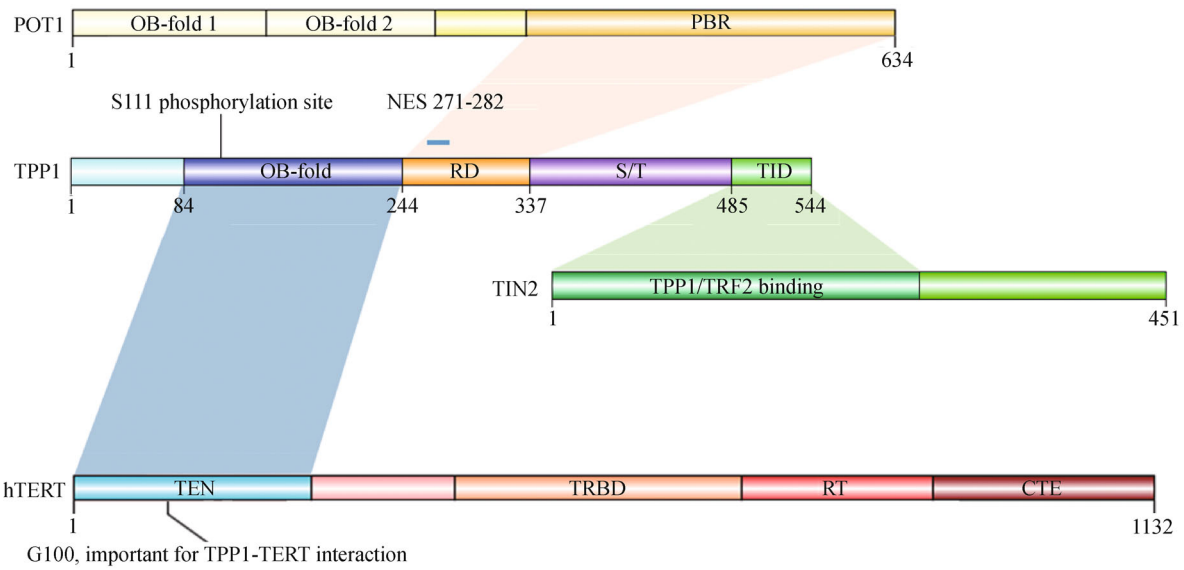

Figure 1 Schematic domains representation of TPP1 and its interacting proteins mainly discussed in this paper (extended and re-edited from Ye et al., 2004). The reported hTERT interaction sites, S111 phosphorylation site and NES in TPP1 were showed. OB-fold, oligonucleotide binding fold; PBR, PTOP(TPP1) binding region; RD, POT1 recruitment domain; S/T, Ser-rich region; TID, TIN2-interacting domain; TEN, telomerase N terminus domain; TRBD, telomerase RNA binding domain; RT, reverse transcriptase domain; CTE, C-terminal extension. G100 site is important for TPP1 and hTERT interactions.

that utilizes the TEBP-α/β heterodimer (or its equivalent) to regulate telomeres.

TPP1 alone do not have binding activity to telomeric DNA. However, association of TPP1 and POT1 through their respective PBR and RD domains can significantly enhance POT1-ssDNA interaction (Wang et al., 2007; Xin et al., 2007). In an elegant study by the Cech's lab, it was found that long telomeric ssDNA substrates were coated by multiple POT1-TPP1 heterodimers to form a compact and well-ordered structure, which contributes to overhang protection and regulation of telomerase activity (Taylor et al., 2011). More recently, the POT1-TPP1 complex was also shown to interact with G-quadruplex DNA, likely generating dynamic G-quadruplex folding and unfolding near the 3′ end through the back and forth movement of the complex along the overhang (Hwang et al., 2012). This model suggests a possible mechanism for the dynamic modulation of TPP1-POT1 interaction with the telomerase. In addition, TPP1 binding to POT1 can also greatly enhance its discrimination against telomeric non-coding RNAs such as TERRA (Azzalin et al., 2007; Nandakumar et al., 2010).

The telomerase, consisting of the catalytic reverse transcriptase subunit TERT and the RNA component TERC, is responsible for the highly regulated and efficient extension of telomeric DNA (Greider and Blackburn, 1989; Lingner et al., 1997; Collins, 2008). Little is known how telomerase activity is regulated in mammalian cells. Initial studies of TPP1 knockdown and mutant phenotypes suggested TPP1 to be a negative regulator of telomere length, presumably through TPP1-POT1 interaction (Liu et al., 2004b). It was therefore surprising when we observed that TPP1 could directly interact with TERT through its OB fold (Xin et al., 2007). The Lei's group also reported that the TPP1-POT1 heterodimer could enhance telomerase processivity by decreasing primer dissociation and increasing the translocation of telomerase (Wang et al., 2007; Latrick and Cech, 2010). These findings have led to increased scrutiny of the TPP1-telomerase interaction, offering insight into TPP1-dependent telomere length regulation. Being both a positive and a negative regulator of telomere length, TPP1 has emerged as a key player in telomere maintenance.

In budding yeast, the telomerase holoenzyme component Est3 (ever shorter telomeres 3) interacts with the telomerase through a domain predicted to be similar to TPP1 OB-fold (Hughes et al., 2000; Lee et al., 2008; Yu et al., 2008; Talley et al., 2011; Yen et al., 2011). *In vitro*, telomerase activity is stimulated in the presence of Est3. Mutation of conserved residues in the surface of putative OB fold in Est3 resulted in shorter telomeres (Lee et al., 2008; Yu et al., 2008). Such conservation suggests that TPP1 and Est3 are functional homologs. Proper TPP1 telomere targeting is crucial to its recruitment activity, as loss of either TPP1 or TIN2 (which tethers TPP1 to telomeres) led to incorrect telomerase recruitment (Abreu et al., 2010). Indeed, only wild type TPP1, not the OB fold deletion mutant, could rescue the recruitment of telomerase to telomeres (Abreu et al., 2010). Further studies from three independent groups in 2012 provided us a higher resolution picture of the specific interaction between TPP1 and TERT. Multiple potential telomerase interacting sites on TPP1 were identified, the loop residues (D166, E168 and K170) (Zhong et al., 2012), the telomerase interaction interface (W167 through F172, as well

as L183 and E215) (Sexton et al., 2012), and the TEL patch (TPP1 glutamate (E) and leucine (L)-rich patch, which includes E168, E169, E171, R180, L183, L212 and E215) (Nandakumar et al., 2012). Interestingly, mutations in the TPP1-TERT interacting region on TERT were found in human disease DC, indicating the physiological importance of such interaction (Zhong et al., 2012; Zaug et al., 2013). If telomerase is activated by TPP1, one might expect a mechanism to reset telomerase activity after the ends are extended. The mammalian Ctc1-Stn1-Ten1 (CST) complex has been proposed to act as a terminator for the telomerase-mediated telomere extension process (Chen et al., 2012a). In fact, Stn1, a subunit of CST, could directly interact with TPP1 and compete with TPP1-POT1 complex for binding to telomeric ssDNA (Wan et al., 2009; Chen et al., 2012a).

A series of studies from the Blasco's group have highlighted the importance of TPP1-dependent telomerase recruitment in development. It has been shown that the telomerase is highly expressed in pluripotent stem cells such as embryonic stem (ES) cells (Flores et al., 2005; Blasco, 2007). During somatic cell reprogramming to generate iPS (induced pluripotent stem) cells, net telomere elongation occurs and depends on telomerase activity (Marion et al., 2009). However, net telomere elongation was abolished in iPS cells derived from TPP1-deficient mouse embryonic fibroblasts (MEFs), suggesting a crucial role for TPP1 in recruiting the telomerase to maintain telomeres *in vivo* (Tejera et al., 2010). Binding of the POT1-TPP1 complex to telomeres will cap the ends and possibly block telomerase access. On the other hand, POT-TPP1 can recruit the telomerase and promote its processivity, thereby positively regulating telomere length. This yin-yang function of the complex underscores the dynamic and complex nature of telomere regulation (Xin et al., 2007).

Protecting telomere integrity

Telomere maintenance is intimately linked to DNA damage response pathways as well as DNA replication. The telosome-centric protein network helps protect telomeres from being recognized as double-strand breaks (DSBs) and from subsequent double-strand repair (DDR) mechanisms such as non-homologous end joining (NHEJ) and homologous recombination (HR) (de Lange, 2005). Short telomeres or TRF2 deficiency can elicit DDR through ataxia-telangiectasia mutated (ATM), the kinase that is primarily responsible for sensing and responding to DSBs (Karlseder et al., 1999; Shiloh, 2003; Herbig et al., 2004; Celli and de Lange, 2005; Verdun and Karlseder, 2006). Disruption of TPP1-POT1 function, on the other hand, leads to replication protein A (RPA) binding and ataxia telangiectasia and Rad 3 related ATR-mediated ssDNA damage pathways (Flynn et al., 2011). In normal cell cycle process, RPA is coated on telomere ssDNAs for replication of telomeric DNA. It was puzzling that how the cells distinguish replicative telomere ends from damaged ones and effectively displace RPA from telomere ssDNAs. The interplay between telomere regulators and DNA replication machineries was highlighted in an elegant study by the Zou's lab. It was demonstrated that RPA displacement and telomere protection was achieved, following DNA replication, through the coordinated action of non-coding RNA TERRA, heterogeneous nuclear ribonucleoprotein A1, and TPP1-POT1 complex (Flynn et al., 2011).

Mice have two POT1 isoforms, Pot1a and Pot1b, both of which interact with TPP1 (Wu et al., 2006). Individual knockout (KO) of the two isoforms in mice led to distinct results, increased chromosomal fusions and aberrant HR for Pot1a KO mice (Hockemeyer et al., 2006; Wu et al., 2006) and G-overhang elongation for Pot1b KO mice (Hockemeyer et al., 2006). Consistent with the idea that TPP1 tethers POT1 to telomeres, TPP1 knockout phenotype is similar to Pot1a/b double knockout phenotype (Kibe et al., 2010). Since TPP1 acts as an important POT1 recruitment factor, TPP1 mutants such as TPP1ΔRD that can strip POT1 from telomeres should also trigger ATR-dependent DDR. However, long-term knockdown of TPP1 elicits primarily ATM-dependent DDR at telomeres (Guo et al., 2007), indicating distinct pathways for TPP1 and POT1 in protecting chromosomal ends and repressing DDR at telomeres in mice. A similar result was obtained using the mouse TPP1 disease mutant cell line – $Tpp1^{acd/acd}$, where telomere de-protection can occur due to dysfunctional TPP1 (Hockemeyer et al., 2007). Indeed, the POT1a-TPP1 complex appears to mainly repress HR at telomeres (as opposed to TRF2-mediated repression of NHEJ).

In 2010, Blasco and her colleagues utilized conditional knockout (CKO) mice and derivative MEFs to make a clearer vision to TPP1's role *in vivo* (Tejera et al., 2010). According with TPP1's telomere capping function, MEFs with deficient TPP1 had more sister chromatid fusions and multi-telomeric signals relating to telomere fragility, which were increasing chromosomal instability. In the meantime, abogation of TPP1 also abolished net telomere elongation in the process of reprogramming into iPSCs. The results offered people strong evidences in TPP1 recruiting telomerase. In stratified epithelia CKO mice, their skin showed severe hyperpigmentation and impaired hair follicle morphogenesis. These syndromes are likely caused by cell cycle arrest, a consequence of TPP1's absence eliciting DDR at telomeres. This model was supported by p53 deficient mice, which overcame the skin problems caused by TPP1 KO.

Telosome assembly and spatial control of telomere proteins

During our studies of core telomere proteins by large-scale immunoprecipitation (IP) and mass spectrometry, we frequently observed all six core telomere proteins together (Liu

et al., 2004b; O'Connor et al., 2004), suggesting that they could form a single complex. Subsequent co-IP and chromatography studies showed that RAP1, TIN2, POT1, and TPP1 could directly or indirectly associate with TRF1 or TRF2, supporting the notion that these six telomeric proteins could form a high-order complex (Liu et al., 2004a) (Fig. 2). We named the complex telosome and it was later renamed to shelterin (de Lange, 2005). Furthermore, we demonstrated that the extreme C terminus of TPP1 mediates its direct interaction with TIN2, as well as regulates the integrity of the telosome complex by enhancing TIN2-TRF2 association. More specifically, TPP1 stabilizes the TIN2-TRF2 and TRF1-TIN2-TRF2 interactions so that TIN2 can bridge both TRF1 and TRF2 subcomplexes, promoting telosome assembly (O'Connor et al., 2006) (Fig. 2).

The three proteins, TPP1, TIN2, and POT1 can localize and interact with each other in both the cytoplasm and the nucleus (Chen et al., 2007). When we looked closer, we found a nuclear export signal (NES) in the RD region of TPP1. The nuclear export function of this NES sequence is independent of RD-mediated TPP1 interaction with POT1 (Chen et al., 2007). Thanks to TPP1's ability to shuttle between the cytosol and nucleus, a regulatory cascade is established. TPP1 regulates POT1 nuclear localization, and TIN2 association with TPP1 in turn promotes POT1 retention in the nucleus. Our recent work on TIN2 has added another level of control to this regulatory loop. We have found that TIN2 can also localize to the mitochondria where it regulates oxidative phosphorylation (Chen et al., 2012b). We determined that the mitochondrial targeting sequences on TIN2 were located in the N terminus of TIN2, overlapping with the region that mediates TPP1 binding. In this case, TPP1 interaction with TIN2 can block TIN2 mitochondrial targeting, thereby enhancing nuclear targeting of TIN2. We have shown that decreased TIN2 targeting to the mitochondria resulted in decreased oxidative phosphorylation and increased glycolysis. These findings further attest to the intricate mechanisms that govern telomere maintenance and other cellular events. In addition to a direct role in recruiting and regulating the telomerase and protecting the integrity of chromosomal ends, TPP1 also can impact mitochondrial function through its interaction with TIN2, providing new avenues to explore the link between telomere maintenance and metabolic control, and the extra-telomeric function of telomere proteins.

Post-translational modification of TPP1

The sensing and repair of DSBs by the DDR pathway is a multi-step and multi-player process. It has been demonstrated that ubiquitination is a critical posttranslational modification event that helps control the assembly of DDR complexes at DSBs. In particular, the E3 ligase RNF8 (RING finger protein 8) is recruited to damage sites and mediates the ubiquitination of histones such as H2A and H2AX (Panier and Durocher, 2009). In fact, it was revealed that one of the RNF8 substrates at the telomeres is TPP1. RNF8 can directly bind and ubiquitinate TPP1 at Lys233 with Lys63 polyubiquitin chain to stablize TPP1 at the telomeres (Rai et al., 2011). In fact, in

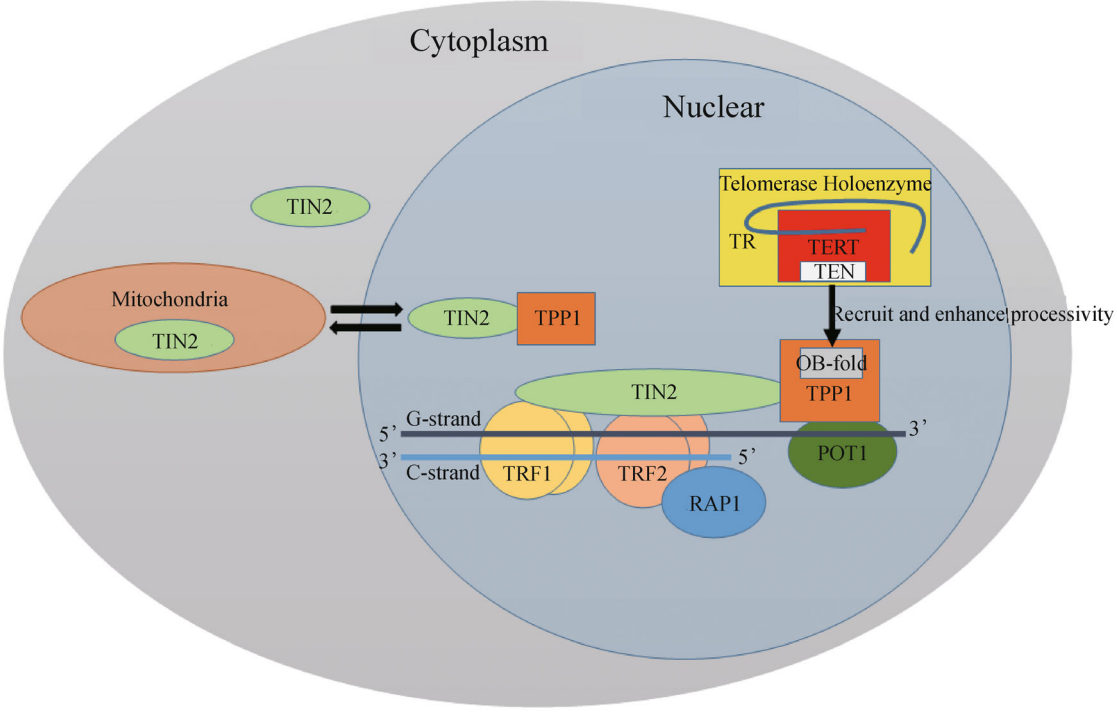

Figure 2 TPP1 functions in telomere capping, telosome assembly, telomerase recruitment and interaction proteins spatial regulation.

our whole-genome studies of interactors of telosome proteins through Bi-molecular Fluorescence Complementation (BiFC) assays, we identified a number of ubiquitin E3 ligases as potential interactors and regulators of TPP1 (Lee et al., 2011). These findings link the E3 ubiquitin ligase such as RNF8 to telomere end protection, and further emphasize the importance of TPP1 in telomere maintenance.

Telomere homeostasis in dividing cells is tightly regulated during cell cycle. Phosphorylation of the TPP1 homolog TEBP-β regulates cell cycle-dependent telomerase recruitment in ciliate (Paeschke et al., 2008), and the telomerase has been shown to extend telomeres during S phase in human cells (Wright et al., 1999; Zhao et al., 2009). These observations prompted us to investigate TPP1 phosphorylation throughout the cell cycle. We found that TPP1 could be phosphorylated at multiple sites in a cell cycle-dependent manner (Zhang et al., 2013). Specifically, Ser111 (S111) within the OB fold underwent phosphorylation during S/G2/M phase of the cell cycle, likely mediated by CDKs. Mutation of Ser111 to Ala was accompanied by decreased telomerase recruitment and shortened telomeres, indicating that phosphorylation of TPP1 Ser111 regulates telomerase access during critical cell cycle stages when telomere elongation occurs. Interestingly, the phosphate group of phosphorylated Ser111 is predicted to be positioned at the same interface formed by D166, E168, E169, E171, and E215, residues required for TPP1-telomerase interaction (Nandakumar et al., 2012; Sexton et al., 2012; Zhong et al., 2012), suggesting that phosphorylated S111, in addition to the previously mapped D/E residues, also participates in interaction with the telomerase.

The state of telomeres is also tightly connected to cell growth signaling. The PI3K/Akt signaling cascade is one of the best-studied pathways in regulating cell growth, survival, and aging (Manning and Cantley, 2007;Carnero, 2010; Kloet and Burgering, 2011). Akt functions downstream of PI3K and has been shown to interact with and phosphorylate hTERT and TRF1 (Kang et al., 1999; Chen et al., 2009; Haendeler et al., 2003; Chung et al., 2012). Our recent work on TPP1 suggests that TPP1 can also be phosphorylated by the Akt kinase (Han et al., 2013). We found that the OB fold could mediate homodimerization of TPP1, a process regulated by Akt. When we inhibited Akt activity using siRNAs or small molecule inhibitors, we observed reduced TPP1 dimerization that was accompanied by decreased telomeric localization of TPP1 and POT1, and increased damage at telomeres. This work suggests previously unknown mechanisms that communicate signals at the plasma membrane, through Akt and TPP1, to the machinery that regulates and protects the telomeres.

Conclusion and outlook

It is interesting to note that our BIFC screening of genome-wide binding partners of TPP1 identified many candidates with no apparent function on telomeres (Lee et al., 2011). And recent findings of TIN2 are prime examples of how telomere proteins may have important roles in diseases and in areas outside of telomeres. For example, TIN2 has been linked to the rare congenital disorder dyskeratosis congenita (Walne et al., 2008; Sarper et al., 2010; Yang et al., 2011; Sasa et al., 2012) and oxidative phosphorylation in the mitochondria (Chen et al., 2012b). Given the intertwined regulatory loops and interactions among telosome components, it is reasonable to speculate a possible role for TPP1 in diseases and other areas. It has been found that altered expression of TPP1 may play a direct role in the pathogenesis of diseases such as rheumatoid arthritis (Qing et al., 2012), B cell chronic lymphocytic leukemia (B-CLL) (Redon et al., 2007), and ulcerative colitis and Crohn's disease (Arnerić and Lingner, 2007). These findings underline the potential of TPP1 as a therapeutic target. In addition, our findings that TPP1 inhibits TIN2 mitochondrial localization and senses Akt activation, suggest that TPP1 may regulate cross-talks between different cellular aging pathways. Further exploring the canonical and extra-telomeric functions of TPP1, therefore, will not only advance our understanding of the molecular activities of TPP1, but also its potential role in diseases and other biologic processes.

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Compliance with ethics guidelines

Sijie ZHANG, Zhenhua LUO, Guang SHI, Dan LIU, Zhou SONGYANG and Junjiu HUANG declare that they have no conflict of interest. This manuscript is a review article and does not involve a research protocol requiring approval by the relevant institutional review board or ethics committee.

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