Review article

Redox regulation of tumor suppressor PTEN in cell signaling

Ying Zhang\textsuperscript{a}, Jiyoung Park\textsuperscript{b}, Seong-Jeong Han\textsuperscript{c}, Sung Yeul Yang\textsuperscript{a}, Hyun Joong Yoon\textsuperscript{a}, Iha Park\textsuperscript{a}, Hyun Ae Woo\textsuperscript{b}, Seung-Rock Lee\textsuperscript{a,}\textsuperscript{*}

\textsuperscript{a} Department of Biochemistry, Department of Biomedical Sciences, Research Center for Aging and Geriatrics, Research Institute of Medical Sciences, Chonnam National University Medical School, Gwangju, 501-190, Republic of Korea
\textsuperscript{b} College of Pharmacy, Graduate School of Pharmaceutical Sciences, Ewha Womans University, Seoul, 120-750, Republic of Korea
\textsuperscript{c} COTDE Inc. 19-3, Ugakgol-gil, Susin-myeon, Cheonan-si, Chungcheongnam-do, 330-882, Republic of Korea

\textbf{ARTICLE INFO}

\textbf{Keywords:} PTEN
Redox regulation
Peroxides
Trx dimerization
Prx dimerization
Prx hyperoxidation

\textbf{ABSTRACT}

Phosphatase and tensin homologs deleted on chromosome 10 (PTEN) is a potent tumor suppressor and often dysregulated in cancers. Cellular PTEN activity is restrained by the oxidation of active-site cysteine by reactive oxygen species (ROS). Recovery of its enzymatic activity predominantly depends on the availability of cellular thioredoxin (Trx) and peroxiredoxins (Prx), both are important players in cell signaling. Trx and Prx undergo redox-dependent conformational changes through the oxidation of cysteine residues at their active sites. Their dynamics are essential for protein functionality and regulation. In this review, we summarized the recent advances regarding the redox regulation of PTEN, with a specific focus on our current state-of-the-art understanding of the redox regulation of PTEN. We also proposed a tight association of the redox regulation of PTEN with Trx dimerization and Prx hyperoxidation, providing guidance for the identification of novel therapeutic targets.

1. Introduction

Reactive oxygen species (ROS) are inevitably generated during aerobic and anaerobic metabolism and have detrimental effects on cellular biomolecules under pathological conditions [1]. Increasing evidence has indicated that ROS, such as H\textsubscript{2}O\textsubscript{2}, are produced and employed in physiological settings to serve as important signaling messengers for coordinating a variety of physiological functions, including proliferation, differentiation, apoptosis, signal transduction, and other critical events [2–5]. To function as signaling messengers, these reactive molecules mainly trigger reversible oxidative post-translational modifications (PTMs) of reactive cysteine residues in regulatory proteins [6]. Because of the lack of enzymes to remove hydroxyl radicals and reactive aldehydes, their aggressive reactivity leads to the irreversible oxidation or degradation of functional proteins [7,8], a mechanism underlying various disorders and pathologies, such as diabetes, obesity, and cancer [9,10].

Signaling through PI3K/AKT is pivotal to cell growth and survival. The interaction of growth factors with receptor tyrosine kinases (RTK) typically activates PI3K. It has been shown that external stimuli, such as insulin, cytokines, neurotransmitters, peptide growth factors, and hormones, can activate the PI3K pathway, which results in ROS generation [5,11–13]. Upon the activation of cells by growth stimuli, classic PI3K family members catalyze the phosphorylation of phosphatidylinositol 4,5-bisphosphate (PIP2) to phosphatidylinositol 3,4,5-trisphosphate (PIP3), a potent activator of 3-phosphoinositide-dependent kinase (PDK) and AKT. PTEN is a nonredundant, plasma-membrane lipid phosphatase that can antagonize PI3K by dephosphorylating PIP3 at position D3 to generate PIP2. Numerous studies have demonstrated that the catalytic activity of PTEN is modulated by ROS, subsequently resulting in its catalytic inhibition [14,15]. Therefore, the oxidative modification of PTEN contributes to augmenting PIP3 levels, resulting in the activation of the PI3K/AKT signaling pathway and protecting cells from oxidative stress-induced cell death.

Endogenous antioxidants, such as thioredoxins (Trxs) and peroxiredoxins (Prxs), can modulate ROS levels and intracellular redox state. Thus, they have become targets for redox modifications [16]. Trxs can catalyze the reversible reduction of protein disulfide bonds using redox-active cysteine residues in their active site. Prxs are a family of thioldependent antioxidants that can scavenge cystolic or mitochondrial peroxides. Trx and Prx undergo redox-dependent modifications of catalytic amino acids that can affect protein functionality or impact subcellular protein targeting, protein-protein interactions, or protein stability. In this review, we focused on recent achievements and novel
A growing body of evidence has indicated that PTEN can exert part of its tumor suppressive functions by controlling cell migration, spreading, focal adhesions, and genomic stability, independent of its phosphatase activities [23,28,31]. For instance, microspherule protein 1 (MSP58)-mediated cellular transformation is inhibited by its physical interaction with the C-terminal domain of PTEN [32]. A recent study showed that cytosolic PTEN could suppress CHD-1 induced trimethyl lysine-4 histone H3 modification by stimulating chromodomain-helical-DNA-binding protein 1 (CHD1) proteasomal degradation, leading to the transcriptional activation of the TNF/NF-κB pathway [33]. Accordingly, in vivo genetic analyses have revealed that PTEN loss is not synonymous with AKT overexpression [34].

Recent studies revealed the existence of translational isoforms of PTEN, PTEN-Long (PTEN-L) and PTEN-β. Like canonical PTEN, PTEN-L is a membrane-permeable lipid phosphatase that is secreted from cells and can be taken up by other cells directly. PTEN-L, also localized in the mitochondria, can regulate mitochondrial functions and energy production by associating with canonical PTEN to increase PTEN-induced putative kinase 1 (PINK1) expression [35,36]. PTEN-β localizes in the nucleolus and negatively regulates ribosomal DNA transcription and ribosomal biogenesis by physically interacting with and dephosphorylating nucleolin [37]. With a high sequence homology to canonical PTEN, PTEN-L, and PTEN-β may be modulated by the same or similar mechanisms.

Disruptions in the regulation of PTEN by a range of molecular mechanisms can generate various dysfunctional PTEN species and/or a spectrum of PTEN levels that can variously contribute to the pathogenesis of inherited syndromes, including Cowden disease, Lhermitte-Duclos syndrome, Bannayan-Zonana Syndrome [38], cancers, and other diseases. These molecular mechanisms include the epigenetic loss and mutation of PTEN; transcriptional regulations; post-transcriptional regulation, including microRNA, the disruption of competitive endogenous RNA (ceRNA) networks, and long non-coding RNAs; post-translational modifications; and the aberrant localization of PTEN. PTEN function is also finely regulated through protein-protein interactions [28,39]. More recently, evidence has shown that PTEN is capable of forming dimers, which has been proven to be a novel mechanism for its functional regulation [40]. The following sections highlight our current understanding of the redox regulation of PTEN in cell biology.

3. Redox regulation of PTEN by peroxides, thioredoxin, and peroxiredoxin

3.1. Oxidative inactivation of PTEN by H2O2

PTEN is sensitive to oxidation because it contains nucleophilic cysteine residues in the active site. It has been reported that the catalytic
activity of PTEN is fine-tuned by the exposure to oxidizing agents in vitro or oxidative stress conditions in cells. The essential active Cys124 residue of human PTEN, surrounded by three basic amino acid residues in the active site pocket, is readily oxidized by forming an intramolecular disulfide with Cys71 [15], resulting in the inhibition of its phosphatase activity. Importantly, H$_2$O$_2$-mediated PTEN oxidation is reversible, which is predominantly reduced by thioredoxin. Conversely, cellular PTEN activity can be protected by the presence of ROS scavengers [41]. We have designed a convenient approach to monitor intra-PTEN disulfide using a mobility shift assay [15,42] (Fig. 2). In this procedure, all free cellular thiols and selenols of proteins are first blocked by alkylation with N-ethylmaleimide (NEM) and the alkylated PTEN confers a higher molecular weight. The proteins were then separated under non-reducing conditions in the presence of sodium dodecyl sulfate (SDS) and subjected to immunobLOTS using antibodies to PTEN. Differences in molecular weight and conformational structure, the oxidized and reduced forms of PTEN can then be visualized [15]. Treatment of HeLa cells with H$_2$O$_2$ resulted in PTEN oxidation in a time-dependent manner, with the maximal oxidation after 10 min of exposure. The oxidized PTEN was then converted to the reduced form, mostly after 120 min of exposure (Fig. 3). This clearly indicates that the oxidation reaction in the cells was reversible. In cells exposed to H$_2$O$_2$, the augmented oxidation of PTEN was accompanied by an elevation in the amount of cellular PIP3 [15] and the downstream activation of AKT through its phosphorylation, both on Ser473 (pAKT Ser473) and Thr308 (pAKT Thr308) (Figs. 3 and 4A). Phosphorylation of both Thr308 and Ser473 residues is required for the full activation of AKT. H$_2$O$_2$-dependent increases in PIP3 abundance and Akt activity were correlated with the oxidative inactivation of PTEN in PTEN-expressing glioma cells, but not in PTEN-null glioma cells, further supporting the notion that oxidative stress regulates PI3K-dependent signaling through the oxidative inhibition of PTEN [43–45].

**Catalytic activity of PTEN is modulated by thioredoxin.** Oxidation-driven PTEN inactivation can be reversed by cellular reducing systems, among which the thioredoxin (Trx) system plays a major role. Trx plays an important role in PTEN activation after H$_2$O$_2$ treatment. It has been demonstrated that Trx is even more efficient than glutathione or glutaredoxin in the reduction of oxidized PTEN [15]. Trx is a ubiquitous protein present in all species from archaeabacteria to human. Functions of Trx have been subjected in many investigations [46]. Primarily, the reduced form of Trx serves as a general protein thiol-disulfide oxidoreductase. The evolutionarily conserved Trp-Cys$_{32}$-Gly-Pro-Cys$_{35}$ catalytic center provides thiol nucleophiles involved in Trx-dependent reducing activity. The N-terminal cysteine of the reduced form Trx initiates a nucleophilic attack on the target protein disulfide and forms a transient mixed disulfide which is subsequently attacked by the C-terminal cysteine of Trx to generate the reduced target protein and oxidized Trx with a disulfide bond. Such oxidized Trx is recycled by Trx reductase (TrxR), which utilizes NADPH as an electron donor [47]. Thus, the Trx system is comprised of Trx, selenoenzyme TrxR, and NADPH [48,49], known to have pivotal roles in the regulation of redox signaling via maintaining the thiol-related redox status balance.

Previous studies have revealed that E. coli and human Trxs act as monomers [50–52]. It has been observed that Trxs derived from vertebrates are more susceptible to oxidation that forms dimers or oligomers via Cys residues [50]. Except for the formation of an active Cys$_{55}$-Cys$_{55}$ disulfide, a non-active Cys$_{62}$-Cys$_{69}$ disulfide bond, dimers, and multimers are also formed under long exposure to air or high levels of H$_2$O$_2$ and diamide [50,51,53,54]. Crystallographic studies have revealed that dimerization through an intermolecular disulfide bond between Cys$_{53}$ and Cys$_{55}$ of two monomers renders Trx unable to carry out its redox activity compared to the crystallized Trx-TrxR complex [50,55]. The homodimerization of Trx started to accumulate 5 min after H$_2$O$_2$ treatment and lasted until 60 min of incubation in HeLa extracts (Fig. 4B). However, the Trx dimer disappeared after 120 min of incubation, showing a similar trend in the reduction kinetics of H$_2$O$_2$-oxidized Trx.

**Fig. 2.** Experimental scheme for monitoring the redox status of PTEN by mobility shift. N-ethylmaleimide (NEM) reacts with reduced PTEN thiols resulting in PTEN-NEM adducts, which increase the protein mass. This additional molecular weight leads to a slower shift on non-reducing SDS-PAGE, generating an additional upper band on the gel. In contrast, if PTEN thiols are oxidized to form a disulfide bond, they will not react with NEM and mobility shift will be observed. This approach allows determination of the ratio between the oxidized (intra-PTEN disulfide, inactive) and reduced (PTEN-red, active) forms of PTEN after exposure to oxidants.

**Fig. 3.** H$_2$O$_2$-induced PTEN reversible oxidation and AKT phosphorylation in HeLa cells. (A) Serum-starved HeLa cells were exposed to 1 mM H$_2$O$_2$ for various times. All samples were alkylated with 10 mM NEM and subjected to non-reducing or reducing SDS-PAGE, followed by immunoblot analysis with PTEN, p-AKT (Ser473), p-AKT (Thr308), total AKT or actin antibodies. (B) The intensity of the oxidized PTEN bands and p-AKT bands from (A) was quantitated using ImageJ software. Red, reduced PTEN; Oxi, oxidized PTEN.
oxidized PTEN. This suggests the H\textsubscript{2}O\textsubscript{2}-induced production of Trx dimers in response to elevated levels of oxidized PTEN. Dimeric Trx has long been considered as a potential cellular regulatory redox signaling molecule that might be a possible target for the development of anticancer drugs [56]. Our studies have reported that cellular Trx is one of the targets of organic hydroperoxides that can induce Trx dimerization and oligomerization, causing the irreversible oxidation of PTEN [49,57].

Trx is overexpressed in many human cancers. It is associated with increased tumor cell proliferation, inhibited apoptosis, and decreased patient survival [58,59]. It has been previously reported that increased levels of Trx1 can bind to PTEN in a redox-dependent manner to inhibit its PtdIns-3-phosphatase activity, resulting in increased Akt activation in cells [60]. The interaction between Trx-1 and PTEN occurs through a disulfide bond between the active site Cys \textsuperscript{51} of Trx-1 and Cys \textsuperscript{172} of PTEN, inhibiting PTEN’s lipid phosphatase activity and increasing tumorigenesis [60]. This provides an additional mechanism for tumorigenesis with a loss of PTEN activity. However, the inactivation of PTEN by Trx can be reversed in the presence of thioredoxin-interacting protein (Txnip). Upon Txnip interaction with Trx through a disulfide bond between Cys \textsuperscript{247} of Txnip and Cys \textsuperscript{172} of Trx, Trx is no longer able to bind and allow for reactivation PTEN [61].

Catalytic activity of PTEN is modulated by peroxiredoxin.

Peroxiredoxins (Prxs) are a ubiquitously expressed family of small non-seleno peroxidases (22–27 kDa) that can catalyze reduction of H\textsubscript{2}O\textsubscript{2}, organic hydroperoxides, and peroxynitrite using reducing equivalents provided by thiol-containing proteins [62]. Mammalian cells possess six isoforms of Prx. They can be classified into three subgroups: four 2-Cys Prx isoforms (Prx1-4), one atypical 2-Cys Prx isoform (Prx5), and one 1-Cys Prx isoform (Prx6) [62]. Although these six mammalian Prx isoform have different individual functions in cellular redox regulation and antioxidant protection, they all can enable to regulate intracellular H\textsubscript{2}O\textsubscript{2} levels by catalyzing peroxide reduction for signaling and metabolism [63]. All mammalian Prx enzymes are homodimers arranged in a head-to-tail orientation. They contain a conserved cysteine residue in the N-terminal region that is the primary site of the oxidation of H\textsubscript{2}O\textsubscript{2}. In the catalytic cycle of 2-Cys Prx proteins, the conserved C\textsubscript{p}-SH (Cys \textsuperscript{51} in Prx1) is selectively oxidized to C\textsubscript{p}-SO\textsubscript{2}H intermediate at low levels of H\textsubscript{2}O\textsubscript{2}. After reacting with the C-terminal-conserved C\textsubscript{p}-SH (Cys \textsuperscript{172} in Prx1) of the other subunit in the homodimer, an intermolecular disulfide is generated. Ultimately, it is specifically reduced by Trx [62,64]. Under normal cellular homeostasis with low H\textsubscript{2}O\textsubscript{2}, rather than displaying peroxidase activity, predominantly low-molecular weight oligomeric 2-Cys Prxs can also protect proteins from degradation. With higher doses of H\textsubscript{2}O\textsubscript{2}, C\textsubscript{p}-SO\textsubscript{2}H generated as an intermediate during catalysis, occasionally undergoes further oxidation to C\textsubscript{p}-SO\textsubscript{3}H catalysis in the presence of Trx, resulting in the inactivation of its peroxidase activity. The generated C\textsubscript{p}-SO\textsubscript{3}H can be reduced back to thiol by sulfiredoxin (Srx) [65,66]. However, further oxidation to C\textsubscript{p}-SO\textsubscript{4}H is irreversible, resulting in Prx degradation [65]. Recently, it has been reported that when Cys \textsuperscript{51} in Prx1 was in an overoxidized form due to oxidative stress or heat shock stress, the formation of high molecular weight complexes was favored [67]. The structural change from low molecular weight oligomer to high molecular weight complexes is accompanied by a functional change from peroxidase to molecular chaperone activity [68,69]. The peroxidase function of Prx can be regulated by various post-translational modifications, including phosphorylation, lysine acetylation, glutathionylation, nitrosylation, and thiol oxidation [70–74].

It has been reported that Prx1 can protect and promote PTEN tumor suppressive function by interacting with PTEN and protecting disulfide bond formation under mild oxidative stress [75]. The lipid phosphatase activity of PTEN was fully preserved by Prx1 in cells under low concentrations of H\textsubscript{2}O\textsubscript{2} (25 μM), where Prx1 was found to interact with PTEN. However, under higher concentrations of H\textsubscript{2}O\textsubscript{2} (500 μM), Prx1 was irreversibly hyperoxidized and dissociated from PTEN [75]. Prx1 preserved PTEN lipid phosphatase activity under oxidative stress at a 1:1 (mol:mol) ratio of Prx1 and PTEN, and could not be further enhanced by excess Prx1, indicating that Prx1 most likely interacts with PTEN as a monomer [75]. Mutational analysis and computational analysis suggested that Prx1 interacts within the C2 domain of PTEN (aa 186–274) and PTEN with the N-terminal of Prx1 (aa 1–21) and the C-terminal of Prx1 (aa 183–199) [75]. Hyperoxidized Prx was observed when cells were exposed to H\textsubscript{2}O\textsubscript{2} for various times using an antibody
specific against cysteine sulfenic acids (Fig. 5A). The basal Prx1 dimer was readily observed in H$_2$O$_2$-untreated HeLa cells because Prxs are predominantly obligate dimers at the time of lysis in the absence of NEM. Prx1 hyperoxidation was increased maximally after 5 min of exposure and then decreased progressively over time (Fig. 5B). In addition, augmentation with oxidized PTEN induced by H$_2$O$_2$ showed kinetics similar to H$_2$O$_2$-induced Prx1 hyperoxidation (Fig. 5C). The temporary inhibition of Prx by hyperoxidation impaired H$_2$O$_2$-scavenging activity.

Inactivated Trx dimerization resulted in a delay in the reduction of PTEN and Prx1. However, whether Prx1 reduced the H$_2$O$_2$-induced intramolecular disulfide bond in PTEN is still unclear. Consequently, the redox regulation of PTEN by H$_2$O$_2$ is mediated through Trx and Prx systems in cell signaling.

### 3.2. Oxidative inactivation of PTEN by organic hydroperoxides

Organic peroxides and hydroperoxides are tumor promoters [76]. The tumor promoting activity of organic hydroperoxides involves in generation of free radical derivatives [77–79]. Cumene hydroperoxide (CuHP) is a stable organic hydroperoxide with oxidizing capabilities [80]. CuHP can induce lipid peroxidation by reacting with adjacent fatty acid side-chains in the presence of transition metal, resulting in continuous generation of lipid hydroperoxides via chain reactions [81,82]. These lipid hydroperoxides, in turn, can generate several ROS, such as alkoxy and peroxy radicals in vitro and in vivo, thereby exerting various types of oxidative damage, including lipid peroxidation, protein modification, and DNA damage [80,83,84]. Tert-butyl hydroperoxide (t-BHP) is extensively metabolized in target issues to form several free radical intermediates, including phenoxyl, peroxy, alkoxy, and alkyl radical derivatives in murine keratinocytes [80], hemoglobin-thiyl and methyl radicals in rat liver stomach [85]. The cytotoxic effects of t-BHP are involved in glutathione depletion [86], hemoglobin oxidative denaturation, hemolysis and erythrocyte membrane lipid peroxidation [87,88], inner mitochondrial membrane permeabilization [89], DNA single strand breakage [90,91], and apoptosis [76]. Stimulation of HeLa cells or recombinant PTEN with either CuHP or t-BHP resulted in PTEN oxidation by forming an intramolecular disulfide between Cys124 and Cys71 in time- and concentration-dependent manners [49,57]. However, these organic hydroperoxides-mediated cellular PTEN oxidation was not reversible, because the cellular Trx was inactivated by dimerization [49,57]. In addition, the ablation of Prx enhanced CuHP-induced PTEN oxidation in MEF cells [49]. Overall, these results imply that PTEN is an important physiological target for organic peroxide-mediated redox signaling. Its irreversible oxidation could play a key role in organic peroxide-induced tumorigenesis.
3.3. Oxidative inactivation of PTEN by lipid peroxide

Lipoxygenases (LOX) can catalyze the production of hydroperoxyeicosatetraenoic acid (HpETE) from arachidonic acid (AA). 15-LOX can metabolize AA to form 15(s)-hydroperoxyeicosatetraenoic acid (15s-HpETE), the oxidative precursor of 15-hydroxyeicosatetraenoic acid (15s-HETE). Human 15-LOX has two isoforms, 15-LOX-1 and 15-LOX-2. 15-LOX-1 is a dual-specificity enzyme that metabolizes AA, principally to 15s-HpETE, and to far smaller amounts of 12s-HpETE. 15-LOX-2 can metabolize AA to 15s-HpETE. It has little or no ability to metabolize AA to 12s-HpETE. These HpETEs are subsequently reduced and transformed to produce eicosanoids known to be important signaling molecules in immune responses and other physiological processes. Increased levels of lipid peroxides have been linked to the pathogenesis of a variety of human diseases, including neurodegeneration, atherosclerosis, type II diabetes, metabolic disorders, solid tumors, and hematologic malignancies, through cellular oxidative damage [92–95]. The ability of lipid peroxides to oxidize PTPs has been reported [96]. PTEN oxidation by unidentified AA metabolites has also been shown [97]. Both 15s-HpETE [98] and 12s-HpETE (Fig. 6) resulted in PTEN oxidation in MEF cells, and Prx III deletion aggravated the 12/15s-HpETE-induced PTEN oxidation in MEF cells. 15s-HpETE-mediated cellular PTEN oxidation was identical to that of H2O2, indicating that an intramolecular disulfide bond between Cys124 and Cys71 was formed [98]. However, 15s-HETE was unable to induce PTEN oxidation in MEF cells [98].

3.4. S-nitrosylation of PTEN by nitric oxide

PTEN is also modified by S-nitrosylation [99–101], a covalent modification of cysteine residues by nitric oxide (NO), which is another crucial redox mechanism that regulates PTEN activity. S-nitrosylation and H2O2-mediated oxidation occur on distinct Cys residues of PTEN. A recent study has shown that NO can induce PTEN S-nitrosylation on Cys83, leading to the suppression of lipid phosphatase activity of PTEN and induction of PTEN protein degradation via the ubiquitin-proteasome system (UPS) through NEDD4-1-mediated ubiquitination [102,103]. Involvement of PARK2 encoding ubiquitin E3 ligase Parkin in PTEN S-nitrosylation has been reported [104]. Depletion of PARK2 enhanced the 5-AMP-activated protein kinase (AMPK)-mediated activation of endothelial nitric oxide synthase (eNOS), leading to increases in NO levels that drove S-nitrosylation and subsequent ubiquitination of PTEN [100]. These findings suggest that the S-nitrosylation of PTEN could serve as a possible new therapeutic target.

4. Concluding remarks

The reversible oxidation of Cys residues in proteins upon cellular oxidants is linked to signaling events. PTEN was discovered as a bona fide tumor suppressor. The importance of its functions in the regulation of cell growth, motility, and the inhibition of apoptosis has been well-established. ROS has been recognized as a secondary messenger that can modify cell signaling by oxidizing protein cysteine thiols.
Challenges have moved from PTEN pleiotropic natural functions toward understanding its regulation. This review concentrated on the redox regulation of PTEN, which is crucially linked to its tumor suppressor function.

Trx and Prx play vital roles in the control of intracellular redox state of PTEN. The participation of H$_2$O$_2$ in intracellular signaling by targeting PTEN, Trx, and Prx, and the regulation of H$_2$O$_2$ concentrations by Prx are depicted schematically in Fig. 7. Growth factors induced the activation of PI3K, resulting in the conversion of PI2P to PI3P. PI3P produced the induction of H$_2$O$_2$ by activating the NOX (Nicotinamide adenine dinucleotide phosphate oxide) complex. The interaction of Prx1 and PTEN is essential for protecting PTEN from oxidation-induced inactivation. PTEN lipid phosphorylation activity was fully protected by Prx1 in cells under low H$_2$O$_2$ exposure, where Prx1 was found to bind PTEN as a monomer [75]. The generated H$_2$O$_2$ mediated the inactivation of cytosolic Prx molecules located nearby through forming intramolecular disulfides. The disulfides were subsequently and specifically reduced by Trx, which in turn received reducing equivalents from NADPH via TrxR. However, when treated with higher concentrations of H$_2$O$_2$, Prx1 is known to become irreversibly hyperoxidized and dissociate from PTEN. H$_2$O$_2$ can inactivate Prx in a two-step peroxidation of the active site Cys-SH to Cys-SO$_2$H, which can be reactivated via an ATP-dependent reduction catalyzed by sulfiredoxin (Srx). Prx inactivation allows the accumulation of local H$_2$O$_2$, which in turn promotes the inactivation of PTEN by forming a disulfide bond. In addition to impaired activity of Prx, the intermolecular disulfide/dimerization of Trx also occurs, causing a loss of Trx reactivity, which in turn, promotes the oxidation of PTEN. This inactivation of PTEN increases the abundance of phosphorylated AKT and sufficiently triggers downstream signaling events. The impaired activity of Trx caused by dimerization provides a mechanism by which Trx activity is transiently inhibited under the conditions of oxidative stress, providing more time for sensing and transmission of oxidative signals. However, further studies are needed to reveal the mechanisms that involved in Trx transient inhibition and the important roles of Trx and Prx in redox signaling closely associated with PTEN redox regulation.

Declaration of competing interest

The author has no competing interests to declare.

Acknowledgments

The present study was supported by the grants (2018R1D1A1B06051438 and NRF-2015R1D1A1A01059571) from the National Research Foundation, Republic of Korea. Jiyoung Park was supported by the Health Fellowship Foundation.

References

[1] C.C. Winterbourn, Reconciling the chemistry and biology of reactive oxygen species, Nat. Chem. Biol. 4 (5) (2008) 278–286.
[2] L. Zhang, X. Wang, R. Cueto, C. Effi, Y. Zhang, H. Tan, X. Qin, Y. Ji, X. Yang, H. Wang, Biochemical basis and metabolic interplay of redox regulation, Redox Biol. 26 (2019) 101284.
[3] S.G. Rhee, S.W. Kang, W. Jeong, T.S. Chang, K.S. Yang, H.A. Woo, Intracellular hydrogen peroxide, J.Am.Soc.Nephrol.:JASN (J.Am.Soc.Nephrol.) 14 (8 Suppl 3) (2003) S211–S215.
[4] S.G. Rhee, Redox signaling: hydrogen peroxide as intracellular messenger, Exp. Mol. Med. 31 (2) (1999) 53–59.
[5] S.G. Rhee, Redox signaling: hydrogen peroxide as intracellular messenger, Exp. Mol. Med. 31 (2) (1999) 53–59.
[6] A. Corcoran, T.G. Cotter, Redox regulation of protein kinases, FEBS J. 280 (9) (2013) 1944–1965.
[7] R.T. Dean, S. Pu, R. Stocker, M.J. Davies, Biochemistry and pathology of radical-mediated protein oxidation, Biochem. J. 324 (Pt 1) (1997) 1–18.
[8] L.J. Yan, R.S. Solahi, Mitochondrial adenine nucleotide translocase is modified oxidatively during aging, Proc. Natl. Acad. Sci. U.S.A. 95 (22) (1998) 12896–12901.
[9] D.P. Jones, Redefining oxidative stress, Antioxidants Redox Signal. 8 (9–10) (2006) 1865–1879.
[10] C.K. Roberts, K.K. Sindhu, Oxidative Stress and metabolic syndrome, Life Sci. 84 (21–22) (2009) 705–712.
[11] T. Finkel, Oxidant radicals and signaling,Curr. Opin. Cell Biol. 10 (2) (2000) 248–253.
[12] S.G. Rhee, Y.S. Bae, S.R. Lee, J. Kwon, Hydrogen peroxide: a key messenger that modulates protein phosphorylation through cysteine oxidation, Sci. STKE : Signal Transduct, Knowl. Environ. 53 (2000) (2000) pe1.
[13] V.J. Thannickal, B.L. Fanburg, Reactive oxygen species in cell signaling, Am. J. Physiol. Lung Cell Mol. Physiol. 279 (6) (2000) L1005–L1028.
[14] J. Kwon, S.R. Lee, K.S. Yang, Y. Ahn, Y.J. Kim, E.R. Stadman, S.G. Rhee, Reversible oxidation and inactivation of the tumor suppressor PTEN in cells stimulated with peptide growth factors, Proc. Natl. Acad. Sci. U.S.A. 101 (47) (2004) 16419–16424.
[15] S.R. Lee, K.S. Yang, J. Kwon, C. Lee, W. Jeong, S.G. Rhee, Reversible inactivation of the tumor suppressor PTEN by H2O2, J. Biol. Chem. 273 (22) (2008) 20336–20342.
[16] H. Sies, C. Berndt, D.P. Jones, Oxidative stress, Annu. Rev. Biochem. 86 (2017) 715–748.
[17] J. Li, C. Yen, D. Liaw, K. Podesypina, S. Bao, S.I. Wang, J. Puc, M. Ciliareis, L. Rodgers, R. McCombie, S.H. Bigner, B.C. Giovannella, M. Ittmann, B. Tycko, H. Hibshoosh, M.H. Wigler, R. Parsons, PTEN, a putative protein tyrosine phosphatase gene mutated in human brain, breast, and prostate cancer, Science (New York, N.Y.) 275 (5308) (1997) 1943–1947.
[18] S. Funamoto, R. Meili, S. Lee, L. Parry, R.A. Firtel, Spatial and temporal regulation of 3-phosphoinositides by PI3-kinase and PTEN mediates chemotaxis, Cell 109 (2002) 611–623.
[19] T. Maehama, G.S. Taylor, J.H. Dixon, PTEN and myotubularin: novel phosphoprotein phosphatases, Annu. Rev. Biochem. 70 (2001) 247–279.
[20] J. Luo, B.D. Manning, L.C. Cantley, Targeting the PI3K-Akt pathway in human cancer: rationale and promise, Canc. Cell 4 (4) (2003) 257–262.
[21] V. Stambolic, A. Suzuki, J.L. de la Pompa, G.M. Brothers, C. Mirtsos, T. Sasaki, H. Wang, Biochemical basis and metabolic interplay of redox regulation, Redox Biol. 26 (2019) 101284.
and 15-(S)-HETE on chronic myelogenous leukemia cell line K-562: reactive oxygen species (ROS) mediate caspase-dependent apoptosis, Biochem. Pharmacol. 74 (2) (2007) 202–214.

[95] D. Trachootham, J. Alexandre, P. Huang, Targeting cancer cells by ROS-mediated mechanisms: a radical therapeutic approach? Nat. Rev. Drug Discov. 8 (7) (2009) 579–591.

[96] M. Conrad, A. Sandin, H. Forster, A. Seiler, J. Frijhoff, M. Dagnell, G.W. Bornkamm, O. Radmark, R. Hooft van Huijsduijnen, P. Aspenstrom, F. Bohmer, A. Ostman, 12/15-lipoxygenase-derived lipid peroxides control receptor tyrosine kinase signaling through oxidation of protein tyrosine phosphatases, Proc. Natl. Acad. Sci. U.S.A. 107 (36) (2010) 15774–15779.

[97] T.M. Covey, K. Edes, F.A. Fitzpatrick, Akt activation by arachidonic acid metabolism occurs via oxidation and inactivation of PTEN tumor suppressor, Oncogene 26 (39) (2007) 5784–5792.

[98] Y. Zhang, J. Park, S.J. Han, Y. Lim, I. Park, J.S. Kim, H.A. Woo, S.R. Lee, Peroxiredoxin III protects tumor suppressor PTEN from oxidation by 15-Hydroperoxy-eicosatetraenoic acid, Oxid. Med. Cell. Longev. 2019 (2019) 2828493.

[99] C.X. Yu, S. Li, A.R. Whorton, Redox regulation of PTEN by S-nitrosothiols, Mol. Pharmacol. 68 (3) (2005) 847–854.

[100] A. Gupta, S. Anjomani-Virmouni, N. Koundouros, M. Dimitriadi, R. Choo-Wing, A. Valle, Y. Zheng, Y.H. Chiu, S. Agnihotri, G. Zadeh, J.M. Asara, D. Anastasiou, M.J. Arends, L.C. Cantley, G. Poulogiannis, PARK2 depletion connects energy and oxidative stress to PI3K/Akt activation via PTEN S-nitrosylation, Mol. Cell 65 (6) (2017) 999–1013 e7.

[101] L. Zha, L. Li, Q. Zhang, X. Yang, Z. Zou, B. Hao, F.M. Marincola, Z. Liu, Z. Zhong, M. Wang, X. Li, Q. Wang, K. Li, W. Gao, K. Yao, Q. Liu, NOS1 S-nitrosylates PTEN and inhibits autophagy in nasopharyngeal carcinoma cells, Cell Death Discov. 3 (2017) 17011.

[102] Y.D. Kwak, T. Ma, S. Diao, X. Zhang, Y. Chen, J. Hsu, S.A. Lipton, E. Masliah, H. Xu, F.F. Liao, NO signaling and S-nitrosylation regulate PTEN inhibition in neurodegeneration, Mol. Neurodegener. 5 (2010) 49.

[103] N. Numajiri, K. Takasawa, T. Nishiyama, K. Ohno, W. Hayakawa, M. Arada, H. Matuda, K. Azumi, H. Kamata, T. Nakamura, H. Hara, M. Minami, S.A. Lipton, T. Uehara, On-off system for PI3-kinase-Akt signaling through S-nitrosylation of phosphatase with sequence homology to tensin (PTEN), Proc. Natl. Acad. Sci. U.S.A. 108 (25) (2011) 10349–10354.

[104] A. Gupta, S. Anjomani-Virmouni, N. Koundouros, G. Poulogiannis, PARK2 loss promotes cancer progression via redox-mediated inactivation of PTEN, Mol. Cell. Oncol. 4 (6) (2017) e1329692.

[105] U. Putz, S. Mah, C.P. Goh, L.H. Low, J. Howitt, S.S. Tan, PTEN secretion in exosomes, Methods (San Diego, Calif.) 77–78 (2015) 157–163.