— **Review** —

Biological Functions of Protein Disulfide Isomerase as a Target of Phenolic Endocrine-disrupting Chemicals

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Endocrine disrupting chemicals (EDCs) are widespread in the environment and suspected of interfering with endocrine homeostasis. Bisphenol A (BPA), 2,2-bis(4-hydroxyphenyl)propane, an EDC, has been widely used throughout the world as an industrial chemical. BPA is a developmental, neural, and reproductive toxicant that mimics estrogen and can interfere with growth and body function. In a recent study, a BPA-binding protein was isolated from P2 fractions of rat brain, and identified as protein disulfide isomerase (PDI). PDI has been studied extensively as a key enzyme involved in the formation of the correct pattern of disulfide bonds in proteins. PDI is also known to be a membrane-associated 3,3′,5-triiodo-L-thyronine (T_3)-binding protein. BPA inhibits the binding of T_3 to PDI and isomerase activity of PDI. PDI has four consecutive domains, **a**, **b**, **b'**, and **a'**, each with a thioredoxin fold. The domains **a** and **a'** have a catalytically active Cys-Gly-His-Cys motif, while **b** and **b'** have substrate-binding sites. To address the chemical structural requirements of BPA for the inhibitory effect of PDI, several BPA analogs were tested, suggesting that the phenolic structure is important for BPA to affect PDI functions. To investigate the biological effects of BPA on PDI functions, GH3 cells were used. GH3 is a rat pituitary tumor cell line and releases growth hormone (GH) via the T_3 receptor. Over-expression of PDI suppresses the T_3-induced release of GH and when the cells were treated with BPA together with T_3, the amount of GH released was much increased. Thus, PDI plays an important role in the hypothalamic-pituitary-thyroid axis. In this review, the possible involvement and biological significance especially in the central nervous system, of PDI as a target of phenolic environmental chemicals, are discussed.

Key words —— endocrine-disrupting chemical, bisphenol A, protein disulfide isomerase, thyroid hormone, protein folding, chaperone

INTRODUCTION

Tens of thousands of man-made chemicals have been introduced into the environment in the last few decades. Some of these compounds are considered to disrupt endocrine homeostasis in humans and wild life.[1] These chemicals, distinguished as endocrine disrupting chemicals (EDCs), bridge many chemical classes and are an integral part of the world economy and commerce. One such chemical, bisphenol A (BPA), is produced mainly as a monomer of polycarbonate plastics. A recent study demonstrated that exposure to BPA in the neonatal or lactational period affected the development of brain.[2–5] Protein disulfide isomerase (PDI, EC 5.3.4.1) was recently identified as a target of BPA. PDI catalyzes thiol-disulfide exchange, thus facilitating the formation and rearrangement of disulfide bonds. By *in vitro* investigation, it was revealed that BPA binds to PDI competitively with 3,3′,5-triiodo-L-thyronine (T_3), and inhibits isomerase activity of PDI.

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BPA

BPA, 2,2-bis(4-hydroxyphenyl)propane, has been widely used throughout the world as an industrial

chemical. This chemical is now utilized in hard, polycarbonate plastics, as well as the epoxy resins used in the linings of food and beverage containers, dental sealants and numerous other consumer products.[6–8] BPA is a developmental, neural, and reproductive toxicant that mimics estrogen and can interfere with growth and body function. Experiments with animals demonstrated that BPA affects the reproductive, neurological and immune systems during critical stages of development. Exposure of rodent fetuses to BPA at a very low dose was found to produce postnatal estrogenic effects: reduced sperm numbers in males, increased prostate gland weight, changes in the development and tissue organization of the mammary gland, disruption of sexual differentiation in the brain, long-term deleterious effects in the vagina, and accelerated growth and puberty in females.[9–15]

EFFECTS OF BPA ON THE CENTRAL NERVOUS SYSTEMS

Recent studies have linked mental conditions ranging from cognitive impairment to autism, to environmental pollutants. Prenatal and neonatal exposure of mice to BPA resulted in impaired development of the brain.[16] BPA also caused aggressive behavior.[17] Chronic exposure of BPA enhanced dopamine D1 receptor-dependent rewarding effects and hyperlocomotion induced by methamphetamines.[5] Dams exposed to BPA either as fetuses or in adulthood exhibited altered maternal behavior.[18] Exposure to BPA in *Xenopus laevis* disrupted Notch signaling.[19, 20] These findings suggest prenatal and neonatal exposure to BPA may cause neuronal toxicity during embryonic development. On the other hand, non-prenatal or non-neonatal effect of EDCs have also recently raised great concern. BPA induced the release of dopamine in a non-genomic manner through guanine nucleotide binding proteins (G-protein) involving the activation of ryanodine-sensitive calcium stores or N-type calcium channels in a rat pheochromocytoma cell line, PC12.[21] At nanomoler concentrations of BPA, a temporary increase in Ca^{2+} was induced in *N*-methyl-D-aspartate (NMDA)-responsive neurons of cultured rat hippocampal neurons in culture.[22] Excessive activation of NMDA receptors drives Ca^{2+} influx, which in turn activates neuronal NO synthase (nNOS) as well as the generation of reactive oxygen species (ROS).[23, 24] It is currently thought that over stimulation of extrasynaptic NMDA receptors mediates this neuronal damage.

PDI

BPA-binding protein was recently isolated from P2 fractions of rat brain using a BPA-Sepharose column,[25] and identified as PDI. It has been demonstrated that BPA inhibits binding of PDI to T_3 *in vitro* (Fig. 1).[25–27] Protein disulfide isomerase was the first endoplasmic reticulum (ER) oxidoreductase to be identified and characterized over 40 years ago.[28] PDI is a multifunctional protein, and its major role is to assist in the folding of proteins containing disulfide bonds. PDI forms disulfide in two ways, oxidation and isomerization (Fig. 2). The oxidation is a simple reaction, generating disulfide from two thiols. The isomerization includes the reduction of disulfide, refold-

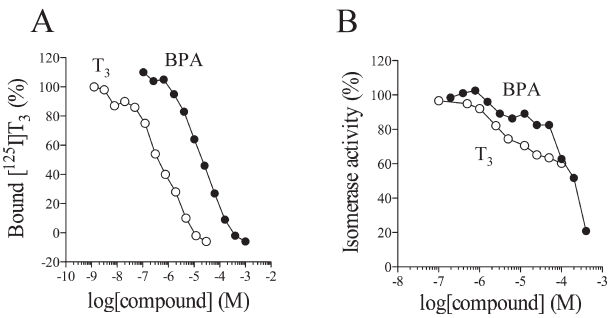

Fig. 1. Inhibition of T_3-binding to PDI by T_3 and BPA (A) and Inhibition of the Isomerase Activity of PDI by T_3 and BPA (B)

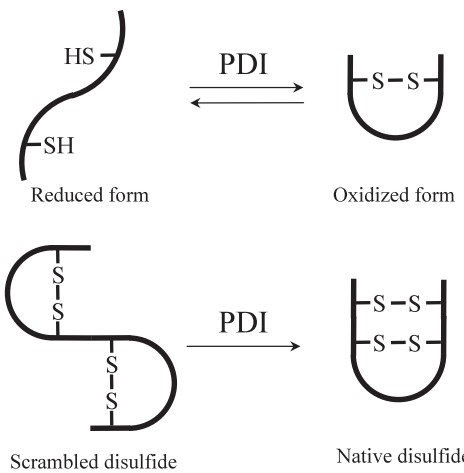

Fig. 2. Oxidation and Isomerase Reactions Catalyzed by PDI

ing, and oxidation of thiols (formation of disulfide). PDI has been studied extensively as a key enzyme involved in the formation of the correct pattern of disulfide bonds in proteins. It is clear that PDI is capable of both the formation and isomerization of disulfide bonds *in vitro*,[29] in yeast[30] and in mammalian systems.[31] PDI is an abundant protein primarily located in the lumen of the ER, and found in all eukaryotic cells.[32] Its presence in the nucleus and on the cell surface[33] suggests additional functions. In addition to having catalytic activity, PDI constitutes the noncatalytic subunit of prolyl 4-hydroxylase (P4H, EC 1.14.11.2)[34,35] and microsomal triglyceride transfer protein (MTP).[36–38] P4H catalyzes the formation of 4-hydroxyproline in collagens and other proteins with collagen-like amino acid sequences by the hydroxylation of proline residues in peptide linkages. In most species, P4H complexes are $\alpha 2\beta 2$ tetramers with PDI representing the β subunits and catalytic activity residing in the α subunits.[39] The role of PDI in P4H complexes is independent of its catalytic activity since active site mutants of PDI can form fully active P4H tetramers.[40,41] PDI functions within the P4H complex to keep the active subunits soluble and in a catalytically active conformation. MTP is a heterodimer containing subunits of PDI and 97-kDa protein.[37] MTP catalyzes the transfer of triglyceride, cholesteryl ester, and phosphatidylcholine between membranes.[42] PDI is necessary to maintain the catalytically active form of MTP and prevent the aggregation of MTP.[37] PDI is also known to be a membrane-associated T_3-binding protein and exhibits chaperone-like activity. PDI has been shown to bind peptides, proteins, and hormones.[43–46] PDI as a thyroid hormone-binding protein was identified as single 55-kDa protein found to be a major T_3-binding protein using an affinity labeling reagent, *N*-bromoacetyl-[^{125}I]T_3, in various cell types[47,48] and in various cellular compartments such as plasma membrane fractions and in the lumen of the endoplasmic reticulum.[48–50] The cloning and sequencing of its cDNA[51,52] revealed similarity to PDI, as experimentally demonstrated later on the basis of enzymatic assays.[44,53] PDI also binds β-estradiol (E_2) as well as T_3. A short segment of the PDI sequence exhibits some similarity to the estrogen receptor, but this sequence does not appear to constitute an independent structural domain.[54] The human PDI segments 120–163 and 182–230 have significant similarity with the estrogen receptor segments 350–392 and 304–349, respectively, located in the estrogen-binding domain. It has been suggested that E_2 binds to this sequence of PDI. A recent report suggested that there are at least three binding sites in PDI,[46] with one site involved in binding protein/peptides, and the remaining two in binding hormones. However, the hormone-binding site of PDI is not understood. The demonstration that BPA had inhibitory effects on T_3 and E_2 binding to PDI implies that BPA could potentially displace T_3 and E_2 from PDI.[25]

THE DOMAIN STRUCTURE OF PDI

PDI has five consecutive domains each with a thioredoxin fold (Fig. 3).[55] Domains **a** and **a'** have a catalytically active Cys-Gly-His-Cys motif, while **b** and **b'** domain do not have catalytic activity. The order of the domains is **a**-**b**-**b'**-**x**-**a'**-**c**, where **x** is a linker region between **b'** and **a'**[56] and **c** is a C-terminal acidic α-helix containing the ER retention signal.[57,58] Substrates of PDI are recognized by the **b'** domain, which contains the peptide-binding region, and reactions are catalyzed by catalytic cysteine residues of **a** and **a'**.[57,59] The **b'** domain is the principal substrate-binding site with specific mutations in a small hydrophobic pocket affecting binding.[56] Ile_{272} in the **b'** domain of human PDI has been identified as crucial to the binding of substrates.[59] While simple isomerization can be accomplished with one catalytic domain in combination with the **b'** domain, where the isomerization is linked to a significant change in the substrate's structure, all domains are required for full enzymatic activity.[60] Additionally, the **b** domain and **x** region, which is a flexible linker, stabilize the structure of **b'**.[61,62] The **c** domain is a putative Ca^{2+}-binding region and also reported to play a critical role in stabilization of the functional conformation of PDI under extreme conditions.[63,64] The specific order of the domains within PDI allows for the dual activity of this enzyme, as a disulfide isomerase and a disulfide oxidase, by establishing an asymmetry in

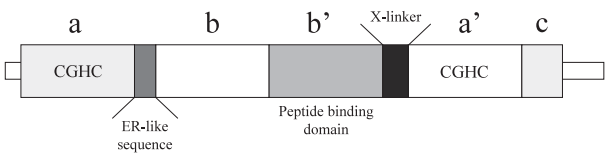

Fig. 3. Domain-based Structure of PDI

oxidation rates between the two active sites.[65]

PROTEIN-FOLDING ACTIVITY OF PDI

For isomerase reactions (reduction of disulfide, refolding, and oxidation of thiols), PDI requires four thioredoxin domains linked in tandem with a C-terminal anionic tail. The two catalytic domains (**a** and **a'**) are located in the N- and C-terminal thioredoxin domains, separated by the two noncatalytic thioredoxin domains (**b** and **b'**). The structure of PDI reveals a C-shaped molecule arranged about the noncatalytic domains, similar to that observed for the prokaryotic isomerase DsbC.[57] Moreover, the multidomain structure of PDI is not really necessary for the simple oxidation of thiols (disulfide formation). The individual **a** and **a'** domains each exhibit 50% of the dithiol-oxidizing ability of full-length PDI. To catalyze isomerization, the reduced (dithiol) form of PDI is also required. Thus, PDI appears to catalyze isomerization simply by reducing incorrect disulfides and allowing another opportunity to form a correct disulfide. This continues until the connection of disulfides is sufficiently stable that it is no longer a substrate for the isomerase activity of PDI. PDI catalyzes the formation and reduction of disulfide bonds and their isomerization in a wide range of proteins *in vitro*, such as insulin, ribonuclease, bovine pancreatic trypsin inhibitor, and immunoglobulin Fab fragments.[66, 67] The substrate most often used for isomerase activity assays of PDI is ribonuclease A (RNase A).[28] T_3 inhibits the PDI catalyzed refolding of reduced RNase A;[43] however, the inhibition is only partial, and saturating T_3 inhibits the isomerase activity by only 40%.[25] E_2 also has been reported to inhibit isomerase activity at a comparable level.[25, 54] Although T_3, E_2 and BPA bind to the same site, the maximum inhibition by BPA was 20%, indicating the inhibitory mechanism of BPA to be different from that of T_3 or E_2.[25]

CHAPERONE ACTIVITY OF PDI

PDI acts as a chaperone by preventing protein aggregation and/or by retaining proteins in the ER, thereby circumventing their degradation by the ubiquitin-proteasome pathway.[68] PDI catalyzes as a chaperone the renaturation of denatured, reduced insulin or of ribonuclease T1 or AIII[69] or alcohol dehydrogenase.[70] BPA, which binds to and inhibits the isomerase activity of PDI, has no significant effect on the chaperone activity.[71] T_3 has no inhibitory effect on the chaperone activity of PDI either.[46] Thus, neither BPA nor T_3 likely influences the interaction of PDI with aggregation-prone substrates. The peptide antibiotic bacitracin has been shown to inhibit both the isomerase[72] and chaperone activity[46] of PDI, and is thought to interact with the peptide/protein-binding site of PDI. The binding of BPA to PDI was not inhibited by bacitracin,[25] indicating BPA's mode of inhibitory action to be different from that of bacitracin.

REQUIRED CHEMICAL STRUCTURE FOR INHIBITION OF PDI ACTIVITY

In 2000–2001, the Ministry of the Environment in Japan listed twenty compounds for health risk assessment. These compounds include phenolic compounds, organotins, and phthalates. In order to identify compounds which inhibit PDI activities, assays of the binding to radioligand and isomerase activity of PDI were performed in the presence of each test chemicals. Furthermore, halogenated derivatives of BPA, namely tetrabromobisphenol A (TBBPA) and tetrachlorobisphenol A (TCBPA), were assessed. TBBPA and TCBPA are widely used as flame retardants in numerous products.[73] Nonylphenol and octylphenol are industrial additives used in a variety of detergents and plastics.[74] 2,4-Dichlorophenol and pentachlorophenol are extensively used in the manufacture of pesticides, herbicides, and wood preservatives.[75] Organotin compounds have been used as agricultural and wood preservative fungicides, antifouling paint biocides, and thermal stabilizers for polyvinyl chloride (PVC). Consequently, organotin contamination is found in various environmental media,[76] wildlife,[77, 78] and food.[79] Phthalate esters, used in the production of various plastics (including PVC), are among the most common industrial chemicals. Their ubiquity in the environment and tendency to bioconcentrate in animal fat and food are well known.[80–83] Competitive binding assays revealed a correlation between chemical structure and inhibition of the binding of T_3 for PDI.[26] Among the 22 compounds, only phenolic compounds, namely BPA, *p*-octylphenol, *p*-nonylphenol, 2,4-dichlorophenol, pentachlorophenol, TBBPA, and TCBPA, specifically prevented T_3

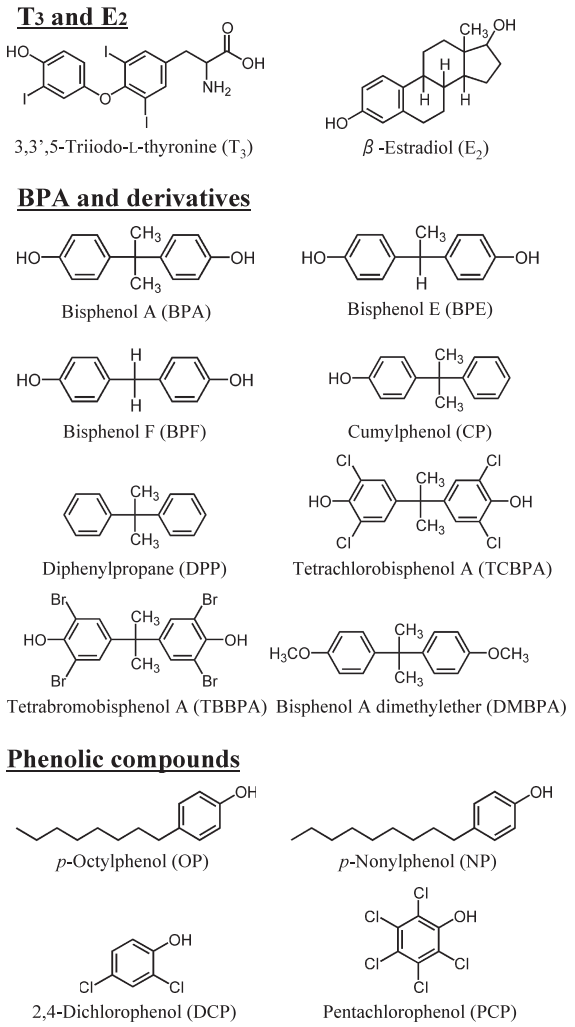

Fig. 4. Chemical Structure of Hormones, BPA Derivatives, and Phenolic Compounds

Table 1. Effects of Environmental Compounds on PDI Activity[26, 99]

Compounds	IC_{50} value for $[^{125}I]T_3$-binding	Inhibition of isomerase activity
T_3	7.60×10^{-7}	Yes
BPAs		
BPA	2.22×10^{-5}	Yes
TCBPA	2.00×10^{-7}	No
TBBPA	1.18×10^{-6}	No
Phenolic compounds		
OP	2.78×10^{-6}	Yes
NP	5.72×10^{-6}	Yes
DCP	3.51×10^{-5}	No
PCP	2.28×10^{-5}	No
PCBs		
PentaCB	ND	No
HexaCB	ND	No
4OH-PentaCB	3.89×10^{-6}	Yes
4OH-HexaCB	3.31×10^{-6}	No

Abbreviations are presented in Figs. 4 and 5.

from binding to PDI (Fig. 4 and Table 1), suggesting the phenolic group(s) of compounds to have important effects on the binding. Among these phenolic compounds, T_3 and non-halogenated compounds including BPA, *p*-octylphenol, and *p*-nonylphenol showed inhibitory effects on the isomerase activity of PDI.[26] However, polyhalogenated phenolic compounds did not inhibit isomerase activity of PDI although they inhibited the binding of T_3 (Fig. 4 and Table 1). PDI has multibinding sites for T_3 and substrates.[46] Therefore, non-halogenated and polyhalogenated phenolic compounds may has different binding mode for PDI. Furthermore, to address the chemical structural requirements of BPA for the inhibitory effect of PDI, several BPA analogs were tested (Fig. 4).[71] Cumylphenol inhibited the binding of BPA to PDI in a dose-dependent manner, but diphenylpropane had little or no inhibitory effect. On the other hand, bisphenol A dimethylether did not prevent BPA from binding to PDI. Inhibition of the binding of bisphenol E and bisphenol F was almost identical to that of BPA, suggesting that substitution of the methyl group at the centre of BPA did not contribute to the affinity for PDI. These findings suggest the hydroxyl group of the phenyl ring(s) to be essential for both the binding affinity and isomerase activity of PDI.

POLYCHLORINATED BIPHENYL (PCB)

PCBs are a class of compounds in which the aromatic biphenyl skeleton carries between one and ten chlorine atoms. Due to their non-flammability, chemical stability, high boiling point and electrical insulating properties, PCBs were used in printed circuit boards, heat carriers, electrical capacitors, and transformers as cooling fluids, flameretardants, hydraulic fluids, adhesives, and plasticizers. PCBs are environmental ccontaminants, still present in the environment due to their high chemical stability, although their production has been halted since 1977. Among toxic man-made chemicals, PCBs are some of the most extensively studied in terms of environmental contamination and toxicological impact. Jacobson *et al.* found that children exposed *in utero* to PCBs have delayed central nervous system functioning, and therefore, PCBs are considered to be developmental neurotoxicants at environmentally relevant concentra-

tions.[84–87] PCBs reduce the circulating levels of thyroid hormone in animals.[88] Because the symptoms of PCB exposure can overlap with those of thyroid hormone dysfunction, several investigators have speculated that the neurological consequences of incidental exposure to PCBs are caused by disruption of the thyroid axis.[89, 90] The most commonly noted neurological abnormalities associated with low level PCB contamination in humans are hyperactivity and impaired learning.[91] Their hydroxylated metabolites have been found in human serum,[92] whole blood,[93] and plasma.[94] Relatively high levels of para-hydroxylated PCB metabolites from penta-, hexa-, and heptachlorinated biphenyls have been found in the plasma of rats exposed to PCBs. *In vivo*, PCBs are converted by cytochrome P450 (CYP) in the liver to hydroxy-PCB (HO-PCB), which an active metabolite.[95, 96] Normally, the hydroxylated metabolites are excreted in feces and/or in urine, by themselves or conjugated to glucuronic acid or sulfate.[97, 98] However, some HO-PCBs are persistent and may directly interfere with the thyroid system. HO-PCBs have possible inhibitory effects on binding of PDI to T_3 (Fig. 5). The binding to T_3 or BPA of PDI is inhibited by 4-hydroxy-2′,3,3′,4′,5′-pentachlorobiphenyl (HO-pentaCB), and 4-hydroxy-2′,3,3′,5,5′, 6′-hexachlorobiphenyl (HO-hexaCB), but not their parental PCB, indicating that the 4-hydroxyl group is essential for binding to PDI (Fig. 5 and Table 1). It is suspected that T_3, BPA, and hydroxylated PCBs share identical binding sites.[99] The manner in which HO-PCBs inhibit the isomerase activity of PDI is the same as that for T_3 or BPA binding. HO-pentaCB but not HO-hexaCB inhibited the isomerase activity of PDI although both compounds bind to PDI, suggesting that substituents at the 3,5-positions of the phenyl ring, besides the 4-hydroxyl group, interfere with the isomerase activity. These findings raise the possibility of the participation of PDI in PCB induced abnormalities of brain development.

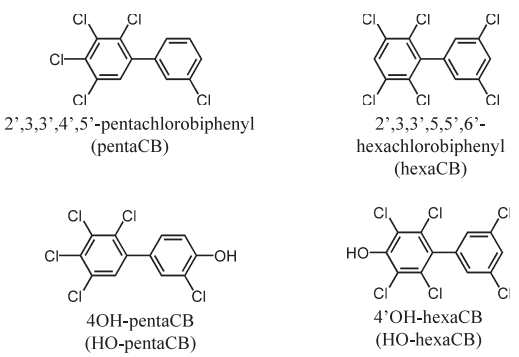

Fig. 5. Chemical Structure of PCBs and Their Metabolites

INVOLVEMENT OF PDI IN GROWTH HORMONE RELEASE IN RAT PITUITARY CELLS

GH3 rat pituitary tumor cells are established somatotrophs, and widely used to study the molecular basis of the activities of thyroid hormone[100, 101] since T_3 stimulates both transcription of the growth hormone (GH) gene, and release of the hormone. Physiological concentrations of thyroid hormone stimulate the rate of GH production in GH3 cells. In GH3 cells, PDI was first identified as a T_3-binding protein a 55-kDa (p55).[48] The p55 protein was then found to be located on the lumenal face of the endoplasmic reticulum and nuclear envelope.[49, 50] Sequencing of cDNA[51] for p55 indicated the coding

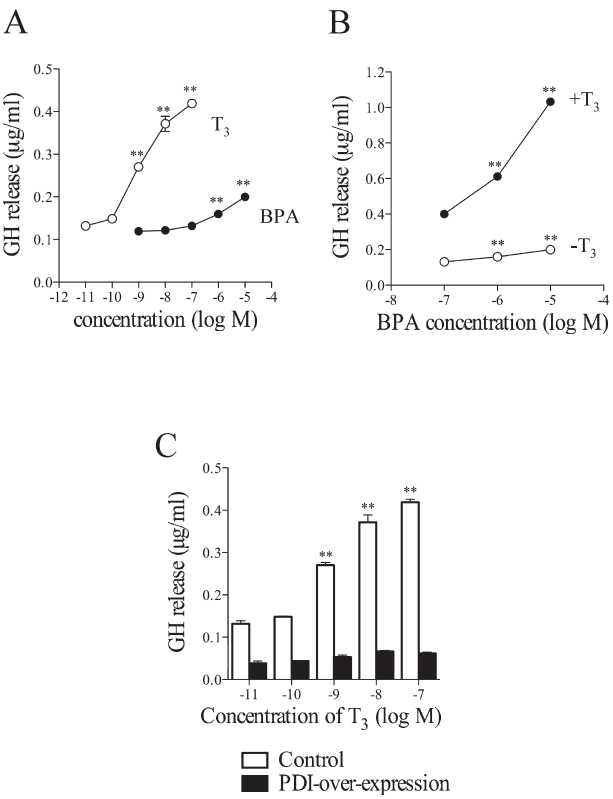

Fig. 6. Amount of GH Released from GH3 Cells

Addition of various concentrations of T_3 and BPA alone (A), addition of various concentrations of BPA in the presence of 10 nM T_3 (B), and addition of various concentration of T_3 to control GH3 cells and GH3 cells overexpressing PDI (C).[27]

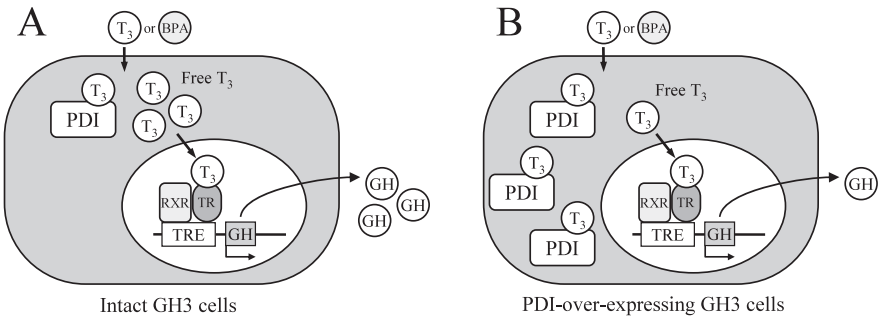

Fig. 7. Possible Mechanism for T_3 and/or BPA-induced Release of Growth Hormone in Mock Treated Cells (A) or PDI-over-expressing GH3 Cells (B)

region to have 85% homology to rat PDI[102] and 98% homology to the β-subunit of human P4H.[103] It is now apparent that these proteins are identical to PDI. BPA has weak agonistic effects on the release of GH at high concentrations in the absence of T_3[27] in GH3 cells (Fig. 6A). The relative potency of BPA was 1/1000 that of T_3, and the magnitude of the response to BPA was much lower than that to T_3. However, when the cells were treated with BPA together with T_3, the response was much greater than that to T_3 alone (Fig. 6B). Forced over-expression of PDI suppressed the T_3-induced release of GH as well as the GH mRNA expresion in GH3 cells (Fig. 6C).[27] Primm and Gilbert suggested that PDI acts as a reserve of T_3 to control its concentration (Fig. 7) in cells. T_3 also down-regulates PDI expression at the post-transcription level.[104] However, the physiological significance of the T_3-binding activity is not well understood.

PDI FAMILY

In humans,[105] there are now 14 known members of the PDI-family in the ER, ranging widely in domain architecture and active-site chemistry; including ERp72, ERp57, P5, PDIp, PDIr, ERp44,[106] ERp28,[107] ERdj5,[108] and ERp18.[109] The functional characterization and differentiation of these family members is far from complete. Among the PDI family, ERp57 shows significant domain similarity with PDI, and has two thioredoxin-like domains with -Cys-Gly-His-Cys- catalytic site motifs in positions corresponding to the thioredoxin-like domains **a** and **a'** of PDI, which altogether consists of domains **a**, **b**, **b'**, and **a'** plus an acidic C-terminal extension **c**.[110] Whereas PDI interacts directly with and also folds non-native proteins, ERp57 is known to act *in vitro* and *in vivo* on glycosylated substrates through its interaction with the ER-resident lectins calnexin and calreticulin.[111, 112] ERp72 also shows domain similarity with PDI, which consists of domains **a**, **a**, **b**, **b'**, and **a'** plus an acidic N-terminal extension **c**.[113] ERp72 is more ERp57-like, and many of the residues that are implicated in ERp57-calreticulin interactions are conserved in ERp72. However, there is no experimental evidence yet to suggest an ERp72-calreticulin interaction. Preliminary study showed that BPA binds to both ERp57 and ERp72, weakly, however, it has no effect on catalytic activity (unpublished data). These observations may suggest that the inhibitory effect of BPA on PDI is quite specific, and also results in abnormal PDI-specific folding of substrates.

CONCLUSIONS

Several reports have described the involvement of PDI as a multifunctional protein in brain impairment, including ischemia,[114, 115] Parkinson's disease,[116, 117] Alzheimer's disease,[117, 118] and Creutzfeldt-Jacob disease.[119] We speculate that EDCs may be responsible at least in part for neurodegenerative disorders via their effects on PDI. Several papers have described the significance of PDI to the physiological activity of thyroid hormone. For example, PDI participates in the inactivation of type 2 iodothyronine 5′-deiodinase, a key enzyme for converting thyroxine (T_4) to bioactive T_3 in the brain.[120] Studies *in vitro* found PDI to be responsible for the multimerization of thyroglobulin, the precursor of thyroid hormone.[121, 122] PDI was also shown to participate in the shedding of α-subunits of thyroid stimulating hormone receptors of thyrocytes.[123] As a cellular T_3-binding protein, Primm and Gilbert proposed that PDI serves as a hormone reservoir to buffer the hormone concen-

tration in cells.[46] Recent study demonstrated that over-expression of PDI in GH3 cells reduced T_3-induced GH mRNA and protein expression.[27] This result suggests that the artificially overexpressed PDI in GH3 cells enhanced the retention of T_3 in the cell, making it difficult for T_3 to enter the nucleus and to activate gene expression (Fig. 7). Thus, PDI plays an important role in the hypothalamic-pituitary-thyroid axis. Thyroid hormones modulate oxygen consumption, the basal metabolic rate, and the metabolism of lipids, carbohydrates and proteins. They also change the rate of synthesis and degradation of a variety of other growth factors and hormones. Consequently, thyroid hormones are essential for growth, development and metabolism in the body.[124] In particular, the actions of thyroid hormones are vital for normal brain development, influencing behavior and cognitive functions in both adults and children.[125] Disruption of the thyroid system during development has been shown to result in permanent changes in neurochemical,[126] morphologic[127] and neurobehavioral end points.[128–130] Further study will hopefully reveal how PDI recognizes its hormonal ligands or phenolic compounds.

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