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Novel Nongenomic Signaling of Thyroid Hormone Receptors in Thyroid Carcinogenesis

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Abstract

The thyroid hormone receptors (TRs) are transcription factors that mediate the pleiotropic activities of the thyroid hormone, T3. Four T3-binding isoforms, TR α 1, TR β 1, TR β 2, and TR β 3, are encoded by two genes, *THRA* and *THRB*. Mutations and altered expression of TRs have been reported in human cancers. A targeted germline mutation of the *Thrb* gene in the mouse leads to spontaneous development of follicular thyroid carcinoma (TR β ^{PV/PV} mouse). The TR β PV mutant has lost T3 binding activity and displays potent dominant negative activity. The striking phenotype of thyroid cancer exhibited by TR β ^{PV/PV} mice has recently led to the discovery of novel non-genomic actions of TR β PV that contribute to thyroid carcinogenesis. These actions involve direct physical interaction of TR β PV with cellular proteins, namely the regulatory subunit of the phosphatidylinositol 3-kinase (p85 α), the pituitary tumor-transforming gene (PTTG) and β -catenin, that are critically involved in cell proliferation, motility, migration, and metastasis. Thus, a TR β mutant (TR β PV), via a novel mode of non-genomic action, acts as an oncogene in thyroid carcinogenesis.

Keywords

thyroid hormone receptor mutants; thyroid cancer; non-genomic action; phosphatidylinositol 3 kinase; pituitary tumor transforming gene; β -catenin; mouse model

1. Introduction

Thyroid hormone receptors (TRs) are members of the nuclear receptor superfamily that mediate the pleiotropic activities of the thyroid hormone, T3, in differentiation, growth, and metabolism (Yen, 2001). There are four major T3-binding receptor isoforms: TR α 1, TR β 1, TR β 2, and TR β 3. They are encoded by two genes, *THRA* and *THRB*, that are located on two different chromosomes. Numerous efforts have been made over the past decade to uncover the underlying mechanism of action of these nuclear receptors. Classically, TRs bind to specific DNA sequences on the promoter of T3-target genes (thyroid hormone response elements, or TREs) to activate or repress basal gene transcription (Yen, 2001). The regulation of their transcriptional activity is complex: it depends not only on T3 binding but also on the type of TREs on the promoter of T3-target genes.

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There are many T3-target tissues, including bone, heart, adipose tissue, liver, pituitary, and brain. The broad spectrum of T3 action is well illustrated by patients with resistance to thyroid hormone (RTH). RTH is a syndrome characterized by reduced sensitivity of tissues to the action of thyroid hormones, and it is inherited in an autosomal dominant manner. The hallmark of RTH is elevated thyroid hormone associated with nonsuppressible thyroid stimulating hormone (TSH). Other clinical signs are goiter, delayed bone growth and maturation, decreased weight, tachycardia, hearing loss, attention deficit disorder, and hyperactivity disorder (Weiss and Refetoff, 2000; Yen, 2003). Numerous mutations in the ligand-binding domain of TR β have been identified in RTH families (Adams et al., 1994; Refetoff et al., 1993). Most patients are heterozygous for the mutation, and the clinical symptoms are mild (Weiss and Refetoff, 2000; Yen, 2003). Only one patient homozygous for a mutant TR β has been reported (Ono et al., 1991). That patient, who died young, displayed extreme RTH with high levels of thyroid hormones and TSH (Ono et al., 1991).

Several findings support the notion that mutations of TRs can be associated with human cancer. Early evidence to suggest that mutated TRs could be involved in carcinogenesis came from the discovery that v-erbA, a highly mutated chicken TR α 1 that has lost the ability to activate gene transcription, leads to neoplastic transformation in erythroleukemia and sarcomas (Sap et al., 1989; Thormeyer and Baniahmad, 1999; Wallin et al., 1992). That male transgenic mice overexpressing v-erbA develop hepatocellular carcinomas is evidence that v-erbA oncoprotein can promote neoplasia in mammals through its dominant negative activity (Barlow et al., 1994). Abnormal expression and somatic mutations of TRs have been observed in human cancers. To date, TR alteration has been reported in thyroid cancer (Wallin et al., 1992; Bronnegard et al., 1994; Puzianowska-Kuznicka et al., 2002), liver cancer (Lin et al., 1999), kidney cancer (Kamiya et al., 2002; Puzianowska-Kuznicka et al., 2000), pituitary tumor (Safer et al., 2001; Ando et al., 2001a; Ando et al., 2001b), and breast cancer (Li et al., 2002; Silva et al., 2002). Knock-in mice harboring a germ-line mutation in TR β (TR β PV), leading to the abolition of T3 binding and dominant negative activity, develop thyroid cancer and pituitary tumor (Furumoto et al., 2005; Suzuki et al., 2002). These studies suggest that partial loss of normal TR function due to reduced expression, or complete loss or alteration of TR activity, provides an opportunity for cells to proliferate, invade, and metastasize. In this context, TR could act as a tumor suppressor.

Taken together, these studies raise two important questions. First, what genes or signaling pathways critical for carcinogenesis are affected by loss of function of TR? Second, by what mechanisms do the mutated TRs alter the activity of the affected genes or signaling pathways to mediate carcinogenesis?

The availability of a mouse model of thyroid cancer harboring the TR β PV mutation provides us with the possibility to explore the role of TR β mutants in tumor progression and metastasis of thyroid cancer. Our earlier studies sought to understand how TR β PV alters signaling pathways by affecting gene expression profiles in the thyroid of the TR β ^{PV/PV} mice (Ying et al., 2003). However, our most recent studies of the TR β PV oncogenic actions in TR β ^{PV/PV} mice showed that this TR mutant not only exerts dominant activity on gene transcription, but also acts via non-genomic action to mediate thyroid carcinogenesis (Furuya et al., 2006; Ying et al., 2006; Furuya et al., 2007a; Guigon et al., 2008). The aim of this review is to highlight recent advances in the understanding of the critical role of the novel non-genomic actions of mutations of TR in thyroid carcinogenesis.

2. Novel modes of non-genomic actions of a TR β mutant

2.1. The TR $\beta^{PV/PV}$ mouse as a model of thyroid cancer

The TR β mutant proteins identified in RTH have either low or no T3-binding affinity and transcriptional activity (Yen, 2003). The creation of a mouse model harboring a C-terminal 14 amino acid frameshift mutation (Kaneshige et al., 2000) led to a better understanding of the molecular mechanisms underlying RTH. This mutation, which was called PV after an RTH patient, leads to a complete loss of T3 binding (TR β mutant) (Meier et al., 1992; Parrilla et al., 1991) and exhibits potent dominant negative activity (Meier et al., 1992) (Figure 1). The TR β^{PV} mouse model faithfully reproduces RTH in humans with the loss of the feedback regulation in the pituitary-thyroid axis (Kaneshige et al., 2000). Consistent with the disorders associated with RTH in humans, TR β^{PV} mice also display growth retardation, thyrotoxic skeletal phenotype, and neurological dysfunction (Kaneshige et al., 2000; O'Shea et al., 2003; Siesser et al., 2005).

Notably, as homozygous TR $\beta^{PV/PV}$ ($TR\beta^{PV/PV}$), but not heterozygous TR $\beta^{PV/+}$ ($TR\beta^{PV/+}$) mice age, they spontaneously develop thyroid cancer (Suzuki et al., 2002). $TR\beta^{PV/PV}$ mice display thyroid hyperplasia as early as 2 months of age, which is followed by follicular thyroid carcinoma (FTC) with progression from capsular and vascular invasion to anaplasia and distant metastases (Suzuki et al., 2002). The similarities in FTC progression between this mouse model and humans provide us with a unique opportunity to identify and characterize the molecular pathways underlying thyroid carcinogenesis and metastasis.

2.2. Overactivation of phosphatidylinositol 3-kinase (PI3K) signaling by TR β^{PV}

Dysregulation of PI3K signaling contributes to abnormal cell growth, cellular transformation in a variety of neoplasms, including thyroid cancer. PI3Ks consist of a catalytic subunit of about 110 kD (p110) and a tightly associated regulatory subunit (p85 α , p85 β or p55 γ). The subunit regulates the association of PI3K with membrane-associated signaling complexes. Upon activation by membrane receptors, PI3K phosphorylates phosphatidylinositol-4,5 biphosphate [PIP2] to form phosphatidylinositol-3,4,5- triphosphate [PIP3]. Through phosphatidylinositol-dependent kinases, the downstream effectors of PI3K, the serine/threonine kinase AKT is phosphorylated and activated to further phosphorylate downstream protein substrates (Neri et al., 2002; Shepherd et al., 1997; Wymann and Marone, 2005). The activity of PI3K is negatively regulated by PTEN (phosphatase and tensin homolog deleted on chromosome 10), a protein phosphatase that dephosphorylates PIP3 to form PIP2 (Eng, 2002).

Recent studies have shown aberrant PI3K/AKT signaling in thyroid carcinogenesis. Inactivating mutations of the tumor suppressor gene *PTEN* were observed in patients with Cowden syndrome, a genetically inherited disorder associated with higher risk to develop several types of tumors, including follicular thyroid carcinoma (Nelen et al., 1997; Steck et al., 1997). Studies of human thyroid cancer by several groups have shown AKT overactivation and overexpression in primary thyroid cancers (Ringel et al., 2001; Miyakawa et al., 2003). Similar to findings in human thyroid cancer, we determined that the PI3K/AKT signaling pathway was overactivated in the thyroid tumors of $TR\beta^{PV/PV}$ mice (Furuya et al., 2006). Consistent with a major role for PI3K signaling in thyroid cancer, the treatment of $TR\beta^{PV/PV}$ mice with the potent PI3K inhibitor LY294002 significantly delayed thyroid tumor progression and blocked metastatic spread (Furuya et al., 2007b).

Most studies aimed at understanding the molecular actions of TR β mutants, including TR β^{PV} , have focused on their interference with the genomic actions of wild-type TR β . By contrast, our study of the molecular mechanisms involved in the overactivation of AKT in thyroid cancer

of $TR\beta^{PV/PV}$ mice led to the identification of a novel non-genomic mode of action of a mutant thyroid hormone receptor that drives thyroid carcinogenesis (Furuya et al., 2006). Indeed, we found that the p85 α regulatory subunit of PI3K physically interacts with TR β 1 or the TR β PV mutant. Notably, p85 α interacted two to three times more strongly with TR β PV than with TR β 1, resulting in a great increase of PI3K activity associated with the overactivation of AKT and phosphorylation of its downstream effectors.

Recent studies have indicated the presence of PI3Ks, their lipid substrates and products, and AKT in the nuclear compartment (Neri et al., 2002; Irvine, 2003). Consistent with these observations, we found that sequestration of TR β PV by p85 α occurred in both the nuclear and the cytoplasmic compartments, suggesting that the regulation of PI3K signaling by TR β PV could occur in both. Indeed, activation of AKT and downstream signaling pathways occurred in the nuclear and cytosolic fraction of thyroid extracts, but with a higher activity in the nuclear fraction (Furuya et al., 2006). Given the critical role of PI3K in regulating multiple signaling pathways, it is likely that its activation by TR β PV in both compartments affects diverse cellular signaling pathways. This hypothesis would be consistent with our earlier studies using cDNA microarrays that showed complex alteration of multiple signaling pathways to be associated with thyroid carcinogenesis in $TR\beta^{PV/PV}$ mice (Ying et al., 2003).

Additionally, we found that the nuclear receptor corepressor (NCoR) was involved in the modulation of TR β PV-induced PI3K activation (Furuya et al., 2007a). NCoR is well known to regulate the genomic actions of nuclear receptors, as it was reported for TR β and TR β mutants *in vitro* and *in vivo*; however, recent studies have also shown that NCoR can be involved in transcription-independent mechanisms. NCoR is found not only in the nucleus, but also in the cytoplasm (Hermanson et al., 2002; Sardi et al., 2006). The redistribution of NCoR to the cytoplasm provides a mechanism for controlling differentiation of neural stem cells into astrocytes (Hermanson et al., 2002). Moreover, cytoplasmic NCoR included in protein complexes can be translocated to the nucleus to regulate astrogenesis (Sardi et al., 2006). These studies led us to evaluate the potential involvement of NCoR in regulating PI3K signaling. Notably, we found that NCoR interacts with p85 α in the nucleus and the cytoplasm, and thereby competes with TR β PV (Figure 2). Experimental alteration of cellular NCoR protein levels by overexpression led to reduced PI3K signaling. Conversely, knocking down cellular NCoR by small interfering RNA (siRNA) approaches increased PI3K activity. In thyroid tumors, cellular NCoR protein abundance was decreased as compared with wild-type thyroids, thereby favoring the interaction between p85 α and TR β PV to activate PI3K signaling (Furuya et al., 2007a). Therefore, NCoR, via protein-protein interaction, is a novel regulator of PI3K signaling that could modulate thyroid tumor progression.

2.3. Stabilization of the pituitary tumor-transforming gene (PTTG) by the TR β PV mutant

PTTG is a mammalian securin that is a critical mitotic checkpoint protein involved in maintaining sister chromatids cohesiveness before entering anaphase (Yu and Melmed, 2001). Its association with separin (a protease) prevents the proteolytic activity of separin from acting on the proper separation of sister chromatids. At each cell cycle, degradation of PTTG by proteasome pathways frees separin, thus leading to chromatid separation at the anaphase. Alteration of normal levels of PTTG may disrupt this orderly process of chromatid segregation with subsequent aneuploidy in the daughter cells (Zou et al., 1999). Indeed, increased expression of PTTG has been demonstrated in human thyroid carcinoma (Heaney et al., 2001; Kim et al., 2005), colon carcinoma (Heaney et al., 2000), and pituitary adenoma (Zhang et al., 1999; McCabe et al., 2003). Aneuploidy is a common feature of thyroid follicular adenoma and carcinoma (Joensuu et al., 1986; Joensuu and Klemi, 1988), and chromosomal instability may underlie the progression to anaplastic thyroid cancer, the more aggressive stage of thyroid cancer.

We discovered chromosomal alterations in seven cell lines derived from thyroid tumors of $TR\beta^{PV/PV}$ mice (Zimonjic et al., 2005). These cell lines exhibit abnormal karyotypes and a variety of structural chromosomal aberrations, including translocations and deletions, raising the possibility that induction of chromosomal aberrations could be one of the key genetic events involved in thyroid carcinogenesis in $TR\beta^{PV/PV}$ mice (Zimonjic et al., 2005). The search for genes underlying the chromosomal aberrations in $TR\beta^{PV/PV}$ mice by cDNA microarray led to the discovery that *Pttg* mRNA levels were significantly increased in thyroid cancer (Ying et al., 2003). In addition, cellular PTTG protein levels were markedly increased in the primary lesions of thyroid as well as lung metastasis of $TR\beta^{PV/PV}$ mice (Ying et al., 2006). Consistently, our cell-based studies further showed that aberrant accumulation of PTTG induced by TR β PV inhibits mitotic progression (Ying et al., 2006). In $TR\beta^{PV/PV}Pttg^{-/-}$ as compared with $TR\beta^{PV/PV}$ mice, there was a consistent decrease in thyroid cell proliferation. The thyroids of $TR\beta^{PV/PV}Pttg^{-/-}$ mice were significantly smaller, and the occurrence of metastasis spread to the lung was less frequent (Kim et al., 2007).

Altogether, these data indicate that abnormal accumulation of PTTG in the thyroids of $TR\beta^{PV/PV}$ mice contributes to thyroid carcinogenesis by affecting cell cycle progression and inducing genetic instability. Still, the molecular mechanisms underlying elevated abundance of PTTG proteins have not been fully elucidated. It is likely that the increase in *Pttg* mRNA levels in $TR\beta^{PV/PV}$ mice (Ying et al., 2003) leads, at least partially, to the aberrant accumulation of PTTG proteins. Because TRs and PTTG are both involved in proteasome-mediated degradation pathways (Dace et al., 2000; Yu et al., 2000), we investigated whether TR β and TR β PV could regulate PTTG protein levels through such mechanisms (Ying et al., 2006). A series of cell-based and molecular studies showed that the DNA binding domain of TR β 1 or TR β PV interacts with the amino-terminal region of PTTG (Ying et al., 2006) and that T3 induces the degradation of TR β 1 concomitantly with that of PTTG via the proteasomal machinery (Ying et al., 2006). In contrast, T3 does not induce TR β PV degradation, consistent with the fact that TR β PV loses T3 binding capacity. Along with the stability of TR β PV, PTTG protein levels are not altered by T3 treatment and remain high (Ying et al., 2006). Therefore, the liganded TR β 1 regulates PTTG stability; this regulatory function is lost in TR β PV that fails to bind T3.

To understand how TR β PV fails to regulate the stability of PTTG as the liganded TR β 1 does, we considered the possibility that the protein complexes TR β 1/PTTG and TR β PV/PTTG recruit proteasome activators differently. It was reported that steroid receptor co-activator-3 (SRC-3) is degraded via 19S proteasome through its physical interaction with proteasome activator 28 γ (PA28 γ), an activator of the trypsin-like activity of the proteasome (Li et al., 2006). Similar to other steroid receptors (Lonard and O'Malley B, 2007), the liganded TR β 1 recruits SRC-3, but the unliganded TR β 1 does not (Ying et al., 2006). TR β PV does not bind SRC-3, whether T3 is present or not (Ying et al., 2006). We therefore tested the possibility of the existence of a differential recruitment of TR β /PTTG and TR β PV/PTTG complexes by SRC-3/PA28 γ . Indeed, we made the remarkable finding that the liganded TR β 1/PTTG recruits SRC-3/PA28 γ through the direct interaction of TR β 1 with SRC-3 (Figure 3A-a), whereas the unliganded TR β 1 and TR β PV fail to recruit SRC-3/PA28 γ (Figure 3A-b) (Ying et al., 2006). Altogether, this study indicates that TR β PV, via protein-protein interaction, affects PTTG degradation by the proteasome pathway and thereby results in mitotic abnormalities, contributing to thyroid carcinogenesis.

2.4. Stabilization and constitutive activation of β -catenin by the TR β PV mutant

β -catenin is a central mediator of the Wnt signaling pathway, which is critical for a variety of cellular processes including cell proliferation, cell apoptosis, and cell migration (Polakis, 2007). Upon increased β -catenin cellular levels and nuclear accumulation, β -catenin together

with the T-cell factor/lymphoid enhancer factor (TCF/LEF) binds to the promoters of downstream target genes (Moon et al., 2002). Induction of these genes has significant effects on tissue development and oncogenesis; abnormal subcellular localization and aberrant accumulation of β -catenin have been reported in a number of human cancers, including thyroid cancers (Garcia-Rostan et al., 1999).

The cellular levels of β -catenin are tightly regulated by two distinct adenomatous polyposis coli (APC)-dependent proteasomal pathways. One includes the glycogen synthase kinase 3 β (GSK3 β)-regulated pathway involving the APC/axin complex (Polakis, 2002) and the other a p53-inducible pathway involving APC/Siah-1 (Liu et al., 2001). An additional mode of β -catenin cellular level regulation is mediated by nuclear receptors, namely retinoid X receptor α (RXR α) and peroxisome proliferator-activated receptor γ (PPAR γ). These nuclear receptors regulate β -catenin protein abundance via APC/GSK3 β /p53-independent mechanisms (Sharma et al., 2004; Xiao et al., 2003).

The finding that the cellular abundance of β -catenin was aberrantly elevated in thyroid tumors of $TR\beta^{PV/PV}$ mice gave us the opportunity to understand how TR β and its TR β PV mutant regulate the cellular levels of β -catenin and, furthermore, what effect β -catenin accumulation has on the activation of β -catenin oncogenic signaling in the thyroids of $TR\beta^{PV/PV}$ mice.

We found that, similar to RXR α and PPAR γ 2, TR β and its mutant TR β PV physically interact with β -catenin *in vitro* and in cells (Sharma et al., 2004; Guigon et al., 2008; Xiao et al., 2003). As reported for RXR α and PPAR γ , β -catenin protein level regulation by TR β and TR β PV occurs via APC/GSK3 β /p53-independent mechanisms. However, in contrast to RXR α and PPAR γ 2, binding of T3 to TR β weakens the interaction of the receptor with β -catenin, thereby leading more uncomplexed β -catenin to be degraded by proteasomal pathways (Guigon et al., 2008) (Figure 3B-a). Importantly, the interaction of β -catenin with TR β PV is not affected by T3, because TR β PV does not bind T3. Therefore, the constitutive association of TR β PV with β -catenin leads to the stabilization of β -catenin (Figure 3B-b).

Although there is a commonality in the early steps of the APC-independent regulation of β -catenin mediated by TR β and by PPAR γ 2 and RXR α in that they all physically interact with β -catenin, ligand binding differently affects the subsequent mode of actions between TR β and PPAR γ 2 or RXR α . The reason for these different modes of nuclear receptor actions in β -catenin cellular level regulation is currently unknown. The A/B domain of RXR (Xiao et al., 2003) and the ligand binding domain of PPAR γ 2 (Liu et al., 2006) were reported to be necessary for their interaction with β -catenin. In contrast, the DNA binding domain of TR β is important in its interaction with β -catenin3 (Guigon et al., 2008). It is thus tempting to speculate that these different interacting regions in TR β , PPAR γ 2, and RXR α may play a role in the different modes of β -catenin regulation.

Our mouse model also gave us the opportunity to study *in vivo* the effect of β -catenin aberrant accumulation on the activity of the β -catenin signaling pathway in the thyroid cancer of $TR\beta^{PV/PV}$ mice. We found that the increased β -catenin cellular levels in the thyroids of $TR\beta^{PV/PV}$ mice was associated with a significant increase in its nuclear translocation and transcriptional activity (Guigon et al., 2008), as shown by the increased cellular abundance of β -catenin phosphorylated on serine 552 (P552- β -catenin) as well as of β -catenin transcriptional downstream targets such as c-myc and cyclin D1, both involved in cell cycle progression, and MT1-MMP (matrix metalloprotease-1) that is involved in cell migration. Given the critical role of β -catenin in mediating epithelial-mesenchymal transition (EMT) (Lee et al., 2006; Hubert et al., 2005), it is very likely that β -catenin constitutive activation in $TR\beta^{PV/PV}$ thyroids plays a role in the tumorigenic progression to metastasis. It would be of interest to determine whether defining features of EMT that were reported in human invasive thyroid

carcinomas such as loss of E-cadherin and gain of vimentin and fibronectin expression (Vasko et al, 2007), are also found in $TR\beta^{PV/PV}$ thyroid cancers.

3. Conclusions and future challenges

Increasing evidence supports the notion that TRs could play an important role in carcinogenesis (Cheng, 2003). The availability of the $TR\beta^{PV/PV}$ mouse as a model of thyroid carcinogenesis provides a valuable tool to understand the role of TR β mutants in tumor progression and metastasis of thyroid cancer. Studies so far have shown that the deleterious effects of TR β mutants in causing RTH are mediated via dominant negative actions at the transcriptional level (Yen, 2003; Cheng, 2005). However, the striking phenotype of thyroid cancer displayed by $TR\beta^{PV/PV}$ mice led to the identification of novel modes of TR β mutant actions that are beyond nucleus-initiated transcription. The direct protein-protein interaction of TR β PV with the PI3K regulatory subunit p85 α increases the activity of PI3K, leading to an activated cascade of downstream signaling to affect cell proliferation, apoptosis, migration, and metastasis. The direct protein-protein interaction of TR β PV with PTTG or β -catenin affects their degradation by the proteasomal pathways, thus leading to PTTG and β -catenin aberrant accumulation. That contributes to genetic instability and constitutively active β -catenin oncogenic signaling to affect cell proliferation and migration. Therefore, the development and progression of thyroid tumors in $TR\beta^{PV/PV}$ mice involve not only impaired transcriptional activity but also derailed post-transcriptional mechanisms. Altogether, our studies uncovered novel molecular mechanisms by which a TR β mutant affects critical signaling pathways for cell proliferation, cell apoptosis, EMT transition and cell migration, to drive thyroid carcinogenesis. It would be of interest to determine whether additional non-genomic mechanisms that are reported to mediate T3/TR actions in other cellular models (Lei et al, 2004; Lei et al, 2008) could also contribute to thyroid tumors in $TR\beta^{PV/PV}$ mice.

A future challenge will be to elucidate whether, in addition to thyroid cancer, these novel modes of action of TR mutants could underlie the development and progression of tumors in other tissues where TR mutations have been reported, such as the pituitary gland and the mammary gland. Of interest, other studies have reported the interaction of wild-type TR β 1 or TR α with p85 α to activate PI3K signaling pathway in pancreatic beta cells, human fibroblasts, vascular endothelial cells, indicating that this mode of PI3K regulation by TRs may well occur in a number of T3-target tissues to regulate key cellular events (Cao et al., 2005; Hiroi et al., 2006; Verga Falzacappa et al., 2007). Given that context, it may be hypothesized that TR mutations could significantly affect PI3K activity and contribute to the development of pathologies. Within that framework, the $TR\beta^{PV/PV}$ mouse model should help advance understanding of the possible effect of TR mutation on the development of human cancer.

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Abbreviations

NCoR	nuclear receptor co-repressor
PI3K	phosphatidylinositol 3 kinase
PPARγ	

	peroxisome proliferator-activated γ
PTEN	phosphatase and tensin homolog deleted from chromosome 10
PTTG	pituitary tumor transforming gene
RTH	resistance to thyroid hormone
RXRα	retinoid X receptor α
SRC-3	steroid receptor co-activator-3
T3	thyroid hormone
TR	thyroid hormone receptor
TSH	thyroid stimulating hormone

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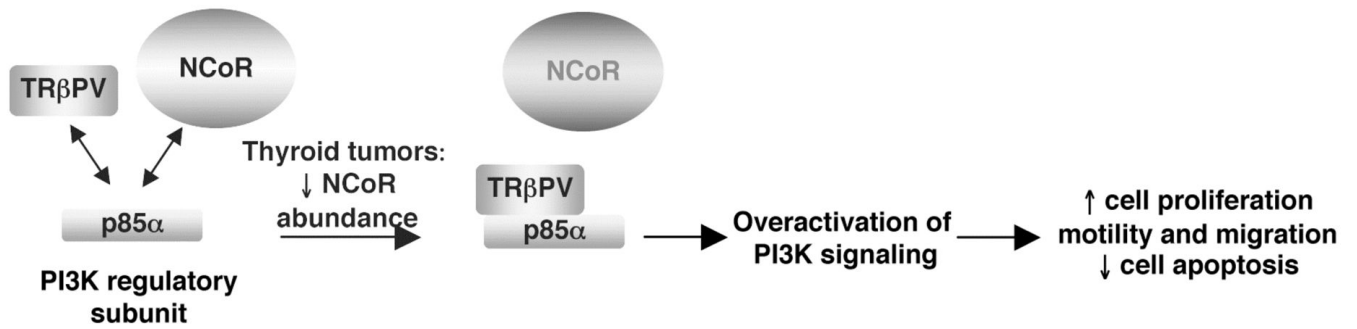


Figure 2. Proposed model for the overactivation of PI3K signaling by TRβPV in the thyroid cancer of $TR\beta^{PV/PV}$ mice

TRβPV physically interacts with the regulatory subunit of PI3K, p85α, to activate PI3K signaling, thereby affecting cell proliferation, motility, migration, and apoptosis (Furuya et al., 2006). TRβPV competes with NCoR to interact with p85α, but in thyroid cancer of $TR\beta^{PV/PV}$ mice, NCoR protein abundance is lower than in normal thyroid, thus favoring the physical interaction of TRβPV and p85α and the overactivation of PI3K signaling (Furuya et al., 2007b).

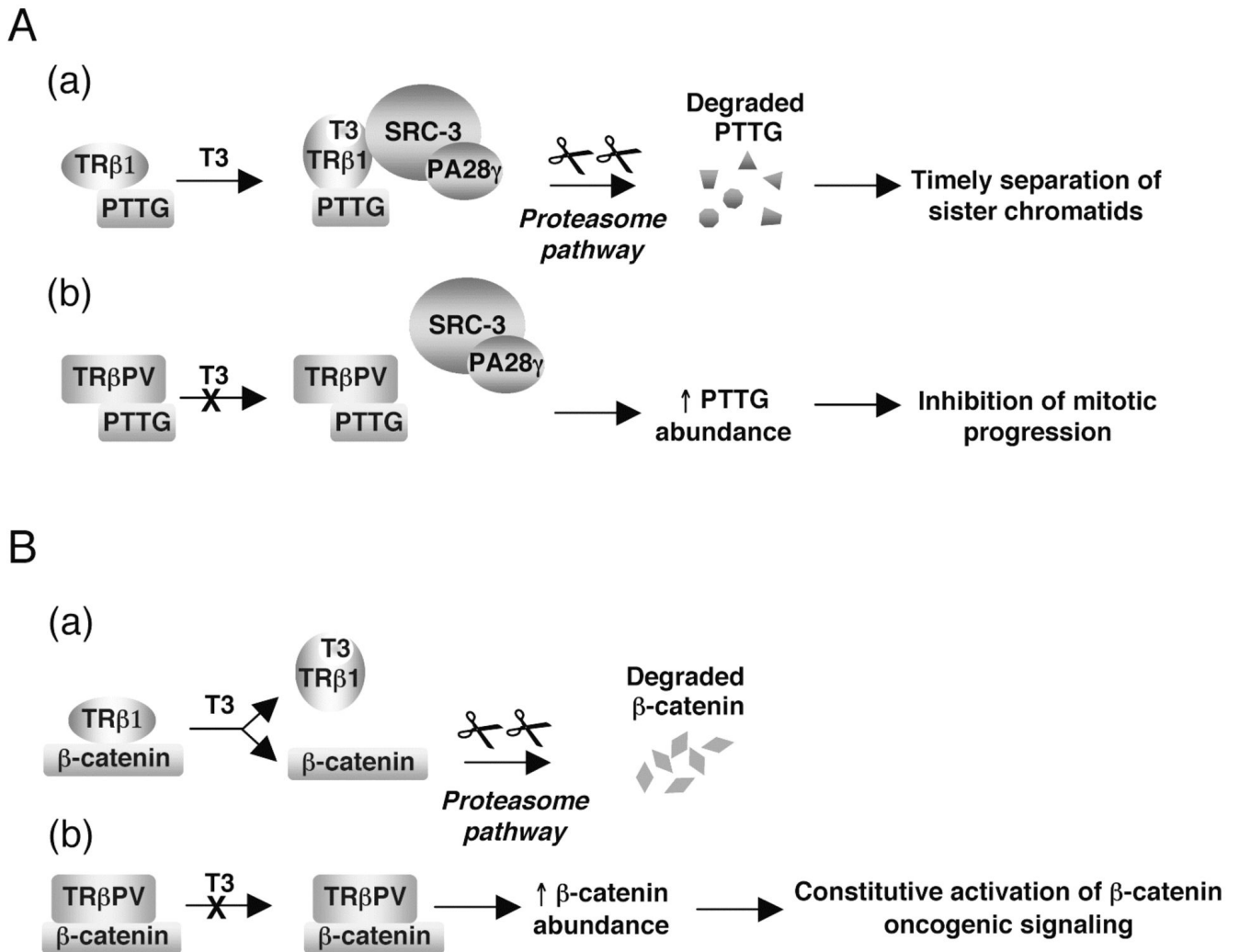


Figure 3. TRβPV, via protein-protein interaction, interferes with proteasomal pathway regulatory function

A) A proposed molecular model for the accumulation of PTTG protein in the thyroid cancer of $TR\beta^{PV/PV}$ mice (a) The direct interaction of TRβ with SRC-3 induced by the binding of T3 induces the formation of TRβ/PTTG/SRC3/PA28γ complexes that activate the degradation of PTTG and lead to timely separation of sister chromatids at anaphase. (b) TRβPV physically interacts with PTTG but as it does not bind T3, it does not interact with SRC-3/PA28γ to activate PTTG degradation. The absence of degradation leads to PTTG accumulation and inhibition of mitotic progression (Ying et al., 2006).

B) A proposed molecular model for the accumulation of β-catenin in the thyroid cancer of $TR\beta^{PV/PV}$ mice. (a) The physical interaction of TRβ with β-catenin is weakened by T3 binding and favors the degradation of β-catenin by proteasomal pathways. (b) TRβPV strongly interacts with β-catenin and the loss of its T3 binding activity inhibits the release of β-catenin from TRβPV/β-catenin complexes in the presence of T3, thereby preventing β-catenin degradation. The absence of β-catenin degradation leads to β-catenin accumulation and constitutive activation of its oncogenic signaling (Guigon et al., 2008).