Journal of Steroid Biochemistry & Molecular Biology 155 (2016) 190–198

Contents lists available at ScienceDirect

Journal of Steroid Biochemistry & Molecular Biology

journal homepage: www.elsevier.com/locate/jsbmb

Organotin compounds cause structure-dependent induction of progesterone in human choriocarcinoma Jar cells

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ARTICLE INFO

Article history:
Received 31 March 2014
Received in revised form 10 October 2014
Accepted 14 October 2014
Available online 18 October 2014

Keywords:
Tributyltin (TBT)
Triphenyltin (TPT)
3β-Hydroxysteroid dehydrogenase type I
Placenta
PPARγ

ABSTRACT

Organotin compounds, such as tributyltin (TBT) and triphenyltin (TPT), are typical environmental contaminants and suspected endocrine-disrupting chemicals because they cause masculinization in female mollusks. In addition, previous studies have suggested that the endocrine disruption by organotin compounds leads to activation of peroxisome proliferator-activated receptor (PPAR)γ and retinoid X receptor (RXR). However, whether organotin compounds cause crucial toxicities in human development and reproduction is unclear. We here investigated the structure-dependent effect of 12 tin compounds on mRNA transcription of 3β-hydroxysteroid dehydrogenase type I (3β-HSD I) and progesterone production in human choriocarcinoma Jar cells. TBT, TPT, dibutyltin, monophenyltin, tripropyltin, and tricyclohexyltin enhanced progesterone production in a dose-dependent fashion. Although tetraalkyltin compounds such as tetrabutyltin increased progesterone production, the concentrations necessary for activation were 30–100 times greater than those for trialkyltins. All tested active organotins increased 3β-HSD I mRNA transcription. We further investigated the correlation between the agonistic activity of organotin compounds on PPARγ and their ability to promote progesterone production. Except for $DBTCl_2$, the active organotins significantly induced the transactivation function of PPARγ. In addition, PPARγ knockdown significantly suppressed the induction of mRNA transcription of 3β-HSD I by all active organotins except $DBTCl_2$. These results suggest that some organotin compounds promote progesterone biosynthesis *in vitro* by inducing 3β-HSD I mRNA transcription via the PPARγ signaling pathway. The placenta represents a potential target organ for these compounds, whose endocrine-disrupting effects might cause local changes in progesterone concentration in pregnant women.

1. Introduction

Organotin compounds, such as tributyltin (TBT) and triphenyltin (TPT), have been used widely as antifouling biocides for ships and fishing nets [1]. There are many reports of the biological effects of organotin compounds, which vary in their toxic effects on eukaryotes. One of the most notable toxicities in sexual development and reproduction is that of TBT- and TPT-mediated endocrine disruption in some species of gastropods [2]. This phenomenon is known as 'imposex'—the superimposition of male genitalia on female animals. Therefore, these organotin compounds are suspected to cause endocrine-disrupting effects in mammals, including humans. Human exposure to organotin compounds may result from consumption of organotin-contaminated meat and fish products or occupational exposure during the manufacture and formulation of organotin compounds or the

Abbreviations: 3β-HSD I, 3β-hydroxysteroid dehydrogenase type I; CL, corpus luteum; DBT, dibutyltin; DMSO, dimethyl sulfoxide; FCS, fetal calf serum; hCG, human chorionic gonadotropin; GR, glucocorticoid receptor; LG, LG100268; LUC, luciferase; MEM, minimal essential medium; PPAR, peroxisome proliferator-activated receptor; RXR, retinoid X receptor; RT-PCR, reverse transcription polymerase chain reaction; Rosi, rosiglitazone; siRNA, small interfering RNA; TBT, tributyltin; TPT, triphenyltin.

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http://dx.doi.org/10.1016/j.jsbmb.2014.10.010
0960-0760/

Table. 1
Tin compounds tested in current study.

Tin compounds	Abbreviation	Purify (%)	CAS No.	MW[a]	ClogP[b]	Source
Trimethyltin chloride	TMTCl	>98	1066-45-1	199.3	−0.51	Aldrich Chemicals
Triethyltin bromide	TETBr	>97	2767-54-6	285.8	1.27	Aldrich Chemicals
Tripropyltin chloride	TPrTCl	>98	2279-76-7	283.4	2.66	Merck
Tributyltin chloride	TBTCl	>95	1416-22-0	325.5	4.25	Tokyo Kasei Kogyo
Triphenyltin chloride	TPTCl	>95	639-58-7	385.5	3.57	Aldrich Chemicals
Tricyclohexyltin hydroxide	TChTOH	>99	13121-70-5	385.2	5.39	Aldrich Chemicals
Trioctyltin hydride	TOTH	>95	869-59-0	459.4	10.3	Tokyo Kasei Kogyo
Butyltin trichloride	$MBTCl_3$	>95	1118-46-3	282.2	0.41	Aldrich Chemicals
Dibutyltin dichloride	$DBTCl_2$	>97	683-18-1	303.8	1.56	Tokyo Kasei Kogyo
Tetrabutyltin	TeBT	>93	1461-25-2	347.2	10.0	Aldrich Chemicals
Phenyltin trichloride	$MPTCl_3$	>98	1124-19-2	302.2	0.77	Aldrich Chemicals
Diphenyltin dichloride	$DPTCl_2$	>96	1135-99-5	343.8	2.06	Aldrich Chemicals

[a] MW represents the molecular weight.
[b] ClogP represents the calculated logPow.

application and removal of organotin-containing paints [3]. The possible exposure of humans to organotins therefore has prompted great concern about potential toxicities.

The placenta plays a vital role in maintaining pregnancy. The production of steroid hormones, such as progesterone and estrogens, is a crucial function of the primate placenta. In humans, by 7 wk of gestation, nearly all progesterone and estrogens in circulation are synthesized by the placenta [4]. In human placenta, steroid biosynthesis is regulated by various steroidogenic enzymes. The enzyme 3β-hydroxysteroid dehydrogenase/isomerase (3β-HSD) catalyzes the conversion of 3-hydroxy-5-ene-steroids (dehydroepiandrosterone and pregnenolone) to 3-oxo-4-ene-steroids (androstenedione and progesterone) [5]. The 3β-HSD enzymes exist in multiple isoforms in humans and rodents. Among these isoforms, type I (3β-HSD I) is expressed exclusively in the placenta. It converts pregnenolone to progesterone to help maintain the uterus in a quiescent state throughout human pregnancy [5]. Whereas placental production of progesterone is required to protect the conceptus during midgestation onward [6], the ingestion of progestins (i.e., natural and synthetic progesterone and testosterone derivatives that produce biologic effects similar to those of progesterone) during pregnancy is associated with an increased risk of hypospadias [7]. Given the pivotal functional roles of 3β-HSD I, the developmental and reproductive toxicities of environmental contaminants known to have endocrine-disrupting effects plausibly might involve placental 3β-HSD I in humans.

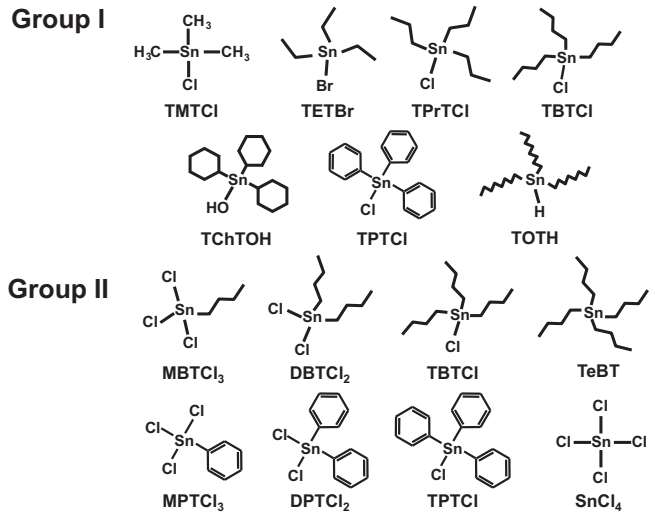

Fig. 1. Structure of tin compounds in the current study.
Group I, comparison of different structures of alkyl and aryl chains in trialkylated and triarylated tin compounds. Group II, comparison of different numbers of alkyl or aryl chains in butyltin and phenyltin compounds. The abbreviation for each compound used is indicated in Table 1.

In a previous study, we demonstrated that exposure to nontoxic concentrations of some organotin compounds dose-dependently increased the mRNA transcription and production of estradiol to increase the catalytic activity of aromatase, which converts androgen to estrogen, and of 17β-HSD I, which converts low-activity estrone to high-activity estradiol [8–10]. We also identified that TBT and TPT act as nanomolar agonists for both the retinoid X receptor (RXR) and peroxisome proliferator-activated receptor (PPAR)γ, which are members of the nuclear receptor superfamily [11]. The promotion of estrogen biosynthesis by the organotin compounds just described involves the activation of RXR rather than PPARγ [9,10].

PPARγ is expressed abundantly in human trophoblast cells and serves as an essential regulator of placental differentiation and endocrine functions [12]. PPARγ is activated by a variety of fatty acids and by a class of synthetic antidiabetic agents, the thiazolidinediones [13]. PPARγ regulates the transcription of genes by heterodimerizing with RXR and by binding to the PPAR response elements in the target gene promoter [14]. Human chorionic gonadotoropin (hCG) is a crucial target gene of PPARγ in human placenta, and its production and mRNA transcription is ligand-dependently controlled by PPARγ [12]. hCG is a luteotropic factor, and its stimulation by hCG governs not only progesterone production in the corpus luteum (CL) during the first trimester [15] but also testosterone production within the fetal testes [16]. Given the pivotal functional roles of hCG in development and reproduction, factors that change PPARγ-mediated transcription in the placenta may greatly alter fetal development by disrupting these endocrine functions. Indeed, we found that some organotin compounds, including TBT and TPT, promote hCG production [11]. Therefore, PPARγ is a crucial molecular target of organotin compounds in the endocrine disruption of human placenta.

To facilitate the application of current knowledge regarding the toxicity of organotin compounds to development and reproduction in humans via the PPARγ signaling pathway, we assessed the possible effects of 12 tin compounds on the production of progesterone and mRNA transcription of 3β-HSD I in human placental cells by using human choriocarcinoma Jar cells. Furthermore, we investigated the correlation between the potency of these compounds as agonists for PPARγ and progesterone production in Jar cells, and we addressed the potential toxicity of organotin compounds as endocrine disruptors in humans.