# CHANGES IN THYROXINE GLUCURONIDATION AND TOTAL SERUM THYROXINE IN RATS TREATED WITH POLYCHLORINATED BIPHENYLS AND DIBENZOFURANS

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## Introduction

Polyhalogenated aromatic hydrocarbons (PHAHs) such as polychlorinated dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs), and biphenyls (PCBs) are persistent environmental pollutants that induce a broad spectrum of toxic effects in animals[1,2,3]. Serum thyroid hormone (TH) decreases have been observed in rats exposed to PHAHs[4,5,6]. Circulating T4 can be decreased through several pathways including synthesis inhibition, alterations in serum transport, and alterations in catabolism. PHAHs are thought to alter serum T4 concentrations by the induction of hepatic T4-glucuronidation. The increase in T4 catabolism is due to induction of UDPGT isoforms through a Ah receptor mediated pathway or a phenobarbital-like response. The present study focuses on the decrease of total serum T4 concentrations and its relationship to induction of hepatic glucuronidation of T4 for a series of PCBs and PCDFs

## Material and Methods

Animals and Treatment: Female Long Evans rats (25 days old) were obtained from Charles River Breeding Laboratories, Raleigh, NC. The animals were housed under controlled conditions. . Water and food were given ad libitum. The animals were dosed by oral gavage for 4 days with the following compounds PCB 126 (.03,.1,.3,1,3,10,30, and 100 ug/kg/day), PCB 105 (.09,.3,.9,3,9,30,90 mg/kg/day), PCB 118 (.03,.1,.3,1,3,10 mg/kg/day), TCDF (.3,1,3,10,30,100 ug/kg/day), 1-PCDF (.3,1,3,10,30,100 ug/kg/day), 4-PCDF (.03,.09,.3,.9,3,9,30,90 ug/kg/day). The animals were killed 24 hours after the final dose. Serum and livers were collected and stored at $-80^0$C. Liver microsomal fractions were prepared as described by DeVito et al, 1993[7]

T4 Assay: Total serum T4 was determined according to the method described by Goldey et al, 1995[8], via an 125I radio-immunoassay (Coat-a-Count kit, Diagnostic Products, Inc.) Samples were counted in gamma counter and calibrated according to standards provided with the kit.

UDPGT Assay: T4 glucuronidation was determined using a modification of the method of Visser et al, 1993[5]. Briefly, 2 mg/ml liver microsomes were incubated 30 minutes with 1 uM T4, 50,000 cpm of the $^{125}$I –labeled compound, in the presence or absence (blank) of 5mM UDPGA in .2 ml 100 mM Tris/HCl (pH 7.8) and 5 mM $MgCl_2$. 1 mM

of 6-Propyl-2-thiouracil was added to prevent deiodination. The reaction is stopped with 0.2 ml of ice cold methanol. The mixture is then centrifuge and 0.2 ml of supernatant was removed and placed in .750 ml of .1 M HCl. The samples are then analyzed by chromatography on Sephadex LH-20 columns

**Statistical Analysis:** Total Serum T4 and UDPGT activity was analyzed independently using a one-way analysis of variance (ANOVA).

Figure 1

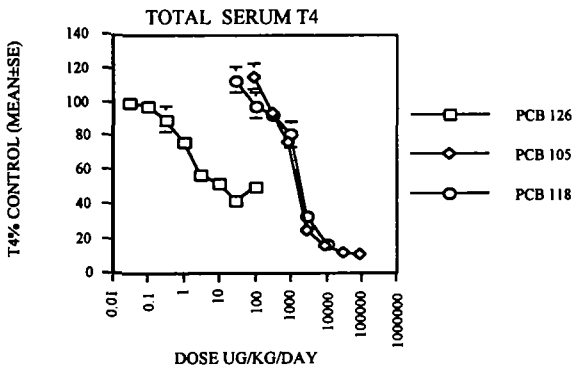



Figure 2

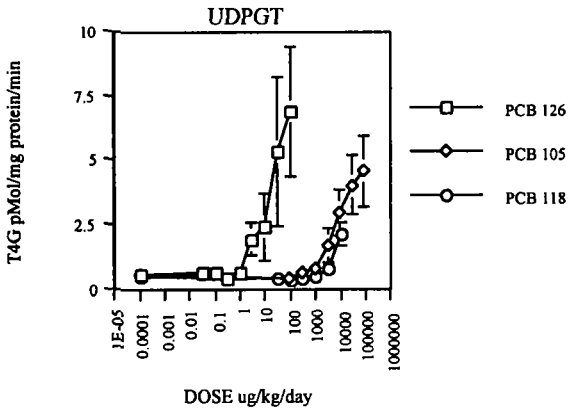


Figure 3

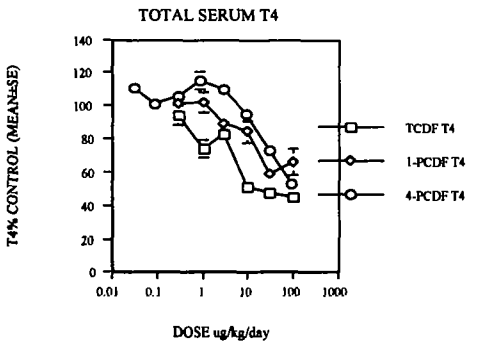
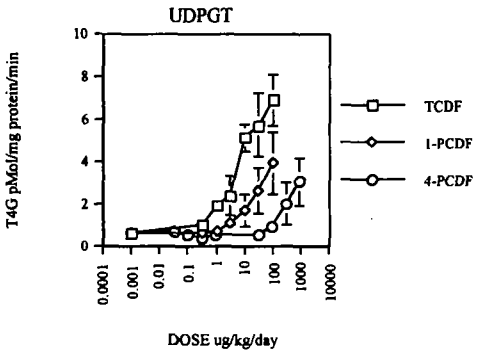



Figure 4




## Results

Analysis of total serum T4 in rats exposed to PCBs 126, 105, and 118 respectively resulted in dramatic decreases. PCB 118 and PCB 105 (Figure 1) produced decreases of up to 85% at the highest doses examined (10 and 90 mg/kg/day). PCB 126 decreased T4 concentrations by 50% at the highest dose examined. Conversely, PCB 126 induced T4-glucuronidation by 13 fold (Figure 2) compared to an 8- and 4-fold induction by PCBs 105 and 118, respectively. The decrease in total serum T4 was not as pronounced in the PCDFs compared to the mono-ortho PCBs. TCDF and 4-PCDF decreased serum total T4 by approximately 50% at the highest doses, while 1-PDCF decreased serum total T4 by only 34% at the highest doses (Figure 3). TCDF produce an 11-fold induction of UDPGT activity at the highest doses examined (Figure 4). 1-PCDF and 4-PCDF gave induced T4-glucurondiation by approximately 6-fold highest doses examined.

## Discussion

All six compounds produce significant decreases in total serum T4 and increases of UDPGT activity. However, no direct relationship was observed between the magnitude in the decreases in serum T4 and the amount of T4-glucuronidation. The most dramatic decreases in total serum T4 occurred in the PCB 105 and 118 exposed animals (>50%). Rats exposed to PCB 126 or the PCDFs had decreases in serum T4 of 50% or less. In contrast, induction of T4-glucuronidation was greatest in rats exposed to PCB 126 and

TCDF. These data suggest that T4-glucuronidation may not fully account fully for the reduction of circulating T4 by the mono-ortho PCBs. Other mechanisms may be involved in the decreases observed with the mono-ortho PCBs. Alternatively, the UDPGT assay may not correctly discriminate between different isoforms of UDPGT and may not reflect *in vivo* conditions.

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