Rationale for Considering Neurotoxic Activities of *Ortho*-Substituted Non-planar PCBs in Risk Assessment

Debdas Mukerjee, National Center for Environmental Assessment, United States Environmental Protection Agency, Cincinnati, OH 45268 USA

Abstract

Ortho-substituted non-coplanar PCBs having weak/no Ah receptor activity are more prevalent in breast milk of mothers, food products, and human tissue samples from industrial countries than coplanar PCBs having high Ah receptor activity. Epidemiological and experimental animal studies suggest that *in utero* and lactational exposures to PCBs may be associated with neurological and behavioral dysfunction. Since current PCB risk assessment involves the usage of toxic equivalency factors derived from Ah receptor activity of chemicals, careful attention must be paid for the role of non-coplanar PCBs in the PCB risk assessment process.

Introduction

Residues of numerous chemicals, including environmental and occupational contaminants and pharmaceutical drugs, have been detected in human breast milk. Because breast milk is the sole source of nutrition and provides complete nutrition to the infant, the composition of the breast milk is of crucial importance. Polychlorinated biphenyls (PCBs) are widely distributed in breast milk of mothers from industrial countries and their levels are about 10,000 times higher than the total concentrations of polychlorinated -dibenzodioxins and -dibenzofurans [1]. Because of their greater prevalence in the environment, ortho-substituted non-coplanar PCBs are more widespread in human milk than coplanar PCBs [2,3]. Considerable concern has surfaced recently that PCBs and related halogenated aromatic hydrocarbons can induce neurological and behavioral dysfunctions in animals and humans[4]. Neurotoxic effects are claimed to have been caused in ecosystems and exposed human populations by these chemicals [5,6].

Intakes of fish, meat and dairy products are the major source of exposure to PCBs in humans. Because of their lipophilicity and persistence, residues of PCBs or their metabolites tend to bioaccumulate in human adipose tissues including mammary tissue. *Ortho*-substituted non-planar congeners, PCB-138, -153, and -180 represent more than 60% [7]; PCB-22, -52,-101,-138, -153, and -180 represent 62.6% [8] and PCB-153 constitutes 30% of the total PCBs in breast milk [9].

The average half-life of PCBs in the blood of offspring of occupationally exposed mothers is 2 - 6 years [10]. PCBs have been detected in the sera [11], adipose tissues [12,13] and bone marrow [14] of breast-fed children. The level of PCBs in infants' blood is influenced by the duration of

lactational exposure. *Ortho*-substituted PCBs have been detected in the brains of rat pups of Aroclor 1254 exposed dams [15].

Effects Associated with PCB Exposed Infants

At high concentrations, PCBs may be associated with neurological disorders [6] including long-term effect on intellectual function of offspring [5,16]. Effects of neonatal exposures to these chemicals are often manifested during the pubertal period. Higher than expected rates of low normal I.Q. scores, poor reading comprehension, memory problems and difficulty in paying attention were reported in 11-year old children who were exposed to PCBs [5]. Although it has also been suggested that *in utero* PCB exposure may have a small negative effect on the neurological condition of the 18- month old toddlers [17], *in utero* PCB exposed mice have been reported to have altered motor function [18,19].

Mechanisms of Neurotoxic Activities of PCBs

The mechanism of neurotoxic effects of PCBs has not yet been conclusively elucidated, but may involve alteration in basic endocrine function that controls neurotransmitter and the organization of the developing brain. Increased exposure to this class of compounds, through intrauterine and breast milk, can affect thyroid function in newborn human infants [20]. Thyroid hormone effects are mediated by all types of PCBs and several different mechanisms are involved. Alteration of normal thyroid physiologic homeostasis is associated with myriad of clinical conditions including developmental, growth, metabolic, neurologic and behavioral functions. Abnormalitites of the thyroid functions are associated with loss in speech, hearing, memory and motor coordination [21]. Irregular thyroid hormone status associated with ototoxicity has been observed during developmental PCB exposure [22]. PCBs affect thyroid activity by increasing development of the endoplasmic reticulum, vacuolization of mitochondria and decreasing the colloidal droplet lysosome interaction [23]. PCBs can mobilize T_4 at doses lower than those causing significant enzyme induction [24]. Non-planar PCB metabolites have greater affinities for the T_4 transport protein, prealbumen (transthyretin). This displacement makes T_4 more vulnerable to metabolism and excretion. Non-planar PCBs can reduce serum T_4 but not T_3 levels. T_4 level is decreased in rat offspring of PCBs exposed dams with normal circulating T_4 during lactation [25]. The induction of phase 2 enzymes, particularly UDP-glucuronyl transferase (UDP-GT), can deplete the T_4 levels by significantly increasing thyroxine metabolism and excretion [26,27]. Non-planar PCB induced T_4 elimination may be due to induction of phase 2 enzymes which enhances bile flow [28]. Hypothyroidism can activate transformation of T_4 to T_3 in the central nervous system cells [29]. This decrease in T_4 during active growth and development of the organs including brain may be responsible for PCB induced functional neurological impairments [30]. Levels of brain neurotransmitter including dopamine can be decreased in prenatally exposed PCBs rodents [31-33] and *ortho*-substituted non-planar congeners are more potent than meta- or para-substituted planar congeners in decreasing dopamine levels in PC12 cells [34]. Uptake and release of brain neurotransmitter including dopamine are controlled by intracellular Ca^{2+} homeostasis, which can be altered in cerebellar granule cells by *ortho*-substituted non-planar congeners [35]. Only ortho-substituted non-planar PCBs have been found to alter intracellular Ca^{2+} -dependent enzymes, including protein kinase C translocation [36]. Endocrine effects including thyroid hormone

disturbances, enzyme induction and abnormal calcium homeostasis seem to be interrelated and are involved in neurotoxic activity of *ortho*-substituted non-planar PCBs.

Exposure and Risk Assessments of Ortho-substituted Non-planar PCBs of Breast-fed Infants

The average level of total PCBs in human milk fat from industrial countries has been suggested to be between 0.5 and 4 μg/g, but the levels are decreased to between 0.5 and 1.5 μg/g if a dozen peaks are used for quantitation [1]. Daily ingestion rate of breast milk for a 5 kg infant is 850 ml and there is 4.5 gm of fat in 100 ml of breast milk [37]. Therefore, a 5 kg infant ingests 38.2 gm fat in breast milk/day. It is reasonable to assume that almost 100% of the ingested PCBs from mothers' milk are absorbed by the infant [38].

The average level of PCBs in human milk fat, in the worst case scenario, is 4 μg/g. 62.6% of the total PCBs in breast milk is ortho-substituted non-planar PCB-22, -52, -138, -153, and -180. [8], the corresponding concentration of ortho-substituted non-planar PCB-22, -52, -138, -153, and -180 is 2.5 μg/g. A 5 kg infant ingests (38.2 gm fat/d X 2.5 μg/g) 95.6 μg/day of *ortho*-substituted non-planar PCB-22, -52, -138, -153, and -180. In other words, *ortho*-substituted non-planar PCBs exposure to this child via mother's milk is 19.1 μg/kg.bw/day.

There is a glaring gap in knowledge concerning the effects of non-planar PCBs not associated with the aryl hydrocarbon receptor, including actions on immature animals, qualitative and quantitative influence of enzyme induction, and subsequent metabolism of the toxicant. Furthermore, little is known regarding their dose-dependant health effects on children. Because of their potency as enzyme inducers and the demonstrated biological actions of the resultant metabolites, low-dose effects of non-planar PCBs are frequently dramatically different from high-dose effects. Consequently, modest exposure may result in different effects than high dose exposure because of multiple actions of individual chemicals. Disruption of normal homeostasis of the thyroid by PCBs is an important consideration in risk assessment. Accuracy of risk assessment has been elusive due to the complexity of actions and interactions of PCB mixtures. No work has been yet done to determine the risk of newborn infants nursed by mothers whose milk contains high levels of these pollutants. For an accurate assessment of risks, models of neurotoxicity need to integrate dose dependent endpoints in multiple neurotoxic and enzymatic systems *in vivo*. There are some *in vitro* assays which are used for detecting neurotoxic activity of *ortho*-substituted non-coplanar PCBs. While screening tests involving *in vitro* methodologies are useful, *in vivo* animal dose response bioassay is more relevant for quantitative risk assessments . There is an urgent need to develop dose response data base on neurotoxic effects of *ortho*-substituted non-coplanar PCBs such that quantitative risk assessments be performed for taking public health preventive measures.

Literature Cited

(1). Jensen, A.A. *Sci.Total Environ.* **1987**, 64:259-293.
(2). Norén, K.; Lundén, A., Sjövall, J., Bergman, A. *Chemosphere* **1990,** 20:935-941.
(3). Norén, K.; Lundén, A. Organohalogen Compds. **1990,** 1:263-266.
(4). Seegal, R.F. *Crit. Rev. Toxicol.* **1996,** 26:709-737.
(5). Jacobson, J.L.; Jacobson, S.W. *Toxicol. Ind. Hlth.* **1996,** 12:435-445.
(6). Rogan, W.J.; Gladen, B.C.; Hung, K-L.; et al. Science **1988,** 241:334-336.
(7). Schultz, E.; Malisch, R. Z. *Anal. Chem.* **1984,** 319:54-59.
(8). Abraham, K.; Alder, L.; Beck, H. et al. *Organohalogen Comps.* **1995,** 26: 63-67.
(9). Schoula, R.; Hajslova, J.; Benko, V.; et al. *Chemosphere* **1996.** 33:1485-1494.
(10). Shirai, J.H.; Kissel, J.C. *Sci. Total Environ.* **1996,** 187:199-210.
(11). Schantz. S. L.; Jacobson, H.E.B.; Humphry, S.W. et al. *Arch. Environ. Hlth.* **1994,** 49: 452-458.
(12). Niessen, K.H.; Ramolla, J.; Binder, G.; et al. *Eur. J. Ped.* **1992,** 142:238-243.
(13). Teufel, M.; Niessen, K.H.; Sartoris, J.; et al. *Arch. Environ. Contam.Toxicol.* **1990,** 19: 646-652.
(14). Scheele, J.M.; Teufel, M.; Niessen, K.H. *Environ. Path. Toxicol. & Oncology.* **1995,** 14: 11-14.
(15). Shain, W.; Overmann, S.R.; Wilson, L.R.;et al. *Arch.Environ.Contam.Toxicol.* **1986,** 15: 687-707
(16). Rogan, W. J.; Gladen, B. *NeuroToxicol.* **1992,** 749-764.
(17). Huisman, M.; Koopman-E-C.; Lanting, C.I. et al., *Early Human Dev.* **1995,**43:165-176.
(18). Tilson, H.A.; Davis, G.J.; McLachlan, J.A.; Lucier, G.W. *Environ. Res.* **1979,** 18:466-474.
(19). Pantaleoni, G.; Fannini, D.; Sponta, A.M.; et al. *Fundam. Appl. Toxicol.* **1988,** 11:440-449.
(20). Pluim, H.; Koppe, J.G.; Olie, K.; et al. *The Lancet* **1992**, 339:1303.
(21). Porterfield, S.P. *Environ. Helath Perspect.* **1994,** 102(Suppl.2):125-130.
(22). Goldey, E.S. *Tox. Appl. Pharm.* **1995,** 135:77-88.
(23). Collins, W.T.; Capen, C.C. *Amer.J. Path.* **1980,** 99:125-142.
(24). Hansen, L.G.; Li, M-H.;Saeed, A. *Arch. Environ. Contam. Toxicol.* **1995**, 29:334-343.
(25). Ness, D.K.; Schantz, S.L.; Moshtaghhian, J.; Hansen, L.G. *Tox. Lett.* **1993,** 68:311-323.
(26). Bastomsky, C.H.; Murthy, P.V.N.; Banovac, K. *Endocrinology* **1976,** 98:1309-1314.
(27). Brouwer, A. *Aquatic Toxicol.* **1989,** 15:99-106.
(28). Roberts, R.J.; Plaa, G.L. *Biochem. Pharmacol* **1969,** 16:827-829.
(29). Köhrle, J.; Brabant, G.; Hesch, R.D. *Horm. Res.* **1987**, 26::58,.
(30). Hendrich, C.E.; Jackson, W.J.; Porterfield, S.P. *Neuroendocrionol* **1984**, 38:429-437.
(31). Agarwal, A.K.; Tilson, H.A.; Bondy, S.C. *Toxicol. Lett.* **1981**, 7:417-424.
(32). Seegal, R.F.; Brosch, K.O.; Bush, B. *Toxicol. Lett.* **1986**, 30:197-202.
(33). Seegal, R.F.; Bush, B.; Brosch, K.O. *NeuroToxicol.* **1991**, 12:55-66.
(34). Shain, W.; Bush, B.; Seegal, R. *Tox. Appl. Pharm.* **1991,** 111:33-42.
(35). Kodavanti, P.R.S.; Shin, D-S.; Tilson, H.G.; Harry, G.J. *Tox. Appl. Pharm.* **1993**, 123: 97-106.
(36). Kodavanti, P.R.S.; Ward, T.R.; McKinney, J.D.; Tilson, H.A. *Arch. Toxicol.* **1996,** 70: 150-157.

(37). ICRP (International Commission on Radiological Protection). Report of the Task Group on Reference Man. Pergamon Press, New York, 1975.
(38). McLachan, M.S. *Tox. Appl. Pharm.* **1993**, 68-72.

[This article has been reviewed by NCEA-Cin., US EPA and approved for publication. Approval does not mean that the contents necessarily reflect policies and views of the Agency]