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REVIEW

BISPHENOL A AND ITS POTENTIAL TOXIC EFFECTS ON LIVING ORGANISMS

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ABSTRACT

Bisphenol A (BPA), 2, 2,-bis (4-hydroxyphenyl) propane, CAS No. 80-05-7, is an industrial chemical that is made by combining acetone and phenol. Food is acknowledged to be a main source of exposure to BPA as a consequence of BPA migration from food containers. Several studies have reported BPA migrations from can surface coatings or plastics into foods and food-simulating liquids at high temperatures**,** and with repeated use of plastic products. Evaluation of BPA confirmed a no-observable adverse effect level (NOAEL) of 5 mg/kg/body weight/day and established a maximum total daily intake (TDI) of 0.05 mg/kg body weight. A metabolism of BPA is characterized mainly by phase II conjugation reactions in the gastrointestinal tract and in the liver. The presence of BPA in the environment can cause serious health problems (endocrine disruptions, neurotoxic, genotoxic and other problems). However, there are controversial opinions about BPA. Based on current knowledge of literature the need for further experimental studies in addressing health of human and animal populations living in different ecosystems may be still useful.

Keywords: bisphenol A, environmental exposure, toxic effects, health

Bisphenol A - characterization and exposures

Bisphenol A (BPA), 2, 2,-bis (4-hydroxyphenyl) propane, CAS No. 80-05-7, is an industrial chemical that is made by combining acetone and phenol (Figure 1). It is extensively used as a monomer in the production of polycarbonate plastics and as a precursor of epoxy resins. Polycarbonate (PC) is widely used in the manufacture of food containers (e.g., milk, water and infant bottles) and epoxy resins are used as the interior protective lining for food and beverage cans. As a result of these food contact uses, minute quantities of BPA can potentially leach out into water or food and consumers may be exposed to BPA through the diet **(ECB, 2008; Aschberger *et al.*, 2010)**.

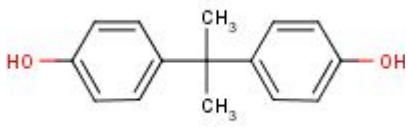

Figure 1 Bisphenol A – 2, 2,-bis (4-hydroxyphenyl) propane**:** Synonyms: BPA; 4,4'-(1-Methylethylidene) bisphenol; 4, 4'-Isopropylidenediphenol; BPA

Food is acknowledged to be a main source of exposure to bisphenols as a consequence of BPA migration from food containers. Several studies have reported BPA migration from can surface coatings or plastics into foods and food-simulating liquids at high temperatures and with repeated use of plastic products (**Fernandez *et al.*, 2007**).

The European Food Safety Authority (EFSA) set a Specific Migration Limit of 3 mg BPA per kg food (3 ppm) for the protection of EU consumers. A recent re-evaluation of BPA confirmed a no-observable adverse effect level (NOAEL) of 5 mg/kg body weight/day and established a maximum total daily intake (TDI) of 0.05 mg/kg body weight **(EFSA, 2008; Hengstler *et al.*, 2011)**. This TDI was based on body weight changes in two- and three-generation studies in mice and rats. These studies and the derivation of the TDI were critized. The Committee of the German Society of Toxicology concluded that the current TDI for BPA is adequate and that available evidence indicates that BPA exposure represents no noteworthy risk to the health of human including newborn and babies **(Hengstler *et al.*, 2011)**.

The presence of BPA in the environment can cause serious health problems. However, there are controversial opinions. For example, BPA has been considered a weak environmental oestrogen, it binds to the oestrogen receptors, α and ß with an affinity being about 10,000 to 100,000-fold weaker than that of 17 ß-oestradiol (**Pennie *et al.*, 1998**). Other

studies with BPA showed the increased susceptibility to cancerous changes **(Jenkins *et al.*, 2009)**, the fluctuations in hippocampal neurogenenesis in young adult mice (**Kim *et al.*, 2011**), neurotoxic effects **(Le *et al.*, 2008)**, genotoxic effects **(Naik and Vijayalaxmi, 2009; Karim and Husain, 2010)** and other health problems **(Fernandez *et al.*, 2007)**.

The aim of this text was to prepare a short overview on the BPA effects, mainly about its various health problems in living creatures, especially about the adverse effects that are still controversial.

Metabolisms and toxicokinetics

The metabolism of BPA is of great importance for the assessment of its toxicity at the cellular and molecular levels as well as for understanding its bioavailability and biological properties. BPA has aqueous solubility (0.5 – 1.3 mmol/L), its metabolism to more water-soluble conjugates is extensive, probably reflecting its lipophilicity (log octanol/water partition coefficient = 2.2 – 3.4) **(Doerge and Fisher, 2010)**. Several metabolic pathways of BPA were observed. Metabolism of BPA is characterized mainly by phase II conjugation reactions in the gastrointestinal tract and in liver. During the first-pass metabolism, the major metabolic pathway involves chemical reaction, rapid conjugation by uridine diphosphate glucuronyltrasferases (UGT) to form BPA-glucuronide. Second minor metabolic pathway involves chemical reaction – sulphatation to form BPA-sulphate by sulfotransferase in rats and humans **(Pottenger *et al.*, 2000; Ye *et al.*, 2006; Mazur *et al.*, 2010)**. It is also proposed other minor metabolite, as BPA-3, 4-quinone **(Zalko *et al.*, 2003)**. The choice of animal models is very important for comparing for example BPA glucuronidation in animals and humans. In these studies there are also other very important parameters, as the mode of administration, dose of BPA, as determining the partitioning of BPA in different organs and tissues, or distribution ratio tissue/blood. After oral exposure to BPA in human body glucuronidation is realized through BPA-monoglucuronide metabolite, in the gut and liver. Enzymes UGT and their isoforms are localized in the endoplasmatic reticulum and catalyse lipophilic compounds as are BPA and others to charge them to water-soluble compounds, glucuronides **(Mazur *et al.*, 2010)**.

Bisphenol A – Endocrine Disruptor

According to the U.S. Environmental Protection Agency (U.S. EPA), endocrine disruptors are defined as the exogenous agents that interfere with the production, release, transport, metabolism, binding, action, or elimination of natural hormones in the body**,** responsible for the maintenance of homeostasis and the regulation of developmental processes. The definition is not limited to the oestrogen system. Endocrine disruption comprises effects on other endocrine systems, including effects associated by androgens, thyroid hormone, prolactin, and insulin, among others **(Wetherill *et al.,* 2007; Aschberger *et al.,* 2010)**. BPA is characterized as a weak environmental oestrogen and also has other endocrine disrupting activities that are mediated via multiple molecular mechanisms **(Alonso-Magdalena *et al.,* 2006; Bouskine *et al.,* 2009)**. Amphibian metamorphosis is dependent on tri-iodo-thyronine (T_3) and active form of thyroid hormones (TH). *Xenopus laevis* provides a useful model for studying BPA effects in vertebrates, as the main BPA metabolites are similar to those produced in mammalians **(Kloas and Lutz, 2006)**. This phenomenon used **Fini *et al.* (2009)**, when metabolism of BPA, a thyroid function disruptor, was investigated in *Xenopus laevis* tadpoles. Six peaks corresponding to BPA metabolites were detected. Their results were compared with standards isolated from rat and human material, and the two main metabolites were identified as BPA-glucuronide and BPA-sulphate. It should be noted, that experimental studies suggest that BPA adversely affects male reproductive system in general and testis in particular. **Nagel *et al.* (1997)** have reported a statistically significant increase in prostate weight of 30% at a BPA dose of 2µg/kg, and 35% increase was similarly noted in the male offspring of mothers dosed with 20µg/kg. They also mentioned a statistically significant decrease (20%) in daily sperm production per gram of testis at dose 20 µg/kg BPA. Reactive oxygen species (ROS) are implicated to be one of the key regulators of insulin signalling and glucose homeostasis in various organs. BPA is a potential testicular toxicant for the induction of oxidative stress and also potent diabetogenic agent. It is important to understand the effects of BPA on insulin signalling and glucose homeostasis in testis **(Ohta *et al.,* 2011; Jain *et al.,* 2011)**. The investigators studied the effects of low doses of BPA on insulin signalling molecules, glucose transporter-2 (GLUT-2) and stereoidogenesis in rat testis. BPA was administered to rats by oral gavages at dose of 0.005; 0.5; 50 and 500µg/kg body weight/day for 45 days. They mentioned a dose-dependent decrease in the activities of antioxidant enzymes, 3-ß-hydroxysteroid dehydrogenase and 17-ß-hydroxysteroid dehydrogenase. Steroidogenic acute regulatory protein and testosterone were also observed. From their

observations, they have concluded that BPA can directly interact with glucose transporter-2 and glucose transporter-8 and impair glucose transport. They reported that persistent exposure to BPA at low doses impairs insulin signalling mechanism, glucose homeostasis and steroidogenesis in testis. Oxidative stress and estrogenicity of BPA contribute to observed effects **(D´Cruz *et al.,* 2011)**.

Neurotoxicity of BPA

Nervous system is very important for all mental, sensory and motor activities, and regulates homeostasis through interaction with the endocrine system. Several neurotoxic endpoints in *in vivo* studies suggested that BPA treatment during development can cause alterations in brain **(Palanza *et al.,* 2008; Moser, 2010; Tian *et al.,* 2010). Palanza *et al.* (2008)** examined both male and female offsprings and found that maternal exposure to BPA affected: behavioural responses to novelty before puberty, and as adults, exploration and activity in a free-exploratory open field, exploration in the elevated plus maze and sensitivity to amphetamine-induced reward in the conditioned place preference test. Their findings are evidence of long-term consequences of maternal exposure to low dose BPA at the level of neurobehavioral development. The EU report **(ECB, 2008)** included review of all studies on the effects on neurological development following prenatal and perinatal exposure to BPA. The neurotoxicity endpoints were evaluated as locomotory and exploratory activity, cognitive, emotional, social, sexual and maternal behaviour, behaviour response to pharmacological challenge, brain morphology, immunohistochemistry and receptor/gene expression. **Aschberger *et al.* (2010)** noted, regarding on neurodevelopmental toxicity, that neural development has become the toxicological endpoint raising most concern for regulatory bodies. For a better understanding of the potential neurotoxicity risk of BPA, BPA should be studied in terms of validity and reliability of the test systems and for their relevance for effects on the behaviour and cognitive development of humans and relevant exposure and exposure routes.

Genotoxicity of BPA

Genotoxic studies of the BPA were realized using *in vitro* and *in vivo* evaluations with controversial results. **Bucher, (2010)** reported that BPA is not a mutagen in *in vitro* tests, nor does it induce cell transformation. BPA has been shown to affect chromosomal structure in *in*

vitro studies, but it is no evidence for *in vivo* studies. BPA is not likely to pose a genotoxic hazard to human. **Audebert *et al.* (2011)** investigated genotoxicity and cytotoxicity of BPA and bisphenol F (BPF) on human cells (*intestinal cell line: LS 174 T, hepatoma cell line: HepG2, and renal cell line: ACHN*) to biotransform BPA and BPF, through the use of a novel genotoxic assay based on the detection of histone H2AX phosphorylation. BPF exhibited an intermediary cytotoxicity, and BPA was most cytotoxic compound tested. BPA and one metabolite of BPF, dihydroxybenzophenone (DHB), were not found to be genotoxic in the cell line examined. Only BPF was clearly genotoxic in HepG2 cells. Their results showed that some human cell lines metabolize bisphenols and established the genotoxic potential of bisphenol F. Monomer BPA, BPA-glycoldyldimethacrylate (Bis-GMA), is used as biomaterial in dentistry. Researchers evaluated the potential toxicological implications of Bis-GMA on murine macrophage cell line RAW 264.7. Their results suggest that caspase-3, caspase-8 and caspase-9 activities play a critical role in Bis-GMA-induced cytotoxicity and genotoxicity *in murine macrophage cell line RAW 264.7 cells* **(Li *et al.*, 2012)**. With view to the continuing presence of different genotoxic substances in the environment the importance of using cytogenetic biomarkers to prevent health disturbances and produce ecological food is still increasing. *In vivo* genotoxic potential of BPA was studied by **Naik and Vijayalaxmi, (2009)** in mouse bone marrow cells using cytogenetical assays, as chromosomal aberrations, micronucleus test and c-mitotic effects. The authors applicated single oral doses of 10; 50 and 100 mg/kg and repeated oral dose of 10 mg/kg for 5 days. Their investigation revealed that although BPA failed to induce conventional chromosomal aberrations and micronuclei, its genotoxic effects were manifested in the form of achromatic lesion and c-mitotic effects in bone marrow cells of Swiss albino mice. Later they studied genotoxic effects BPA and octylphenol (OP) in rats using comet assay. Adult male Wistar albino rats were divided into several groups as follows: BPA125, resp. OP125 (received 125 mg/kg b.w., n = 6), BPA250, resp. OP250 (received 250 mg/kg b.w., n = 6) and were orally administrated for 4 weeks. They observed significant differences in the BPA250 and OP250 ($P < 0.05$ and $P < 0.01$, respectively), compared with the control group. Oral administration of BPA and OP may posses a genotoxic risk in rats in high doses of tested chemicals and may not be so critical in low doses **(Ulutaş *et al.*, 2011)**. Furthermore, there is a need of studies for exploring mechanisms of genotoxic potential BPA *in vivo*, because findings from oral studies are limited in their interpretation and in risk assessment context, they noted.

CONCLUSION

Based on current knowledge of literature it may be concluded that there is a need for further experimental studies with BPA in addressing health of human and animal population living in different ecosystems.

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