# Discussion of Bisphenol A as an Environmental Endocrine Disruptor: The Low Dose Effect and Governmental Regulations Concerning its Use and Disposal: A Literature Review

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**Abstract:**

The disruptive and potentially harmful effects of naturally occurring and man-made endocrine disrupting compounds found in the environment are a topic of considerable debate within government, industry and the general public. Bisphenol A (BPA) is of particular concern due to its incorporation in many consumer products and its potential for leeching. Scientific study continues with attempts to identify and quantify risk associated with this chemical, in order to support industry and regulatory actions. The issue of greatest concern with regards to BPA is the effects of routine exposure to very low concentration of the chemical. The effects of this phenomenon, called Low Dose Effects, raise a great deal of controversy as it is difficult to accurately assess the health outcomes from these exposures. This paper gives a basic understanding of what constitutes Low Dose effects and also examines several studies conducted to determine the health outcomes as a result of exposure to low dose BPA.

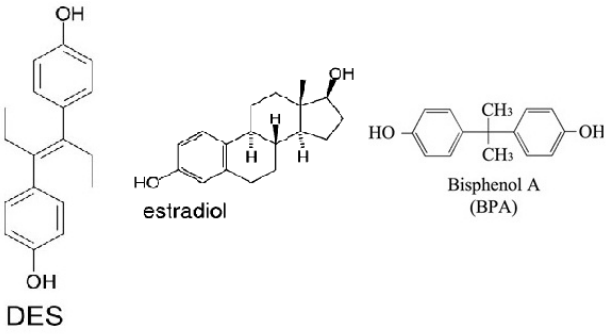
Regulatory activities seek to mitigate risk through enactment of legislation to control the use of and exposure to these compounds. As a result the regulatory agencies in Canada and the U.S. have banned the sale of certain consumer products containing BPA and have imposed strict limits on concentration in industrial effluents and waste water drainage. This paper outlines the conditions and limits put in place by both the Canadian and U.S. government regarding the presence of BPA in effluents and consumer products.

**Keywords:** Endocrine disruption, male infertility, low dose effect

## Introduction

The subject of endocrine disrupting chemicals is currently one of the more controversial issues in environmental toxicology. The prospect that humans are constantly being exposed in the environment to natural and artificial chemicals that modulate the way the endocrine system works (and by extension many other systems in the body) is a troubling thought. One of the most important concerns is the effects this is having on human reproduction. As perceived decreases in sperm counts and increases in infertility (defined as the inability to produce pregnancy while in a stable relationship and engaging in sexual intercourse without contraception for greater than a year (Pflieger-Bruss, Schuppe, & Schill, 2004)) continue to be published, the theories that this may be due to endocrine modulators are increasing in popularity. (Richter et al., 2007) Therefore, increases in research and regulations regarding these chemicals are imperative to ensuring the health of the public. Bisphenol A is a chemical of particular concern due to its presence in many consumer products and also the large number of contradictory results among studies. Along with Diethylstilbestrol (DES), a known estrogenic toxin, BPA has a chemical structure very similar to that of natural estradiol and this is important in considering the possible affects of exposure to this chemical.

**Figure 1** The similar chemical structures of Diethylstilbestrol and Bisphenol A as compared to Estradiol result in their estrogen mimicking activity at the body's estrogen receptors. ((Morais-de-Sa, Pereira, Saraiva, & Damas, 2004) (Hashimoto, Okada, & Imaoka, 2008)(Zhao et al., 2007))




A review of several studies (discussed in the Low Dose Effect section) illustrates the ongoing controversy surrounding the issue of exposure to endocrine disrupting chemicals and more importantly the effects of low dose exposures. (vom Saal & Hughes, 2005) Focusing on the issue of the low dose effect, five studies are analyzed that propose either the finding of adverse effects or the lack thereof as a result of exposure of mice or rats to BPA at low doses. While DES is an endocrine disrupting chemical, it can not be considered alongside BPA as it has well documented toxic effects, and as such it is used in all of the studies discussed as a positive control for adverse effects of exposure. The controversial topics of what a low dose is, the concept of environmentally relevant doses (Richter et al., 2007), appropriate animal model selection (Pflieger-Bruss et al., 2004), the need for positive control groups (vom Saal & Hughes, 2005), and the influence of funding sources (vom Saal & Hughes, 2005) are examined in the analysis of these journal articles. The conclusion that there are in fact adverse health effects related to low dose or environmentally relevant exposures is becoming a more prominent finding but still demands significant further research to reinforce this finding. (vom Saal & Hughes, 2005)

Finally, the details of current governmental regulations regarding these chemicals are discussed. Canada was the first country to perform an in depth risk assessment and subsequently place various bans on the use, sale, import and disposal of BPA and DES. The restrictions on BPA placed by both governments focus specifically on the sale of BPA containing consumer products (i.e. Baby bottles), and the BPA levels found in industrial effluents and waste water. (Health Canada, 2008) With regards to DES, there have been long standing restrictions limiting the use of this drug to only non-food producing animals and even then it is highly regulated. (Health Canada, 2003b) Both Canada and the United States have extensive regulations with regards to BPA and DES and are pressing for continued research on the subject.

## The Low Dose Effect

The most controversial issue surrounding the potential human health effects of endocrine disrupting chemicals is the topic of the low dose effect.(vom Saal & Hughes, 2005) Low dose commonly refers to a dosage used in studies that would be considered environmentally relevant. (Richter et al., 2007) In other words, doses resulting in test animal serum levels of the endocrine-disrupting chemical being similar to those observed in human serum from routine environmental exposure. (Richter et al., 2007) This dose is determined based on the usual reference dose and the governmentally set "safe dose". (vom Saal & Hughes, 2005) The standard reference dose or lowest observed adverse

effect level (LOAEL) for BPA prior to 1997 was 50mg/kg/day for studies involving rats. (Richter et al., 2007) This value is contested based on evidence that effects can be observed at much lower concentrations. (vom Saal & Hughes, 2005) The U.S. Environmental Protection Agency's (EPA) "safe dose" limit for BPA is set by dividing the LOAEL by 3 log factors or safety factors (i.e. 1000), which gives a value of 50µg/kg/day based on the pre-existing reference dose. (Richter et al., 2007) The issue with BPA is that studies have been published with both no observed effects and significant observed effects in both mice and rats for dosages below the "safe dose". (Pflieger-Bruss et al., 2004)

A review published in 2005, (vom Saal & Hughes, 2005) discusses the existence of both results in published studies but proceeds to make several important points regarding the proportions of the opposing results. In the review by vom Saal(vom Saal & Hughes, 2005), the author discusses a large inconsistency in the data with regards to the source of funding for each study. Vom Saal *et al.* contend that of the studies published at the time of their publication, all those that were funded by industry sources found no effects for low dose exposures. Conversely, the authors point out that of the studies funded by the government, greater than 90% found significant observable effects from low dose exposures to BPA.

**Table 1** Source of funding and results of low dose BPA studies. Also compared based on CD-SD rat use as an animal model; due to its low sensitivity to BPA this may affect results. (vom Saal & Hughes, 2005)

| | All studies | | CD-SD rat studies | | All studies except CD-SD rats | |
|---|---|---|---|---|---|---|
| **Source of funding** | **Harm** | **No harm** | **Harm** | **No Harm** | **Harm** | **No Harm** |
| Government | 94 (90.4) | 10 (9.6) | 0 (0) | 6 (100) | 94 (96) | 4 (4) |
| Chemical corporations | 0 (0) | 11 (100) | 0 (0) | 3 (100) | 0 (0) | 8 (100) |

Values shown are no. (%)

As shown by the above table the proportion of results that observed harmful effects from low dose BPA exposures far outweigh the number of studies that had the opposite finding.

A review of 4 other low dose BPA studies revealed similarly conflicting results. A study completed by Pflieger-Bruss *et al.* (2004)outlined the common effects of xeno-estrogen compounds, (i.e. BPA and DES) in animal studies as hypospadias, cryptorchidism, decreased sperm count and testicular tumours. This study went on to compare peri- and post- natal exposures to BPA at low doses (2.4µg/Kg/day) in rats, with the observed results being reduced serum leutenizing hormone and testosterone in the postnatally exposed animals and reduced testosterone in adulthood for the perinatally exposed rats. (Pflieger-Bruss et al., 2004)A high dose (300µg/Kg/day) was also administered to the rats with no observed effect.(Pflieger-Bruss et al., 2004) Ultra low dose (0.1µg/Kg/day) tests were also conducted using mice exposed for 28 straight days, with the following results: decreased testicular and epididymal sperm counts, and decreased weights of the testes and seminiferous vesicles. (Pflieger-Bruss et al., 2004)

Two of the other studies reported opposing findings in rats and mice respectively.(Cagen,et al., 1999b)(Cagen, et al., 1999a) The study conducted on rats (Cagen et al., 1999b), used an oral dosing method via the animals' drinking water. Test groups were set up with 0 (control), 0.01, 0.1, 1.0, and 10 ppm of BPA dissolved in their drinking water. A positive control group was also tested using 0.1 ppm DES. Female mice were given these dosages for a period of 7 weeks, starting 2 weeks prior to mating and continuing through to 22 days of lactation. (Cagen et al., 1999b) The males and offspring were not dosed.(Cagen et al., 1999b) Offspring from the F1 generation were sacrificed at 90 days and tissues were analyzed with the following results: BPA exposed rats showed no effects on growth, survival or reproductive parameters (including weights of testes, prostate and preputial gland; sperm count; daily sperm production and testes histopathology). (Cagen et al., 1999b) The DES exposed rats showed a decrease in body weight, body weight change and food consumption in the adult female rats, as well as a decrease in the number of pups per litter and an increase in gestational length. (Cagen et al., 1999b)

The second study with a result of no effect was also published by Cagen *et al.* (1999a) but this study was conducted with mice in lieu of the previously tested rats. A different method of administration was also used in this study. The maternal mice were given oral deposit doses of 0.2, 2, 20 and 200 µg/Kg/day of BPA and a positive control group was given 0.2µg/Kg/day of DES.(Cagen et al., 1999a) These doses were given on gestational days 11 through 17 and the pups of the F1 generation were sacrificed at 90

days old.(Cagen et al., 1999a) Tissue collection and analysis revealed no significant dose response in the BPA test groups.(Cagen et al., 1999a)

The last publication reviewed was itself a review of several studies that found dose response effects from low dose BPA exposures. (Richter et al., 2007) It emphasized several common issues with low dose BPA studies, those being: the selection of appropriate animal strain, the use of a positive control, and the relevance of the dosing method. (Richter et al., 2007) As outlined in the vom Saal and Hughes (2005) article, this article discussed the possible effects of an inappropriate animal model for this type of study. For example CD-SD rats have much lower sensitivity to estrogenic compounds and would therefore register no observed effects in low dose BPA studies where other strains of rat may produce significant dose response effects. (Richter et al., 2007) This consideration must be heeded in order for any study on low dose BPA effects to have significant validity. (vom Saal & Hughes, 2005) In order to determine a strain's sensitivity to estrogenic compounds with hopes of validating the results of a low dose exposure study, a positive control must be used (vom Saal & Hughes, 2005).

DES and Ethynylestradiol are commonly used as positive controls in BPA or other estrogenic compound exposures as they have well characterized and replicated adverse effects. (Odum et al., 2002)(Richter et al., 2007) Despite their known ability to cause dose response effects, it is still necessary to consider if the method of dosing is appropriate with the positive control chemicals as well as the experimental chemical. Certain chemicals can be more or less biologically active depending on the route of administration, as with ethynylestradiol which has a low bioactivity if administered orally. (Richter et al., 2007) It is also important to consider method of dosing with regards to environmental relevance, as this can affect validity of the results when compared to normal human exposure. (Richter et al., 2007) For example, oral administration (such as: oral deposit, oral gavage, drinking water) represents a much more accurate model of regular human exposure and allows for more accurate results given that the drugs will be undergoing the same metabolic processes in the animal models as they would if ingested by a human. (National Toxicology Program (U.S.), 2008) This is an important consideration that adds to the validity of a study.

In conclusion, as a small sample of the numerous studies out there, the five articles reviewed provide compelling evidence to both support and refute the evidence that low dose BPA exposure can have adverse effects on human health. While there appears to be more evidence in support of the argument that there are observable effects at and below the "safe dose", more testing is surely needed prior to these findings being considered absolute. The other considerations voiced by these articles, including appropriate animal model and route of exposure, should also be further tested to determine the most biologically relevant methods to use for a study on the effects of estrogenic endocrine disruptors, specifically BPA.

## Governmental Regulations

As testing continues on the effect of environmental endocrine disruptors on the various physiological systems of the human body, government agencies have begun to take notice of the potential for possible health risks and have responded accordingly. In 1996 the U.S. Food Quality Protection Act

contained amendments to the U.S. Federal Food, Drug and Cosmetic Act and amendments were also made that year to the Safe Drinking Water Act. (Stokes, 2004)These amendments require that the U.S. Environmental Protection Agency (EPA) implement the new laws to monitor the endocrine disrupting effects of new chemicals. (Stokes, 2004) In 1998 the EPA proposed the Endocrine Disruptor Screening Program (EDSP) to satisfy these conditions. (Stokes, 2004) The EDSP outlines how the EPA uses both in vitro and in vivo testing to identify chemicals that might adversely affect humans and ecologically important species. (Stokes, 2004)

Bisphenol A is a chemical commonly used in the production of polycarbonate plastics and resins. (National Toxicology Program (U.S.), 2008) Polycarbonate plastics are used to make a wide range of consumer products including: water and infant bottles, impact-resistant safety equipment, compact disks and even some medical equipment. (Stokes, 2004) It is also used in the canning process to generate the protective epoxy coating inside canned food containers to prevent contamination arising from leaching of the metal container into the food (Pflieger-Bruss et al., 2004). This widespread use increases the potential for human exposure on a regular basis. (Richter et al., 2007) As a result, in 2008 Canada became the first country in the world to perform a risk assessment on BPA with the participation of industry and other stakeholders, as well as to conduct a 60 day public comment period on the decision to

ban certain BPA containing products. (Health Canada, 2008) Based on the findings that there were some minor health risks associated with BPA containing baby bottles as well as the public concerns gathered during the comment period, the Canadian government banned the import, sale and advertisement of BPA containing polycarbonate baby bottles in June 2009. (Health Canada, 2009; Health Canada, 2009) In addition to this ban, regulations have been proposed and are under review in a response to the findings that significant amounts of BPA were being found in wastewater and sludge treatment plants. (Health Canada, 2008) These proposed regulations limit the amounts of BPA effluent that is allowed to enter the environment from an industrial source. (Environment Canada, 2009)This limit will be 1.75µg/L in the industrial effluent released from any facility with the exception of wastewater from the treatment of intake water. (Environment Canada, 2009) The proposed implementation date of this regulation is no later than April 2011. (Environment Canada, 2009)

Diethylstilbestrol is a synthetic estrogen used in both human and animal medicine. From 1948 to 1977 DES was used in France to prevent miscarriage and pregnancy related bleeding in pregnant women. (Health Canada, 2003a) Diethylstilbestrol was subsequently recognized as a genotoxin and carcinogen to both humans and animals. (Health Canada, 2003b) As a result, the following regulations were created in Canada: prohibition of the sale of DES or other stilbene compounds for administration to food producing animals; prohibition of the sale of animals treated with these drugs for use as food; prohibition of the sale of food products from animals treated with these drugs; and prohibition of the sale of food products containing residues of these drugs. (Health Canada, 2003b) A full prohibition of the sale of DES and other stilbene compounds was proposed, but there is still sufficient use of these drugs in a veterinary context to prevent this. For example Diethystilbestrol is still used as an effective veterinary treatment for estrogen responsive incontinence in spayed female dogs and to prevent pregnancy in dogs and cats. (Health Canada, 2003b)

In conclusion, both Canada and the United States have imposed strict regulations on the use, content and disposal of BPA in consumer products and waste by-products based on preliminary results concerning its effects on human health. Further research is needed to fully understand the hazardous nature of this chemical in order to be able to set regulatory limits that protect against possible adverse health effects, with special attention to possible low dose effects. As a well characterized hazardous chemical, the strict regulations against the use of DES appear to protect the public from its adverse health effects, while still allowing for its limited range of beneficial uses. DES maintains its uses in veterinary medicine and laboratory testing as a positive control. As more evidence develops on the effects of low dose exposures to BPA, a review of the current regulations will be needed to ensure the ongoing protection of the public against the endocrine disrupting effects of this chemical.

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