Current Science International
ISSN: 2077-4435

Volume : 08| Issue : 01| Jan.- Mar. 2019
Pages: 109-118

Assessment of Urinary Bisphenol A Levels in a Sample of Obese and Non-Obese Egyptian Children

Mai M. Youssef[1], Ebtissam M. Salah El-Din[1], Mones M. Abu Shady[1], Jihan Hussein[2], Iman A. El-Ashmawy[1], Iman H. Kamel[1], Rania N. Sabry[1], Fatma Alzaree[1], Dina A. Salah[1] and Ehsan A. Badawy[2]

[1]Child Health Department, [2]Medical Biochemistry Department, Medical Division, 33 El Buhouth St., (Former El-Tahrir St.) 12622, Dokki, Giza, Egypt.

Received: 24 Jan. 2019 / Accepted 25 Feb. 2019 / Publication date: 05 Mar. 2019

ABSTRACT

Background: Bisphenol A (BPA) is used extensively worldwide in the manufacture of plastic polymers. The environmental obesogenic hypothesis suggests that early life exposure to BPA may increase the risk for weight gain later in childhood. ***Objective:*** To investigate the relationship between urinary BPA concentrations and obesity indicators in a sample of obese and non-obese Egyptian children and adolescents. ***Methods:*** Total urinary BPA and urinary creatinine concentrations were estimated in a random sample of 166 Egyptian children and adolescents of both sexes and different social levels. Their age ranged from 6-16 years. Measurement of anthropometric parameters was performed and children were classified according to body-mass index z-score into two groups; obese and non-obese. ***Results:*** Bisphenol A was detected in all urine samples of children and adolescents with a GM 0.67ng/ml. About 77% of children had very low concentration of urinary BPA below 1.3ng/ml. No significant difference in concentrations of unadjusted urinary BPA between obese and non-obese children. The log transformed urinary BPA and the adjusted log transformed urinary BPA for urinary creatinine showed significant higher levels in non-obese children and adolescents. No correlation between obesity indices (such as BMI-z score, waist and hip circumferences) and urinary BPA could be detected. ***Conclusion:*** Egyptian children in this study were extensively exposed to BPA with wide range of variability. The association between urinary BPA concentrations and indicators of obesity could not be confirmed. Future large prospective studies with multiple measurements of BPA, detailed measures of nutrient intake and energy expenditure are greatly recommended.

Keywords: Urine bisphenol A (BPA), Obesity, Egyptian children.

Introduction

Endocrine-disrupting chemicals (EDCs) are a class of chemicals that could disturb homeostasis or modify the action of endogenous hormones, increasing the risk of non-communicable diseases throughout life. It is claimed that, children are more susceptible to harmful effects of EDCs owing to their greater exposure and rapid development. The most sensitive times of exposure to EDCs is during critical periods of development, such as during fetal development, perinatal life, childhood and puberty (Biro and Wien, 2010). Bisphenol A (BPA) is one of the endocrine-disrupting compounds; used in the manufacture of polycarbonate plastics and epoxy resins and as an additive in thermal paper. Humans are exposed to BPA via plastic containers of food and beverages, canned food, medical devices, dental sealants and the printing of thermal paper used for cash register receipts and tickets (Biedermann *et al.,* 2010; Geens *et al.,* 2011).

BPA can leach from containers into consumer products, which presents many opportunities for human exposure, most commonly via ingestion and, to a lesser extent, via inhalation and dermal routes (Vandenberg *et al.*, 2010). Once ingested, BPA is efficiently absorbed (95%) and is rapidly and mainly excreted in urine as BPA conjugates. The urinary concentration of total (free plus conjugated) BPA has often been used to evaluate the exposure level of BPA from all sources (Zoller *et al.,* 2012). Detectable BPA levels have also been measured in placental and amniotic fluids (Balakrishnan *et al*., 2010), and human breast milk (Vandenberg *et al.,* 2007) suggesting that exposure starts in utero and may continue postnatally via breastfeeding .

Corresponding Author: Iman H Kamel, Department of Child Health, National Research Centre (NRC), El-Buhouth St., Dokki, Cairo, Egypt P.O. Box: 12622
E-mail:imankamelh@gmail.com

Bisphenol A (BPA) is an endocrine-disrupting chemical that can mimic or partly mimic estrogens, androgens and thyroid hormones. It affects the endocrine system by competing with endogenous hormones by binding to receptors and modifying the synthesis, metabolism and action of these hormones (Yoon *et al*., 2014). BPA may potentiate the risk of reduced fertility in males (Maffini *et al*., 2006) and precocious puberty in females (Rasier *et al*., 2006). It has been associated with increased incidence of cardiovascular diseases and diabetes (Rezg *et al*., 2014) and could present a possible role in the development of cancer (Keri *et al*., 2007). BPA has been labeled as a xenobiotic which has an obesogenic, diabetogenic behavior and act as an antagonist to thyroid hormone action. (Decherf & Demeneix, 2011; Moriyama *et al*., 2002).

Childhood obesity is one of the major public health challenges (WHO, 2004). It has been defined as an abnormal or excessive fat accumulation that may impair health causing life-threatening diseases (WHO, 2014). Obesity is considered an important risk factor for cardiovascular diseases, certain cancers, respiratory ailments, and it adversely affects the health-related quality of life. In addition, obese children are at a much higher risk of being obese in adulthood (Zoller *et al.,* 2012).

It has been observed that the worldwide prevalence of childhood obesity had increased by 47.1% between 1980 and 2013 (Ng *et al.,* 2014). Egypt has a high prevalence of obese individuals among both children and adults. In adults the prevalence is about 35.3% which represents the highest prevalence worldwide. In children, the prevalence is about 10.2% which is slightly less than the highest level of obese children observed in the United States (Afshin *et al.,* 2017).

Obesity occurs when energy consumption exceeds energy expenditure. However, the current obesity epidemic can't be completely explained by excess food consumption and inadequate physical activity (Baillie-Hamilton, 2002; Grun and Blumberg, 2006). Other risk factors might play a role in this epidemic.

Research evidence suggests that exposure to environmental obesogen-chemicals that promote lipid accumulation and adipogenesis-may have a role in increasing obesity risk, especially when exposure occurs in utero and early postnatal life (Tang-Peronard *et al.,* 2011; Romano *et al.,* 2014). In vitro studies have shown that BPA enhances adipocyte cell differentiation, leading to excess fat accumulation (Masuno *et al.,* 2005). Rodent studies have also found BPA exposure to increase adipose tissue mass and promote weight gain (Miyawaki *et al.,* 2007). In humans, epidemiologic studies have demonstrated association of higher BPA levels with higher risk of overweight and obesity in both children and adults (Carwile *et al.,* 2011; Trasande *et al*., 2012; Wang *et al*., 2012; Do *et al*. 2017; Eng *et al*., 2013and Bhandari *et al*.,2013). However, existing epidemiological studies have reached inconclusive findings (Oppeneer and Robien, 2014).

The aim of the current research is to investigate the relationship between urinary BPA concentrations and obesity indicators in a sample of obese and non-obese Egyptian children and adolescents.

Subjects and Methods

Study design and population

The study was a cross sectional one that included 166 apparently healthy children and adolescents (94 boys and 72 girls) who were randomly selected from public and private primary, and preparatory schools in Giza. Their age ranged from 6-16years old. Children were excluded if they had a history of liver disease, renal diseases, thyroid disorders, endocrinal and genetic obesity or children whose parents or guardians refuse to participate.

Ethical considerations:

Ethical Approvals were obtained from The Medical Research Ethical Committee of the National Research Center (NRC) N0 16 368, the Egyptian Ministry of Education and the directors of schools that participated in the research from 2012-2014. Written consents were obtained from the parents and oral one from each student to be involved in the research.

Methods

Each child enrolled in the study was subjected to the following:

-Physical Examination:

Children were subjected to thorough clinical examination that included chest, heart, abdominal, and central nervous system examination.

-Anthropometric measurements:

Weight, height, waist & hip circumferences were measured. The height was measured to the nearest 0.1 cm using a Holtain portable anthropometer, and the weight was determined to the nearest 0.01 kg using a Seca scale Balance with the subject dressed in minimal clothes and without shoes. The BMI was calculated as weight (in kilograms) divided by height (in meters) squared. Body mass index (BMI) is a measure used to determine childhood overweight and obesity. Obesity is defined as a BMI at or above the 95th percentile for children and teens of the same age and sex or greater than 2 standard deviations above the WHO Growth Reference median. (WHO, 2014). BMI- Z-score was calculated based on the WHO growth standards with the help of Anthro-plus Program for personal computers. Children with BMI- Z-score >2 were considered obese (WHO, 2009).

Biochemical investigations:

Each child gave a morning spot urine sample which stored at -70 until assays for estimation of urinary BPA concentration. The concentration of total species of urinary BPA was determined by HPLC, Agilent technologies 1100 series, equipped with a quaternary pump (G131A model), according to the method described by Matsumoto *et al.* (2003) . Briefly, urine sample (50 mL) was buffered with 30 mL of 2.0 M sodium acetate buffer (pH 5.0) and hydrolyzed enzymatically with β-glucuronidase/sulfatase (4.414/168 U/mL) for 3 hours at 37°C in a shaking water bath. After hydrolysis, 100 mL of 2 N hydrochloric acid was added, and the hydrolysate was extracted once with 5 mL of ethyl acetate. After centrifugation, 4mL of supernatant was transferred to a new tube and evaporated with N2 gas stream. The residue was dissolved with 400 ul of acetonitrile / water (60/40) v/v; 40 ul of the solution was then injected onto HPLC. Estimation of BPA was carried out by reversed phase HPLC column (150×4.6, particle size 5 μm) and mobile phase consisted of tetrahydrofuran/water/acetonitrile mixture (17.5/65/17.5) v/v with flow rate 1.5 ml/min and florescent detector was set at 275 and 300 (excitation and emission). Serial dilutions of standards were injected onto HPLC and their peak areas were determined. A linear standard curve was constructed by plotting peak areas versus the corresponding concentrations. The concentration in samples was obtained from the standard curve. The limit of detection (LOD) was calculated with the method recommended by EPA (2004). The LODs of BPA in urine was 0.3ng/ml.

Determination of urinary creatinine:

Urinary creatinine was determined using kinetic kit according to the method of Bartels (1972). BPA concentration was adjusted to the urinary creatinine concentration to correct for the urine dilution (Barr *et al.,* 2005).

Statistical analysis:

Statistical Package for the Social Sciences (SPSS) version 21 (SSPS Inc., Pennsylvania, USA) was used for data analysis. The BPA concentration was adjusted to the urinary creatinine concentration to correct for the urine volume. As the range between minimum and maximum values of urinary BPA was very wide (0.3 to 18.9 ng/ml); the geometric mean was calculated to overcome the problem of outliers. Urinary BPA and BPA/Creatinine levels were log transformed to improve normality of the distribution. Urinary BPA, was categorized into quartiles from < 1.3ng/ml to > 4.9ng/ml. The data of obese and non-obese subjects were expressed as mean ± standard deviation and were compared by use of student's t-test. Pearson's correlation analysis was conducted to evaluate the correlation between the normally distributed variables. All used tests were considered significant at $p<0.05$.

Results

The total number of the clinically studied population was166, of which 94 were males and 72 were females. They were classified according to BMI-z score into two groups: The obese group were 46 children and adolescents and the non- obese group were 120 children and adolescents. Means± SD values of age and anthropometric indices are shown in table 1.

Table 1: Age and anthropometric measurements of the studied sample

	BMI group	Mean	SD	t-test	p
Age (years)	Non obese	10.97	2.34	-2.190	0.059
	Obese	11.95	2.18		
Weight (Kg)	Non obese	39.48	14.56	-9.735	0.000*
	Obese	66.53	18.76		
Height (cm)	Non obese	142.52	19.77	-2.966	0.003*
	Obese	151.98	13.75		
BMI (weight(kg)/Height (m2)	Non obese	18.62	3.28	-15.181	0.000*
	Obese	28.19	4.29		
BMI-z score	Non obese	0.33	1.17	-13.235	0.000*
	Obese	2.78	.69		
Waist c (cm)	Non obese	65.24	9.88	-11.526	0.000*
	Obese	86.74	12.13		
Hip c (cm)	Non obese	79.61	14.71	-7.875	0.000*
	Obese	99.38	12.88		

P < 0.05 is significant

Table 2 shows urinary BPA concentrations of the studied children and adolescents. Bisphenol A was detected in all examined urine samples. Means, median and geometric means values of the total urinary BPA, urinary BPA/ gm creatinine and LOG of adjusted urinary BPA/gm creatinine are shown in table 2.

Table 2: Urinary BPA concentrations of the studied children and adolescents

BPA	Mean	SD	Median	Geometric Mean
Total urinary BPA ng/ml	1.28	1.48	0.68	0.67
Urinary BPA/ gm creatinine	798.91	1038.24	453.52	436.76
Log urinary BPA/gm creatinine	2.6402	0.49	2.66	2.59

.

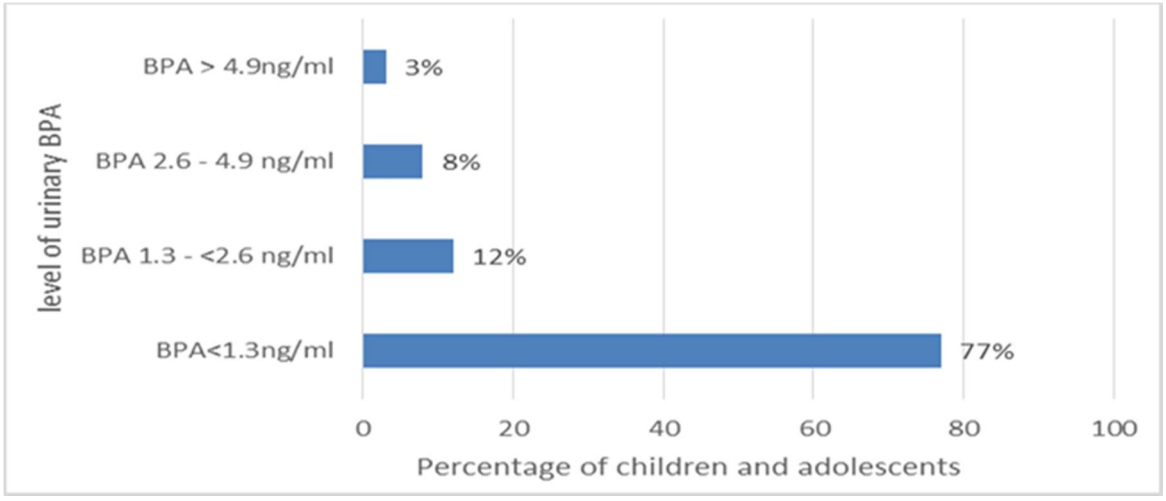

Fig. 1: Distribution of children and adolescents along urinary BPA quartiles

Table 3 shows a comparison of urinary BPA levels in obese and non-obese children. No significant difference could be detected between the two groups as regards total urinary BPA. The mean LOG total BPA in obese children (-0.28 ± 0.44 ng/ml) was significantly lower than that in non-obese children (-0.12 ± 0.47ng/ml) (p=0.047) and the mean LOG of adjusted BPA/gm creatinine in obese children (2.44 ± 0.53) was significantly lower than that in non-obese children (2.72 ± 0.53) (p=0.004).

Table 3: Comparison of BPA levels in obese and non-obese subjects

Parameter	non-obese (n=120) Mean ± SD	Obese (n= 46) Mean ± SD	P-Value
Total BPA (ng/ml of urine)	1.43±2.31	0.92± 1.34	0.164
LOG Total BPA	-0.12±0.47	-0.28-0.44	0.047*
Log BPA/ gm creatinine	2.72± 0.53	2.44±0.53	0.004*

P < 0.05 is significant

Table 4 and 5 demonstrate the correlation between urinary BPA concentrations with indicators of obesity in obese and non-obese group respectively. No significant association could be detected in both groups.

Table 4: Correlation of urinary BPA concentrations with indicators of obesity in obese group

		LOG TOTAL BPA	Log BPA/creatinine g
Body mass index	Pearson Correlation	-.270	-.066
	Sig. (2-tailed)	.330	.816
BMI-z score	Pearson Correlation	-.222	-.142
	Sig. (2-tailed)	.158	.371
Waist c	Pearson Correlation	-.265	-.227
	Sig. (2-tailed)	.094	.154
Hip c	Pearson Correlation	-.172	-.140
	Sig. (2-tailed)	.282	.383
Waist to height ratio	Pearson Correlation	-.047	.069
	Sig. (2-tailed)	.770	.668

*. Correlation is significant at the 0.05 level (2-tailed).

Table 5: Correlation of urinary BPA concentrations with indicators of obesity in non- obese group

		LOG TOTAL BPA	Log BPA/creatinine g
Body mass index	Pearson Correlation	-.042	-.037
	Sig. (2-tailed)	.921	.932
BMI-z score	Pearson Correlation	-.112	-.178
	Sig. (2-tailed)	.261	.074
Waist c	Pearson Correlation	-.072	-.140
	Sig. (2-tailed)	.472	.155
Hip c	Pearson Correlation	-.078	-.132
	Sig. (2-tailed)	.438	.181
Waist to height ratio	Pearson Correlation	.056	.142
	Sig. (2-tailed)	.577	.156

*. Correlation is significant at the 0.05 level (2-tailed).

Discussion

Children are more susceptible to the harmful effects of environmental pollutants owing to the rapid growth and development of their body systems and their longer lifespan. Consequently, they become more vulnerable to develop chronic diseases in adulthood (WHO 2004, 2011). In this cross-sectional study, the relationship between BPA exposure and obesity in childhood and adolescence was investigated. The study population was randomly recruited 166 children and adolescents of both sexes and their age ranged from 6 to16 years.

In the current study, BPA was detected in all examined urine samples of children and adolescents. This is going with many previous studies reported the detection of BPA in the blood and

urine of most people worldwide indicating its universal exposure (Zhang *et al*., 2011; Olsen *et al*., 2012 and Ko *et al*., 2014). The concentration of total urinary BPA in this study ranged from 0.3 to 18.9 ng/ml, signifying diverse exposure of Egyptian children to BPA. The majority of the studied sample had BPA concentrations below 1.3ng/ml of urine. The geometric mean (GM) of total urinary BPA in the current study was relatively close to the GM value of previous Egyptian study included girls aged 10-13 years old (Nahar *et al.*, 2012). (GM was 0.67 ng/ml in the current study versus 0.84 ng/ml in the previous one). However, this GM is much lower than that reported for children in European countries, Canada, or Australia. In a German study, a GM of urinary BPA was reported as 2.22 ng/ml in children aged 3- 14 years (Becker *et al*. 2009). In an Australian study conducted on children from 0-15 years old, the GM of the total urinary BPA was 2.57 ± 4.66 ng/ml (Heffernan *et al*., 2013). In Europe, children from six European countries aged 5- 12 years were included in a large study estimating urinary BPA. A GM of urinary BPA was 1.97µg/L which is still higher than that in the current study (Covaci *et al*, 2015). The least GM of urinary BPA recorded in a Canadian Survey of children 6 to 19 years old, was 1.3 ng/ml, as twice as the value of GM of the current study (Bushnik *et al*., 2010). Differences in BPA levels in different countries may be attributed to genetic variability in metabolism or to life style differences. Nevertheless, researchers should be very cautious in comparing measured concentrations among different populations, due to differences in the way of sampling, the sensitivity of the assays and the analytical methods that were used (Volkel *et al*., 2005; Calafat *et al*., 2008).

The growing prevalence of obesity in children and adults is pushing scientists to look for the possible role of endocrine disrupting chemicals such as BPA in the development of obesity. In the current study, there was no significant difference in concentrations of unadjusted urinary BPA between obese and non-obese children. However, the log transformed urinary BPA and the adjusted log transformed urinary BPA for urinary creatinine showed significant higher levels in non-obese children and adolescents compared to the obese group (table 3). No correlation between obesity indices (such as BMI-z score, waist and hip circumferences) and urinary BPA could be detected whether in obese or non-obese subjects (table 4 and 5). The levels of urinary BPA in this study (GM was 0.67 ng/ml and 77% of subjects had levels below 1.3 ng/ml) which were below those from most previous studies may explain lack of association. Li *et al*. (2013) reported a dose-response relationship; higher urinary BPA levels were significantly associated with elevated fat mass index in girls and lower urinary BPA concentration was associated with lower percentage of trunk fat in girls.
Previous studies had reported controversial results about the association of BPA and obesity. In accordance with the current study, some studies failed to find significant association between urinary BPA concentration and obesity indices. A study in Eastern China found no significant difference in urinary BPA concentrations between normal weight and overweight/obese school children (Wang *et al*. 2014). Eleven endocrine disrupting chemicals including BPA were determined in 49 obese and 27 non-obese Indian children. No correlation between BPA levels and body mass index could be detected (Xue *et al*., 2015).

Other studies have reported negative association. In the American study concerned with hormonal and environmental agents affecting U.S. children, urinary BPA was higher among prepubertal girls with body mass index less than 85th reference percentile than those at or above the 85th percentile (Wolff *et al.,* 2007). In a prospective cohort study involved 297 mother-child pairs from Ohio, urinary BPA concentrations were estimated in samples collected from pregnant mothers during the second and third trimesters and their children at 1 and 2 years of age. Each tenfold increase in prenatal or early-childhood BPA concentrations was associated with a modest and nonsignificant decrease in child BMI. These inverse correlations were stronger in girls than in boys (Braun *et al.,* 2014).

Most of adult and children studies have reported a positive association between urinary BPA concentration and indicators of obesity. Body mass index was considered as an outcome in two studies; they have found positive association between increasing levels of urinary BPA and increased BMI in children (Wang *et al* 2012; Bhandari *et al*., 2013). The same pattern of association was found in multivariable analyses examining the association between urinary BPA concentration quartiles and BMI- z score (Trasande *et al*., 2012). In an American study, BMI, waist circumference and waist – height ratio were used as adiposity measures. They have found that higher BPA levels are associated

with BMI at or above the 95th percentile and with waist circumference–to–height ratio equal or more than 0.5 (Eng *et al*., 2013).

It is apparent that, the possible evidence of obesogenic effect of BPA in humans is not conclusive. Results of different studies are inconsistent with negative, positive or no association at all. This discrepancy may be due to: Initially, the cross-sectional design of most of previous epidemiological studies. The nature of the cross-sectional studies restricts their predictive power and can't infer a causality. Obesity is mainly due high caloric diet, physical inactivity in addition to genetic predisposition (Caporossi and Papaleo, 2017). BPA exposure could be a demonstrative for other variables related to obesity, such as high consumption of packaged and canned foods, bad dietary behavior and family income level (Trasande *et al*., 2012). Use of plastic bottles, microwave plastic wares and consumption of canned food was found to be excessive in Egyptian children from high socioeconomic class (Gabr *et al*., 2017). Socioeconomic background and life style factors may predispose to the higher concentrations of urinary BPA in non-obese children in the current study.
Secondly, the method of assessment of BPA exposure followed in most previous studies was analysis of one spot urine sample. This method was reported to show a moderate sensitivity for predicting individual's BPA exposure level (Mahalingalah *et al*., 2008). However, the short half- life of BPA in human body might mask the true exposure level, resulting in weakening of the association between BPA levels and obesity indicators (Wang *et al*., 2012).

There were some study limitations which should be taken in consideration. The small sample size which restricts generalization of results on Egyptian children and adolescents. The cross-sectional study design combined with one morning spot urine sample could not give an accurate measure of long-term BPA exposure, consequently didn't confirm the association between BPA exposure and obesity. Lack of assessment of confounding variables as life style variables, dietary habits, family income level and exposure to other chemical pollutants represent additional points of limitations.

Conclusion

BPA was detected in all of the assessed urine samples with a wide range of variability indicating widespread and diversity of exposure to this endocrine-disrupting chemical. Mean of urinary BPA concentration was significantly higher in non-obese children and this may be attributed to social background and life style factors. No association could be detected between urinary BPA concentrations and indicators of obesity whether in obese or non-obese children and adolescents. However, we can't exclude this association owing to the small sample size and cross-sectional study design. Further large prospective studies with serial measurements of BPA, detailed measures of nutrient intake and energy expenditure, as well as long-term follow-up are greatly recommended.

Ethical considerations

Ethical Approvals were obtained from The Medical Research Ethical Committee of the National Research Center (NRC) N0 16 368, the Egyptian Ministry of Education and the directors of schools that participated in the research from 2012-2014. Written consents were obtained from the parents and oral one from each student to be involved in the research.

Conflicts of interest

There are no conflicts of interest

Funding

The study was a part of a project supported financially by National Research Centre Egypt **(grant no. 10010403).**

Acknowledgements:

The authors thank all the candidates who participated in the study and their parents.

References

Afshin, A., M.H. Forouzanfar and M. Reitsma, et al., 2017. Health Effects of Overweight and Obesity in 195 Countries over 25 Years. N Engl J Med 2017; 377:13-27 DOI, 7(2):52-61 Research Article. ISSN: 0975-7384 CODEN (USA): JCPRC5 52.

Baillie-Hamilton, P.F., 2002. Chemical toxins. A hypothesis to explain the global obesity epidemic. J Altern Complement Med., 8(2):185-192.

Balakrishnan, B., K. Henare, B. Eric et al., 2010. Transfer of bisphenol A across the human placenta. American Journal of Obstetrics and Gynecology, 202(4): 393.e1-393.e7

Barr, D.B., L.C. Wilder, S.P. Caudill, A.J. Gonzalez, L.L. Needham and J.L. Pirkle, 2005. Urinary creatinine concentrations in the U.S. population implications for urinary biologic monitoring measurements. Environ Health Perspect, 10.1056/NEJMoa1614362.

Bartels, H., M. Böhmer and M. Heierli, 1972. Serum creatinine determination without protein precipitation. Clin Chim Acta, 37:193-7.

Becker, K., T. Goeen, M. Seiwert et al., 2009. Ger ES IV: phthalate metabolites and bisphenol A in urine of German children. Int J Hyg Environ Health; 212:685–692. doi: 10.1016/j.ijheh..08.002

Bhandari R, Xiao J& Shankar. A Urinary bisphenol A and obesity in U.S. children. Am J Epidemiol. 2013; 177(11):1263–1270 .

Biedermann, S., P. Tschudin and K. Grob, 2010. Transfer of bisphenol A from the misprinted paper to the skin. Anal. Bioanal. Chem., 398, 571–576.

Biro, F. and M. Wien, 2010. Childhood obesity and adult morbidities. Am J Clin Nutr., 91(5):1499S–1505S.

Braun, J.M., B.P. Lanphear, A.M. Calafat, et al., 2014. Early-life bisphenol A exposure and child body mass index: a prospective cohort study. Environ Health Perspect., 122(11):1239-1245.

Bushnik, T., D. Haines, P. Levallois et al, 2010. Lead and bisphenol A in the Canadian population, catalogue no. 82-003-XPE. Health Rep., 21:1–13.

Calafat, A.M., X. Ye, L.Y. Wong, et al., 2008. Exposure of the U.S. population to bisphenol A and 4-tertiary-octylphenol: 2003–2004. Environ Health Prospect, 116(1):39–44

Caporossi, L and B. Papaleo, 2017. Bisphenol A and Metabolic Diseases: Challenges for Occupational Medicine. Int J Environ Res Public Health, 25;14(9)

Carwile, J.L. and KB. Michels, 2015. Urinary bisphenol A and obesity: NHANES 2003-2006. Environ Res., 111(6):825–830.

Covaci, A., E. Den Hond, T. Geens et al., 2015. Urinary BPA measurements in children and mothers from six European member states: Overall results and determinants of exposure Environmental Research, 141: 77–85.

Decherf, S. and B.A. Demeneix, 2017. The Obesogen Hypothesis: A shift of focus from the periphery to the hypothalamus. J. Toxicol. Environ. Health, B 14, 423–448.

Do, M.T., V.C. Chang, M.A. Mendez and M. De Groh, 2017. Urinary bisphenol A and obesity in adults: results from the Canadian Health Measures Survey. Health Promot Chronic Dis Prev Can., 37(12):403-412. doi: 10.24095/hpcdp.37.12.02.

Eng, D.S, J.M. Lee, A. Gebremariam, et al., 2013. Bisphenol A and chronic disease risk factors in U.S. children. Pediatrics, 132(3): E637–45.

Gabr, A.A., N.N. Mahfouz, M.M. Abu Shady, M.M. Youssef, E.M. Salah El-Din, A.H. Kamhawy et al., 2017. Socioeconomic position as a risk factor for BPA exposure in a sample of Egyptian children. Journal of Applied Pharmaceutical Science,7 (12): 084-089.

Geens, T., D. Aerts, C. Berthot, J.P. Bourguignon, et al., 2012. A review of dietary and non-dietary exposure to bisphenol-A. Food Chem Toxicol., 50:3725–3740.

Grun, F. and B. Blumberg, 2006. Environmental obesogens: organotins and endocrine disruption via nuclear receptor signaling Endocrinology,147 (6 Suppl):S50-5.

Heffernan, A.L., L.L. Aylward, L.M.Toms, G. Eaglesham, P. Hobson, P.D. Sly and J.F. Mueller, 2013. Age-related trends in urinary excretion of bisphenol A in Australian children and adults: evidence from a pooled sample study using samples of convenience. J Toxicol Environ Health A.,76(18):1039-55.

Keri, R.A., S.M. Ho, P.A. Hunt, K.E. Knudsen, A.M. Soto and G.S. Prins, 2007. An evaluation of evidence for the carcinogenic activity of bisphenol A. Reprod Toxicol., 24: 240–252.

Ko, A., M.S. Hwang J.H. Park, H.S. Kang, H.S. Lee and J.H Hong, 2014. Association between Urinary Bisphenol A and Waist Circumference in Korean Adults. Toxicological Research, 30(1):39-44.

Li, J., H. Lai, S. Chen, H. Zhu and S. Lai, 2016. Gender differences in the associations between urinary bisphenol A and body composition among American children: The National Health and Nutrition Examination Survey, 2003-2006. J Epidemiol., 27(5):228-34. doi: 10.1016/j. je.2016.12.001.

Maffini, M.V., B.S. Rubin, C. Sonnenschein and A.M. Soto, 2006. Endocrine disruptors and reproductive health: the case of bisphenol-A. Mol Cell Endocrinol., 254-255: 179–186.

Mahalingaiah, S., J.D. Meeker, K.R. Pearson, A.M. Calafat, X. Ye, J. Petrozza and R. Hauser, 2008. Temporal variability and predictors of urinary bisphenol a concentrations in men and women. Environ Health Persp, 116:173–178.

Masuno, H., J. Iwanami, T. Kidani, K. Sakayama and K. Honda, 2005. Bisphenol A accelerates terminal differentiation of 3T3-L1 cells into adipocytes through the phosphatidylinositol 3-kinase pathway. Toxicol Sci., 84(2):319–27 .

Matsumoto, A., N. Kunugita, K. Kitagawa, et al., 2003. Bisphenol a levels in human urine. Environ Health Perspect, 111: 101–114. 10.1289/ehp.5512.

Miyawaki, J., K. Sakayama, H. Kato, H. Yamamoto and H. Masuno, 2007. Perinatal and postnatal exposure to bisphenol A increases adipose tissue mass and serum cholesterol level in mice. J Atheroscler Thromb,14(5):245–52

Moriyama, K., T. Tagami, T. Akamizu, T. Usui and M. Saijo et al., 2012. Thyroid hormone action is disrupted by bisphenol A as an antagonist. J. Clin. Endocrinol. Metab., 87, 5185–5190.

Nahar, M.S., A.S. Soliman, J.A. Colacino et al., 2012. Urinary bisphenol A concentrations in girls from rural and urban Egypt: a pilot study Environmental Health 2012; 11:20. https://doi.org /10.1186/1476-069X-11-20.

Ng, M., T. Fleming, M. Robinson, et al., 2014. Global, regional, and national prevalence of overweight and obesity in children and adults during 1980-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet, 384:766-781.

Olsen, L., L. Lind and P.M. Lind, 2012. Associations between circulating levels of bisphenol A and phthalate metabolites and coronary risk in the elderly. Ecotoxicology and Environmental Safety, 80:179- 183.

Oppeneer, S.J. and K. Robien, 2012. Bisphenol A exposure and associations with obesity among adults: a critical review Public Health Nutrition, 18(10):1847–1863.

Rasier, G., J. Toppari, A.S. Parent and J.P. Bourguignon, 2006. Female sexual maturation and reproduction after prepubertal exposure to estrogens and endocrine disrupting chemicals: a review of rodent and human data. Mol Cell Endocrinol., 254-255: 187–201.

Rezg, R., S. El-Fazaa, N. Gharbi and B. Mornagui, Bisphenol, 2014. A and human chronic diseases: current evidences, possible mechanisms, and future perspectives. Environ Int., 64: 83–90

Romano, M.E., D. Savitz and J.M. Braun, 2014. Challenges and future directions to evaluating the association between prenatal exposure to endocrine-disrupting chemicals and childhood obesity. Curr Epidemiol Rep., 1(2):57–66.

Tang-Peronard, J.L., H.R. Andersen, T.K. Jensen and B.L. Heitmann, 2011. Endocrine-disrupting chemicals and obesity development in humans: a review. Obes Rev., 12(8):622-636.

Trasande, L., T.M. Attina and J. Blustein, 2012. Association between urinary bisphenol A concentration and obesity prevalence in children and adolescents. JAMA., 308(11):1113–1121.

Vandenberg, L.N., I. Chahoud, J.J. Heindel, V. Padmanabhan, F.J.R. Paumgartten and G. Schoenfelder, 2010. Urinary, circulating, and tissue biomonitoring studies indicate widespread exposure to bisphenol A. Environ. Health Perspect., 118, 1055–1070.

Vandenberg, L., R. Hauser, M. Marcus, N. Olea and V. Welshons, 2007. Human exposure to bisphenol A (BPA). Reprod Toxicol., 24: 139–177.

Volkel, W., N. Bittner and W. Dekant, 2005. Quantitation of bisphenol A and bisphenol A glucuronide in biological samples by high performance liquid chromatography-tandem mass spectrometry. Drug Metab. Dispos., 33:1748–1757.

Wang, B., H. Wang and W. Zhou, 2014. Exposure to bisphenol A among school children in eastern China: a multicenter cross-sectional study. J Expo Sci Environ Epidemiol., 24:657–664.

Wang, H., Xhou, C.X. Tang, et al., 2012. Association between bisphenol A exposure and body mass index in Chinese school children: a cross-sectional study. Environ. Health,doi:10.1186/1476-069X-11-79.

Wolff, M.S, S.L. Teitelbaum, G. Windham, et al., 2014. Pilot study of urinary biomarkers of phytoestrogens, phthalates, and phenols in girls. Environ. Health Perspect.2007; 115, 116–121.

World Health Organization (WHO). Global nutrition targets 2025: Childhood overweight [policy brief]. Geneva

World Health Organization (WHO), 2009. Anthro Plus for personal computers. Manual Software for assessing growth of the world's Children and adolescents. Geneva. 2009 (http://www.who.int/growth).

World Health Organization (WHO), 2011. Summary of principles for evaluating health risks in children associated with exposure to chemicals. Children's Environmental Health. World Health Organization, Geneva 2011.World Health Organization (WHO). Children's health and the environment. A global perspective. World Health Organization, Geneva.2004.

Xue J., Q. Wu, S. Sakthivel, et al., 2015. Urinary levels of endocrine-disrupting chemicals, including bisphenols, bisphenol A diglycidyl ethers, benzophenones, parabens, and. triclosan in obese and non-obese Indian children. Environ. Res., 137:120-8. doi: 10.1016/j.envres.2014.12.007.

Yoon, K., S.J. Kwack, H.S. Kim and B.M. Lee, 2014. Estrogenic endocrine-disrupting chemicals: molecular mechanisms of actions on putative human diseases. J Toxicol Environ Health B Crit Rev.,17: 127–174.

Zhang, Z., H. Alomirah and H.S. Cho., et al., 2011. Urinary bisphenol A concentrations and their implications for human exposure in several Asian countries. Environmental science & technology, 45(16):7044-7050.

Zoeller, R.T., T.R. Brown, L.L. Doan et al., 2012. Endocrine-disrupting chemicals and public health protection: a statement of principles from The Endocrine Society. Endocrinology, 153, 4097–4110. doi: 10.1210/en.2012-1422 .