Food and Chemical Toxicology 125 (2019) 562–565

Contents lists available at ScienceDirect

Food and Chemical Toxicology

journal homepage: www.elsevier.com/locate/foodchemtox

Serum Bisphenol A concentrations in men with idiopathic infertility

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ARTICLE INFO

Keywords:
Bisphenol A
Endocrine disruptors
Male infertility
Semen quality

ABSTRACT

Background: Bisphenol A (BPA) has been associated with male reproductive dysfunction. However, few studies have assessed BPA according to the cause of male infertility.
Aim: To investigate serum BPA concentrations in infertile men according to infertility cause.
Patients and methods: Men with infertility (n = 55) [non-obstructive azoospermia (n = 23), cryptorchidism (n = 12), varicocele (n = 20)] compared with fertile men (n = 25). Serum BPA concentrations were measured along with clinical and hormonal assessment.
Results: BPA was detected in all men, with no difference between infertile and control groups [median (IQR) 0.19 (0.45) vs. 0.18 (0.28) ng/ml, p = 0.689] or among the infertility cause [azoospermia 0.30 (0.69), cryptorchidism 0.12 (0.39), varicocele 0.17 (0.23) ng/ml, p = 0.316]. High concentrations of BPA (> 3 ng/ml) were observed only in infertile men. A negative correlation was observed between BPA concentrations and AMH ($r = -0.320$, $p < 0.01$).
Conclusions: Although male infertility cannot be attributed to exposure to BPA, high concentrations of BPA could contribute to infertility.

1. Introduction

Infertility is defined as the inability to conceive after frequent and unprotected sexual intercourse for more than a year, a condition that currently affects 15% of couples worldwide (Juul et al., 1999; Sharlip et al., 2002; Wong and Cheng, 2011). Approximately 50% of the cases are attributed to the male partner (Irvine, 1998; Juul et al., 1999; Sharlip et al., 2002; Wong and Cheng, 2011). Specific causes are directly linked to only 23% of all male infertility cases, with environmental factors, such as exposure to endocrine disruptors, being one of the major causes of the remaining cases (Wong and Cheng, 2011).

An endocrine disruptor (ED) is defined by the US Environmental Protection Agency as "an exogenous agent that interferes with synthesis, secretion, transport, metabolism, binding action, or elimination of natural blood-borne hormones that are present in the body and are responsible for homeostasis, reproduction, and developmental process" (Diamanti-Kandarakis et al., 2009; Kabir et al., 2015). Bisphenol A is one of the most investigated and, for many authors, one of the most potent ED (Maffini et al., 2006). It has estrogenic and anti-androgenic properties and it is ubiquitous in the environment and consumer products. Nevertheless, human studies demonstrating BPA's endocrine disrupting effects have been sparse (Kuehn, 2007; vom Saal et al., 2007). In males, high concentrations of BPA have been associated with low serum follicle stimulating hormone (FSH), concentrations (Hanaoka et al., 2002), low free androgen index (FAI) (Mendiola et al., 2010), low sperm concentration, sperm vitality and sperm motility (Meeker et al., 2010; Li et al., 2011; Knez et al., 2014), and increased sperm DNA damage (Meeker et al., 2010). Furthermore, seminal but not plasma BPA was negatively associated with sperm concentration, sperm count and morphology (Vitku et al., 2016). Positive association was demonstrated between urinary BPA and total testosterone (tT), luteinizing hormone (LH) and estradiol (E_2) (Lassen et al., 2014). Finally, male BPA concentrations may affect embryo quality during *in vitro* fertilization (IVF) cycles, although no association was observed with time-to-pregnancy (Buck Louis et al., 2014).

To our knowledge, no studies of the effect of BPA exposure to semen quality have been conducted to date, in well-defined groups of men with infertility. In the present study, we assessed the association between plasma BPA concentrations, a biological marker of BPA exposure, and semen quality, in infertile men of specific etiology, such as non-

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https://doi.org/10.1016/j.fct.2019.02.016
Received 17 December 2018; Received in revised form 21 January 2019; Accepted 5 February 2019
Available online 07 February 2019
0278-6915/

Table 1
Demographic, testis volume, sperm and blood chemistry parameters of all men studied.

	Controls	INOA	Varicocele	Cryptorchidism
n	25	23	20	12
Demographic parameters				
Age (years)	30.7 ± 1.7	34.6 ± 1.6	37.4 ± 1.3	33.7 ± 1.2
Clinical parameters				
Testis, right (ml)	23.4 ± 2.1	13.4 ± 1.3	21.6 ± 1.1	13.8 ± 1.8
Testis, left (ml)	22.8 ± 2.3	13.1 ± 1.3	21.2 ± 1.1	15.2 ± 1.6
Semen parameters				
Semen volume (ml)	5.2 ± 0.7	3.7 ± 0.5	3.4 ± 0.4	3.3 ± 0.3
Sperm number (10^6/ml)	65.0 ± 11.3	0 ± 0	12.7 ± 3.9	2.8 ± 1.6
Sperm motility (%)	57.8 ± 2.6	0 ± 0	18.4 ± 4.0	5.5 ± 2.1
Sperm morphology (%)	17.1 ± 1.5	0 ± 0	10.1 ± 3.3	9.6 ± 5.5
Hormones and endocrine disruptor				
FSH (mU/ml)	2.6 ± 0.8	22.6 ± 2.1	8.6 ± 1.3	19.9 ± 3.6
LH (mU/ml)	3.2 ± 0.6	9.5 ± 1.0	5.9 ± 1.0	9.2 ± 2.2
Prolactin (ng/ml)	6.8 ± 1.0	13.4 ± 2.6	11.8 ± 2.1	10.0 ± 1.3
Total testosterone (ng/dl)	620.0 ± 119.4	458 ± 38.2	445.9 ± 53.0	423.7 ± 30.7
AMH (ng/ml)	11.1 ± 0.7	6.0 ± 0.8	6.6 ± 0.6	6.3 ± 1.1
Inhibin B (pg/ml)	133.0 ± 8.8	33.7 ± 5.6	65.6 ± 6.3	38.7 ± 5.1
BPA (ng/ml)	0.4 ± 0.1	1.2 ± 0.5	0.6 ± 0.4	0.2 ± 0.1

Data are given as mean ± standard error of the mean (SEM).
AMH: anti-Müllerian hormone; BPA: Bisphenol A; FSH: follicular stimulating hormone; INOA: idiopathic non-obstructive azoospermia; LH: luteinizing hormone.

obstructive oligo-astheno-teratozoospermia (OAT), cryptorchidism and varicocele.

2. Patients and Methods

Study characteristics. Cross-sectional, case-control, clinical study.

Study population. Fifty-five infertile men, attending the outpatient clinics of the Unit of Reproductive Endocrinology, First Department of Obstetrics and Gynecology, Aristotle University of Thessaloniki, Greece, were prospectively recruited and studied from January to December 2017.

General inclusion criteria were: i) infertility, defined as not achievement of a pregnancy after 12 months of unprotected intercourse, ii) two spermiograms showing azoospermia (absence of sperm in the semen) or OAT, sperm concentration $< 15 \times 10^6$/ml or motility type a and b < 32% or normal morphology < 4%) with time interval of at least 70 days. Of these two spermiograms, the first was used for the data analysis and the second for confirmatory reasons.

Specific inclusion criteria were: i) men with grade II (palpable) or grade III (visible) varicocele and OAT (varicocele group), ii) history of unilateral or bilateral cryptorchidism, independently of the age of its surgical correction (cryptorchidism group); iii) men with no apparent cause of azoospermia, following the conduction of the standard diagnostic procedure.

Exclusion criteria were: i) presence of obstructive azoospermia [confirmed by normal spermatogenesis in testicular sperm extraction (TESE) procedure], ii) presence of causes of azoospermia or OAT other than INOA, cryptorchidism or varicocele (such as Kallmann syndrome, Klinefelter syndrome, testicular cancer).

Twenty-five fertile men from the general population, with normal semen parameters, whose partners were in the first trimester of pregnancy, were used as controls. The control men were recruited from the Obstetrics outpatient clinics. All participants provided information on demographic characteristics and occupational history, through an in-person interview.

3. Methods

All participants underwent andrologic evaluation, including history, physical examination, serum profile of hormones FSH, LH, prolactin, tT, sex hormone-binding globulin (SHBG), anti-Müllerian hormone (AMH) and inhibin B (InhB) and, at least, two spermiograms. All infertile men underwent a standard diagnostic procedure (personal and family history, general and external genitalia clinical evaluation, hormonal assessment, spermiograms, seminal culture, testicular ultrasound, karyotype, Yq microdeletions) as clinically indicated. Informed consent was obtained from all participants, and the study protocol was approved by the Bioethics Committee, Aristotle University of Thessaloniki, Greece.

Hormone assays. Fasting blood samples were obtained at 09:00 and centrifuged for 20 min. The serum was separated and stored at −80 °C until analysis was performed.

All assays were performed using immunoenzymatic techniques IVD and RUO. Serum concentrations of FSH, LH, tT and prolactin were measured using the Immulite 2000 immunoassay system, (Siemens Healthineers Tarrytown, NY, USA).

Serum concentrations of the specific analytes AMH and Inh B were measured commercial two-site sandwich-type enzyme linked immunosorbent assays (Diagnostic Systems Laboratories, Inc. Webster, TX, USA). According to the manufacturer the inter-assay and intra-assay coefficients of variation were 4.8% and 2.4% for AMH and 6.2% and 3.5% for Inh-B, respectively.

Determination of BPA in sera of patients and controls was performed using a competitive immunoassay (IBL International GmbH, Hamburg, Germany). According to the manufacturer the intra- and interassay coefficients of variation ranged 5.5–14.0 and 4.3–5.2%, respectively.

Semen analysis. Semen was collected by on-site masturbation, in a sterile plastic specimen cup, after 3–7 days of abstinence. Spermiogram was performed in all subjects twice. The samples of men with azoospermia were centrifuged at 600 g for 10 min. Sperm concentration, motility and morphology were evaluated according to the World Health Organization (WHO) criteria (WHO, 2010).

Statistical analysis. Data are expressed as median (Interquartile Range - IQR). Kruskal-Wallis test was used for the comparison of numerical parameters among groups, with Mann-Whitney as *post hoc* test. Correlation between study parameters was made by Spearman *rho* test. A p value of less than 0.05 was considered statistically significant. Statistical analysis was performed with SPSS 21 for Windows (SPSS Inc., USA).

4. Results

Demographic characteristics, clinical, hormonal and sperm features in fertile and infertile men, accordance with the cause of infertility are presented in Table 1.

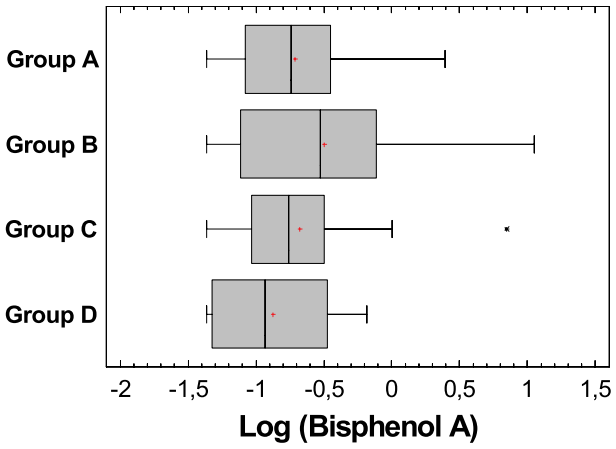

Fig. 1. Comparison of serum Bisphenol A concentrations in Group A: Fertile (Controls), [95% CI -0.920 to −0.507]; Group B: INOA (Idiopathic non-Obstructive Azoospermia), [95% CI -0.807 to −0.193]; Group C: Varicocele), [95% CI -0.932 to −0.426]; and Group D: Cryptorchidism, [95% CI: −1.174 to −0.581] (ANOVA, p > 0.230). Boxes represent the interquartile range; lines inside boxes represent the median value; cross represents mean marker; whiskers represent the lowest and highest observations, respectively.

BPA was detected in all infertile and control men. There was no difference in BPA concentrations between infertile and control men [median (IQR) 0.19 (0.45) vs. 0.18 (0.28) ng/ml, p = 0.689] (Fig. 1) or among the cause of infertility [azoospermia 0.30 (0.69), cryptorchidism 0.12 (0.39), varicocele 0.17 (0.23) ng/ml, p = 0.316]. However, BPA concentrations > 3 ng/ml were observed in the group of men with infertility. Furthermore, a weak but significant negative correlation was observed between BPA concentrations and AMH (r = −0.320, p = 0.02) in the whole group of subjects (Fig. 2).

5. Discussion

Although there are indications that ED have adverse effects on the male reproductive system, the evidence is limited. In the present study, we assessed the association between plasma BPA concentrations and semen quality in infertile men of specific etiology. We found no difference in serum BPA concentrations between infertile and fertile men, but we observed very high concentrations of BPA only in men with azoospermia.

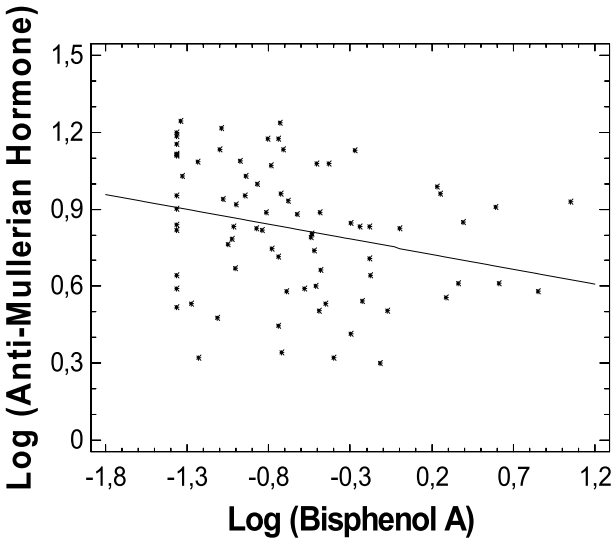

Fig. 2. Correlation between Bisphenol A and anti-Müllerian Hormone concentrations of all men participating in the study (r = -0.320, p = 0.02).

Few studies assessed the impact of BPA in Greek populations. This is the first study indicating 100% exposure to BPA in serum samples collected from 80 Greek adult men. Tzatzarakis et al. demonstrated that BPA was detected in the 41.2% of hair samples collected from urban population and in the 14.8% of hair samples collected from rural population (Tzatzarakis et al., 2015). Furthermore, BPA was detected in the 90% of urine samples from men in Athens and in almost all urine samples (99%) of Greek children of pre-school age (Myridakis et al., 2016).

Animal studies have shown that exposure to BPA could have detrimental effects on the male reproductive system (Talsness et al., 2000; Tohei et al., 2001; Williams et al., 2001; Han et al., 2004; Herath et al., 2004; Foster, 2006; Richter et al., 2007; Salian et al., 2009). As a potent ED, BPA has been shown to have both estrogenic and anti-androgenic properties, which provides biological plausibility for an adverse effect of BPA on the male reproductive system. However, human studies examining this effect have been limited. The results from this study provide evidence that increased BPA concentrations could explain some cases of men with otherwise idiopathic infertility. As we did not find association between serum BPA concentrations and reduced semen quality parameters among infertile men, male infertility cannot be attributed to exposure to BPA; nevertheless, high concentrations of BPA could contribute to infertility.

These results are in accordance with some (Vitku et al., 2016), but not all previous evidence (Meeker et al., 2010; Li et al., 2011; Knez et al., 2014). The discrepancies could be attributed to the biological specimen where BPA was measured, as many studies measured BPA in urine or seminal plasma. Additionally, the sample selection may result in different results: some studies selected men from the general population (Lassen et al., 2014), while others from highly exposed groups (Li et al., 2011) or from couples attending infertility clinics (Meeker et al., 2010; Knez et al., 2014).

However, this study has certain limitations. It is demonstrated that non-persistent chemicals, such as phthalates and BPA, show large intra-individual and day-to-day variations (Frederiksen et al., 2013). Furthermore, it is evident that there is a gap between time of exposure and manifestation of effects which may be more than 20 years. Humans are also exposed to a large number of chemicals (Suk et al., 2002), which, although they have different modes of action, could affect the same hormone system in a dose-additive manner (Hass et al., 2007; Rider et al., 2010).

The mechanisms of the adverse effect of BPA on the male reproductive system have not been clarified. BPA binds to both estrogen receptors (ERα and ERβ), with 10-fold higher affinity to ERβ. Given that the affinity of BPA for ERs is 10,000- to 100,000-fold weaker than that of E_2, it had been considered to be a very weak environmental estrogen (Vandenberg et al., 2007). However, it became evident that BPA can stimulate cellular responses at very low concentrations, not only by binding to the classic, genomic (nuclear) ERs, but through non-genomic (membrane-associated) mechanisms (vom Saal et al., 2007). BPA alters endocrine function through multiple pathways (Wetherill et al., 2007). Animal studies have reported adverse reproductive effects in males exposed to low concentrations of BPA in early life or in adulthood (Meeker et al., 2010; Richter et al., 2007; Cao et al., 2012; Wisniewski et al., 2015). In humans, BPA has been measured in maternal and fetal plasma and placental tissue at birth (Vandenberg et al., 2007; Diamanti-Kandarakis et al., 2009), in the amniotic fluid (Ikezuki et al., 2002; Schonfelder et al., 2002) and in the milk of lactating mothers (Sun et al., 2004; Ye et al., 2006; Kuruto-Niwa et al., 2007). These data indicate that the fetus and neonate are readily exposed to this chemical (Vandenberg et al., 2007; Diamanti-Kandarakis et al., 2009; Calafat et al., 2008; Geens et al., 2009). Finally, BPA has been detected in the follicular fluid of women IVF (Ikezuki et al., 2002). In any case, the impact of BPA on spermatogenesis (Li et al., 2011) has to be assessed in large, appropriately designed studies.

In conclusion, this study provided evidence that although male

infertility cannot be associated with exposure to BPA, high concentrations of BPA could contribute to infertility. Further research is required in order to explore ED actions and to establish methods to avoid exposure of the population to these substances.

Declaration of interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

Acknowledgments

Funding was received from National and Kapodistrian University of Athens, and Aristotle University of Thessaloniki, Greece, to (G.M. and D.G.G). The funding sources played no role in the study design; in the collection, analysis, and interpretation of data; in the writing of the report; or in the decision to submit the report for publication.

Transparency document

Transparency document related to this article can be found online at https://doi.org/10.1016/j.fct.2019.02.016.

References

Buck Louis, G.M., Sundaram, R., Sweeney, A.M., Schisterman, E.F., Maisog, J., Kannan, K., 2014. Urinary bisphenol A, phthalates, and couple fecundity: the longitudinal investigation of fertility and the environment (LIFE) study. Fertil. Steril. 101 (5), 1359–1366.

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

Cao, J., Mickens, J.A., McCaffrey, K.A., Leyrer, S.M., Patisaul, H.B., 2012. Neonatal Bisphenol A exposure alters sexually dimorphic gene expression in the postnatal rat hypothalamus. Neurotoxicology 33 (1), 23–36.

Diamanti-Kandarakis, E., Bourguignon, J.P., Giudice, L.C., Hauser, R., Prins, G.S., Soto, A.M., Zoeller, R.T., Gore, A.C., 2009. Endocrine-disrupting chemicals: an Endocrine Society scientific statement. Endocr. Rev. 30 (4), 293–342.

Frederiksen, H., Kranich, S.K., Jorgensen, N., Taboureau, O., Petersen, J.H., Andersson, A.M., 2013. Temporal variability in urinary phthalate metabolite excretion based on spot, morning, and 24-h urine samples: considerations for epidemiological studies. Environ. Sci. Technol. 47, 958–967.

Foster, P.M., 2006. Disruption of reproductive development in male rat offspring following in utero exposure to phthalate esters. Int. J. Androl. 29, 140–147.

Geens, T., Roosens, L., Neels, H., Covaci, A., 2009. Assessment of human exposure to Bisphenol-A, Triclosan and Tetrabromobisphenol-A through indoor dust intake in Belgium. Chemosphere 76 (6), 755–760.

Han, X.D., Tu, Z.G., Gong, Y., Shen, S.N., Wang, X.Y., Kang, L.N., Hou, Y.Y., Chen, J.X., 2004. The toxic effects of nonylphenol on the reproductive system of male rats. Reprod. Toxicol. 19, 215–221.

Hanaoka, T., Kawamura, N., Hara, K., Tsugane, S., 2002. Urinary bisphenol A and plasma hormone concentrations in male workers exposed to bisphenol A diglycidyl ether and mixed organic solvents. Occup. Environ. Med. 59 (9), 625–628.

Hass, U., Scholze, M., Christiansen, S., Dalgaard, M., Vinggaard, A.M., Axelstad, M., Metzdorff, S.B., Kortenkamp, A., 2007. Combined exposure to anti-androgens exacerbates disruption of sexual differentiation in the rat. Environ. Health Perspect. 115, 122–128.

Herath, C.B., Jin, W., Watanabe, G., Arai, K., Suzuki, A.K., Taya, K., 2004. Adverse effects of environmental toxicants, octylphenol and bisphenol A, on male reproductive functions in pubertal rats. Endocrine 25, 163–172.

Ikezuki, Y., Tsutsumi, O., Takai, Y., Kamei, Y., Taketani, Y., 2002. Determination of bisphenol A concentrations in human biological fluids reveals significant early prenatal exposure. Hum. Reprod. 17 (11), 2839–2841.

Irvine, D.S., 1998. Epidemiology and aetiology of male infertility. Hum. Reprod. 13 (Suppl. 1), 33–44.

Juul, S., Karmaus, W., Olsen, J., 1999. Regional differences in waiting time to pregnancy: pregnancy-based surveys from Denmark, France, Germany, Italy and Sweden. The European Infertility and Subfecundity Study Group. Hum. Reprod. 14 (5), 1250–1254.

Kabir, E.R., Rahman, M.S., Rahman, I., 2015. A review on endocrine disruptors and their possible impacts on human health. Environ. Toxicol. Pharmacol. 40 (1), 241–258.

Knez, J., Kranvogl, R., Breznik, B.P., Voncina, E., Vlaisavljevic, V., 2014. Are urinary bisphenol A levels in men related to semen quality and embryo development after medically assisted reproduction? Fertil. Steril. 101 (1), 15–221 e5.

Kuehn, B.M., 2007. Expert panels weigh bisphenol-A risks. Jama 298 (13), 1499–1503.

Kuruto-Niwa, R., Tateoka, Y., Usuki, Y., Nozawa, R., 2007. Measurement of bisphenol A concentrations in human colostrum. Chemosphere 66 (6), 1160–1164.

Lassen, T.H., Frederiksen, H., Jensen, T.K., Petersen, J.H., Joensen, U.N., Main, K.M., Skakkebaek, N.E., Juul, A., Jorgensen, N., Andersson, A.M., 2014. Urinary bisphenol A levels in young men: association with reproductive hormones and semen quality. Environ. Health Perspect. 122 (5), 478–484.

Li, S., Zhou, Z., Miao, M., He, Y., Wang, J., Ferber, J., Herrinton, L.J., Gao, E.,, Yuan, W., 2011. Urine bisphenol-A (BPA) level in relation to semen quality. Fertil. Steril. 95 (2), 625–630.

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

Meeker, J.D., Ehrlich, S., Toth, T.L., Wright, D.L., Calafat, A.M., Trisini, A.T., Ye, X., Hauser, R., 2010. Semen quality and sperm DNA damage in relation to urinary bisphenol A among men from an infertility clinic. Reprod. Toxicol. 30 (4), 532–539.

Mendiola, J., Jorgensen, N., Andersson, A.M., Calafat, A.M., Ye, X., Redmon, J.B., Drobnis, E.Z., Wang, C., Sparks, A., Thurston, S.W., Liu, F., Swan, S.H., 2010. Are environmental levels of bisphenol a associated with reproductive function in fertile men? Environ. Health Perspect. 118 (9), 1286–1291.

Myridakis, A., Chalkiadaki, G., Fotou, M., Kogevinas, M., Chatzi, L., Stephanou, E.G., 2016. Exposure of preschool-age Greek children (RHEA cohort) to bisphenol A, parabens, phthalates, and organophosphates. Environ. Sci. Technol. 50 (2), 932–941.

Richter, C.A., Birnbaum, L.S., Farabollini, F., Newbold, R.R., Rubin, B.S., Talsness, C.E., Vandenberg, J.G., Walser-Kuntz, D.R., vom Saal, F.S., 2007. In vivo effects of bisphenol A in laboratory rodent studies. Reprod. Toxicol. 24, 199–224.

Rider, C.V., Furr, J.R., Wilson, V.S., Gray Jr., L.E., 2010. Cumulative effects of in utero administration of mixtures of reproductive toxicants that disrupt common target tissues via diverse mechanisms of toxicity. Int. J. Androl. 33, 443–462.

Salian, S., Doshi, T., Vanage, G., 2009. Neonatal exposure of male rats to Bisphenol A impairs fertility and expression of sertoli cell junctional proteins in the testis. Toxicology 265, 56–67.

Schonfelder, G., Wittfoht, W., Hopp, H., Talsness, C.E., Paul, M., Chahoud, I., 2002. Parent bisphenol A accumulation in the human maternal-fetal-placental unit. Environ. Health Perspect. 110 (11), A703–A707.

Sharlip, I.D., Jarow, J.P., Belker, A.M., Lipshultz, L.I., Sigman, M., Thomas, A.J., Schlegel, P.N., Howards, S.S., Nehra, A., Damewood, M.D., Overstreet, J.W., Sadovsky, R., 2002. Best practice policies for male infertility. Fertil. Steril. 77 (5), 873–882.

Suk, W.A., Olden, K., Yang, R.S., 2002. Chemical mixtures research: significance and future perspectives. Environ. Health Perspect. 110, 891–892.

Sun, Y., Irie, M., Kishikawa, N., Wada, M., Kuroda, N., Nakashima, K., 2004. Determination of bisphenol A in human breast milk by HPLC with column-switching and fluorescence detection. Biomed. Chromatogr. 18 (8), 501–507.

Talsness, C., Fialkowski, O., Gericke, C., Merker, H.J., Chahoud, I., 2000. The effects of low and high doses of bisphenol A on the reproductive system of female and male rat offspring. Congenital. Anom. 40, 94–107.

Tohei, A., Suda, S., Taya, K., Hashimoto, T., Kogo, H., 2001. Bisphenol A inhibits testicular functions and increases luteinizing hormone secretion in adult male rats. Exp. Biol. Med. 226, 216–221.

Tzatzarakis, M.N., Vakonaki, E., Kavvalakis, M.P., Barmpas, M., Kokkinakis, E.N., Xenos, K., Tsatsakis, A.M., 2015. Biomonitoring of bisphenol A in hair of Greek population. Chemosphere 118, 336–341.

Vandenberg, L.N., Hauser, R., Marcus, M., Olea, N., Welshons, W.V., 2007. Human exposure to bisphenol a (bpa). Reprod. Toxicol. 24, 139–177.

Vitku, J., Heracek, J., Sosvorova, L., Hampl, R., Chlupacova, T., Hill, M., Sobotka, V., Bicikova, M., Starka, L., 2016. Associations of bisphenol A and polychlorinated biphenyls with spermatogenesis and steroidogenesis in two biological fluids from men attending an infertility clinic. Environ. Int. 89–90, 166–173.

vom Saal, F.S., Akingbemi, B.T., Belcher, S.M., Birnbaum, L.S., Crain, D.A., Eriksen, M., Farabollini, F., Guillete, L.J., Hauser, R., Heindel, J.J., Ho, S.M., Hunt, P.A., Iguchi, T., Jobling, S., Kanno, J., Keri, R.A., Knudsen, K.E., Laufer, H., LeBlanc, G.A., Marcus, M., et al., 2007. Chapel Hill bisphenol A expert panel consensus statement: integration of mechanisms, effects in animals and potential to impact human health at current levels of exposure. Reprod. Toxicol. 24 (2), 131–138.

Wetherill, Y.B., Akingbemi, B.T., Kanno, J., McLachlan, J.A., Nadal, A., Sonnenschein, C., Watson, C.S., Zoeller, R.T., Belcher, S.M., 2007. In vitro molecular mechanisms of bisphenol a action. Reprod. Toxicol. 24, 178–198.

WHO Press, World Health Organization, 20 Avenue Appia, 1211 Geneva 27, Switzerland.

Williams, K., McKinnell, C., Saunders, P.T., Walker, M., Fisher, J.S., Turner, K.J., Atanassova, N., Sharpe, M., 2001. Neonatal exposure to potent and environmental oestrogens and abnormalities of the male reproductive system in the rat: evidence for importance of the androgen-oestrogen balance and assessment of the relevance to man. Hum. Reprod. Update 7, 236–247.

Wisniewski, P., Romano, R.M., Kizys, M.M., Oliveira, K.C.,, Kasamatsu, T., Giannocco, G., Chiamolera, M.I., Dias-da-Silva, M.R., Romano, M.A., et al., 2015. Adult exposure to bisphenol A (BPA) in Wistar rats reduces sperm quality with disruption of the hypothalamic-pituitary-testicular axis. Toxicology 329, 1–9.

Wong, E.W., Cheng, C.Y., 2011. Impacts of environmental toxicants on male reproductive dysfunction. Trends Pharmacol. Sci. 32 (5), 290–299.

Ye, X., Kuklenyik, Z., Needham, L.L., Calafat, A.M., 2006. Measuring environmental phenols and chlorinated organic chemicals in breast milk using automated on-line column-switching-high performance liquid chromatography-isotope dilution tandem mass spectrometry. J. Chromatogr. B. Analyt. Technol. Biomed. Life Sci. 831 (1–2), 110–115.