Endocrine Disrupting Chemicals - the X Factor in Different Pathologies

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Endocrine disruptors (ED) are exogenous agents that interfere with the normal function of the endocrine system and they are considered environmental chemicals with estrogen-like and/or anti-androgenic activity with important impact on the reproductive axis. They act via nuclear receptors, non-nuclear steroid receptors, nonsteroidal receptors, orphan receptors, and different enzyme pathways involved in the biosynthesis and/or metabolism of steroids. The molecules identified as ED and sources of exposure are diverse and vary worldwide, including from natural chemicals found in human and animal food (the main source) up to synthetic chemicals, such as as solvents, plasticizers, pesticides, fungicides, pharmaceuticals etc. ED are incriminated in the occurrence of malignant tumors, birth defects, attention deficit disorders, cognitive impairment, brain development, deformations of the body (including limbs), disturbance of sexual development, menstrual irregularity, precocious puberty, feminizing or masculinizing effects, abortion, not least obesity and decreased fertility. The principles of action are still studied and controversial, therefore, it is difficult to determine the minimum level at which adverse effect occurs and further studies are required.

Key words: endocrine disruptors, endocrine systems, sources, pathology, outcome

Endocrine disruptors (ED) are exogenous agents that interfere with the synthesis, secretion, transport, metabolism, binding or elimination of hormones responsible for homeostasis, reproduction and development of the body [1-3]. They are considered environmental chemicals with estrogen-like and/or anti-androgenic activity with important action over the reproductive axis, which can cause infertility. Any system from the human body that has a hormonal control can be affected by ED [2,4-7]. Because of the complexity of the endocrine system, the mechanisms of action for ED are hard to identify. Therefore, they act via nuclear receptors, non-nuclear steroid receptors, nonsteroidal receptors (e.g.: neurotransmitter receptors such as $C_{10}H_{12}N_2O$ (serotonin), $C_8H_{11}NO_2$ (dopamine), $C_8H_{11}NO_3$ (norepinephrine)), orphan receptors (aryl hydrocarbon – Ahr, the most studied protein in the interaction with ED), enzyme pathways involved in the biosynthesis and/or metabolism of steroids, interfering with cell signaling pathways and hormonal regulated gene expression [1,3].

Estrogen receptors (ER) are coded by two different genes located on chromosome 6 in the locus 6q25.1 for ERα and chromosome 14 in the connection between loci 14q11.1 and 14q11.2 for ERβ. As a result, the utilization of different promoters ends in multiple variants that code for the same protein, 66 kDa for ERα and 55 kDa for ERβ, but the use of various promoters allows a fine tissue-specific regulation of ER expression, allowing the modulation of transcript synthesis and regulating their stability and translational efficiency. In addition, several splice variants were identified, such as ERα-46 and ERα-36. ERα-46 is deleted from the N-terminal part of the protein and, consequently, inhibits the transcriptional activity of ERα-66 in various types of cells. ERα -36, a more recent discovery, lacks both the N- and C-terminal domains, resulting in a form with none of the two activation functions. ERα-36 is capable of acting as a dominant negative form of ERα-66 being also found anchored to the plasma membrane where it can modulate the activation of intracellular signaling pathways, such as the PI3K/Akt or MAPK.

As a result, various tissues express both ER subtypes in variable quantities. A strong expression of ER's is met in tissues related to female reproduction (e.g.: ovary, womb, mammary gland). In both sexes, lung, hepatic, fat, osseous, nervous tissues, and endothelial cells express both receptors with variable levels [1-4].

The molecules identified as ED and sources of exposure are diverse and vary worldwide (table 1). The list includes synthetic chemicals used as solvents / lubricants (($C_{12}H_{10n}Cl_n$ (polyclorinated biphenyls-PCBs) , $C_{12}H_{10-n}Br_n$ (polybrominated biphenyls- PBBs), $C_{12}H_4Cl_4O_2$ (dioxins)), plastics ($C_{15}H_{16}O_2$ (bisphenol A (BPA)) and $C_{12}H_{10}O_4S$ (bisphenolS-BPS)), plasticizers (phthalates), pesticides (($C_{16}H_{15}Cl_3O_2$ (methoxychlor), $C_9H_{11}Cl_3NO_3PS$ (clorpyrifos), $C_{14}H_9Cl_5$ (dichlorodiphenyl-trichloroethane-DDT), $C_8H_{14}ClN_5$ (atrazine), $C_{22}H_{19}Cl_2NO_3$ (cypermethrin), $C_{12}H_8Cl_6O$ (dieldrin)], fungicides ($C_{12}H_9Cl_2NO_3$ (vinclozolin)) and pharmaceuticals ($C_{18}H_{20}O_2$ (diethylstilbestrol), $C_{20}H_{24}O_2$ (ethinyl estradiol)) [1-4]. Natural chemicals found in human and animal food (phytoestrogens including $C_{15}H_{10}O_5$ (genistein) and $C_{15}H_8O_5$ (cumestrol)) can act as ED [8].

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Additionally, heavy metals and metalloids can have estrogenic activity [1,3]. However, food is the main source through which people are exposed to these substances, even indirectly many of these compounds being liposoluble and being found accumulated in the fatty tissue of animals that people consume (for instance, both wild salmon and the breeding one has been shown to contain synthetic organic compounds) [2,9]. There is a continuously updated database which includes all inscriminated compounds with possible ED activity, with a total of about 1038 substances until 2016 [10].

ED are also incriminated in the occurrence of malignant tumors (e.g.: breast, prostate), birth defects, attention deficit disorders, cognitive impairment, brain development, deformations of the body (including limbs), disturbance of sexual development, menstrual irregularity, precocious puberty, feminizing or masculinizing effects, abortion, not least obesity and decreased fertility [2,10,11]. Intrauterine development can be disrupted causing irreversible effects and they can induce transgenerational defects [3]. Other target organs are: the neuroendocrine system, thyroid, cardiovascular system, mammary gland, fat, pancreas, ovary and uterus, testis and prostate [1]. However, there are studies which confirm that some substances considered ED's are in fact safe [12]. Isoflavones and plant extracts from soy and red clover do not promote the growth of human cancer cells but decrease cell proliferation and increase apoptosis and cell cycle arrest [10,12].

Infertility, defined as the inability to conceive after one year of unprotected sexual contacts, has a global prevalence of approximately 9% [13]. In infertile couples, the female cause is predominant in 38, and 20% in males (usually pathologies related to testis and prostate) [14]; the mixed cause represents 27% and infertility without a cause is considered to be around 15% [15]. The role of ED in the increasing incidence of unknown cause infertility is partilly demonstrated by consistent detection of ED residues in human serum, semen and follicular fluid [16].

The principles of action are still studied and controversial, therefore, it is difficult to determine the minimum level at which adverse effect occurs [15-17]. Current data conlcuded that the most important effects of disruptors appear following exposure to low environmental doses than at high doses and can cause receptor down-regulation and cytotoxicity [17]. Some studies suggested that $C_{12}H_5Br_5O$ (PBDE-47) (flame retardant agent) induces a negative imapct on the reproductive system and thyroid gland of female mice in doses to which humans are typically exposed [17]. The effects of ED can be significant in vulnerable individuals due to factors such as genetic background, exposure time and pre-existing pathologies that unables the epuration of this substances, like chronic kidney disease with or without dialysis [3,4,18-24].

Other factors involved in modulating these actions are age at time of exposure, latency (usually consequences are late not immediate) and compensatory mechanisms of elimination/inactivation of ED (which may be altered in liver or kidney diseases) [1,3,25]. There have been described the *windows of susceptibility*, where adverse effects can be drastic and irreversible, including congenital abnormalities (intrauterine exposure), while in a non-critical window mild functional deficits and diseases with adulthood onset may appear [1,3]. In addition, the effects of different types of ED compounds may be additive or synergistic [1].

Endocrine disrupting compounds may affect not only the individual but also future generations by altering factors that regulate gene expression [1,3]. Although there is chronic exposure to ED through inhalation or skin contact, food intake remains the major route of contact (meat, fish, dairy products and vegetables) as well as water and other beverages. Human food may contain environmental pollutants such as heavy metals and pesticide residues and additives in addition to anabolic steroids used in processing and food production [26-28]. Most individuals have detectable traces of these substances in serum or urine [3,26-28]. As an example, recent studies have shown that plastic packaging is an important source of ED in the human diet. Repeated exposure of these to ultraviolet light (UV), heat, acids / alkalis can cause polymers split into monomers such as phthalates and BPA which are found later in food or drinks. Some of these ED are replaced with thermally stable analogues (BPA-free products), but these also contain BPS which have both genomic and non-genomic disrupting effects at low picomolar concentrations [26-28]. Another example is phyto-estrogens, with benefits over the cardiovascular system and related to menopause, for which the minimum effective doses are not yet determined. Studies have shown that in children who are soy-based formula milk fed, serum levels of phytoestrogens are 13000-22000 higher than endogenous estrogen levels, causing concerns because of the possible adverse effects on brain, body morphology, functionality of reproductive organs and on fertility [3,26-28]. Taking all of this into consideration, the main objective of this review was to synthesize current knowledge regarding the ED for a better understanding of the sources, mechanism of action, effects on different hormone-regulated organs and long term consequences.

The most common EDs

Bisphenol A

It is one of the most investigated and potent ED [25,26]. It is used extensively especially in plastic polycarbonate and epoxy resins (Table 1), exposure being widespread (the largest study revealed the presence of BPA in urine samples of 92.6% of American men) [26-28].

Bisphenol A has been considered the prototype of non-steroidal ED which interferes with nuclear estrogen receptors in targeted tissues [28,29].The presence of BPA in women has been associated with decreased serum levels of estradiol, reduced egg production, low number of mature oocytes and normal fertilized oocytes fertilizer [28]. Multiple studies in children have shown an intake of BPA above the limit considered safe of 50μg/ kg/ day [30]. Other worrying effects include changes in the dopamine and reward system leading to impulsivity, hyperactivity and increased susceptibility to drug addiction [31]. High level of BPA is incriminated in the occurrence of endometriosis in experimental studies. Other potential mechanisms are inhibition of apoptotic activity in the breast and increased breast density [3,28-31].

Phthalates

Phthalates (butilbenzilphthalate (BBP), di-N butilphthalate (DBP), di-2etilhexilphthalate (DEHP)) are commonly used as plasticizers in polyvinyl chloride (PVC) products, in personal care products (cans for food, adhesives, perfumes, eyeshadow), toys (notably DEHP), interior construction and biomedical devices [32-34]. However, phthalates are not covalently bonded to plastic and can therefore be released into the environment over time and with usage. Exposure is primarly oral and inhalation. Daily intake in the general population is about 30 μg/kg [29,30,35]. Phthalates are a major group of antiandrogenic substances considered toxic to the reproductive system [36]. They can exercise their anti-

androgenic action by inhibiting testosterone synthesis in Leydig cells (a result of a dysfunction of cytochrome CYP 17) and they can disrupt gene expression that regulate cholesterol homeostasis, lipid and insulin signaling, and also decreasing testosterone levels [37-40]. Rats exposed to phthalates developed multiple abnormalities: cryptorchidism, small testes, micropenis, ductus deferens and epididymis damage, atrophy of the seminiferous tubules [30,39]. Reduced anogenital distance occurs and also an increased incidence of hypospadias [40]. In women, phthalates have been associated with premature sexual development [3,41].

Alkylphenols

The most frequent are ethoxylate-nonylpnehol (most commercial) and octylfenol ethoxylate. These compounds undergo metabolic cleavage in the environment and lose ethylene oxide chains to become alkylphenols. Unlike most exogenous chemicals that usually become less toxic by biodegradation, alkylphenols increase their toxicity during this process [30,34,42]. Their effects were studied on rats, being incriminated in: low weight of reproductive organs, delayed testicular descensus, reduced spermatogenesis, Sertoli cell apoptosis, inhibition of testosterone biosynthesis [30,43].

Chlorinated hydrocarbons

Organochlorine compounds represent an important category of ED because of their prolonged retention in the body [33,44]. They include dichlorodiphenyltrichloroethane (DDT) and its metabolites, gamma-hexachlorociclo-hexane (gammaHCH), polychlorinated biphenyls (PCBs), polychlorinated dibenzofurans (PCDFs) and poly-chlorinated dibenzo-p-dioxins (PCDDs). Although the production and use of DDT, PCBs and related compounds has been banned in industrialized countries, they are still available in biological samples. Organochlorine products bioaccumulate in fatty tissue [28,33,44]. However, DDT and metabolites, gamma-HCH, PCB and PCDD can be found in fluids such as semen, cervical mucus and follicular fluid [32,42]. The major routes of contamination are meat, milk and milk products, mother milk and medication for ectoparasitic diseases (gamma-HCH – lindane) [27], with a half-life of 7-11 years in the body [45]. PCBs can mimic the actions of estrogen and are associated with low sperm count, impaired sexual organs, changes in testosterone and estrogen, precocious puberty, testicular volume reduction, reduced enzyme activity (for example 3β hydoxysteroid dehydrogenase (3 βHSD)) [46]. Dioxins cause alteration of breast development with increased susceptibility for breast cancer (inhibit cyclooxygenase 2 via AHR). DDT induce sexual precocity and reduced fertility in daughters of exposed women [2].

Different effects of EDs

Female gonadal axes anomalies caused by ED

Pathologies of the female reproductive system where the activity of ED has been incriminated are endometriosis, uterine and ovarian diseases such as premature ovarian failure and polycystic ovarian syndrome [47]. Preantral primordial follicles and exposure to ED can affect folliculogenesis, causing meiotic aberrations (e.g.: aneuploidy) or follicular atresia [47]. Granulosa and theca cells, which are crucial for the development of oocytes and ovarian steroidogenesis are a target for ED action. PCB alter the synthesis of GnRH and decreases its release. On the other hand, DDT and BPA stimulates it [3,47].

Endometriosis

This represents a estrogen-dependent gynecological disorder with an incidence of 10-15% which can cause infertility, having a multifactorial etiology (genetic, hormonal, immunological, environmental factors) [48,49]. Higher levels of BPA and phthalates were revealed in women with this condition [48]. Moreover, women exposed to diethylstilbestrol (DES) in utero may have an 80% higher risk of endometriosis than those unexposed [50]. Experimental studies claim that exposure of mice to BPA or 2,3,7,8-Tetrachlorodibenzodioxin (TCDD) causes a uterine endometriosis phenotype and that women with endometriosis have significantly higher concentrations of TCDD and PCB in the peritoneal fluid, therefore being responsible for chronic inflammation that can stimulate endometrial cells derived from retrograde menstruation [3,51,52]. Other studies provide evidence that among infertile women, the mean expression of estrogen receptor alpha (ERα) and beta (ERβ), androgen receptor (AR) and pregnane X receptor (PXR) was significantly higher compared to the fertile patients. Patients with endometriosis have higher levels of peroxisome proliferator-activated receptor γ (PPARγ) than all women with other causes of infertility [51,52].

Polycystic ovary syndrome (PCOS)

This condition is a heterogeneous syndrome characterized by persistent anovulation, oligo or amenorrhea and hyperandrogenism affecting 5-8% of women of childbearing age often leading to infertility [1,3]. Women with PCOS have higher risk of being associated with insulin resistance, diabetes, chronic kidney disease, endometrial cancer and infertility by anovulation [15,53-55]. Recent studies point to the higher levels of BPA in biological fluids of women with PCOS and its role in the pathogenesis of hyperandrogenism and hyperinsulinemia. It seems that the mother's exposure to BPA during pregnancy may determine the development of PCOS in female offspring [55]. A presumed etiology for PCOS is excessive prenatal exposure to testosterone resulting both from genetic predisposition to a tesosterone hypersecretion and exposure to environmental factors that increase embryonic tesosterone [56,57]. One of the ED associated with PCOS is BPA [47]. BPA was measured in serum, follicular fluid and also in fetal serum and amniotic fluid confirming the transplacental passage [47]. Studies confirm that increased BPA concentration in the follicular fluid of PCOS patients may play an important role in its pathogenesis by down-regulating the expression of aromatase in granulosa cells [53,58]. However, increased BPA may be a result rather than a cause of PCOS because women with this pathology have a higher circulating level of tesosteron than healthy ones, and increased concentrations of androgens lowers the clearance of BPA [59].

Premature ovarian failure (POF)

POF represents the cessation of normal ovarian function before 40 years, with an incidence of 1% in women of childbearing age and increasing prevalence in modern society [60]. Exposure to BPA in utero mice resulted in impaired oocytes [61]. In women exposed to DES in utero studies have reported an earlier age of menopause. These observations are consistent with the low follicular reserve and the low number of ovulated oocytes occurring in mice exposed to DES after ovulation induction with gonadotropin [61]. Regarding ovarian steroidogenesis, some compounds are incriminated in influencing this process. For example,

Table 1
MAIN ENDOCRINE DISRUPTORS AND THEIR INCRIMINATED ACTIONS [1-62]

Endocrine disruptor (ED)	Sources	Possible mechanism	Incriminated actions
Bisphenol A (BPA)	-plastic polycarbonate epoxy resins waterbottles,plastic containers, CDs,electronics,medical equipment (dental fillings), nail polish	interferes with nuclear estrogen receptors in targeted tissues	-decreased serum levels of estradiol -reduced production of eggs -low number of mature oocytes -low number of normal fertilized oocytes -changes in the dopamine and rewardsystem (impulsivity, hyperactivity, increased susceptibility to drug addiction) -occurrence of endometriosis in experimental studies -inhibition of apoptotic activity in the breast and increased breast density -role in the pathogenesis of polycystic ovary syndrome (PCOS)
Phthalates butilbenzilphthalate (BBP) di-n-butilphthalate (DBP) di2etilhexilphthalate (DEHP)	Polyvinyl chloride products, personal care products (cans for food, adhesives, perfumes, eyeshadow), toys (notably DEHP), interior construction, biomedical devices	anti-androgenic action by inhibiting testosterone synthesis in Leydig cells, can disrupt gene expression that regulate cholesterol homeostasis, lipid and insulin signaling=> lowtestosterone	In rats: -cryptorchidism, -small testes, -micropenis, -ductus deferens and epididymis damage, -atrophy of the seminiferous tubules, -reduced anogenital distance occurs -increased incidence of hypospadias In women: premature sexual development
Alkylphenols	surfactants in products such as detergents, disinfectants, cosmetics, spermicides and pesticides	unknown	In rats: -low weight of reproductive organs, -retarded testicular descensus, -reduced spermatogenesis, -Sertoli cell apoptosis, -inhibition of testosterone biosynthesis
Chlorinated hydrocarbons	Insecticides: dichlorodiphenyltrichlorethane (DDT), lindane, chlorobenzilat, metoxychlor	Endocrine effects through estrogen receptor and AhR	-mimic the actions of estrogen Are associated with: - low sperm count, -impaired sexual organs, -changes in testosterone and estrogen, -precocious puberty, -testicular volume reduction, -reduced enzyme activity: 3β hydoxysteroid dehydrogenase(3βHSD) Dioxins: alteration of breast development with increased susceptibility for breast cancer DDT:sexual precocity and reduced fertility in daughters of exposed women

TCDD decreases the expression of the luteinizing hormone (LH) receptor stimulated by follicle-stimulating hormone (FSH). The main metabolite of DDT, p,p'-dichloro-diphenyldichloroethylene (DDE) stimulates vascular endothelial growth factor and insulin growth factor-1 (IGF-1) expression in the luteinizing granulosa in patients with in vitro fertilization suggesting a possible role in steroidogenesis and infertility. Other preliminary studies have shown that BPA decreases proliferation and aromatase expression induced by FSH receptor PPARγ activation and increases IGF-1 and IGF type 1 receptor in granulosa-like tumor cells in humans. These data suggest that ED may have local effects on ovarian function [1].

Others conditions

The ability of synthetic chemicals to alter the reproductive function has been clearly demonstrated by the consequences of diethylstilbestrol (DES) used by pregnant women [51,52]. The daughters of women who were treated with DES had a higher incidence of cervicovaginal cancers, low fertility and an increased rate of ectopic pregnancies, breast cancer and early menopause [47,62].

Further research is needed to improve current knowledge about known ED. The studies should further investigate the possible transgenerational effects and genetic changes caused by ED, and the potential negative effect in patient that require dialysis [63-67].

Scientist research ED implication in a wide range of pahologies such as breast cancer, testicular and prostate cancer, chronic autoimmune thyroiditis or thyroid neoplasm [68], cancer [69-71], obesity, diabetes mellitus, chronic kidney disease, secondary hypertension, arteriopathy, skeletal system pathology and others [72-80].

Conclusions

Additional studies are needed in oder to understand the effects of ED on different organs, especially the reproductive system. The essence is that one in six couples have reproductive problems and the data above illustrates the role of ED in female procreation underlining how sensitive the embryo, fetus, toddler, teenager and adult are to ED. To reduce the risk of ED exposure disorders in the next generation, the society must reduce contamination of air, water and soil, also improve public education regarding ED.

References

1.DIAMANTI-KANDARAKIS, E., BOURGUIGNON, J.P., GIUDICE, L.C., HAUSER, R., PRINS, G.S., SOTO, A.M., ZOELLER, R.T., GORE, A.C., Endocr. Rev., **30**, nr. 4, 2009, p. 293
2.MUNTEANU, C., HOTETEU, M., Balneo-Res. J., **2**, nr. 4, 2011, p. 115
3.MARQUES-PINTO, A., CARVALHO, D., Endocr. Connect., **2**, nr. 3, 2013, p. R15
4.HART, R.J., Physiol. Rev., **96**, nr. 3, 2016, p. 873
5.POIANA, C., NEAMÞU, M.C., AVRAMESCU, E.T., CARSOTE, M., TRIFÃNESCU, R., TERZEA, D., NEAMTU, O.M., FERECHIDE, D., DÃNCIULESCU MIULESCU, R., Rom. J. Morphol. Embryol., **54**, nr. 3 Suppl, 2013, p. 717
6.CAPATINA, C., GHINEA, A., DUMITRASCU, A., POIANA, C., Int. J. Diabetes Dev. Ctries., **36**, nr. 4, 2016, p. 393
7.PAUN, D.L., POIANA, C., PETRIS, R., RADIAN, S., MIULESCU, R.D., CONSTANTINESCU, G., ORBAN, C., Chirurgia (Bucur)., **108**, nr. 6, 2013, p. 900
8.BENNETAU-PELISSERO, C., Curr. Opin. Clin. Nutr. Metab. Care., **19**, nr. 6, 2016, p. 477
9.NENDZA, M., WENZEL, A., MULLER, M., LEWIN, G., SIMETSKA, N., STOCK, F., ARNING, J., Environ. Sci. Eur., **28**, nr. 1, 2016, p. 26
10.SIKKA, S.C., WANG, R., Asian J. Androl., **10**, nr. 1, 2008, p. 134
11.GHEORGHISAN-GALATEANU, A., TERZEA, D.C., CARSOTE, M., POIANA, C., J. Ovarian Res., **6**, nr. 1, 2013, p. 28
12.REITER, E., GERSTER, P., JUNGBAUER, A., Gynecol. Endocrinol., **27**, nr. 12, 2011, p. 1037
13.BOIVIN, J., BUNTING, L., COLLINS, J.A., NYGREN, K.G., Hum. Reprod., **22**, nr. 6, 2007, p. 1506
14.PLACER, J., SALVADOR, C., PLANAS, J., TRILLA, E., LORENTE, D., CELMA, A., LOPEZ, M.A., MOROTE, J., J. Endourol., **29**, nr. 3, 2015, p. 332
15.BRETVELD, R., BROUWERS, M., EBISCH, I., ROELEVELD, N., Scand. J. Work Environ. Health., **33**, nr. 1, 2007, p. 13
16.YOUNGLAI, E.V., FOSTER, W.G., HUGHES, E.G., TRIM, K., JARRELL, J.F., Arch. Environ. Contam. Toxicol., **43**, nr. 1, 2002, p. 121
17.VANDENBERG, L.N., COLBORN, T., HAYES, T.B., HEINDEL, J.J., JACOBS, D.R. JR., LEE, D.H., SHIODA, T., SOTO, A.M., VOM SAAL, F.S., WELSHONS, W.V., ZOELLER, R.T., MYERS, J.P. Endocr. Rev., **33**, nr. 3, 2012, p. 378
18.CHECHERITA, I.A., SMARANDACHE, D., DAVID, C., CIOCALTEU, A., ION, D.A., LASCĂR, I., Rom. J. Morphol. Embryol., **53**, nr. 1, 2012, p. 7
19.DAVID, C., PERIDE, I., NICULAE, A., CONSTANTIN, A.M., CHECHERITA, I.A., B.M.C. Nephrol., **17**, nr. 1, 2016, p. 131
20.ROSSELLI, M., DUBEY, R.K., IMTHURN, B., MACAS, E., KELLER, P.J., Human Reproduction, **10**, nr. 7, 1995, p. 1786
21.VOM SAAL, F.S., AKINGBEMI, B.T., BELCHER, S.M., BIRNBAUM, L.S., CRAIN, D.A., ERIKSEN, M., FARABOLLINI, F., GUILLETTE, L.J. JR., 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., MCLACHLAN, J.A., MYERS, J.P., NADAL, A., NEWBOLD, R.R., OLEA, N., PRINS, G.S., RICHTER, C.A., RUBIN, B.S., SONNENSCHEIN, C., SOTO, A.M., TALSNESS, C.E., VANDENBERGH, J.G., VANDENBERG, L.N., WALSER-KUNTZ, D.R., WATSON, C.S., WELSHONS, W.V., WETHERILL, Y., ZOELLER, R.T., Reprod. Toxicol., **24**, nr. 2, 2007, p. 131
22.EUSTACHE, F., MONDON, F., CANIVENC-LAVIER, M.C., LESAFFRE, C., FULLA, Y., BERGES, R., CRAVEDI, J.P., VAIMAN, D., AUGER, J., Environ. Health Perspect., **117**, nr. 8, 2009, p. 1272
23.ISVORANU, I., PERIDE, I., RADULESCU, D., NICULAE, A., SINESCU, R.D., CHECHERITA, I.A., Rev. Chim.(Bucharest), **66**, no. 9, 2015, p. 1316
24.CHECHERITĂ, I.A., DAVID, C., STOICA, L., POPESCU, P., CIOCALTEU, A., LASCĂR, I., Rom. J. Morphol. Embryol., **52**, nr. 2, 2011, p. 533
25.PRICOP, C., BRANISTEANU, D.D., ORSOLYA, M., PUIA, D., MATEI, A., CHECHERITA, I.A., Int. Urol. Nephrol., **48**, nr. 2, 2016, p. 183
26.ANWAY, M.D., CUPP, A.S., UZUMCU, M., SKINNER, M.K., Science, **308**, nr. 5727, 2005, p. 1466
27.SETCHELL, K.D., ZIMMER-NECHEMIAS, L., CAI, J., HEUBI, J.E., Am. J. Clin. Nutr., **68**, nr. 6 Suppl, 1998, p. 1453S
28.DINSDALE, E.C., WARD, W.E., Nutrients, **2**, nr. 11, 2010, p. 1156
29.SANTANGELI, S., MARADONNA, F., OLIVOTTO, I., PICCINETTI, C.C., GIOACCHINI, G., CARNEVALI, O., Gen. Comp. Endocrinol., **245**, 2017, p. 122
30.KNEZ, J., Reprod. Biomed. Online, **26**, nr. 5, 2013, p. 440
31.CALAFAT, A.M., YE, X., WONG, L.Y., REIDY, J.A., NEEDHAM, L.L., Environ. Health Perspect, **116**, nr. 1, 2008, p. 39
32.JEDDI, M.Z., RASTKARI, N., AHMADKHANIHA, R., YUNESIAN, M., Environ. Monit. Assess, **188**, nr. 9, 2016, p. 534
33.AL-SALEH, I., SHINWARI, N., ALSABBAHEEN, A., J. Toxicol. Sci., **36**, nr. 4, 2011, p. 469
34.EHRLICH, S., WILLIAMS, P.L., MISSMER, S.A., FLAWS, J.A., YE, X., CALAFAT, A.M., PETROZZA, J.C., WRIGHT, D., HAUSER, R., Hum. Reprod., **27**, nr. 12, 2012, p. 3583
35.VANDENBERG, L.N., CHAHOUD, I., HEINDEL, J.J., PADMANABHAN, V., PAUMGARTTEN, F.J., SCHOENFELDER, G., Environ. Health Perspect., **118**, nr. 8, 2010, p. 1055
36.SUZUKI, T., MIZUO, K., NAKAZAWA, H., FUNAE, Y., FUSHIKI, S., FUKUSHIMA, S., SHIRAI, T., NARITA, M., Neuroscience, **117**, nr. 3, 2003, p. 639
37.GRADY, R., SATHYANARAYANA, S., Curr. Urol. Rep., **13**, nr. 4, 2012, p. 307
38.FOSTER, P.M., Crit. Rev. Toxicol., **35**, nr. 8-9, 2005, p. 713
39.BARLOW, N.J., PHILLIPS, S.L., WALLACE, D.G., SAR, M., GAIDO, K.W., FOSTER, P.M., Toxicol. Sci., **73**, nr. 2, 2003, p. 431
40.FOSTER, P.M., Int. J. Androl., **29**, nr. 1, 2006, p. 140
41.CRINNION, W.J., Altern. Med. Rev., **15**, nr. 3, 2010, p. 190
42.CHOU, Y.Y., HUANG, P.C., LEE, C.C., WU, M.H., LIN, S.J., J. Pediatr. Endocrinol. Metab., **22**, nr. 1, 2009, p. 69
43.DUMITRESCU,G.,PET,I., DRONCA,D.,AHMADI,M., PETRESCU CIOCHINA,L.,FILIMON,N.M.,POPESCU,R.,Rev.Chim.(Bucharest),**69,**no.1,2018,p.140
44.WHITE, R., JOBLING, S., HOARE, S.A., SUMPTER, J.P., PARKER, M.G., Endocrinology, **135**, nr. 1, 1994, p. 175
45.SCHLEBUSCH, H., WAGNER, U., VAN DER VEN, H., AL-HASANI, S., DIEDRICH, K., KREBS, D., J. Clin. Chem. Clin. Biochem., **27**, nr. 9, 1989, p. 663
46.*** W.H.O., 2016, http://www.who.int/mediacentre/factsheets/fs225/en/, accesed December 2016
47.MANDRIOLI, D., BELPOGGI, F., SILBERGELD, E.K., PERRY, M.J., Environ. Health, **15**, nr. 1, 2016, p. 97
48.FAROON, O.M., KEITH, S., SMITH-SIMON, C., DE ROSA, C.T., 2003, http://www.inchem.org/documents/cicads/cicads/cicad55.htm, accessed December 2016
49.SMARR, M.M., KANNAN, K., BUCK LOUIS, G.M., Fertil. Steril., **106**, nr. 4, 2016, p. 959
50.CRAIN, D.A., JANSSEN, S.J., EDWARDS, T.M., HEINDEL, J., HO, S.M., HUNT, P., IGUCHI, T., JUUL, A., MCLACHLAN, J.A., SCHWARTZ, J., SKAKKEBAEK, N., SOTO, A.M., SWAN, S., WALKER, C., WOODRUFF, T.K., WOODRUFF, T.J., GIUDICE, L.C., GUILLETTE, L.J. JR., Fertil. Steril., **90**, nr. 4, 2008, p. 911
51.REDDY, B.S., ROZATI, R., REDDY, B.V., RAMAN, N.V., B.J.O.G., **113**, nr. 5, 2006, p. 515

52.MISSMER, S.A., HANKINSON, S.E., SPIEGELMAN, D., BARBIERI, R.L., MICHELS, K.B., HUNTER, D.J., Fertil. Steril., **82**, nr. 6, 2004, p. 1501
53.SIGNORILE, P.G., SPUGNINI, E.P., MITA, L., MELLONE, P., D'AVINO, A., BIANCO, M., DIANO, N., CAPUTO, L., REA, F., VICECONTE, R., PORTACCIO, M., VIGGIANO. E., CITRO, G., PIERANTONI, R., SICA, V., VINCENZI, B., MITA, D.G., BALDI, F., BALDI, A., Gen. Comp. Endocrinol., **168**, nr. 3, 2010, p. 318
54.CASERTA, D., BORDI, G., CIARDO, F., MARCI, R., LA ROCCA, C., TAIT, S., BERGAMASCO, B., STECCA, L., MANTOVANI, A., GUERRANTI, C., FANELLO, E.L., PERRA, G., BORGHINI, F., FOCARDI, S.E., MOSCARINI, M., Gynecol. Endocrinol., **29**, nr. 5, 2013, p. 444
55.AZZIZ, R., WOODS, K.S., REYNA, R., KEY, T.J., KNOCHENHAUER, E.S., YILDIZ, B.O., J. Clin. Endocrinol. Metab., **89**, nr. 6, 2004, p. 2745
56.FRANKS, S., MASON, H., WILLIS, D., Mol. Cell. Endocrinol., **163**, nr. 1-2, 2000, p. 49
57.RUTKOWSKA, A., RACHON, D., Gynecol. Endocrinol., **30**, nr. 4, 2014, p. 260
58.FRANKS, S., MCCARTHY, M.I., HARDY, K., Int. J. Androl., **29**, nr. 1, 2006, p. 278
59.TAKEUCHI, T., TSUTSUMI, O., IKEZUKI, Y., TAKAI, Y., TAKETANI, Y., Endocr. J., **51**, nr. 2, 2004, p. 165
60.WANG, Y., ZHU, Q., DANG, X., HE, Y., LI, X., SUN, Y., Gynecol. Endocrinol., **33**, nr. 1, 2017, p. 21
61.HUNT, P.A., KOEHLER, K.E., SUSIARJO, M., HODGES, C.A., ILAGAN, A., VOIGT, R.C., THOMAS, S., THOMAS, B.F., HASSOLD, T.J., Curr. Biol., **13**, nr. 7, 2003, p. 546
62.BALINT,O., SECOSAN,C., GRIGORAS,D., ILINA,R.,LEVAI,C.M.,SAS,I., Mat.Plast, **54**, no.3, 2017, p.549
63.SUSIARJO, M., HASSOLD, T.J., FREEMAN, E., HUNT, P.A., PLoS Genet., **3**, nr. 1, 2007, p. e5
64.HATCH, E.E., TROISI, R., WISE, L.A., HYER, M., PALMER, J.R., TITUS-ERNSTOFF, L., STROHSNITTER, W., KAUFMAN, R., ADAM, E., NOLLER, K.L., HERBST, A.L., ROBBOY, S., HARTGE, P., HOOVER, R.N., Am. J. Epidemiol., **164**, nr. 7, 2006, p. 682
65.CHANDRA, A., MARTINEZ, G.M., MOSHER, W.D., ABMA, J.C., JONES, J., Vital Health Stat. 23, nr. 25, 2005, p. 1
66.YOUNGLAI, E.V., FOSTER, W.G., HUGHES, E.G., TRIM, K., JARRELL, J.F., Arch. Environ. Contam. Toxicol., **43**, nr. 1, 2002, p. 121
67.ANWAY, M.D., SKINNER, M.K., Endocrinology, **147**, nr. 6 Suppl, 2006, p. S43
68.BARBU, C.G., FLORIN, A., NEAMŢU, M.C., AVRAMESCU, E.T., TERZEA, D., MIRON, A., DANCIULESCU MIULESCU, R., POIANA, C., FICA, S., Rom. J. Morphol. Embryol., **56**, nr. 2, 2015, p. 527
69.POIANA, C., NEAMTU, M.C., AVRAMESCU, E.T., CARSOTE, M., TRIFĂNESCU, R., TERZEA, D., NEAMŢU, O.M., DANCIULESCU MIULESCU, R., Ro. J. Morphol. Embryol., **54**, nr. 1, 2013, p. 201
70.NITIPIR, C., BARBU, M.A., POPA, L.G., MIHAI, M.M., RADU, I., MIREA, D., GIURCANEANU, C., SCAUNASU, R.V., Farmacia, **63**, nr. 6, 2015, p. 805
71.CARSOTE, M., PAUN, S., NEAMTU, M.C., AVRAMESCU, E.T., IOSIF, C., TERZEA, D., CONSTANTINOIU, S., DANCIULESCU MIULESCU, R., NEAMTU, O.M., POIANA, C., Rom. J. Morphol. Embryol., **53**, nr. 2, 2012, p. 401
72.NEAGU, T.P., TIGLIS, M., BOTEZATU, D., ENACHE, V., COBILINSCHI, C.O., VÂLCEA-PRECUP, M.S., GRINESCU, I.M., Rom. J. Morphol. Embryol., **58**, nr. 1, 2017, p. 33
73.PERIDE, I., CHECHERITA, I.A., SMARANDACHE, D.R., RADULESCU, D., SINESCU, R.D., NICULAE, A., PRICOP, C., Rom. J. Morphol. Embryol., **56**, nr. 2 Suppl, 2015, p. 777
74.NEAGU, T.P., ŢIGLIS, M., COCOLOS, I., JECAN, C.R., Rom. J. Morphol. Embryol., **57**, nr. 4, 2016, p. 1215
75.NEAGU, T.P., ENACHE, V., COCOLOS, I., TIGLIS, M., COBILINSCHI, C., TINCU, R., Rom. J. Morphol. Embryol., **57**, nr. 2, 2016, p. 437
76.CHECHERITA, I.A., SMARANDACHE, D., RĂDULESCU, D., PERIDE, I., BRATU, O., CIOCÂLTEU, A., SEBE, I., LASCĂR, I., Chirurgia (Bucur.), **108**, nr. 5, 2013, p. 736
77.NEAGU, T.P., SINESCU, R.D., ENACHE, V., ACHIM, S.C., TIGLIS, M., MIREA, L.E., Rom. J. Morphol. Embryol., **58**, nr. 2, 2017, p. 603
78.PALMER, J.R., WISE, L.A., HATCH, E.E., TROISI, R., TITUS-ERNSTOFF, L., STROHSNITTER, W., KAUFMAN, R., HERBST, A.L., NOLLER, K.L., HYER, M., HOOVER, R.N., Cancer Epidemiol. Biomarkers Prev., **15**, nr. 8, 2006, p. 1509
79.NEAGU, T.P., TIGLIS, M., POPP, C.G., JECAN, C.R., Rom. J. Morphol. Embryol., **57**, nr. 3, 2016, p. 1051
80.BURUIANA, A., DUMITRU, N., GHEMIGIAN, A., PETROVA, E.N., Rev. Chim.(Bucharest), **68**, no. 7, 2017, p. 1557

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