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Bisphenol Exposure and its Impact on Multi-System Pathological Changes in Laboratory Animal Models - A Review

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Abstract

Bisphenol A (BPA), a synthetic compound found in polycarbonate plastic widely used in food and drink packaging, medical devices, thermal paper, and dental materials, presents health risks due to contamination in various sources. Due to its mass productions and widespread applications, the presence of BPA is ubiquitous in the environment. It accumulates in human tissues, exhibiting hormone-like properties by binding to estrogen receptors, affecting body weight, and influencing carcinogenesis. BPA interacts with GPR30, impacting metabolism and cancer progression, and may disrupt male reproductive function through androgen receptor binding. Key transcription factors, including PPARγ, C/EBP, Nrf2, HOX, and HAND2, contribute to BPA's effects on fat, liver, cardiovascular health, and cancer. Additionally, epigenetic changes such as DNA methylation, histone modification, and altered microRNA expression contribute to the pathological effects of BPA. Research studies using *in vitro* cell lines, rodent models, and epidemiological analysis have convincingly shown the increasing susceptibility to cancer at doses below the oral reference dose set by the Environmental Protection Agency for BPA. The purpose of this review is to analyze the mechanism of action of bisphenol A, with a special focus on multi systemic pathological changes in animal models.

Key words: Bisphenol A, receptors, epigenetics, metabolism, pathology.

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The World Health Organization's International Program on Chemical Safety defines endocrine-disrupting chemicals (EDCs) as exogenously derived substances capable of adversely affecting the endocrine system, thereby posing risks to life forms and populations This diverse class, comprising synthetic or naturally occurring compounds (Caserta *et al*., 2021), due to their small size and lipophilic nature, readily traverse cell membranes and accumulate in adipose tissue, with humans, positioned higher in the food chain, facing significant exposure (Mukhopadhyay *et al*., 2022).

EDCs interfere with hormone synthesis, elimination, transportation, metabolism, and action, acting through various receptors and pathways associated with reproductive and endocrine systems, potentially impacting growth, development, the immune system, neurological functions, and reproductive health. These substances can replace natural hormones, and synthetic chemicals not only disrupt hormone production but also affect circulation and peripheral effects. Clinical challenges, including infertility, endometriosis, diabetes, and various cancers, are linked to EDC exposure (Fenichel *et al*., 2013). While EDCs may target any endocrine gland, many interfere with the hypothalamic-pituitary-gonadal axis, and diverse chemicals affecting adipose tissue regulation may contribute to obesity, metabolic syndrome, and diabetes (Ghosh *et al*., 2022).

Bisphenol A

Bisphenol A(BPA) is an established EDC because of its potential as an estrogen mimic

and was first established as ED in the 1930s. In endocrine receptor dependent pathway, even the low concentrations of BPA had potency to act similar to that of estradiol because of the presence of structural phenol.

BPA is the most widely available of the myriad forms of EDC, which appear white, crystalline synthetic solid substance that has been used to synthesize polysulfones, polycarbonates, polyether ketones, polyesters, and a key family of epoxy resins. It is classed as a plasticizer because of its characteristics (Mukhopadhyay *et al*., 2022). As a result, it is regularly used in infant bottles, metal-based food cans, beverage containers, ophthalmic lenses, health and dental materials, electronics and electric utilities, water pipelines, and thermal receipts, flame retardants (Yilmaz *et al*., 2020).

Global Production

The global bisphenol A market is projected to reach approximately 7,348K tons by the end of 2023, increasing at a CAGR of around 3% per year in the period 2017-2023. Regionally, the largest global bisphenol A market was North East Asia, which accounted for about 50% of the total in volume terms. India Bisphenol demand stood at 2.08 Thousand Tonnes in FY2021 and is forecast to reach 16.73 Thousand Tonnes by FY2030, growing at a healthy CAGR of 26.09% until FY2030.Based on studies, the United States Environmental Protection Agency (EPA) set a reference dose of 50 µg/kg bw/day for BPA.

To establish an acceptable level of human exposure to BPA, the National Toxicology Program (NTP) of the United States Environmental Protection Agency (EPA) convened a committee to investigate the broad range of biological effects associated with chronic exposure. Based on numerous experimental studies, the no-observed-adverse-effects level (NOAEL) for BPA was determined to be 5 mg/kg b. wt/day, while the lowest-observed-adverse-effects level (LOAEL) was set at 50 mg/kg b. wt/day. As a respond to a refined risk assessment of BPA and its unwanted health effects, the European Food Safety Authority (EFSA) decreased the tolerable daily intake (TDI) to 4 µg/kg bw/day from the 50 µg/kg bw/day in 2015 (EFSA Panel on Food Contact Materials & Aids, 2015).

Metabolism of BPA / Toxicokinetics

BPA is absorbed through oral exposure and metabolized in the liver predominantly via glucuronidation (90%) and sulfation (10%), rendering the conjugated forms inactive (Almeida *et al*., 2018). Unconjugated, bioactive BPA is found in human body fluids and tissues at trace levels. After excretion into bile, unconjugated BPA and glucuronic acid separate, with unconjugated BPA undergoing re-absorption into the bloodstream, leading to enterohepatic circulation and extended elimination. BPA's half-life depends on glucuronidase presence in various organs. This lipophilic compound has an affinity for adipose tissue and gradual release into other tissues. Liver enzymes, including uridine diphosphate glucuronosyl transferase and phenol sulfotransferase, perform phase II conjugation, forming BPA glucuronide (BPAG) and trace BPA-sulfate conjugates. Glucuronidas-

Physicochemical and acute toxicological Properties of BPA

Property	Value
Molecular weight	228.18 g/mol
Boiling point	220°C at 4 mm Hg; 398°C at 760 mm Hg
Melting point	150-157°C
Specific gravity	1.060- 1.195 g/ml at 20-25°C
Solubility in water	120-300 mg/Lat 20-25°C
Vapor pressure	8.7 X 10-^{10} -3.96 X I*0-^{1}*mm Hg at 20-25°C
LD_{50} rat, oral	3,300-4,240 mg/kg
LD_{50} mouse, oral	2,500-5,200 mg/kg

es UGT2B15 and UGT1A9 play significant roles in this conversion. BPA is primarily cleared as a glucuronide in urine (94.6% elimination rate) (Provencher *et al*., 2014). Altered UGT activity results in elevated unconjugated BPA levels and its estrogenic mimicry, contributing to various adverse effects, including hyperinsulinemia, obesity, cardiovascular issues, thyroid disorders, hypertension, polycystic ovary syndrome (PCOS), reproductive abnormalities, and cancer in both animal and human studies.

BPA metabolism exhibits gender-based differences, leading to variations in blood BPA concentrations. Men tend to have higher blood BPA levels, possibly due to androgens' ability to inhibit glucuronidase, leading to faster overall metabolism (Gerona *et al*., 2013). Factors like renal clearance and organ accumulation also influence total BPA levels. Women, with greater fat stores, tend to accumulate BPA in their tissues, while men often exhibit higher urinary clearance rates (Caporossi and Papaleo, 2015). Men typically have more glucuronoconjugate intermediates in their urine, while women show higher levels of sulfoconjugate intermediates, suggesting a lower clearance rate in females (Kim *et al*., 2003). Research has indicated widespread BPA presence in human tissues, with the highest levels found in the brain (up to 2.36 ng/g), adipose tissue (1.12–12.28 ng/g), and hepatic tissue (0.77–3.35 ng/g). Additionally, elevated levels of total BPA (1.1 ng/mL) and unconjugated BPA (0.4 ng/mL) have been reported in breast milk (Wang *et al*., 2017).

Interaction of BPA with Humans

Human exposure to BPA can be categorized into two main pathways: environmental exposure and food-related exposure. BPA exposure can occur through ingestion, inhalation, and skin absorption. Environmental exposure can result from BPA leaching into the environment due to various factors, including industrial processes and waste disposal practices. On the other hand, food-related exposure is linked to the interaction between BPA and the food chain, including bioaccumulation and the use of BPA in food packaging materials. In contrast to some other EDC, BPA has a relatively short half-life, approximately 5.3 hours (Basak *et al*., 2021).

Under normal physiological conditions, BPA glucuronide, a conjugated form of BPA, can be efficiently cleared from the body due to specific enzymatic processes (Encarnação *et al*., 2019). This conjugation process renders BPA hydrophilic and biologically inactive, facilitating its excretion through urine. However, this initial metabolic process triggers BPA activation in organs such as the placenta, lungs, liver, and kidneys through the enzyme beta-glucuronidase. The accumulation of BPA, particularly in fat-rich organs like the ovaries, could account for the heightened adverse effects of BPA on female reproductive health as compared to male reproductive health.

Molecular Pathogenesis

The molecular pathogenesis of bisphenol A (BPA) involves intricate mechanisms that disrupt normal cellular functions and homeostasis.

Receptor Interactions: BPA interacts with several hormone receptors, including estrogen receptors (ERα and ERβ), androgen receptors (AR), thyroid hormone receptors (TRα and TRβ), glucocorticoid receptors (GR), estrogen-related receptor gamma (ERRγ), and G-protein-coupled receptor 30 (GPR30). These interactions can lead to agonistic or antagonistic effects on receptor activation, disrupting the normal hormonal signaling pathways.It has been demonstrated that bisphenols inhibit follicle development, prompt hypercholesterolemia, and potentially trigger pre-adipocyte proliferation. These effects can be attributed to the activation of the peroxisome proliferator-activated receptor gamma (PPARγ), aryl hydrocarbon receptor (AHR), and the pregnane X receptor (Basak *et al*., 2021).

Gene Expression Regulation: Upon binding to hormone receptors, BPA can modulate gene expression patterns within cells. It acts as a transcriptional regulator, influencing the expression of genes involved in various physiological processes such as cell proliferation, differentiation, and metabolism. Dysregulation of gene expression by BPA can lead to aberrant cellular responses and functional alterations. Prenataly exposure to small doses of BPA results in suppression of the Anti-mullerian Hormone gene, which is considered to be due to

BPA's influence on the decreased granulosa cell function and consequent acceleration of apoptotic activity. Another probable cause may be the reduced expression of IGF1R, IGF1, and IGF2 genes following BPA exposure, that appears to inhibit ovarian cell formation and proliferation. These may lead to abnormal hormone level and ovarian architexture by dysfunctional HPO-axis (Dong *et al*., 2011).

Intracellular Signaling Pathways: BPA activates intracellular signaling cascades through receptor-mediated mechanisms. Activation of these signaling pathways can regulate cell proliferation, survival, and differentiation. Dysregulation of these pathways by BPA can contribute to cellular dysfunction and pathological conditions. During its non-genomic actions, BPA is known to activate the extracellular governed kinase/mitogen-activated protein kinase (ERK/MAPK), phosphatidylinositol 3-kinasesserine/threonine protein kinase (PI3K-AKT), and cytoplasmic Ca2+-dependent signalling processes (Marino *et al*., 2012). Following the activation of intracellular Ca2+ signaling, the agonistic action of BPA is mediated through a non-classical estrogen receptor called GPCR30 (Alonso-Magdalena *et al*., 2012).

Oxidative Stress: BPA exposure is associated with increased production of reactive oxygen species (ROS) and oxidative stress within cells. ROS accumulation can damage cellular components such as proteins, lipids, and DNA, leading to cellular dysfunction and tissue damage. Moreover, oxidative stress induced by BPA can further exacerbate its toxic effects and contribute to the development of various diseases.

Mitochondrial Dysfunction: BPA has been shown to impair mitochondrial function, including mitochondrial membrane potential, ATP production, and respiratory chain activity. Mitochondrial dysfunction can disrupt cellular energy metabolism, increase oxidative stress, and induce apoptosis. Dysfunction in mitochondrial activity by BPA can contribute to tissue damage and the development of metabolic disorders.

Epigenetic Modifications: BPA exposure has been linked to alterations in epigenetic mechanisms, including DNA methylation, histone modifications, and microRNA expression. These epigenetic changes can persistently modulate gene expression patterns, leading to long-term alterations in cellular function and phenotype. Dysregulation of epigenetic processes by BPA may contribute to the development of diseases such as cancer, metabolic disorders, and neurodevelopmental abnormalities. Sabry *et al.* (2022) BPA can induce apoptosis through a pathway that does not involve miR-21 signaling, but under specific conditions, it may act through miR-21 and programmed cell death 4 (PDCD4) to induce antiapoptotic and pro-oncogenic effects in cells.

BPA and Cancer

Exposure to BPA has been linked to an increased cancer risk. BPA can stimulate the growth of hormone-dependent tumors by binding to hormone receptors and directly affecting the activity of oncogenes and tumor-suppressor genes. In breast cancer cells, BPA exposure has been shown to downregulate the p53 gene, along with its downstream proapoptotic regulator Bcl2-associated X protein (BAX). Additionally, BPA activates pro-survival signaling pathways such as the Phosphoinositide 3-kinase/Protein kinase B/mechanistic target of the rapamycin (PI3K/Akt/mTOR) pathway, as well as factors downstream of mTOR like Eukaryotic translation initiation factor (e1F4B, e1F4E) leading to resistance against anti-cancer drugs. Conversely, BPA decreases the expression levels of suppressor genes like Phosphatase and tensin homolog (PTEN), Tuberous sclerosis 1 (TSC1), and Tuberous Sclerosis Complex 2 (TSC2). BPA exposure disrupts the cell cycle and causes DNA damage by activating CTNNB1, initiating the Catenin Beta 1 (CTNNB1), which is the initiator of the aberrant constructed CTNNB1-nuclear factor kappa-B1 (NFKB1)-AR-insulin-like growth factor-1 (IGF1)-Twist-related protein 1 (TWIST1) pathway, potentially leading to lymphomagenesis. In oncological contexts, BPA interferes with mammary gland morphogenesis, contributing to breast cancer development. Moreover, it's associated with oral cancer, possibly due to its direct activation of estrogen receptors in oral mucosa and salivary glands. This activation triggers cell proliferation, invasion, and migration via G-protein-related signaling. Prolonged BPA exposure induces preneoplastic changes in murine oral mucosa. In vitro studies

demonstrate BPA-induced expression of metalloproteinases and growth factors, promoting cell proliferation and angiogenesis while reducing apoptosis (Della Rocca *et al.,* 2023).

Adverse Health Impact of BPA in Human beings

The population studies report the association of high levels of free BPA in body fluids with various disorders. Male reproductive system - BPA was shown to deteriorate sexual function as sexual desire, erectile ability and ejaculation intensity and caused premature ejaculation, infertility and poor sperm quality (Tian *et al*., 2018).

Studies that proved estrogenic characteristics of BPA were reported as early as 1936 (Ullah *et al.,* 2018). With the increase in research studies, the link between BPA and the production of oxidative stress became clearer. Oxidative stress can be induced by disruption of prooxidants/antioxidants balance of cells because of many environmental contaminants including BPA (Pirozzi *et al*., 2020). Male infertility is associated with oxidative stress.

Female reproductive system –BPA may play a role in pathogenesis of ovarian insufficiency, polycystic ovary syndrome, decreased fertility (Ozel *et al*., 2019) and increased risk of adverse pregnancy outcomes like recurrent miscarriage, spontaneous preterm birth and preterm premature rupture of membranes.

BPA introduction led to increment in post-implantation loss, fatal mutations, damage to DNA and apoptosis, DNA mutation, error in DNA replication and genomic instability can happen if there is no repairing of oxidative DNA prior to DNA replication. Development disorders- Prenatal exposure to BPA increased the risk of low birth weight, rapid infant weight gain, childhood obesity and high child's BMI (Yang *et al*., 2022). Decreased growth of height in boys and shortened anogenital distance (AGD) (Mammadov *et al*., 2018) has also been linked to prenatal BPA exposure. Maternal exposure to BPA has been linked to neurodevelopmental and behavioral disorders in offspring and also associated with abnormalities of onset of puberty (Jensen *et al*., 2019).

Metabolic disorder -BPA exposure is linked to increased risk of obesity, Type-2 diabetes, cardiovascular diseases, insulin resistance and abnormalities of bone metabolism (Amin *et al*., 2019). Current epidemiological studies have revealed that higher urinary BPA concentration in humans is associated with various types of cardiovascular diseases, including angina, hypertension, heart attack, coronary and peripheral arterial diseases (LaKind *et al*., 2014).

Respiratory system- BPA exposure is positively associated with lung function damage and the odds of persistent wheeze in children (Spanier *et al*., 2014). A significantly positive relationship between urinary BPA levels and asthma in children has been reported (Youssef *et al*., 2018). BPA exposure has been linked to the development of obstructive sleep apnea syndrome (OSAS).

Liver- Urinary bisphenol A concentrations are associated with abnormal liver function in the elderly. BPA exposure increases the risk of suspected nonalcoholic fatty liver disease (NAFLD) (Zhou *et al*., 2023).

Immune function- BPA exposure has been found to be positively associated with the increased risk of aeroallergies and the occurrence of autoimmune thyroid disease (Chailurkit *et al*., 2016). Mental diseases –A positive correlation between BPA and autism spectrum disorder severity has been reported (Minatoya *et al*., 2018).Cancer –BPA exposure may be related to development of prostate cancer, lung cancer and breast cancer (Tse *et al*., 2017).

Effect of BPA in animal studies

Body and Organ weight

Karnam *et al*. (2015) noted that dose-dependent reductions in feed consumption and body weight in male Wistar rats exposed to BPAfor 60 days underscoring its negative influence on growth. In studies focusing on the male reproductive system, Grami *et al*. (2020) found a decreased testicular weight in Wistar rats. Prenatal BPA exposure at doses of 0.250 mg/kg observed by Christiansen *et al*. (2014) resulted in a significant reduction in male anogenital distance, endocrine signalling disruptions in prenatal sexual development. In rats, liver weight declined in studies conducted by Kourouma *et al*. (2014) at 25mg/kg

b.wt. for 30 days.

Gurmeet *et al*. (2014) reported an increase in lung and liver weight with BPA exposure. In male rats, Kaur *et al*. (2018) found significant decreases in testis and epididymis weight, accessory sex organs, and body weight due to BPA exposure @ 20mg kg^{-1} for 45 days. Nah *et al*. (2011) noted a decline in ovarian weight in female rats treated with various BPA doses, and Markey *et al*. (2005) observed estrogenic responses, such as increased uterine wet weight, luminal epithelial height in CD-1 mice.

Clinical Signs

Multiple studies have revealed a spectrum of behavioral and physiological changes in animals exposed to BPA. Yamasaki *et al*. (2002) observed soft stools, decreased mobility, reduced respiration, and lower body temperature in rats given high oral doses of BPA. Samova *et al*. (2018) noticed reduced feed intake, decreased responsiveness, hair thinning, and other behavioral changes in mice exposed to varying BPA doses. Kaur *et al*. (2021) found various behavioral alterations, including weakness, hyperactivity, irritability, and increased mortality in BPA-exposed rats. Amaravathi *et al*. (2017) noted reduced feed intake, anxiety, and hair discoloration in female rats in a dose-dependent manner.

Haematology

Yamasaki *et al*. (2002) observed decreased hemoglobin and hematocrit values in female rats treated with BPA (600- 1000mg/kg b.wt. /day in olive oil orally for 28days). Sujan *et al*. (2020) found that BPA at doses of 50, 200, and 600 mg/kg body weight for 90 days led to significant decreases in Hb, PCV, TEC, MCV, MCH, and MCHC levels, along with a significant increase in TLC values. Similarly, Karnam *et al*. (2015) observed a significant reduction in Hb, PCT, and TLC levels, accompanied by an increase in the percentage of neutrophils and a decrease in the percentage of lymphocytes. Pal *et al*. (2017) found a substantial reduction in the total white blood cell (WBC) count in rats exposed to higher doses of BPA(120mg/kg) for 4 weeks, attributed to BPA-induced oxidative stress affecting leucopoiesis.

Serum Biochemistry

Studies investigated the effects of BPA on various biochemical parameters in experimental animals. Higashihara *et al*. (2007) observed decreased serum total cholesterol, glucose, and albumin in female rats given BPA at doses of 20 mg/kg per day. Amaravathi *et al*. (2017) reported significant decreases in total protein and glucose in male and female rats exposed to high BPA doses (500 mg/kg b.wt). Yamasaki *et al*. (2002) noted increased levels of aspartate transaminase (AST), alkaline phosphatise (ALP) in male rats, along with decreased albumin and A: G ratio in female rats exposed to BPA. Karnam *et al*. (2015) reported significant increases in AST, ALT, ALP, creatinine, uric acid, and decreased levels of glucose, total protein, and globulin due to BPA @ 200,600 mg/kg b. wt. and decreased GT was observed after 30 days exposure to BPA at doses of 2, 10, 50 mg/ kg B.Wt (Kourouma *et al.,* 2014).

Antioxidant Parameters

Kabuto *et al*. (2004) noted increased catalase and GPx activity in the liver and kidney, as well as elevated TBARS in the brain, kidney, and testes of mice exposed to BPA during embryonic/fetal life at 50 mg/kg/day. Kourouma *et al*. (2014) reported decreased catalase (CAT), SOD, and GSH-PX levels in liver. Kamel *et al*. (2018) noted elevated MDA levels in the liver and testes, accompanied by reduced SOD and GSH levels. In another study by Panpatil *et al*. (2020), BPA exposure led to reduced SOD, GPx, and CAT activity in the liver and kidney, along with increased MDA and nitric oxide (NO) levels. Similar reductions in antioxidant enzyme levels in the liver and kidney of rats were reported by Eweda *et al*. (2020).

Li *et al*. (2018) found that BPA increased testicular MDA levels and decreased GSH-Px and SOD activities, which could be alleviated by Vitamin C. Kaur *et al*. (2021), using a 20 mg/kg body weight BPA dose for 45 days, noted significant decrease in SOD, CAT, GR, GPx, and GSH in testis and epididymis tissue homogenate due to BPA exposure.Top of Form

Hormonal Assay

Hormones like luteinizing hormone (LH), testosterone, and Follicular Stimulating Hormone

(FSH) are crucial for regulating reproductive functions. LH stimulates testosterone production, vital for male reproductive development, while FSH, in conjunction with testosterone, supports sperm growth and release.

Bisphenol A (BPA) has been found to disrupt Leydig cells, reducing testosterone production in rats (Mazroa, 2011 and Kamel *et al*., 2018) hinting at hypogonadotropic hypogonadism (Nakamura *et al*.,2010).

BPA exposure is also reported to affect female hormone levels. Zhou *et al*. (2023) found altered hormone concentrations in female rats, including increased testosterone and progesterone. Fernández *et al*. (2009) noted higher estradiol and lower progesterone levels due to high BPA doses, affecting hypothalamic-pituitary function. Ropero *et al*. (2008) observed varying effects on insulin, depending on the BPA dose. Additionally, Amaravathi *et al*. (2017) noted increased T3, T4, and E2 levels with higher BPA. Sujan *et al*. (2020) reported lower T4 and estradiollevels in rats.

Semen Analysis

Spermatozoa are highly susceptible to oxidative stress due to the abundance of polyunsaturated fatty acids in their cell membranes, making them vulnerable to damage by free radicals. This sensitivity results in lipid peroxidation, a process that adversely affects various sperm parameters, including motility, concentration, morphology, and overall quality.

Campos *et al*. (2019) reported that adult rats exposed to BPA at doses of 0.5 mg/kg and 5 mg/kg body weight experienced a significant 40% decrease in both daily and total sperm production. Chioccarelli *et al*. (2020) found that BPA negatively affected sperm motility and viability because it inducing chromatin condensation abnormalities during sperm maturation in the epididymis, consistent with Karnam *et al*. (2015) reported that significant reductions in epididymal sperm count (25 million/mL) at doses of 200 and 600 mg/kg body weight for 28 days. Grami *et al*. (2020) found that reduced sperm density, motility, viability, and an increase in morphological abnormalities (up to 30%).

Kourouma *et al*. (2014) reported dose-dependent reductions in epididymal sperm count (below 45 million/mL) and increased abnormalities, including acrosome vesicles, caps, and deformed nuclei in spermatids (2, 10, and 50 mg/kg body weight/day for 20 days. Mazroa (2011) documented a significant increase in sperm abnormalities in BPA-exposed rats, including coiled tails, bent tails, double tails, detached heads, and retained cytoplasmic droplets, suggesting potential challenges during sperm maturation. Higher BPA doses, such as 50 mg/kg, led to more kinked and detached head sperm upto 35%, potentially resulting from DNA damage during spermatogenesis.

Gross Pathology

Karnam *et al*. (2014) reported severe hemorrhagic lesions in various organs, including the lung, liver, kidney, spleen, stomach, intestine, and brain, in rats exposed to acute toxicity at 3375 mg/kg b.wt and mild congestion, slight liver enlargement, and brain hemorrhage in rats exposed to subchronic toxicity at 50 mg/kg b.wt. Amaravathi *et al*. (2012) noted moderate liver enlargement with paleness, lung edema, and emphysema, as well as enlarged spleen, heart, and testis at doses of 250 and 500 mg/kg b.wt. Sujan *et al*. (2020)found that bilateral hypertrophy, ventricular infarction and haemorrhages over the pericardium in heart, petechial haemorrhages, congestion, mutifocal infarction and hepatomegaly in liver, focal infarct and subcapsular petechiae in kidney, moderate congestion in brain, severe consolidation in lungs and slight increase in size of spleen@ 200, 500 mg/kg b.wt for 90 days.

Histopathology

Liver

Amaravathi *et al*. (2017) and Liu *et al*. (2022) observed various liver abnormalities resulting from BPA exposure, including degenerated hepatocytes, mononuclear cell aggregates, perivascular and periductular infiltration of mononuclear cells, periductular fibrosis, binucleated hepatocytes, and changes such as anisokaryosis and karyomegaly. These changes progressed to mild vesicular fatty changes in paracentral hepatocytes, increased apoptotic bodies, bile duct proliferation with dysplasia, proliferating blood capillaries, and endothelial

cell proliferation(Khan *et al*., 2021). Haroun *et al*. (2019) observed disarrangement of hepatic plates, dilatation, and congestion of blood sinusoids, congested dilated central veins, swollen hepatocytes with severe vacuolar degeneration, and marked inflammatory cellular infiltrations in the portal area in response to BPA exposure at 20 mg/kg body weight for 15 consecutive days (Alazzouni and Hassan, 2016). Poormoosavi *et al*. (2018) noted necrotic alterations of hepatocytes and increased lymphocytic infiltration and proliferation of Kupffer cells following treatment with BPA (Kanwal *et al*., 2018)

Kidney

Hassan *et al.* (2013) proposed that BPA-treated rats displayed renal tissue effects due to accumulated BPA metabolites and impaired renal excretion, resulting in marked congestion, tubular epithelial degeneration and necrosis.

Kidney showed congested glomeruli, intertubular hemorrhages, extensive degenerative changes in tubular and collecting ductular epithelial cells with cast formation in tubules, and hyalinized and thickened blood vessels with vacuolation in tunica intima and media. Edema of glomeruli and proliferation of mesangial cells (Amaravathi *et al*., 2017). Similar finding by Nafea Al-Sayed *et al*. (2021) observed distorted glomeruli, narrow bowman's space, mildly edematous proximal tubular lining, intra-tubular casts at 320 mg/kg b.wt. for three months. Additionally, Haroun *et al*. (2019) found that BPA exposure at 25 mg/kg b.wt. for six weeks resulted in atrophied renal corpuscles with contracted glomerular tufts and widening of the urinary space with luminal dilatation. Saleh *et al*. (2023) reported that glomerular tuft splitting, glomerular epithelium loss, and capsular space widening at 40 mg/kg b.wt. for 30 days.

Reproductive Organs

Testis

Exposure to BPA has been associated with extensive detrimental effects on the testes and sperm of rats in multiple studies. Grami *et al*. (2020) reported severe testicular damage, including disruptions in spermatogenic cells, abnormal seminiferous tubules, and the disruption of germinal membranes. Nafea Al-Sayed *et al*. (2021) observed thickened basement membranes, sclerotic tubules, a reduced germinal layer, and marked reduction in spermatogenesis. In a study by Amaravathi *et al*. (2012) reported congested blood vessels, sperm granuloma formation, and necrosed cells, disturbed germinal epithelial cells and changes in Leydig cell numbers and perivascular edema (Morgan *et al*., 2014), Sertoli cell degeneration (Acaroz *et al*., 2019).

Epididymis

The epididymis, consisting of the head (caput), body (corpus), and tail (cauda) segments, plays a vital role in sperm maturation and storage. It also functions as a protector against oxidative stress by eliminating reactive oxygen species (ROS) and releasing antioxidant enzymes into the epididymal lumen, crucial for maintaining sperm integrity.

Mazroa (2011) observed a decrease in the epithelial height in the cauda epididymis duct, along with structural changes. Tolba *et al*. (2018) reported irregularities in epididymal tubules and degenerative changes in principal cells, including cell alterations and hypertrophy. El-ghazzawy *et al*. (2011) observed stereocilia degeneration and cytoplasmic vacuolations in the cauda epididymis.

Accessory Sex Gland

Amaravathi *et al*. (2012) observed that, there were degenerative changes in seminal vesicle epithelial cells, characterized by hyperplasia of the lining epithelium, eosinophil infiltration, and papillary projections and prostate gland showed hyperplasia, desquamation of epithelial cells, acini shrinkage, thickened septa, eosinophilic infiltration, and prostate carcinoma-like changes, with these effects becoming more pronounced over time.

Uterus and Ovary

Newbold *et al*. (2007) reported enduring adverse effects of neonatal BPA exposure in rodents, resulting in conditions like cystic ovaries, cystic endometrial hyperplasia, and oviduct lesions, particularly in high-dose groups (200 mg/kg

b.wt). Chao *et al*. (2012) and Zhang *et al*. (2014) found that postnatal BPA exposure in rodents and lambs resulted in a decrease in ovarian follicular reserve, an increase in antral atretic follicles, a higher incidence of multiple oocyte follicles, and reduced ovarian weight, indicating potential disruptions in ovarian function over a period of two months.

Sujan *et al*. (2020) conducted histopathological examinations of rat ovarian tissue exposed to a 50 mg/kg b. wt of BPA over 90 days, revealing degenerative changes in granulosa cells within ovarian follicles. Amaravathi *et al*. (2014) found endometrial polyps. Alazzouni and Hassan (2016) observed hypertrophied glands and cell hyperplasia in the uteri of BPA-exposed rats, along with deteriorated and atretic ovarian follicles. Schonfelder *et al*. (2004) found reduced thickness in the uterine epithelium and cavities within the luminal endometrial epithelium in rats exposed to BPA at 50 mg/kg body weight during gestation days 6 through 21.Top of Form

Lung

Amaravathi *et al*. (2012) found that animals exposed to BPA exhibited various lung abnormalities, including congestion, perivascular and bronchiolar lymphoid aggregates, mononuclear cell (MNC) infiltration between alveoli, mild to moderate emphysema, peribronchial fatty infiltration, widening of the interstitial space, desquamated bronchial epithelial cells, bronchitis, increased hyperplasia of bronchiolar epithelium, hyperplasia of alveolar epithelial cells with papillary projections, and the presence of giant cells in focal areas (Saleh *et al*., 2023).

Brain

Morgan *et al*. (2014) found significant neurological changes included submeningeal hemorrhages, capillary proliferation, swollen neurons with central chromatolysis, congestion, spongiosis in the white matter, and endothelial cell proliferation in cerebral capillaries. Demyelinating changes, gliosis, perineuronal satellitosis and neuronophagia were evident in later stage. In the cerebellum, there were hemorrhages and ghost cell appearance of purkinje cells, along with changes like demyelination and alterations in granular layer structure..These brain alterations were linked to BPA's estrogen-like properties, potentially affecting neuronal development through estrogenic mechanisms (Stump *et al*., 2010).

Other Organs

Yamasaki *et al*. (2002) and Amaravathi *et al*. (2017) observed that Adrenal cortical vacuolation, haemorrhage and degenerative changes. Additionally, in gastrointestinal tract showed squamous epithelial hyperplasia in the stomach, lacteal dilatation in the duodenum, and mucosal hyperplasia in the cecum, inflammatory cell infiltration in BPA-treated rats.

In BPA treated groups heart showed Myocardial cell hyaline degeneration (Acaroz *et al*., 2019), Focal myocardial necrosis, Intermuscular hemorrhages, endothelial cell proliferation (Amaravathi *et al*., 2017) along with pancreas showed hemorrhages, acinar epithelial vacuolar degeneration, fatty infiltration, islets of langerhans atrophy, and ductular epithelium hyperplasia. In thyroid congestion, hemorrhages, irregular acini, adenocarcinoma formation, and inflammatory cell infiltration and in lymphoid organs varying degrees of lymphocyte depletion and hemorrhages were evident in three months at 250, 500 mg/kg b.wt.

Immunohistochemistry

In mammary glands, BPA from embryonic day 9 through PND 1 @ 0, 2.5, 25, 250 or 1000μg/Kg b.wt. daily for 90 days induced an increase in hyperplastic ducts that were estrogen-sensitive, marked by Ki-67 and ER positivity (Murray *et al*., 2007). In male reproductive organs, BPA prompted enhanced apoptosis in spermatogenic cells and altered the relative expression of Bax and Bcl2 (Li *et al*., 2016). Additionally, BPA caused increased immunoreactivity of Fas, Caspase-3, and Fas Ligand in Leydig cells, primary spermatocytes, and spermatids (Sheng *et al*., 2012). Amaravathi *et al*. (2017) noted that BPA exposure led to increased expression of Bcl2 and VEGF in various tissues, including hepatocytes around the central vein, oviduct epithelium, and endometrial glands. Furthermore, BPA exposed rats showed increased expression of ER-β receptors in ovarian follicular and stromal cells and uterine endometrial and luminal cells,

emphasizing its estrogenic impact (Moustafa and Ahmed, 2016). A study on nephrotoxicity in rats indicated strong Bax immunostaining in the kidney cortex following BPA exposure, which was alleviated by gallic acid treatment (Saleh *et al*., 2023). Moreover, BPA exposure showed positive anti-caspase 3 activity in the liver and kidney and positive anti-estrogenic activity in the pancreas (Alazzouni and Hassan, 2016).Top of Form

Conclusion

Widespread exposure to Bisphenol A (BPA) persists due to its extensive use in polycarbonate plastic production and its release into food and beverages. Recognized as a significant risk factor for endocrine, immune, and oncological diseases, BPA has been banned in certain products, including cosmetics and baby bottles. However, conflicting results on its toxic effects exist, attributed to varying concentrations and experimental models. Non-oral administration, diverse doses, and limited animal test subjects further complicate toxicological and epidemiological interpretations. Efforts to elucidate BPA's molecular mechanisms reveal its binding to specific hormone receptors (ERs, ARs, and thyroid hormone receptors), directly regulating gene expression. Non-genomic actions involve membrane-associated ERs and/or GPR30, activating signal transduction pathways and key transcription factors related to growth, differentiation, and energy metabolism. Upstream pathways may contribute to stable, inheritable modifications by regulating epigenetic enzymes, sustaining BPA-induced effects. Alternative compounds like bisphenol S (BPS) and bisphenol F (BPF) have been explored but exhibit similar or heightened risks. BPS, for instance, proves as effective as BPA in promoting breast cancer and poses additional reproductive harm. Consequently, limiting plastic consumption and advocating for BPA-free products remain the most effective practices to mitigate its harmful effects.

References

Acaroz, U., Ince,S., Arslan-Acaroz,D., Gurler,Z., Demire,H.H., Kucukkurt,I., Eryavuz,A., Kara,R., Varol, N., and Zhu,K., (2019) Bisphenol-A induced oxidative stress, inflammatory gene expression, and metabolic and histopathological changes in male Wistar albino rats: protective role of boron. *Toxicol. Res*., **8**(2): 262-269.

Alazzouni, A. S and Hassan,B.N., (2016) Evaluation of anti-estrogen drug and stem enhance in amelioration of histopathological effects of bisphenol a on vital organs in murine model: histological and immunohistochemical studies. *Int. J. Pharm. Bio Sci.*, **7**(2): 478-491.

Almeida, S., Raposo,A., Almeida-Gonzalez, M. and Carrascosa,C., (2018) Bisphenol A: Food exposure and impact on human health. C*ompr. Rev. Food. Sci. Food. Saf.*, **17**(6): 1503-1517.

Alonso-Magdalena, P., Ropero,A.B., Soriano,S., Garcia-Arevalo,M., Ripoll,C., Fuentes,E., Quesada, I., and Nadal,A., (2012) Bisphenol-A acts as a potent estrogen via non-classical estrogen triggered pathways. *Mol. cell. Endocrinol.,* **355**(2): 201-207.

Amaravathi, P., Srilatha,C., Ramadevi,V., Sreenivasulu,D., Prasad, P.E., and SujathaK., (2017) Hepatotoxic Effect of Bisphenol A in Rats, an Immunohistochemical and Ultrastructural Study. *J. Anim. Res.*, **7**(5): 871-878.

Amaravathi, P., Srilatha,C.H., Ramadevi,V., Sreenivasulu,D., Prasad, P.E., and Sujatha,K., (2012) Pulmonary and genotoxicity of Bisphenol-A in Wistar albino rats. *Curr. Biotica.*, **6**(1): 53-60.

Amin, M.M., Ebrahim, K., Hashemi, M., Shoshtari-Yeganeh, B., Rafiei, N., Mansourian, M., and Kelishadi, R., (2019) Association of exposure to Bisphenol A with obesity and cardiometabolic risk factors in children and adolescents. *Int. J. Environ. Health Res.,* **29**(1): .94-106.

Basak, R., Banerjee, A., and Rai,U., (2021) December. Demystifying bisphenol a-induced alterations in hypothalamic-pituitary-ovarian functions leading to polycystic ovarian syndrome. *Proc. Zool. Soc*., **4**(4): 466-478.

Campos, P.D., Oliveira,I.M., Sena de Souza,J., Da Conceicao,R.R., Giannocco,G.,. Chiamolera, M.I, Silva,M.R.D.D., Romano, M.A., and Romano,R.M., (2019) Maternal bisphenol A exposure disrupts spermatogenesis in adult rat offspring. *J. Toxicol. Environ. Health.,* **82**(3): 163-175.

Caserta, D., Costanzi,F., De Marco,M.P., Di Benedetto,L., Matteucci,E., Assorgi,C., Pacilli,M.C., Besharat,A.R., Bellati, F., and Ruscito,I., (2021) Effects of endocrine-disrupting chemicals on endometrial receptivity and embryo implantation: a systematic review of 34 mouse model studies. *Int. J. Environ. Res.,* **18**(13): 6840.

Chailurkit, L.O., Aekplakorn, W., and Ongphiphadhanakul, B., (2016) The association of serum bisphenol A with thyroid autoimmunity. *Int. J. Environ. Res. Public Health.,***13**(11): 1153.

Chao, H.H., Zhang,X.F., Chen,B., PanB., Zhang,L.J., Li,L., SunX.F., Shi Q.H., and ShenW., (2012) Bisphenol A exposure modifies methylation of imprinted genes in mouse oocytes via the estrogen receptor signaling pathway. *Histochem. Cell. Biol*., **137**: 249-259.

Chioccarelli, T., Manfrevola,F., Migliaccio,M., Altucci,L., Porreca,V., Fasano, S., and CobellisG., (2020) Fetal-

Perinatal exposure to bisphenol-A affects quality of spermatozoa in adulthood mouse. *Int. J. Endocrinol*., 2020.

Christiansen, M.N., Chik,J., LeeL., Anugraham,M., Abrahams, J.L., and Packer,N.H., (2014) Cell surface protein glycosylation in cancer. *Proteomics.*, **14**(4-5): 525-546.

Della Rocca, Y., Traini, E.M., Diomede, F., Fonticoli, L., Trubiani, O., Paganelli, A., Pizzicannella, J. and Marconi, G.D., (2023) Current evidence on Bisphenol A exposure and the molecular mechanism involved in related pathological conditions. *Pharmaceutics*, **15**(3): 908.

Dong, S., Terasaka, S., and Kiyama,R., (2011) Bisphenol A induces a rapid activation of Erk1/2 through GPR30 in human breast cancer cells. *Environ. pollut.*, **159**(1): 212-218.

El Ghazzawy, I.F., Meleis,A.E., Farghaly, E.F., and Solaiman,A., (2011) Histological study of the possible protective effect of pomegranate juice on bisphenol-A induced changes of the caput epididymal epithelium and sperms of adult albino rats. *Alex. J. Med*., **47**(2): 125-137.

Encarnação, T., Pais,A.A., Campos, M.G., and Burrows,H.D., (2019) Endocrine disrupting chemicals: Impact on human health, wildlife and the environment. *Sci. prog.,* **102**(1): 3-42.

Eweda, S.M., Newairy,A.S.A., Abdou, H.M., and Gaber,A.S., (2020) Bisphenol A-induced oxidative damage in the hepatic and cardiac tissues of rats: The modulatory role of sesame lignans. *Exp. Ther. Med*., **19**(1): 33-44.

Fenichel, P., Chevalier, N., and Brucker-Davis,F., (2013) Bisphenol A: an endocrine and metabolic disruptor. *Ann. Endocrinol.*, **74**(3): 211-220.

Fernández, M., Bianchi,M., Lux-Lantos,V., Libertun,C., (2009) Neonatal exposure to bisphenol a alters reproductive parameters and gonadotropin releasing hormone signaling in female rats. *Environ. Health Perspect.*, **117**(5): 757-762.

Gerona, R.R., Woodruff,T.J., Dickenson,C.A., PanJ., Schwartz,J.M., Sen,S., Friesen,M.W., Fujimoto, V.Y., and Hunt,P.A., (2013) Bisphenol-A (BPA): BPA glucuronide, and BPA sulfate in midgestation umbilical cord serum in a northern and central California population. *Environ. Sci. Technol.,* **47**(21): 12477-12485.

Ghosh, A., Tripathy, A., and Ghosh,D., (2022). Impact of endocrine disrupting chemicals (EDCs) on reproductive health of human. *Proc. Zool. Soc. Lond.,***75**(1): 16-30.

Grami, D., Rtibi,K., Hammami,I., Selmi, S., De Toni,L., Foresta,C., Marzouki, L., and Sebai,H., (2020) Protective action of eruca sativa leaves aqueous extracts against bisphenol a-caused in vivo testicular damages. *J. Med. Food*. **23**(6): 600-610.

Gurmeet, K.S.S., Rosnah,I., Normadiah,M.K., Das,S., and Mustafa,A.M., (2014) Detrimental effects of bisphenol A on development and functions of the male reproductive system in experimental rats. *EXCLI J*. **13**:151.

Haroun, M.R., Zamzam,I.S., Metwally, E.S., and EL-Shafey,R.S., (2019) Effect of vitamin c on bisphenol a induced hepato& nephrotoxicity in albino rats.*Egypt. J. Forensic* Sci. Appl. Toxicol., **16**: 57-85.

Hassan, A.H., Ismail,A.A., and Khudir,A.N., (2013) Effects of pre-and postnatal exposure to Bisphenol-A on the reproductive efficacy in male albino rats. *J. Kerala. Univ.,* **11**(3): 158-172.

Higashihara, N., Shiraishi,K., Miyata,K., Oshima,Y., Minobe, Y., and Yamasaki,K., (2007) Subacute oral toxicity study of bisphenol F based on the draft protocol for the "Enhanced OECD Test Guideline no. 407". *Arch. Toxicol*., **81**: 825-832.

Jensen, T.K., Mustieles, V., Bleses, D., Frederiksen, H., Trecca, F., Schoeters, G., Andersen, H.R., Grandjean, P., Kyhl, H.B., Juul, A., and Bilenberg, N., (2019) Prenatal bisphenol A exposure is associated with language development but not with ADHD-related behavior in toddlers from the Odense Child Cohort. *Environ. Res.,* **170**:398-405.

Kabuto, H., Amakawa, M., and Shishibori,T., (2004) Exposure to bisphenol A during embryonic/fetal life and infancy increases oxidative injury and causes underdevelopment of the brain and testis in mice. *Life. sci.,* **74**(24): 2931-2940.

Kamel, A. H., Foaud, M.A., and Moussa,H.M., (2018) The adverse effects of bisphenol A on male albino rats. *J. Basic. Appl. Zool*., **79**(1): 1-9.

Kanwal, Q., Qadir,A., Amina, Asmatullah, Iqbal H.H., and MunirB., (2018) Healing potential of Adiantum capillus-veneris L. plant extract on bisphenol A-induced hepatic toxicity in male albino rats. *Environ. Sci. Pollut. Res.,* **25**: 11884-11892.

Karnam, S.S., Ghosh, R.C., and Mondal,S., (2014) Effect on body weight and feed consumption of Bisphenol A induced subacute toxicity in rats. *Explor. Anim. Med. Res.,* **4:** 228-234.

Karnam, S.S., Ghosh,R.C., Mondal, S., and Mondal,M., (2015) Evaluation of subacute bisphenol–A toxicity on male reproductive system. *Vet. world.,* **8**(6): 738.

Kaur, S., Saluja, M., and Bansal,M.P.,(2018) Bisphenol A induced oxidative stress and apoptosis in mice testes: Modulation by selenium. *Andrologia*., **50**(3): 12834.

Kaur, S., Saluja,M., Aniqa, A., and Sadwal,S., (2021) Selenium attenuates bisphenol A incurred damage and apoptosis in mice testes by regulating mitogen-activated protein kinase signalling. *Andrologia*., **53**(3):13975.

Kim, Y.H., Kim,C.S., Park,S., Han,S.Y., Pyo, M.Y., and Yang,M., (2003) Gender differences in the levels of bisphenol A metabolites in urine. *Biochem. Biophys. Res. Commun.,* **312**(2): 441-448.

Kouroumaa, A., Tamayob, Y.M.L., and Kedia,Y., (2014) Correlation between Epididymal Semen Quality Parameters and Malondialdehyde Levels in Adults SD rats Liver after Exposure to Bisphenol A. *IJSR*, **3**(6): 0201458

LaKind, J.S., Goodman, M., and Mattison,D.R., (2014) Bisphenol A and indicators of obesity, glucose metabolism/ type 2 diabetes and cardiovascular disease: a systematic review of epidemiologic research. *Crit. Rev. Toxicol*., **44**(2): 121-150.

Li, J., Bach,A., Crawford,R.B., Phadnis-Moghe,A.S., Chen,W., D'Ingillo, S., and Kaminski,N.E., (2018) CLARITY-BPA: Effects of chronic Bisphenol A exposure on the immune system: Part 1–Quantification of the relative number and proportion of leukocyte populations in the spleen and thymus. *Toxicol*., **396**: 46-53.

Li, J., Sheng,N., Cui,R., Feng,Y., Shao,B., Guo,X., Zhang, H., and Dai,J., (2016) Gestational and lactational exposure to bisphenol AF in maternal rats increases testosterone levels in 23-day-old male offspring. *Chemosphere*., **163**:552-561.

Liu, R., Liu,B., Tian,L., Jiang,X., Li,X., Cai,D., Sun,J., Bai, W., and Jin,Y., (2022) Exposure to bisphenol A caused hepatoxicity and intestinal flora disorder in rats. *Int. J. Mol. Sci.,* **23**(14): 8042.

Mammadov, E., Uncu, M., and Dalkan, C., (2018) High prenatal exposure to bisphenol A reduces anogenital distance in healthy male newborns. *J CLIN RES PEDIATR E*., **10**(1): 25.

Marino, M., Pellegrini,M., La Rosa, P., and Acconcia,F., (2012) Susceptibility of estrogen receptor rapid responses to xenoestrogens: Physiological outcomes. *Steroids*., **77**(10): 910-917.

Markey, C.M., Wadia,P.R., Rubin,B.S., Sonnenschein,C., and SotoA.M., (2005) Long-term effects of fetal exposure to low doses of the xenoestrogen bisphenol-A in the female mouse genital tract. *Biol. Reprod*., **72**(6): 1344-1351.

Mazroa, S.A. (2011) Effect of bisphenol A on the cauda epididymis of adult male albino rats and the possible protective role of quercetin: a histological and immunohistochemical study. *Egypt. J. Histol*., **34**(2): 377-390.

Minatoya, M., Itoh, S., Yamazaki, K., Araki, A., Miyashita, C., Tamura, N., Yamamoto, J., Onoda, Y., Ogasawara, K., Matsumura, T., and Kishi, R., (2018) Prenatal exposure to bisphenol A and phthalates and behavioral problems in children at preschool age: the Hokkaido Study on Environment and Children's Health. *Environ. Health Prev. Med.,* **23**(1): 1-11.

Morgan, A.M., El-Ballal,S.S., El-Bialy, B.E., and El-Borai,N.B., (2014) Studies on the potential protective effect of cinnamon against bisphenol A-and octylphenol-induced oxidative stress in male albino rats. *Toxicol. Rep*., **1**: 92-101.

Moustafa, G.G. and AhmedA.A., (2016) Impact of prenatal and postnatal exposure to bisphenol A on female rats in a two generational study: Genotoxic and immunohistochemical implications. *Toxicol. Rep*., **3**: 685-695.

Mukhopadhyay, R., Prabhu,N.B., Kabekkodu, S.P., and Rai,P.S., (2022) Review on bisphenol A and the risk of polycystic ovarian syndrome: an insight from endocrine and gene expression. *Environ. Sci. Pollut. Res.,* **29**(22): 32631-32650.

Murray, T.J., Maffini,M.V., Ucci,A.A., Sonnenschein, C. and Soto,A.M., (2007) Induction of mammary gland ductal hyperplasia and carcinoma in situ following fetal bisphenol A exposure. *Reprod. Toxicol*., **23**(3): 383-390.

Nafea Al-Sayed, M., Mohamed Sherif,M., Mohamed Al-Fadaly,N., Ibrahim Hasan A., and SayedA.E.R., (2021) Histopathological and histochemical study of the effects of vitamin c on bisphenol a toxicity in albino rats. *Al-Azhar. Med. J*., **50**(4): 2901-2914.

Nah, W.H., Park, M.J., and Gye,M.C.,(2011) Effects of early prepubertal exposure to bisphenol A on the onset of puberty, ovarian weights, and estrous cycle in female mice. *Clin. Exp. Reprod. Med.,* **38**(2): 75.

Nakamura, D., Yanagiba,Y., Duan,Z., Ito,Y., Okamura,A., Asaeda,N., Tagawa,Y., Li,C., Taya,K., Zhang S.Y., and NaitoH., (2010) Bisphenol A may cause testosterone reduction by adversely affecting both testis and pituitary systems similar to estradiol. *Toxicol. Lett*., **194**(1-2): 16-25.

Newbold, R.R., Jefferson, W.N., and Padilla-Banks,E., (2009) Prenatal exposure to bisphenol a at environmentally relevant doses adversely affects the murine female reproductive tract later in life *Environ. Health. Persp*., **117**(6): 879-885.

Özel, Ş., Tokmak, A., Aykut, O., Aktulay, A., Hançerlioğulları, N., and Engin Ustun, Y., (2019) Serum levels of phthalates and bisphenol-A in patients with primary ovarian insufficiency. *Gynecol. Endocrinol.,***35**(4): 364-367.

Pal, S., Sarkar,K., Nath,P.P., Mondal,M., Khatun, A., and Paul,G., (2017) Bisphenol S impairs blood functions and induces cardiovascular risks in rats. *Toxicol. rep.,* **4**:560-565.

Panpatil, V.V., KumariD., Chatterjee,A., Kumar,S., Bhaskar,V., Polasa, K., and Ghosh,S., (2020) Protective effect of turmeric against bisphenol-a induced genotoxicity in rats. *J. Nutr. Sci. Vitaminol*., **66**(Supplement): 336-S342.

Pirozzi, C., Pontoni, L., Fabbricino, M., Bogush, A., and Campos, L.C., (2020) Effect of organic matter release from natural cork used on bisphenol a removal from aqueous solution. *J. Clean. Prod.*, **244**:118675.

Poormoosavi, S.M., Najafzadehvarzi,H., Behmanesh, M.A., and Amirgholami,R., (2018) Protective effects of Asparagus officinalis extract against Bisphenol A-induced toxicity in Wistar rats. *Toxicol. Rep.*, **5**: 427-433.

Provencher, G., Bérubé,R., Dumas,P., Bienvenu,J.F., Gaudreau, E., Bélanger,P., and Ayotte,P., (2014) Determina-

tion of bisphenol A, triclosan and their metabolites in human urine using isotope-dilution liquid chromatography–tandem mass spectrometry. *J. Chromatogr.*, **1348**: 97-104.

Ropero, A.B., Alonso-Magdalena,P., Garcia-Garcia,E., Ripoll,C., Fuentes, E., and Nadal,A., (2008) Bisphenol-A disruption of the endocrine pancreas and blood glucose homeostasis. *Int. j. androl.*, **31**(2): 194-200.

Sabry, R., Williams, M., Werry, N., LaMarre, J., Favetta, L.A., (2022) BPA Decreases PDCD4 in Bovine Granulosa Cells Independently of miR-21 Inhibition. *Int. J. Mol. Sci.,*8276.

Saleh, S.M., Mahmoud,A.B., Al-Salahy, M.B., and Mohamed Moustafa,F.A., (2023) Morphological, immunohistochemical, and biochemical study on the ameliorative effect of gallic acid against bisphenol A-induced nephrotoxicity in male albino rats. *Sci. Rep.,* **13**(1): 1732.

Samova, S., Patel,C.N., Doctor,H., Pandya,H.A., and Verma,R.J., (2018) The effect of bisphenol A on testicular steroidogenesis and its amelioration by quercetin: an in vivo and in silico approach. *Toxicol. res.,* **7**(1): 22-31.

Schönfelder, G., Friedrich,K., Paul, M., and Chahoud,I., (2004) Developmental effects of prenatal exposure to bisphenol A on the uterus of rat offspring. *Neoplasia.,* **6**(5): 584-594.

Sheng, Z.G., Tang,Y., Liu,Y.X., Yuan,Y., Zhao,B.Q., Chao, X.J., and Zhu,B.Z., (2012) Low concentrations of bisphenol a suppress thyroid hormone receptor transcription through a nongenomic mechanism. *Toxicol. appl. pharma.,* **259**(1): 133-142.

Spanier, A.J., Fiorino, E.K. and Trasande, L., (2014) Bisphenol A exposure is associated with decreased lung function. *J. Pediatr.*, **164**(6):.1403-1408.

Stump, D.G., Beck,M.J., Radovsky,A., Garman,R.H., Freshwater,L.L., Sheets,L.P., Marty,M.S., Waechter, J.M. Jr, Dimond,S.S., Van Miller, J.P., and Shiotsuka,R.N., (2010) Developmental neurotoxicity study of dietary bisphenol A in Sprague-Dawley rats. *Toxicol. Sci.,* **115**(1):167-182.

Sujan, K.M., Hoque,E., Haque,M.I., Mustari,A., Miah, M.A., and Islam,M.K., (2020) Ameliorating effects of black seed oil on bisphenol-A induced abnormality of blood, hormone profile and gonadal histology of female mice. *J. Adv. Biotechnol. Exp. Ther.*, **3**(1): 43-48.

Tian, Y., Zhou, X., Miao, M., Li, D.K., Wang, Z., Li, R., Liang, H., and Yuan, W., (2018) Association of bisphenol A exposure with LINE-1 hydroxymethylation in human semen. *Int. J. Environ.*, **15**(8): 1770.

Tolba, A.M., and Mandour,D.A., (2018) Histological effects of bisphenol-A on the reproductive organs of the adult male albino rat. *Eur. J. Anat.*, **22**(2): 89-102.

Tse, L.A., Lee, P.M.Y., Ho, W.M., Lam, A.T., Lee, M.K., Ng, S.S.M., He, Y., Leung, K.S., Hartle, J.C., Hu, H., and Kan, H., (2017) Bisphenol A and other environmental risk factors for prostate cancer in Hong Kong. *Environ Int.*, **107**:1-7.

Ullah, A., Pirzada, M., Jahan, S., Ullah, H., Shaheen, G., Rehman, H., Siddiqui, M.F., and Butt, M.A., (2018) Bisphenol A and its analogs bisphenol B, bisphenol F, and bisphenol S: Comparative in vitro and in vivo studies on the sperms and testicular tissues of rats. *Chemosphere*, **209**:508-516.

Wang, Z., Liu, H., and Liu,S., (2017) Low-dose bisphenol A exposure: A seemingly instigating carcinogenic effect on breast cancer. *Adv Sci.,* **4**(2): 1600248.

Yamasaki, K., Sawaki,M., Noda,S., Imatanaka, N., and Takatsuki,M., (2002) Subacute oral toxicity study of ethynylestradiol and bisphenol A, based on the draft protocol for the 'Enhanced OECD Test Guideline no. 407'. *Arch. Toxicol*., **76**:65-74.

Yang, Y., Shi, Y., Chen, D., Chen, H., and Liu, X., (2022) Bisphenol A and its analogues in paired urine and house dust from South China and implications for children's exposure. *Chemosphere*, **294**: 133701.

Yilmaz, B., Terekeci,H., Sandal, S., and Kelestimur,F., (2020) Endocrine disrupting chemicals: exposure, effects on human health, mechanism of action, models for testing and strategies for prevention. *Rev. Endocr. Metab. Disord*., **21**:127-147.

Youssef, M.M., El-Din, E.M.S., AbuShady, M.M., El-Baroudy, N.R., Abd El Hamid, T.A., Armaneus, A.F., El Refay, A.S., Hussein, J., Medhat, D., and Latif, Y.A., (2018) Urinary bisphenol A concentrations in relation to asthma in a sample of Egyptian children. *Hum. Exp. Toxicol.,* **37**(11): 1180-1186.

Zhang, K.S., Chen,H.Q., Chen,Y.S., Qiu,K.F., Zheng,X.B., Li,G.C., Yang, H.D., and WenC.J., (2014) Bisphenol A stimulates human lung cancer cell migration via upregulation of matrix metalloproteinases by GPER/EGFR/ERK1/2 signal pathway. *Biomed. Pharmacother.,* **68**(8): 1037-1043.

Zhou, R., Jin,S., Jin,C., Sun,Y., Zhang,L., Yan, J., and Jiang,H., (2023) Association of urinary bisphenol A with chronic obstructive pulmonary disease–related diseases: evidence from the National Health and Nutrition Examination Survey database (2005–2016). *Environ. Sci. Pollut. Res.,* **30**(12): 33170-33180.