

Endocrine Disruption and Perspective Human Health Implications: A Review

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Citation

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Abstract

Over the last decades, many tone of man - made chemicals have been produced and released into the environment. Many of these environmental contaminants have the ability to modulate the action of hormones by interacting with hormone receptors and mimic or antagonize the production and activities of endogenous hormones are called endocrine disruptors. The public health implications of these so called endocrine disruptors have been the subject of scientific debate, media interest and policy attention over the past several years. In this article we review the background of environmental endocrine disruption, developmental effects due to early exposure of Endocrine disrupting compounds (EDCs), assessing mode of action of endocrine disruptors, disruption of female hormonal function and its potential effects, and current evidence of possible impacts on reproduction system and neurobehavioral function. Finally, worldwide health policy activities that are relevant to endocrine-disrupting compounds in the environment are discussed.

INTRODUCTION

Current widespread interest in a range of health effects potentially associated with endocrine-disrupting compounds (EDCs) has made exposure assessment for these compounds a priority. Studies of potential health effects associated with EDCs have been hampered by lack of information about the major sources of exposure to EDCs. Furthermore, because many EDCs act additively through a common mechanism of action or have antagonistic or other interactive effects by operating at different points in cell signaling systems, consideration of exposure to mixtures is critical in studies of health effects. Hormones play a crucial role in guiding normal cell differentiation in early life forms, and so exposure to endocrine disrupting substances in the egg or in the womb can alter the normal process of development. Mature animals can also be affected, but in the developing organism it is especially vulnerable. Exposure at this sensitive time may cause effects that are not evident until later in life, such as effects on learning ability, behavior, reproduction and increased susceptibility to cancer and other diseases. Many pesticides are now suspected of being endocrine disruptors-compounds that can lead to an increase in birth defects, sexual abnormalities and reproductive failures ¹. Specific effects found in wildlife and laboratory studies on mammals, reptiles, amphibians and fish include abnormal blood hormone levels, reduced fertility, altered

sexual behavior, modified immune system, masculinization of females, feminization of males, undescended testicles, reduced penis size and testis, altered bone density and structure, cancers of the male and female reproductive tract, and lastly, malformed fallopian tubes, uterus and cervix. Other than pesticides some of the industrial chemicals also cause endocrine disruption like phthalates, a class of chemicals used as plasticizers, have attracted special attention because of their high-volume production, environmental presence and possible association with adverse reproductive health outcomes in females ²³. According to recent biomonitoring studies exposure of the general population to phthalates is widespread ⁴⁵.

Wildlife will be especially vulnerable to the endocrine disrupting effects of pesticides, because these chemicals are deliberately released into the environment. Humans exposed occupationally are at increased risk, and there are studies linking exposure to endocrine-disrupting compounds which comprise a diverse group of compounds and their observed health effects include estrogenic, androgenic, anti-estrogenic and antiandrogenic effects and disruption of thyroid function, the immune system, sexual differentiation of the brain during fetal development and motor function. EDCs may also have carcinogenic effects ⁶ and significant decrease of testis weight, epididymal sperm counts, sperm motility and marker testicular enzymes for testosterone biosynthesis ⁷

. The developing offspring is the most sensitive target of endocrine disruption. Many man-made chemicals can cross the placental barrier, thereby allowing the mother's body burden to be shared with her developing offspring. Further, intake occurs as nursing animals drink fat-loving chemicals that are bound into fat-rich milk. In egg laying species, the chemicals are transported from mother to the egg yolk where the chemicals cause irreversible damage during incubation. These concentrated doses of chemicals during embryonic, fetal and early postnatal development can be the highest exposures encountered throughout life. Again, the timing for such exposures is of concern since much of the neural, reproductive, and immune development continues long after birth or hatching ⁸ .

FETAL LIABILITY AND DEVELOPMENTAL EXPOSURE

In a recent USEPA summary report ⁹ , defined liability applied to risk assessment as a four-component system: (1) susceptibility or sensitivity of the human or ecological receptors; (2) differential exposures of the receptors; (3) differential preparedness of the receptor to withstand the insult from exposure; (4) differential ability to recover from these effects. All of these components are pertinent to systems undergoing development from the fetus through childhood. The developing child (fetus to prepuberty) is particularly susceptible to environmental insult, because development is a highly integrated process in which high rates of proliferation and extensive differentiation are coordinated with each other and with programmed cell death. Rapid growth rates allow for mutagenic and epigenetic alterations as cells proliferate. Some pesticides and other industrial chemicals can disturb development of the endocrine system and of the organs that respond to endocrine signals in organisms indirectly exposed during prenatal and/or early postnatal life; effects of exposure during development are permanent and irreversible. The risk to the developing organism can also stem from direct exposure of the offspring after birth or hatching. According to Buckley et al., ¹⁰ the frequency of household insecticide use by mothers during pregnancy is associated with an increased risk for non-Hodgkin's lymphoma. An association has also been demonstrated for mothers exposed in the course of household pest extermination. They have also established odd ratios for various immunopathological and histological parameters, as well as for different age groups, to determine whether the risks observed might be associated with one of these variables in particular. The most statistically significant association was with direct childhood

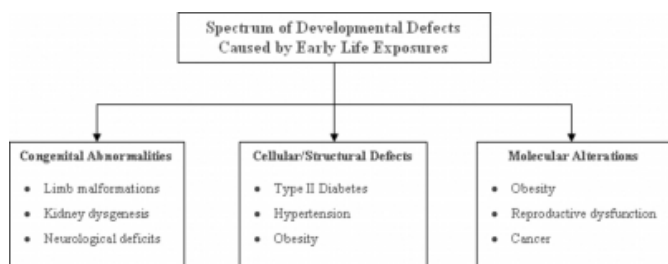
exposure ($P < 0.01$ for B and T cell lymphoma, large-cell lymphoma in the 0-6 age group, and $P < 0.05$ for Burkitt lymphoma and acute lymphoblastic leukemia in the 6+ age group). According to these authors, direct postnatal exposure to pesticides is significantly associated with non-Hodgkin's lymphoma in children. In addition, transgenerational exposure can result from the exposure of the mother to a chemical at any time throughout her life before producing offspring due to persistence of endocrine-disrupting compounds in body fat, which is mobilized during egg laying or pregnancy and lactation. Many of these compounds can directly bind to hormone receptors, block or in some way alter the activity of endogenous compounds that are synthesized by the endocrine system, thereby initiating or blocking receptor-activated gene transcription ¹¹ . Other exogenous compounds act indirectly on hormonal homeostasis by altering steroidogenesis, hormone transport on binding proteins, receptor numbers on target organs or hormone metabolism. For example, polychlorinated biphenyls (PCBs) interfere with thyroid function through a variety of mechanisms, including increased metabolism of T_4 (thyroxine), interference with T_4 delivery to the developing brain by displacement from the carrier protein, and interference with the conversion of T_4 to T_3 (triiodothyronine) ¹² . Changes in thyroid hormone levels can adversely affect neurobehavioral functions, fertility, pregnancy outcome, and postnatal development in humans and animals ¹³ .

For many years now, there has been public concern raised about the potential health effects of pesticides on the developing fetus and in childhood. Depending on stage of development, the fetus is selectively sensitive to particular chemical toxicants. There is an increasing concern for adverse health outcomes after developmental early life exposure to environmental compounds that trouble the endocrine system is explained in Fig. 1. Gestational and prenatal exposures to EDCs may have long-term effects on the endocrine system that can influence tumor development later in life. Alexander et al., ¹⁴ recently demonstrated that in utero exposure to compounds is associated with an elevated risk of acute leukemia in infants. The carbamate pesticide propoxur is one environmental chemical identified where exposure was highly correlated with leukemia in the offspring. Parental cigarette smoking has also been linked to childhood cancer. In a dose response study conducted by Wolf et al., ¹⁵ , in which testosterone propionate was administered to rat dams during late gestation (gestational day 14-19), inhibition of nipple development and increased

anogenital distance in female offspring were among the most sensitive indicators of masculinisation. However, exposure to environmental compounds in utero—a proliferative period in human development has long been thought to be critically involved in the causation of cancers in children and young adults. Although a wide range of potentially harmful agents may be involved, there is unequivocal evidence for two environmental agents: ionizing radiation and the estrogen agonist diethylstilbestrol (DES) ¹⁶. Transgenerational effect includes that the endocrine disruptors may not only impact the individual directly exposed, but also future generations. DES in mice can be passed to subsequent generations even though they are not directly exposed. The increased susceptibility for tumors was seen in both the granddaughters and grandsons of mice who were developmentally exposed to DES ¹⁷. Mechanisms involved in the transmission of disease were shown to involve epigenetic events. Kappers et al., ¹⁸ reported that two endocrine disrupting compounds, caused fertility defects in male rats that were passed down to nearly every male in subsequent generations. This study suggests that the two compounds may have caused changes in the developing male germ cells and that endocrine disruptors may be able to reprogram or change the expression of genes without mutating DNA.

Figure 1

Figure 1: Spectrum of developmental defects



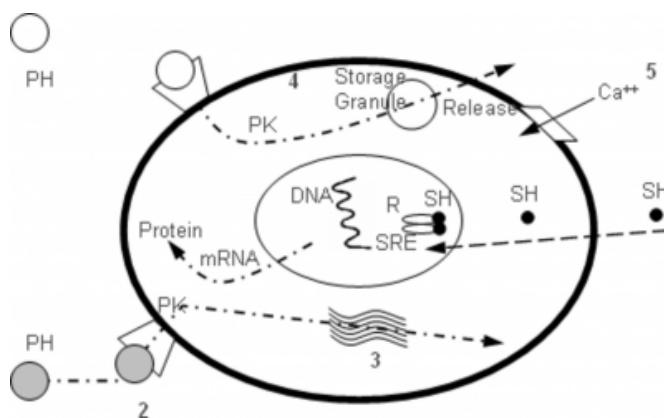
ASSESSING MODE OF ACTION AND POTENTIAL HEALTH IMPLICATIONS

Information regarding mode of action of endocrine disruptor improves the scientific basis for risk decisions. The processes involved in the mode of endocrine disruptors are much more complicated with the interconnection of nervous, endocrine and immune systems. Endocrine-disrupting compounds are known to act at multiple sites through multiple modes of action, but the mechanism of action including putative exposure-response relationships are poorly understood ¹⁹. It is necessary to explore the mechanisms of action from multiple points of view. Numerous substances, such as dioxins, polychlorobiphenyls

(PCBs), DDT (dichlorodiphenyltrichloroethane) and phthalates have been suspected of working through endocrine-disruptive mechanisms ²⁰. Environmental agents can alter endocrine function through a variety of mechanisms and some of the better-studied cellular target sites are included in Fig. 2.

Figure 2

Figure 2: Environmental agents can alter endocrine function at cellular level through a variety of mechanisms.



Some of the better-studied cellular target sites include (1) steroid hormone receptor-mediated changes in protein synthesis and/or mitosis, alterations that occur after exposure to (2) compounds that interfere with membrane receptor binding, (3) steroidogenesis, (4) compounds that interfere with the synthesis of other hormones and (5) compounds that alter the flux of ions across the membrane within certain types of hormone-secreting cells. Ca⁺⁺=calcium ion, SH=steroid hormone in blood or cytoplasm, PH=peptide hormone, R=steroid hormone receptor, SRE=steroid response element, PK=protein kinase.

Many pesticides have now been found to have estrogenic or anti-androgenic activity, and some bind to the androgen or estrogen receptors. Those which have been found to bind to the estrogen receptor include: ortho-phenylphenol 1, DDT and metabolites (although the anti-androgenic properties of p'p' DDE may be of greater importance); methoxychlor; chlordecone 2, dieldrin, endosulfan; 1-hydroxychlordecone (a metabolite of chlordane), and toxaphene ²¹. In recent years, there has been growing concern over the possibility of hormonal disruption at low levels of exposure. The traditional risk assessments of environmental contaminants are based on the extrapolation of high-dose data in experimental systems to estimate the effects at much lower doses in humans. However, the validity of such extrapolations is frequently challenged if one considers that

low levels of exposure. Some of these can induce estrogenic effects at relatively low levels. For example, administration of methoxychlor to the new born rat at a dose level of 0.5 µg per day caused accelerated puberty and accelerated loss of fertility. The dose levels at which effects are noted are fairly low ²².

Ahmed ²³ has reported extensive study into the effects of pesticides on immune system functioning; they noted that several pesticides which are commonly used in residential and agricultural areas may suppress normal human immune responses. It was found that endocrine disruptors are not merely affects the 'reproductive hormones', but also affect the functioning of several non reproductive tissues, notably the immune system. Pyrethroids can elicit a range of immunotoxic and neurotoxic effects in humans and other mammals, and their exposure may contribute to reproductive dysfunction, developmental impairment and cancer ²⁴. Organophosphates can interfere with the immune system and exert immunotoxic effects directly or indirectly. Immunotoxicities are varied and include pathology of immune organs, and decreased humoral and/or cell mediated immunity, decreased non-specific immunity, decreased host resistance, hypersensitivity and autoimmunity ²⁵. Permethrin and cypermethrin showed dose-dependent cytotoxicity. The most toxic pyrethroid was cypermethrin followed by permethrin and natural pyrethrin. This study confirms that the cell toxicity was dependent on the chemical structure and pyrethroids without an ?-cyano group shows the weakest physiological effect ²⁶. Treatment with cypermethrin caused significant decreases in ejaculate volume, sperm concentration, total sperm output, sperm motility and plasma testosterone ²⁷. Synthetic halogenated chemicals increase liver tumors after early life-stage exposure. Recently, a prototypical endocrine-disrupting compound, 2, 3, 7, 8 - tetrachlorodibenzo-p-dioxin (TCDD), has been shown to be a developmental toxicant of the mammary gland in rodents ²⁸. No significant changes were detected in nipple development and anogenital distance of female offspring at any dose level tested for EDC. In contrast to males, sexual differentiation of females is largely hormone-independent, yet still susceptible to hormonal disruption, e.g., masculinisation of the female fetus by exposure to androgens ²⁹.

DISRUPTION OF FEMALE HORMONAL FUNCTION

Study on endocrine disruption is an emerging field; researches are needed to give deep perception in endocrine

disruptors. Immature organisms are especially susceptible to endocrine disruption because the reproductive system is under development and relatively small changes in endogenous hormone levels may result in permanent structural and functional changes ³⁰. The development and the function of female reproductive tract depend upon hormone concentrations and balance. The endocrine disruptors modulate the hormonal function in the body and, in particular, affect the steroid hormones. Changes in effective concentrations of hormones can occur if an endocrine disruptor binds to a specific hormone receptor. This chemical substance may then either mimic the hormone or block the normal biological response by occupying the receptor site. Alternatively, endocrine disruptors may able to react directly or indirectly with the hormone structure to alter its function, modulate the number of hormone receptor, change the way of hormone synthesis and their affinities for specific molecules.

INTERFERENCE WITH HORMONE SYNTHESIS;

Some EDCs, such as fenarimol, prochloraz, and other imidazole fungicides possess the ability to inhibit estrogen biosynthesis through CYP19 aromatase inhibition in vitro, preventing the conversion of androgens to estrogens. Vinggaard et al., ³¹ hypothesized that compounds which can inhibit aromatase activity in vitro may be able to cause local changes in estrogen and androgen concentrations in vivo. Aromatase induction is a physiological mechanism to deactivate xenobiotics, which does not inevitably cause a toxic effect. Fenvalerate interrupted calcium homeostasis and FSH-stimulated P4 biosynthesis through calcium signal pathway which is able to block the FSH surge and ovulation in rats ³².

INTERFERENCE WITH HORMONE STORAGE AND RELEASE

Catecholamine hormones (e.g. norepinephrine) are stored in granular vesicles of chromomaffin cells within the adrenal medulla and within presynaptic terminals in the central nervous system. Therefore, they can be released quickly on demand. In contrast, steroid hormones are not stored intracellularly within secretory granules, but are readily synthesized after gonadotropin stimulation of the gonads. Norepinephrine is critical for the preovulatory increase in the pulsatile release of GnRH (gonadotropin releasing hormone) and the subsequent ovulatory surge of LH. Fenvalerate directly or indirectly, or secondarily alter Ca²⁺ homeostasis in ovarian cells, it may impair the reproductive endocrine functions. Furthermore, accumulating evidences indicate

calcium ions modulate gonadotropin-stimulated steroidogenesis by granulosa cells ³³.

INTERFERENCE WITH HORMONE TRANSPORT AND CLEARANCE

Steroid hormones in the bloodstream do not float around freely, but are bound to carrier proteins, such as SHBG (Sex-Hormone Binding Globulin) and albumin. Because only free hormones can be biologically active, increases or decreases in the concentration of SHBG will have a major impact on the available and active steroid hormone concentrations in blood. Estrogens are known to increase the synthesis of SHBG in the liver and thus increase the SHBG concentration in plasma, whereas androgens decrease these concentrations. Generally, clearance occurring in liver and the clearance rate is different for each hormone and is influenced by compounds that alter liver enzyme activity involved in hormone clearance. Many pesticides induce the liver enzymes monooxygenase and UDP-glucuronosyltransferase, resulting in increased clearance of the pesticide itself for detoxification purposes. Marty et al., ³⁴ demonstrated a weak response following exposure to 50 and 100 mg/kg body weight of phenobarbital, with a decrease in T₄ but no effect on TSH or thyroid histology. They recently showed that thyrotoxic effects can also be observed following exposure to toxicants which act via induction of liver enzymes and subsequent clearance of T₄ ³⁵. In this study, T₄ concentration was the most sensitive endpoint, with TSH and thyroid histology having lower sensitivity.

INTERFERENCE WITH HORMONE RECEPTOR AND BINDING

Hormones travel from their point of release in the bloodstream to particular tissues where they convey their messages. For the message to be interpreted, hormones bind to receptors. Hormone and receptor have a precise fit, so that only a specific type of hormone can bind to a specific receptor. A number of environmental agents may alter this process by mimicking the natural hormone (agonists) or by inhibiting receptor binding (antagonists). The latter mechanism is based on complete or partial blocking of the specific receptor. Regarding the estrogen receptor, this mechanism only applies when the endocrine disruptor concentration is high, because the affinity of endocrine disruptors for the estrogen receptor is usually many times lower than that of 17- β -estradiol. Three different mechanisms are involved usually during interference with hormone receptor by EDCs which includes (1) binding and activating the estrogen receptor; (2) binding without

activating the estrogen receptor; and (3) binding with other receptors ³⁶. For example pyrethroid compounds (permethrin, cypermethrin, fenvalerate and deltamethrin) at high concentrations inhibited the binding of estradiol to rat uterus cystolic estrogenic receptors (ERs) in a competitive binding study and suggested that signaling pathways other than ERs are involved in mediating pyrethroid induced or inhibited MCF-7 cell proliferation and pyrethroid insecticides may alter the conformation ER without binding to it.

INTERFERENCE WITH THE THYROID FUNCTION

Normal thyroid hormone blood levels are essential for growth and development of tissues and for the maintenance of tissue and organ function. Large numbers of environmental chemicals have been reported to affect production, transport, or metabolism of thyroid hormones, some acting by more than one mechanism ³⁷. Zaidi et al., ³⁸ found that TSH was elevated, total T₃ suppressed, and T₄ marginally decreased in pesticide formulators. Long term exposure to Ammonium perchlorate which is an inorganic ion used as a primary ingredient in rocket fuel, paints, fertilizers and lubricants which has recently been detected in drinking water supplies in the United States ³⁹, the thyroid axis is significantly altered as evidenced by decreased thyroxin in serum and a subsequent rise in TSH.

INTERFERENCE WITH THE CENTRAL NERVOUS SYSTEM

The central nervous system (CNS) is very important in the integration of hormonal and behavioral activity. Disturbances in these finely tuned mechanisms can severely impair normal adaptive behavior and reproduction. Since many pesticides are known to be neurotoxic, it is conceivable that these compounds can disrupt the coordinating activity of the CNS by disrupting brain cell functions. Also, pesticides can alter the hypothalamic and pituitary function and thus secretion of GnRH, LH, and FSH in a more direct manner by modifying the feedback of endogenous hormones. In relation to the early development of the female CNS and reproductive systems the more accepted theory is that which occurs in the absence of any significant gonadal hormone actions. However, female mice knockout of an aromatase gene (ArKO) (deficient in aromatase activity) and therefore unable to aromatize androgen to estrogen, suggesting that estradiol is required for the development of the neural mechanisms controlling this behavior in mice ⁴⁰.

POTENTIAL EFFECTS ON FEMALE HORMONE DISRUPTION

Large numbers of environmental chemicals have been reported to affect production, transport, or metabolism of hormones, some acting by more than one mechanism. Endocrine disruption may result in disturbances in the reproductive system, such as modulation of hormone concentrations, ovarian cycle irregularities, and impaired fertility. The organochlorine pesticide hexachlorobenzene (HCB) is a worldwide persistent organic pollutant and has been detected in various tissues and human fluids including serum and ovarian follicular fluid. Any environmental compound mimicking or antagonizing steroid hormone action could presumably alter the glycosylation of LH and FSH, thereby reducing their biological activity⁴¹. Endocrine disruptors with estrogenic properties may be able to block ovulation similar to contraceptive pills. Human fertility is a delicate process that can be influenced by many factors, such as hormonal imbalance caused by pesticides. However, in most studies it is not clear whether impaired fertility is due to hormonal imbalance or to other toxic effects. The pubertal female rat assay is designed to detect alterations in thyroid hormone status, hypothalamic–pituitary–gonadal (HPG) function, inhibition of steroidogenesis ERs and antiestrogens

⁴² ·

REPRODUCTION AND NEUROBEHAVIORAL EFFECTS

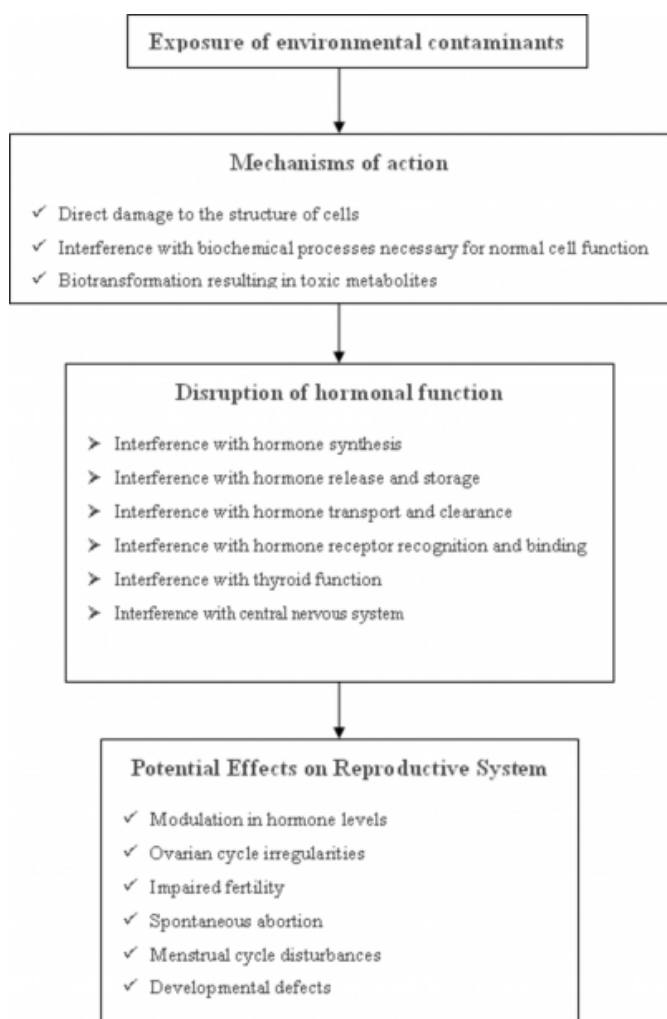
Endocrine-disrupting compounds are among the most complex environmental health threats known today. Reproductive toxicity begins with parental exposure to toxicants. Preconception, conception, prenatal, and postnatal periods are all windows of opportunity for adverse reproductive outcomes. By mimicking natural hormones such as estrogen and testosterone, these chemicals can interact with the body's endocrine system and exert toxic effects that may lead to reproductive and developmental abnormalities or cancer. The reproductive hazards of a chemical can affect men, women, the fetus and postnatal development. Moniz et al.,⁴³ reported that there was a delay in vaginal opening, suggested that perinatal fenvalerate exposure may induce alterations on sexual maturation in females and the only external signal of puberty is canalization of the vagina, which is initially imperforated, and later becomes patent as a consequence of estrogenic stimulation. Transgenerational effects on reproductive capacity may be a common feature of endocrine-disrupting chemicals. Pesticides may cause hormonal imbalance and

reproductive toxicity, especially in female through several different mechanisms: direct damage to the structure of cells, interference with biochemical processes necessary for normal cell function, and biotransformation resulting in toxic metabolites (Fig. 3). Anway et al.,⁴⁴ reported very recently that pregnant rats treated with the estrogenic pesticide methoxychlor or the anti-androgenic fungicide vinclozolin have subfertile offspring, and that the male offspring pass the defect through the male germ line for at least four generations. They also reported that there is an altered pattern of DNA methylation in the male germ line. Developmental treatment with Diethylstilbestrol (DES) caused an alteration in gene imprinting as demonstrated by the penetrance of a tumor suppressor gene in the uterus. Thus, in several model systems gene imprinting by estrogen is associated with later uterine disease. The heritability of these changes can be seen by the transmission of disease to the next generation. Mice exposed to DES early in development, when mated to control males, produced offspring that also had increased risk for vaginal adenocarcinomas. Thus, the 'granddaughters' of the treated mice expressed the same rare cancer as the daughters. The implications for transmission of epigenetic traits are obvious, as is the possibility for transgenerational transmission of the DES effect in humans⁴⁵. Since 1997 a large number of peer reviewed journal articles has been showing that bisphenol-A harm in animals at levels to which the average human is exposed. Bisphenol A is another chemical that similar to methoxychlor, has the ability to bind estrogen receptors and initiate cellular responses similar to those caused by estradiol. Recent findings include chromosomal damage in developing oocytes in mouse ovaries and abnormalities in the entire reproductive system in male mice, including a decrease in testicular sperm production and a decrease in fertility. In addition fetal exposure to bisphenol A increases the rate of postnatal growth and decreases the age at which females mature sexually. These females also have mammary gland abnormalities and appear pre-cancerous by the time the female reaches young adulthood. Bisphenol A also causes abnormal brain development, and change in brain function and behavior, similar to methoxychlor⁴⁶. Ohi et al.,⁴⁷ studied the adverse effects of Fipronil (phenylpyrazole) on reproductive function in Wistar rats and found that exposure lengthened the estrous cycle, decreased plasma progesterone and estradiol levels, and reduced the pregnancy index. Timing of exposure, duration, dose and susceptibility, or genotype of parent and fetus or child can play a role in the

outcome observed. For each adverse reproductive effect, there appears to be a critical window of exposure. These concepts in reproductive and developmental human toxicity were discussed in detail in a series of workshops conducted by USEPA held in June 2000 ^{48,49}.

Figure 3

Figure 3: Potential effects of environmental contaminants on female reproduction



Neurodevelopmental abnormalities are exceedingly difficult to monitor, yet the evidence suggests that further investigation of time trends and causes is urgently needed. Chemical interference with hormone actions, particularly during neurodevelopment, and consequent effects on cognitive behavior are a current concern. Approximately 27% (696 of 2,588) of the workers who sprayed pure pyrethroids reported having experienced symptoms such as abnormal facial sensations (paresthesia), dizziness, headache, nausea, loss of appetite, blurred vision, and tightness of the chest. Eight of these workers were diagnosed with mild acute pyrethroid poisoning, characterized in part

by listlessness and muscular fasciculations. GABA receptors are the target sites for several insecticides; including chlorinated cyclodienes (e.g. endosulfan) and phenylpyrazoles (e.g. fipronil). Parmigiani et al., ⁵⁰ administered the widely used pesticide methoxychlor to pregnant mice and the offsprings were examined for neurochemical changes in the dopaminergic system in the basal ganglia area of the brain. Neurons that use dopamine as a neurotransmitter (the dopaminergic neural system) are involved in the control of locomotor activity and exploration. The basal ganglia were studied because one of the major impacts of methoxychlor on behavior is to increase exploratory activity to novel stimuli. This is also the area of the brain where degeneration occurs in Parkinson's disease, as well as associated changes in behavior. A change in behavior was associated, particularly in females, with a decrease in dopamine receptors in the basal ganglia. Males exposed to methoxychlor also showed an increase in territorial behavior, which is associated with very low levels of exposure to this pesticide. Low dose effects on the rat female reproductive system following exposure to a single administration of PBDE (Polybrominated diphenyl ethers) used as a flame retardants, observed a decrease in serum thyroxine (free) and serum estradiol concentrations in offsprings causes some neurobehavioural alterations such as nerve excitability and reproduction abnormalities ⁵¹. Nerve excitability was assessed by presenting two sequential electrical stimuli of equal intensity and duration to the median nerve area of the wrist and recording the median nerve activity at the lateral side of the elbow. Lazarini et al., ⁵² also reported during the administration of deltamethrin 0.08 mg/kg of body weight, to a Wistar rat, persistent changes in behavior and/or biochemistry, including learning, motor activity, sexual behavior and they observed more learning behavior in males, zero effect on locomotion frequency in males or females. There is a relationship between exposure to certain neurotoxic pesticides such as pyrethroids during pregnancy and subsequent problems in children. Such a connection is suspected between learning and developmental disorders in children ⁵³. Certain pesticides may also interrupt the neurological development process particularly during critical period and induce harmful effects on sensory, motor and cognitive functions for example Aziz et al., ⁵⁴ examined both the behavioral and biochemical changes by the administration of deltamethrin 1.0 mg/kg to the Druckrey rat. The effects were delayed surface righting reflex, during 6 and 12 weeks postnatal; increased acetylcholine esterase activity; increased GAP-43,

regarding immunohistochemistry both % area and total number of positive cells were decreased. Experimental studies have shown that low-dose neonatal exposure to pesticides, including those in the organophosphorus category, may cause irreversible changes in the cerebral functions of adult animals ⁵⁵. Recent data concerning the effects of organophosphorus insecticides on the early development of the nervous system have prompted Health Canada and the United States Environmental Protection Agency (USEPA) to re-evaluate the toxicity of these products. It was on the basis of these new evaluations that severe usage restrictions were recently imposed for chlorpyrifos, an insecticide widely used in residential landscape maintenance.

In perhaps the most thought provoking recent work, Winrow et al. ⁵⁶ demonstrated that inhibition of neuropathic target esterase (NTE) activity, either genetically or by OPs that inhibit NTE, results in a neurological hyperactivity phenotype in mammals (mice). Possible linkage of attention deficit/hyperactivity disorders in children to pesticide exposure in early childhood was hypothesized by these authors. This limited review of neurodevelopmental toxicants and their possible effects in children clearly indicate the need to conduct toxicogenomics studies together with neurobehavioral assessments of children and their exposures to pesticides.

HEALTH POLICY ACTIVITIES

Recent activities (particularly by the USEPA and OECD) have been aimed at defining tiered testing strategies, validating screening and short-term tests and evaluating the feasibility of modifying existing test guidelines to encompass the detection of endocrine disrupting compounds. Finally, it is essential that a rigorous evaluation of assay is carried out prior to incorporation into a testing strategy and it is important to define how the information provided by these can be used in hazard identification and possibly for risk assessments ⁵⁷. At present, little effort is being made by public authorities to inform and sensitize the public about the rational use of pesticides. Accordingly, any major changes in the framework currently governing pesticide use should be accompanied by a comprehensive awareness campaign on health and environmental risks, so that the public fully understands the reason behind the changes. Furthermore, annual awareness campaigns should be developed by the relevant government departments. Regulatory agencies should recognize that the current endpoint of most tests to assess the risk of pesticides and

other pollutants (carcinogenicity, acute toxicity, and immediate mutagenicity) have led to the misconception that these chemicals do not pose a threat to the health of wildlife, domestic animals, or humans. Low-level exposures to endocrine-disrupting compounds are ubiquitous in today's environment. Persistent chemicals such as DDT, PCBs and dioxins are detectable in nearly 100% of human blood samples, and even some of the shorter-lived potential endocrine disruptors are frequently detected in general population survey of residues in blood or urine ⁵⁸. The ubiquitous nature of the exposures combined with the nontrivial potential health effects justifies further research, education and preventive action to reduce human exposures to endocrine disruptors. A great deal of work on endocrine disruptors is taken by some of the international organizations are illustrated in Table 1.

Figure 4

Table 1: Organizations and some policy activities concerning endocrine disruptors

Activity	Lead organizations	Web site
Screening and testing chemicals for endocrine effects	USEPA (United States Environmental Protection Agency)	www.epa.gov/scipoly/ospendo/index.htm
	Organization for Economic Cooperation and Development, Europe	www.oecd.org/ehs/endocrin.htm
Prevention of pesticides and toxic substances	USEPA (United States Environmental Protection Agency), EDSTAC (Endocrine Disrupting Screening and Testing Advisory Committee), Final Report - 2006	www.epa.gov/scipoly/ospendo/history/finalrpt.htm
	Environment Canada	www.ec.gc.ca/eds/fact/index.htm
Research on mechanisms, wildlife effects and human epidemiology	USEPA (United States Environmental Protection Agency)	www.epa.gov/endocrine
Endocrine Disruptors Research Invention	Organization for Economic Cooperation and Development, Europe	www.oecd.org/ehs/est/endodrp.pds
Education and advocacy for action on endocrine disruptors	World Wild life Fund for Nature, Canada	www.wwfcanada.org/hormone-disruptors/index.html
	Canadian Association of Physicians for the Environment	www.capec.ca
Endocrine Disruptor Information Guide for Researchers & Global Information Network on Chemicals	National Institute of Health, Japan	www.nihs.go.jp/hsr/endocrine-e/index.html www.nihs.go.jp/GINC/index.html

The US Food Quality Protection Act (FQPA) of 1996 was dramatically altered the regulation of pesticides and other environmental pollutants in the United States. This Act, which amended both the Federal Insecticide, Fungicide and Rodenticide Act and the Federal Food Drug and Cosmetics Act, requires the USEPA to consider a number of factors relative to the toxicology of pesticides, including the cumulative effects of exposure to pesticides having a 'common mechanism of toxicity'. The methods for identifying pesticides having a common mechanism of

toxicity, and the ramifications of such a determination, are only now beginning to be clarified. Finally, implementation of the US FQPA (1996) by USEPA is expected to lead to reduction of the overall level of pesticide residuals in food. Included in this effort is review of tolerances of pesticide levels in foods and efforts directed at increased protections for infants and children through additional acute, chronic, and neurotoxicity testing⁵⁹. It is also essential to continue for examining the transgenerational effects in animal studies because some pollutants require metabolism *in vivo* to exert hormonal effects and other developmental effects. The OECD endocrine disruptor activity was initiated in 1996. The focus of the activity was to provide information on national and regional activities concerning endocrine disruptors, develop appropriate test guidelines and harmonize risk assessment approaches. The quality of the scientific data used to support these and other risk decisions is an important component of scientifically based risk assessment⁶⁰. In response to the 1996 legislative mandate for an endocrine screening and testing program, the USEPA formed Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC), which proposed a tiered screening and testing strategy for EDCs in its final report (1998). The EDSTAC proposal includes: (1) a process to prioritize chemicals for evaluation and tiered; (2) screening (Tier 1) and (3) testing (Tier 2) batteries. Under the legislation, some 80,000 existing chemicals and new chemicals were undergoing various screens and/or tests for their potential estrogenicity, as well as other hormonal activities. The integrated computational approach was reported to set priorities for these chemicals for experimental screening and testing. Prioritization would include an estimation of the chemicals ability to interact with steroid hormone receptors using either quantitative structure activity modeling (QSAR) for chemicals that bind steroid receptors or high-throughput prescreening (HTPS) using hormone-dependent gene expression assays, among several other factors⁶¹.

CONCLUSION

The potential effects of endocrine-disrupting compounds (EDCs) on human health and the proven effects of EDCs on wildlife are a major concern among the public and the scientific communities. Ongoing efforts around the world to develop accurate biological monitoring of blood and urine for numerous chemical toxicants will improve exposure assessment in epidemiological studies and may eventually provide tools for physicians to assess risk to individuals⁶². As these tests become standardized and widely available,

they will be useful clinically, just as blood lead testing has helped to facilitate a range of interventions that have resulted in a major reduction in lead poisoning.

Continue the Endocrine Disruptor Screening Program validation work, but combine this with a continuing assessment of the quality and efficiency of the screening assays being validated, as well as continuing improvement of the screening battery by adoption of better, more predictive assessment technology. Improve the Endocrine Disruptor Screening Program's prioritization process by developing a process that considers the potential to cause biological effects, rather than just exposure potential. Developing high-throughput pre screening (HTPS) assays to determine the cumulative risk assessment of compounds having endocrine disrupting activity. Developing new assessment and biomarkers to determine exposure and toxicity levels, especially how mixture of chemicals have impact on individuals or populations.

Different *in vivo* and *in vitro* assay systems are currently used by regulatory authorities for the evaluation of reproductive and developmental toxicity. There is a need to reassess adjuvant used in pesticide formulations for their endocrine activity. Compounds showing effects on the endocrine system or reproduction, such as alkylphenolic surfactants, should be phased out.

Transgenerational exposure, hormonal activity, functionality, and delayed expression of effects must be addressed when determining the hazards of exposure to persistent chemicals already in the environment and of new compounds that might be released in the future. Mechanisms to inform the public about health risks and pesticide alternatives are also needed. Finally, knowledge development is another important goal, so that the risks and health effects of pesticides can be measured more accurately.

The public is routinely informed that the EDCs have been tested, that there are studies demonstrating the absence of their risk, and that regulatory agency adequately protect public health. Clearly significant changes are needed to bring current regulatory practices into conformity with new scientific information. We propose that testing for health effects at doses within the range of human exposure (currently not done) with respect to long-latency effects of developmental exposure throughout the lifespan (currently not done) be required prior to the introduction of any compound intended for use in commerce. Much of the research data reviewed here is necessarily descriptive. With

the rise of genomics and toxicogenomics and the introduction of sensitive immunoassay and analytic methods capable of being used to measure toxicants levels in humans, mechanism-based human studies are at the horizon. With these tools, population toxicologists can assess differences and similarities between childrens and adults response to pesticide exposure.

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