

Towards Less Confusing Terminology in Endocrine Disrupter Research

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Abstract

Recognition that environmental contaminants can interact with hormone receptors and mimic or antagonize the actions of endogenous hormones has led to introduction of the terms *endocrine disruptor*, *endocrine disrupter*, *hormonally active chemicals*, and *hormone mimics* into the scientific and lay press. Reports suggesting a link between exposure to endocrine toxic chemicals and increasing rates of hormone dependent cancers (breast and prostate), developmental effects in the male reproductive tract, falling sperm counts, and endometriosis has resulted in an explosion of research, regulatory actions and policy changes aimed at better understanding the hazards posed by these chemicals and restricting their use. With increasing concern there has been worldwide action to develop testing strategies to allow for early identification of chemicals with endocrine disrupter activity. However, despite an expanding literature and numerous expert panel meetings there continues to be controversy surrounding how to best define endocrine disrupters resulting in ambiguous use of the term and confusion in the literature along with the publication of contentious lists of chemicals with purported endocrine toxic activity. Herein we argue in favor of a more restrictive definition, adoption of a less ambiguous term, and development of a classification system to enhance more effective communication and facilitate appropriate allocation of limited resources in this highly charged area of toxicology.

Introduction

A number of naturally occurring and synthetic chemicals have been shown to exert adverse effects upon the endocrine system across animal classes including mammals (WHO 2002). Within the last decade the field of chemical disruption of the endocrine system has been an active area of research that has captivated the scientific world, and captured the attention of governments, policy makers as well as the media.

In 1996, endocrine disruption was identified as one of the six high-priority research topics within the United States Environmental Protection Agency (Kavlock, 1999). In the same year, the USEPA designated a special task force, the Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC) that was assigned the task of making recommendations for the development of testing and screening programs for endocrine disruptors (EDSTAC, 1998). Likewise, the Organization for Economic Co-operation and Development (OECD) also established a special activity for endocrine disrupter testing and assessment (OECD, 1996). Subsequently, the World Health Organization tasked the International Programme on Chemical Safety with preparing a report describing the Global Assessment of the Scientific literature on endocrine disrupting chemicals (WHO 2002). Indeed, recognition that chemicals in the environment poses the ability to interact with hormone receptors and mimic their activity has been regarded as one of the top five most significant developments in endocrinology of the last century (Schwartz, 2002). An electronic data base (PubMed) search using the search terms endocrine disrupter, endocrine disruptor, endocrine disruption, hormonally active chemicals, endocrine modulation, endocrine mediated toxicity, and hormone mimics yielded 1075 relevant citations. Despite this high level of research activity a globally accepted definition of endocrine

disrupters has yet to be advanced thus leading to miscommunication. Furthermore, the literature has become muddled with chemicals purported to be endocrine disrupters without any reasonable criteria to define the chemicals as such. An acceptable definition and consistent terminology are therefore needed to focus attention on priority chemicals and issues and thus insure the efficient use of limited resources in this high priority area.

The endocrine system functions to maintain the body in a homeostatic state that is optimal for the biochemical reactions necessary to support life. Within the last decade various terms have been used to identify exogenous chemicals that alter the function(s) of the endocrine system. Some of these include: *endocrine disrupters*, *endocrine disruptors*, *endocrine modulators*, *hormone mimics*, and *hormonally active agents* (EDSTAC, 1998). As there are many terms in the literature, there is also substantial disagreement regarding the definition of endocrine disrupting chemicals. We propose that the most commonly used definition of an endocrine disruptor, provided by Kavlock et al., (1996), although widely accepted, is too inclusive and virtually includes all chemicals thus creating ambiguity in both the scientific as well as the lay community. We suggest that a functional definition is essential from which useable criteria to categorize agents as endocrine toxicants can be developed. Development and implementation of a classification system will enhance accurate communication across disciplines of the effects of chemical agents on an organism. Less confusing terminology and improved communication is essential to evidence based decisions and the allocation of limited resources necessary to either restrict or eliminate the use of potentially hazardous chemicals. Therefore, we propose an alternative term for endocrine disrupters, one that takes into account their toxicity and suggest a more restrictive definition. In addition, a classification system is proposed as a potential means

of achieving agreement, advancing discussion, and better communicating what is and what is not known about a chemicals endocrine toxicity.

Defining Endocrine Disrupters

A functional definition of endocrine disrupters should provide a frame work consisting of a set of statements, conditions and attributes under which a class of substances can fall. An ideal definition would neither be too restrictive or overly inclusive; one that avoids being excessively broad or too narrow. Within the literature the most frequently referred to definition of an endocrine disrupter is that of “*an exogenous agent that interferes with the production, release, transport, metabolism, binding, action or elimination of natural hormones in the body responsible for the maintenance of homeostasis and the regulation of developmental processes*” (Kavlock et al, 1996). Similarly the Natural Resources Defense Council (NRDC) defines an endocrine disrupter as “*synthetic chemicals that when absorbed into the body either mimic or block hormones and disrupts the body’s normal functions through altering normal hormone levels, halting or stimulating the production of hormones, or changing the way hormones travel through the body*” (NRDC, 1998). Although widely accepted, these definitions are ambiguous and overly inclusive. We propose that these definitions are overly inclusive due to an inability to discriminate between *alteration* and *adverse effect* on the endocrine system. Since the endocrine system functions to maintain the body’s homeostasis in a constantly changing environment and thus responds readily to a vast array of signals it is hard to imagine any chemical or even physical agent that would be excluded by the above definitions. The Kavlock definition fails to discriminate between chemicals that would adversely affect an organism’s endocrine system and any innocuous exogenous agent that serves to disrupt the physiological balance of the body

either by direct interaction with hormone receptors or indirectly through changes induced in other organ systems. Under such a definition, one could easily classify such innocuous things as a change in room temperature, consumption of a meal, and day light as endocrine disrupters as each is known to induce changes in circulating thyroid hormone (van der Sluijs Veer et al., 1992), insulin (Sheehan, 2004), and melatonin (Stevens et al., 2001) levels, respectively. Indeed, it is readily appreciated that stressful situations alter endocrine status as shown by changes in circulating adrenal steroid levels (McEwen et al., 1995). Moreover, there are many chemicals that with exposure induce changes in circulating hormone levels that are unrelated to any direct toxicity to the endocrine system but are rather the consequence of systemic toxicity. For example, dibromochloropropane exposure has been associated with infertility, complete loss of all sperm in the ejaculate or lowered sperm counts (Meistrich et al., 2003). However, this compound does not act as an endocrine toxicant other than it is directly toxic to spermatogenic cells. Ideally, endocrine altering agents should include *specific chemicals* that disturb an organism's endocrine system beyond the range that would maintain normal physiological function. There have been improvements in presenting a more restrictive definition than the ones previously mentioned. The World Health Organization (WHO) identifies an endocrine disrupters as "*an exogenous substance or mixture that alters function(s) of the endocrine system and consequently causes adverse health effects in an intact organism, or its progeny, or (sub)populations*" (WHO, 2002). EDSTAC, Canada Centre for Occupational Health and Safety (CCOHS), IUPAC, and International Programme for Chemical Safety (IPCS) all agree with the aforementioned definition. We propose that the above definition is a more appropriate definition that readily accounts for the fact that many alterations of the endocrine system are not necessarily deleterious, and fall within a normal range that poses no hazard to the organism.

Inclusion of the adverse effect criterion has been criticized because it is thought to fail to account for some unique features of endocrine toxicity. Specifically, exposure to endocrine toxicants can induce small disturbances in endocrine function, especially during certain developmental stages (Pryor et al., 2000), that have profound effects later in life. For example, *in utero* exposure to estrogenic compounds has been shown to alter prostatic weight in rodents (vom Saal et al., 2002). Similarly, although unusual dose response curves have been reported for several chemicals (Welshons et al., 2003; Palanza et al., 2002) and selected endpoints it is not known how widespread these effects are or what the biological significance of these effects may be. A potentially more serious consideration is a chemical-induced change in the homeostatic set point that may render the individual more sensitive to a future chemical insult and induce an adverse effect that might otherwise have been tolerated. However, this hypothesis has yet to be tested experimentally and demonstrated to be a serious issue requiring attention in the risk assessment process. Rather than establish these chemicals as endocrine toxicants these effects focus attention on the data gap and the need for further research. Regardless, we propose that each of these issues can be easily resolved even with inclusion of the adverse effect criterion in the definition. Nonetheless there continues to be significant disagreement within the scientific community regarding the inclusion of *adverse health effects* as a criteria within the definition. Such controversy has been difficult to resolve due to the fact that the definition provided is not suitable for the physiological effects it attempts to define, and would be more fitting if used in association with the more appropriate term of *endocrine toxicant*.

The utilization of the term *endocrine toxicants* couples the affected system (endocrine) with an adverse health effect in order to discriminate between endocrine toxicants (established risk) and hazardous chemicals (hazard demonstrated but risk unknown). Fundamentally, the term would prove successful in distinguishing between endocrine disruption risk (alteration of hormone levels beyond natural bounds) and endocrine modulation (adjustment within the natural bounds). Prior to this point, the literature suggests that both terms could be used interchangeably (Tattersfield & al., 1997) in disregard to the blatant ambiguity such loose usage of these seemingly different terms would create. The common utilization of the term *endocrine disrupter* by the media, regulatory groups, academic scientists, and non-governmental organization is emotive and saddled with regulatory intentions. In addition, it is also ineffective in communicating divergent meanings. In effect, such a term has lead to confusion regarding the potential for chemicals to interact with physiological systems and to induce changes in endocrine homeostasis vs. induction of adverse health effects. The term endocrine toxicant clearly outlines a substance's endocrine toxicity, as a chemical exhibiting detrimental effect through an endocrine mechanism. Some of the difficulty with the definition and term endocrine disrupter arises from the classification of chemicals as endocrine disrupters in the absence of a clear set of criteria. Therefore, we propose a hierarchical classification system to rationally categorize chemicals on the basis of the evidence for the ability of a chemical to change endocrine homeostasis in humans as well as wildlife and fish, and endocrine function as modeled by *in vitro* systems.

Proposed classification System

We propose that a suitable method of categorizing endocrine toxicants would be based on a classification system similar to that used by IARC to categorize chemical carcinogens. In our proposed system chemicals would be classified within one of three classes based on the available published data. Herein it is proposed that chemicals can be classified into different categories depending on their effects on the endocrine system. Class I toxicants would include those chemicals for which there is evidence of adverse effects in human populations. For example, 2,3,7,8 tetrachlorodibenzo-*p*-dioxin (TCDD) would be classified as a Class I endocrine toxicant due the reported adverse effects that have resulted from human exposure. Accidental exposure in the Seveso, Italy chemical spill led to detrimental effects on metabolism, cytogenesis and immunologic function (Baccarelli et al., 2003). In addition, occupational exposure by Vietnam veterans has been associated with an imbalance in thyroid hormone and TSH levels (Pavuk et al., 2003). Additional chemicals that would fit in this category include but are not limited to: dioxin like polychlorinated biphenyls (PCBs) and *p,p'*-dichlorochlorophenylethylene (*p,p'*-DDE). For further examples and more information, refer to Table 1. In this proposal Class II endocrine toxicants would consist of chemicals for which there exists experimental animal data or evidence from wildlife and fish studies indicating that the chemical has the potential to induce adverse effects on the endocrine system, but there is little evidence of exposure in humans, or evidence of adverse effects in humans is inconsistent. For example, perchlorate would be identified as a Class II endocrine toxicant due to its potential to induce hypothyroidism by hindering iodine uptake in animal models (Clewell et al., 2003). However, its effects on humans even at levels up to 36 times those of normal exposure have been physiologically insignificant (Greer et al., 2002). Other chemicals that we suggest are included in this category include but are not limited to: methoxychlor, mirex, bisphenol-A, and perfluorooctane sulfonate (PFOS), and di-butyl

phthalate. Finally, Class III endocrine toxicants would be agents for which there exists *in vitro* data but no experimental animal data of adverse effects on endocrine homeostasis. For example, the metal cadmium has been shown to poses estrogenic activity in a culture system (Stoica et al., 2000) but evidence of estrogenic effects in whole animals is lacking. The use of such a classification system would provide specificity as well as applicability by ensuring that each chemical compound is categorized according to the available data, and its relevance to the human population. This proposed system would effectively decrease ambiguity and amalgamate the scientific, regulatory and public sectors regarding the topic of endocrine disruption.

Summary and Conclusions

While the area of endocrine disruption has achieved substantial recognition in the scientific and lay communities, the term *endocrine disrupter* and its definition have entailed confusion and ambiguity in communication between scientists, policy makers, the government and representatives of the media. The commonly referred to Kavlock definition of an endocrine disruptor, while achieving the broadest level of acceptance, it is too inclusive and fails to take into consideration the hazardous effects necessary for the classification of such chemicals. We propose that the definition advanced by the WHO and several other organizations including EDSTAC, CCOHS, IUPAC and IPCS which have adopted the concept of adverse effects, and thus is a much more suitable definition for the term *endocrine toxicants*. The implementation of this new term in replacement of endocrine disrupter would serve to discriminate between substances that are detrimental to endocrine homeostasis and those that only modulate endocrine expression within a non-hazardous normal range. Finally, use of a classification system that encompasses human exposure together with experimental animal data and *in vitro* studies to

categorize chemicals based on their potency establishes a groundwork that discriminates between chemicals that are *hazards* (potentially having an effect on endocrine mechanisms), and those that are *risks* (exhibit adverse effects on the human endocrine system due to sufficiently high exposure).

In the absence of a globally accepted definition the terminology and communication of the endocrine toxic effects of environmental contaminants will remain confusing and potentially misleading. To advance the study of endocrine toxicants and enhance existing research and regulatory efforts it will be necessary to build consensus amongst scientists from widely divergent backgrounds and thus assumptions as well as policy, regulatory, and non-governmental organizations. Therefore, the proposal to include adverse effects in the definition and adopt a classification system for endocrine toxicants is advanced as a potential solution to the impasse that currently exists within the scientific, government, and lay communities.

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Table I. Proposed classification scheme for endocrine toxic chemicals.

CLASS	COMPOUND	RATIONALE
Class I (evidence of human exposure, and documented adverse effects in human populations)	TCDD	<ul style="list-style-type: none"> • Human exposure incident in Seveso Italy reported long-term effects of : spontaneous abortion, cytogenetic abnormalities, congenital malformation, impaired liver function and lipid metabolism, as well as neurologic and immunologic impairment (Baccarelli et al., 2003) • Human exposure study of Ranch Hand among Vietnam veterans indicated adverse effects on thyroid hormone metabolism and function resulting from increased levels of thyroid stimulating hormone (Pavuk et al., 2003)
	PCB	<ul style="list-style-type: none"> • Endocrine effects include: depletion of thyroid hormone levels leading to hypothyroidism and neurodevelopment disorders associated with hypothyroidism (U.S. Department of Health and Human Services, 2000) • Human exposure incidents include consumption of fish from the Great Lakes and contaminated rice in Yu Shu, Japan and Yu Chang, Taiwan (Aoki, 2001) • Human health effects included irregular menstrual cycles and altered immune responses, as well as increased levels of blood thyroxine and triiodothyronine (Aoki, 2001)
	<i>p,p'</i> -DDE	<ul style="list-style-type: none"> • Animal studies have reported and increased incidence of liver tumors in mice and hamsters, and thyroid tumors in female rats form oral exposure to DDE (USEPA, 2003) • <i>p,p'</i> has been shown to enhance aromatase enzyme activity (at normal levels in human follicular fluid) leading to enhance basal and FSH-stimulated granulosa cell aromatizing enzyme activity (Younglai et al., 2004) • DDE has been demonstrated to decrease duration of lactation in women of the United States and Mexico (Gladen et al., 1995) • Controversy exists regarding <i>p,p'</i> DDE concentration and potential adverse effects of sperm quality and motility (Hauser et al., 2002) • Investigation of human effects showed that <i>p,p'</i> DDE did not effect serum hormone levels such as LH, FSH and SHBG (Cocco et al., 2004)
Class II (animal data indicating that the chemical has the potential to induce	Methoxychlor	<ul style="list-style-type: none"> • Posses estrogenic properties that has lead to effects on the ovaries, uterus and mating cycle in females, and the testes and prostate in males (USEPA, 2002) • Facilitates the onset of puberty in female rats, and has been shown to induce irregular estrous cyclicity and histopathological alterations in the reproductive tract and anterior pituitary,

<p>adverse effects on the endocrine system but no evidence of adverse effects in humans)</p>		<p>also shown to delay the onset of puberty in male rats (Masutomi et al., 2003)</p> <ul style="list-style-type: none"> • Exposure of male rats during testes development affects embryonic testis cellular composition, germ cell numbers, and germ cell survival and appears to be a reduced spermatogenic capacity (Cupps et al., 2003) • Evidence of thymic atrophy in rat pups (Takeuchi, 2002)
	<p>Mirex</p>	<ul style="list-style-type: none"> • Eating Mirex can have adverse effects on the thyroid and on reproduction in animal models (U.S. Department of Health and Human Services, 1995) • Has been shown to be responsible for tumor promotion in female mice through the ovarian hormone 17-beta estradiol (Porter et al., 2002)
	<p>Perchlorate</p>	<ul style="list-style-type: none"> • Perchlorates affect/hinder the uptake of iodine and therefore could potentially lead to hypothyroidism (Clewell et al., 2003) • Amount of thyroidal iodine inhibited at levels of 9 and 36 times normal exposure were physiologically insignificant (Greer et al., 2002)
	<p>PFOS</p>	<ul style="list-style-type: none"> • Rodent studies demonstrate that PFOS can have detrimental effects on development possibly affecting thyroid function (Inoue et al., 2004) • Exposure lead to decrease n the serum levels of thyroxine (T4) and triiodothyronin (T3) in treated rats dams (Thibodeaux et al., 2003)
	<p>DDT</p>	<ul style="list-style-type: none"> • Large dose intoxication leads to affects on the nervous system resulting in tremors and shivers, as well as on reproduction (U.S. Department of Health and Human Services, 2002) • Human research has indicated adverse affects on DNA as well as apoptosis of human peripheral blood mononuclear cells (Yanez et al., 2004 and Ivan et al., 2004) • Animal research has demonstrated that <i>p,p'</i> DDT induces apoptosis of thymocytes and a dose-dependent thymic atrophy in rats (Teoburbi et al., 1998) • Cumulative exposure does not appear to have effects on human serum hormone levels (Cocco et al., 2004)
<p>Class III Evidence of adverse effects in animal models or <i>in vitro</i> but human exposure is too low or not expected to occur</p>	<p>BPA</p>	<ul style="list-style-type: none"> • Exposure of male rats to environmentally low levels lead to a reduction in daily sperm production 5 weeks later (Sakaue et al., 2001) • BPA proved to be equally as potent as 17-beta estradiol in activating the transcription factor CREB (Quesada et al., 2002) • Debate exists on BPA's effects on the development of the male reproductive tract in animal models (Safe et al., 2001)

	Nonyl Phenol	<ul style="list-style-type: none"> • Has been shown to cause extensive proliferation of lobular development and mammary glands in rats as a weakly estrogenic substance (Odum et al., 1999) • Studies on male rats demonstrated adverse effects on sexual development: reduction of testicular spermatid count, decreased size of testes, epididymis, seminal vesicle and ventral prostate (Chapin et al., 1999 and Lee, 1998)
	Di-butyl Phthalate	<ul style="list-style-type: none"> • Controversy exists regarding the effects of this compound on sexual development: Sharpe et al. (1995) reported a reduction in testis size, whereas other studies have refuted these findings (Ashby et al., 1997) • Accordingly, di-butyl phthalate has been reported to possibly affect sperm motility (Wang et al., 2004) • Exposure at high levels has shown to cause “rapid and reversible diminution of the expression of several proteins required for cholesterol transport and steroidogenesis in the fetal testis, resulting in decreased testosterone synthesis and consequent male reproductive maldevelopment” (Thompson et al., 2004)