Endocrine Disrupting Chemicals: Overview of recent published literature

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Introduction

This report is an overview of key recent publications concerning the health and environmental effects of Endocrine Disrupting Chemicals (EDCs). The term EDCs is normally applied to describe chemicals that are synthetic agents that mimic or block hormones resulting in the potential disruption of critical endocrine processes including reproduction, growth and development. The review is based on the peer-reviewed papers, abstracts and conference proceedings that have been published in the period January – March 2010.

The papers were selected because they address research areas that are considered of direct relevance to the health and environmental effects of EDCs. The papers are presented under six subject areas that are outlined below. When considered appropriate, some papers appear under more than one of the subject areas.

Section 1 – HUMAN EXPOSURE MEASUREMENT AND MODELLING: Papers relating to the measurement or modelling of consumer and/or environmental exposure to EDCs and/or mixtures of EDCs; the development of human biomarkers of exposure to or effect of EDCs and/or mixtures of EDCs.

Section 2 - HEALTH EFFECTS: Papers on the influence of EDCs and/or mixtures of EDCs on health, disease and dysfunction; assessment of the influence of genetic and epigenetic factors on human susceptibility to the effects of EDCs and/or mixtures of EDCs:
   a.) Reproductive/developmental health
   b.) Carcinogenic effects
   c.) Other effects

Section 3 – BIOLOGICAL MECHANISMS and TOXICITY TESTING: Papers on the biochemical and toxicological mechanisms underlying the effects of EDCs and/or mixtures of EDCs, including oestrogenic, androgenic and thyroid effects; other mechanisms including free radical generation.

Section 4 – ENVIRONMENTAL EFFECTS: Papers relating to the effects following environmental exposure to EDCs and/or mixtures of EDCs that are specific to terrestrial and aquatic organisms:
   a.) Aquatic effects
   b.) Terrestrial effects

Section 5 – RISK ASSESSMENT, RISK MANAGEMENT and RISK COMMUNICATION: Papers relating to risk assessment and risk management of EDCs and/or mixtures of EDCs and risk communication issues.

Section 6 – MISCELLANEOUS: Other papers considered of general interest or potential relevance to the study of EDCs and/or mixtures of EDCs that do not relate to the above categories.

The papers presented herein were identified using a series of structured searches of the following on-line databases: Medline, Toxline, Biological Sciences, Scopus and Environmental Sciences and Pollution Management. The search terms used were “endocrine disrupt” and “human or environment or exposure”; thus ensuring any relevant abstracts containing these terms in their title, abstract or descriptors were identified. The paper abstracts
were reviewed and categorised by an experienced IEH Scientist to confirm their relevance before inclusion in this report.
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Review of papers

1. HUMAN EXPOSURE MEASUREMENT AND MODELLING

An inter-laboratory comparison exercise was conducted by Heath and colleagues in Slovenia, France, Germany, Austria, Sweden, Luxembourg, Denmark and Romania under the EU COST Action 636: Xenobiotics in Urban Water Cycle. The exercise demonstrated that both GC-MS and LC-MS/MS (gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry respectively) procedures can be utilised to analyse natural and synthetic hormones (17α-ethinylestradiol, 17β-oestradiol and oestrone) in various water sources sampled including, tap water, river water and waste treatment plant influent and effluent (Heath et al., 2010).

The endocrine disruptor Bisphenol A (BPA) is a major by-product generated during the thermal degradation of polycarbonate (PC). Sala and colleagues reported a method of reducing BPA production through thermal treatment at lower degradation temperatures in nitrogen atmospheres, in the presence of copper chloride (CuCl₂; Sala et al., 2010). Other novel methods reported during this period include yeast bio-assays that have been employed for the direct measurement of oestrogenic activity in wastewater influent and effluent (Balsiger et al., 2010) and also for assessment of thyroid hormone receptor ligand activity of industrial chemicals and environmental pollutants (Shiizaki et al., 2010). Furthermore, a biosensor has been developed to detect estrogenic mycotoxins in milk. The authors report that as the method is rapid and readily automated it could be utilised as a screening method to detect for estrogenic activity in milk products, prior to more detailed analysis (Valimaa et al., 2010). Yang et al. identified circulating thyroxine (T4) as a novel biomarker for detection of non dioxin-like polychlorinated biphenyls, which may aid in overall risk assessment processes in thyroid endocrine disruption (Yang et al., 2010).

Kuch et al. (2010) discussed the possibility of estrogenically active substances present in obsolete landfills being able to leach into groundwater; landfill sites containing polychlorinated biphenyls or waste from gas works were considered to be of particular risk. Sample analysis indicated that oestrogenically active substances were present in the groundwater below obsolete landfills; however, the total activity was ascribed to a number of individual compounds, some of which could not be quantified. It was therefore not possible to assess potential human exposure levels and identify specific remediation measures to employ (Kuch et al., 2010). Similarly, wastewaters from four health care facilities from a single municipality in Texas, USA, were evaluated for eight steroid hormones, octylphenol (OP), nonylphenol (NP), sixteen nonylphenol ethoxylates (NPEOs) and 10 octylphenol ethoxylates (OPEOs). The total mass loading of hormones into wastewater was measured as between 2.5mg d⁻¹ and 138mg d⁻¹, however, the total mass loading of alkylphenol ethoxylates (NPEO + OPEO) was seen to be considerably higher, ranging from 1.8g d⁻¹ to 54g d⁻¹ (Nagarnaik et al., 2010).

The association between exposure to organophosphate pesticides and disruption of thyroid gland function has been noted in animals, however, evidence in humans is less frequently observed. In a longitudinal study of floriculture workers occupationally exposed to organophosphate pesticides, increased levels of thyroid hormones, including thyroid stimulating hormone, were reported one day following pesticide application (Lacasana et al., 2010). A study in India exploring a possible association between recurrent miscarriages and exposure to environmental organochlorine pesticides reported increased levels of blood
hexachlorocyclohexane in women that also had recurrent miscarriages (defined as three or more consecutive miscarriages before 20th week of gestation), as compared with women who progressed to full term live births (Pathak, et al., 2010). In a review, Ranjit and colleagues provide an assessment of how ethnic origin in the USA population may lead to disproportionate exposure to EDCs, affecting birth outcome (Ranjit et al., 2010).

Endocrine disruptive effects in wild populations of chub (Leuciscus cephalus) have been evaluated in five French rivers, representative of different pollution contexts over two seasons. A number of biomarkers were evaluated within the study, with the use of brain aromatase being identified as a potential biomarker of endocrine disruption in vitro and in vivo (Hinfray et al., 2010). In addition a study by Hirsch and co-workers demonstrated use of the fruit fly, Drosophila melanogaster, as a model for distinguishing endocrine disruption induced behavioural changes following exposure to Pb2+ from the direct behavioural effects of Pb2+ on neural mechanisms (Hirsch et al., 2010).

2. HEALTH EFFECTS

a. Reproductive/developmental health

The association between exposure to pesticides and decreased sperm count was investigated in a group of Spanish males. The authors identified a positive correlation (p=0.009) between blood concentration of endosulfan sulphate and a reduced sperm count. However, it should be noted that this study was criticised in its selection criteria and for the analytical methods employed; as the full article is in Spanish, no further details were available for inclusion here (Avivar Oyonarte et al., 2010). A Turkish study reported a positive correlation (p<0.001) between increased plasma levels of phthalates (specifically mono-(2-ethylhexyl)-phthalate or MEHP, and di-(2-ethylhexyl)-phthalate or DEHP; which is thought to have anti-androgenic or oestrogenic effects – or both) and patients with pubertal gynecomastia (DEHP: 4.66 ± 1.58 µg/ml; MEHP: 3.19 ± 1.41 µg/ml), compared with age matched controls (DEHP: 3.09 ± 0.90 µg/ml; 1.37 ± 0.36 µg/ml; Durmaz et al., 2010).

Perchlorate is used therapeutically in the treatment of hyperthyroidism as it inhibits thyroidal iodine uptake, causing reduced production of thyroid hormone. As a consequence of its pharmacologic uses, low levels of Perchlorate have been identified in the environment, ground water and food stuffs. In their report, Leung and colleagues highlight the potential impact on normal thyroid function following low exposure to environmental Perchlorate, which is of particular concern to the developing foetus and infants (Leung et al., 2010). Evidence to support BPA as a potential endocrine disruptor in humans is sparse. In an occupational cohort study of male workers exposed to BPA, sexual function was ascertained through in-person interviews. Li and colleagues reported evidence that occupational exposure to BPA was associated with male sexual dysfunction in a dose dependent manner. In addition, reduced sexual function was found to be evident at a higher frequency within one year of occupational BPA exposure when compared with non-exposed workers. (Li et al., 2010).

Exposure to polychlorinated biphenyls (PCBs) during and after pregnancy has been reported to result in decreased foetal and infant growth and development. However, the effects of exposure to PCBs during the pre-conception period are largely unknown. Murphy and colleagues assessed maternal serum PCB levels at two developmentally-sensitive time periods during pregnancy in relation to birth weight. The authors report a reduction in birth weight in the highest tertile of preconception anti-estrogenic polychlorinated biphenyl (PCB) concentration in maternal serum, as compared to the lowest tertile (Murphy et al., 2010). In a review, Schell and colleagues summarise findings from several of their studies carried out to assess whether exposure to persistent organic pollutants (POPs) and heavy metals, is related to thyroid function and sexual maturity (Schell et al., 2010).

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In further reviews, Chalupka & Chalupka provide a detailed assessment of the evidence for an impact of environmental and occupational exposure to EDCs on reproductive health (Chalupka & Chalupka, 2010), while Fisch and colleagues review epidemiologic data compiled to investigate whether the rising incidence of hypospadias in males is associated with exposure to EDCs such as phthalates and BPA (Fisch et al., 2010).

b. Carcinogenic effects

The association between development of hormone-related cancers, including prostate and breast cancer, and exposure to organochlorine (OC) pesticides was evaluated in a study of US adults. The authors reported a significant association between serum concentrations of some OC pesticides and risk for prostate cancer but not for breast cancer. It is stated that further study is needed to confirm these findings (Xu et al., 2010).

3. BIOLOGICAL MECHANISMS and TOXICITY TESTING

In the months Jan-Mar 2010, a large number of articles relating to mechanisms of EDC activity have been published. For ease of reading, the articles are summarised in the order of human related and then animal related studies.

Disruption of cytokine secretion in human placenta may result in implantation failure or even pregnancy loss. The persistent organic pollutant para-Nonylphenol (p-NP) exhibits estrogen-like activity and has previously been shown to induce apoptosis in human placental cells. Bechi and colleagues investigated the effect of p-NP on cytokine secretion in human placenta using in vitro cultures of human placenta (from first trimester). Low doses of p-NP (≤ nanomolar range) were shown to affect cytokine release (both increase and decrease of selected cytokines) which the authors state raises considerable concern with respect to maternal exposure to p-NP in pregnancy (Bechi et al., 2010). In a further study assessing cytokine function, the effects of EDCs on the immune system were investigated through exposure of human dendritic cells to nonylphenol (NP) and 4-octylphenol (4-OP). Cytokine expression was shown to be affected by both NP and 4-OP which the authors proposed occurred via a mechanism mediated through the oestrogen receptor (Hung et al., 2010). However, in contrast, it has been reported that NP and 4-OP do not act via the oestrogen receptor in affecting monocyte –derived chemokines in a human monocyte cell line (THP-1) (Yeh et al., 2010). During this reporting period, Kwintkiewicz and colleagues demonstrated dose dependent inhibitory effects of BPA on 17β-oestradiol secretion and on growth factors including steroidogenic factor-1 in cultured human granulosa cells; these effects were proposed to be mediated by peroxisome proliferator activated receptor-γ (Kwintkiewicz et al., 2010). In the oestrogen-dependent MCF-7 human breast carcinoma cell line, synthetic pyrethroids suggested to have oestrogenic activity in vitro, did not affect oestrogenic activity as determined by evaluation of cell proliferation. Assessment of the same synthetic pyrethroids in an in vivo rat model supported these findings, with no effect on puberty onset and vaginal opening being shown (Laffin et al., 2010). Environmental EDCs, including BPA, have recently been identified as risk factors in the development of malignant tumours. Zhu and colleagues reported that exposure to several commonly occurring environmental EDCs potently promoted the invasion and metastasis of neuroblastoma cells (SK-N-SH cells through a number of possible routes, including a β oestrogen receptor dependent pathway; Zhu et al., 2010).

Although the utility of human cell and epidemiology studies in assessing EDCs is generally acknowledged, laboratory animal studies are still required to elucidate mechanisms of action. Induction of spermatogenic apoptosis has been identified in prepubertal rat testes following a
single exposure to di(n-butyl) phthalate; activation of oestrogen receptors has been proposed to be integral to the mechanism of action (Alam et al., 2010). The androgen receptor has been found to be a target for disruption by vinclozolin; in rats exposed *in utero*, post-pubertal prostatitis and reduced sperm production via a reversible androgen receptor mediated pathway has been reported (Cowin et al., 2010). Findings from a further study indicate that the effects of vinclozolin may be mediated through interference of the methylation pattern of imprinted genes in male mouse sperm (Stouder, et al., 2010). The food contaminant semicarbazide has been evaluated for endocrine disrupting activity using an integrated *in vivo*/*in vitro* approach. Disruption of vaginal opening and preputial separation were apparent in female and male rats respectively, with decreases in serum oestrogen and dehydrotestosterone levels also apparent (Maranghi et al., 2010). Pregnant rats exposed to an elution product from nitrile-butadiene rubber (4,4’-Butylidenebis(6-t-butyl-m-cresol; BBBC) produced offspring with altered levels of monoamines and associated metabolites, increased serum testosterone and aromatase activity, suggesting BBBC may have endocrine disrupting activity in addition to functional brain development effects (Satoh et al., 2010). Ündeğer and colleagues demonstrated that the herbicide pendimethalin may affect oestrogen, progesterone and androgen receptors in female rats (Ündeğer et al., 2010).

The impact of dioxins (2,3,7,8-Tetrachlorodibenzo-p-dioxin; TCDD) transferred through mother’s milk on the male reproductive system has been investigated in a mouse model. TCDD exposure during lactation was associated with decreased body weight, body length and anogenital distance when compared with control animals; oxidative stress and p53 were proposed to be factors in the mode of action of TCDD (Jin et al., 2010). Thyroid activity has been reported to be disrupted in juvenile female rats following exposure to the thiadiazole fungicides, saisentong (Zhang et al., 2010a) and thiadiazole copper (Zhang et al., 2010b) both of which have structural similarities to N,N-methylene-bis (2-amino-1,3,4-thiadiazole), a teratogen. Interestingly Verner and colleagues report impaired metabolism of BPA or 4-n-Nonylphenol when in combination with commonly occurring drugs such as salicylic acid and carbamazepine in incubated rat liver microsomes (Verner et al., 2010).

Endocrine disrupting effects have been reported in Japanese quail embryos (*Coturnix japonica*) following exposure to 16α-lactone-oestradiol (Mattsson & Brunström, 2010). Studies in ewes (*Ovis aries*) have revealed gender dependent sensitivity in bone tissue following exposure to polychlorinated biphenyls from conception through to one week before delivery, with male foetuses having greater sensitivity in trabecular bone mineral content and females having smaller trabecular cross-sectional area than their respective controls. Furthermore, overall, cortical bone was shown to be more sensitive than trabecular bone (Gutleb et al., 2010). In a further study, accumulation of di(2-ethylhexyl)phthalate (DEHP) was demonstrated in ewes in pregnancy (Herreros et al., 2010). The authors also report that although the presence of DEHP in plasma increased with age due to prolonged exposure, most increases in DEHP appear to be in pregnancy due to mobilisation of body fat reserves.

## 4. ENVIRONMENTAL EFFECTS

### a. Aquatic effects

Aluminium has been reported to have endocrine disrupting effects in mature female Nile tilapia (*Oreochromis niloticus*) fish, including decreases in ovarian protein concentration (Correia et al., 2010). Other metals such as cadmium have also been proposed to have endocrine disrupting properties (Planelló et al., 2010). An investigation of the effects of 17α-ethynylestradiol in mummichog (*Fundulus heteroclitus*) has shown disruption of normal development (Peters et al., 2010). Zebrafish species have been found to be sensitive to a number of chemicals with respect to reproductive function including tribromophenol (Deng et al., 2010); 17β-oestradiol (Jin et al., 2010a); different concentrations of fluorotelomer alcohols.
(Liao et al., 2010) and vinclozolin (Smolinsky et al., 2010). Raised temperature and/or increased photoperiod were shown to cause an increased transcription of oestrogen-responsive genes in zebrafish following exposure to ethinylestradiol, nonylphenol or a mixture of the two, suggesting that seasonal changes should be taken in to consideration when evaluating endocrine disrupting effects (Jin et al., 2010b). Meng and colleagues reported that $\alpha$-oestrogen receptor expression is low in male zebrafish and question whether measurement of vitellogenin, commonly used as a biomarker of oestrogenic activity, is an appropriate measure in male fish (Meng et al., 2010). However, conservation of the vitellogenin gene has been identified in different species of sea turtle, suggesting a level of importance in this species at least (Zaccaroni et al., 2010). During this reporting period the use of roach and perch species (Allner et al., 2010) and wild grey mullet (Aoki et al., 2010) have also been evaluated as possible indicator species of variations, as alternatives to zebrafish. In addition, fathead minnows have been used by Hyndman and colleagues in a study to assess the effects of exposure timing on biomarker expression. The authors conclude that a number of endpoints are necessary to fully establish the biological consequences following exposure of fish to environmental EDCs (Hyndman et al., 2010).

As an alternative to fish species, EDCs have been shown to have adverse effects in amphibian species. Exposure of the male African clawed frog (Xenopus laevis) to atrazine, was shown to result in complete feminisation and chemical castration (Hayes et al., 2010). Conversely, a study evaluating intersex in different species of frogs identified that a phase of intersex during sexual development is normal in some species, highlighting the importance of longitudinal studies when using frogs (Storr-Méndez & Semlitsch, 2010). Evidence of sensitivity to EDC effects in Echinoderm species has also been described (Sugni et al., 2010).

Creusot and colleagues have proposed the use of the human pregnane X receptor bioassay as a potential sensor for emerging aquatic pollutants that are suspected to act as ligands for nuclear receptors; the authors suggest that this assay may provide complimentary information to that already obtained on any oestrogenic, androgenic or dioxin-like activity (Creusot et al., 2010). It has been suggested that biodegradation of alkylphenol polyethoxylates is affected by the presence of trace elements in the water, such as octylphenol and therefore may ultimately affect the ecotoxicity of the water (Hotta et al., 2010).

EDCs have been detected in a number of different bodies of water in a range of countries including France (Kinani et al., 2010), Spain, (Brix et al., 2010), Belgium (Nadzialek et al., 2010); Greece (Arditsoglou & Voutsia, 2010), Czech Republic (Jedlickova et al., 2010), and San Francisco Bay (Brar et al., 2010), Newark Bay (Bugel et al., 2010) and South Carolina (Truman et al., 2010) in the USA.

Scott and colleagues have reviewed the role of dihydroxyprogrenone, a maturation inducing steroid, in male fish (Scott et al., 2010). Mohapatra and colleagues have reviewed the fate of BPA in treated effluent, concluding that the presence of heavy metals and possible thermodynamics of BPA in waste water and waste water sludge (Mohapatra et al., 2010) have major impacts on BPA removal; in a similar vain, the presence of bicarbonate in water has been suggested to increase the degradation rate of BPA (Pétrier et al., 2010).

b. Terrestrial effects

An investigation of dairy farm shed effluents in New Zealand has shown the presence of steroid oestrogens and conjugated oestrogens, indicating the potential impact of agricultural waste on oestrogen load released to soil and aquatic environments (Gadd et al., 2010).
5. RISK ASSESSMENT, RISK MANAGEMENT AND RISK COMMUNICATION

The regulatory implications of screening and testing of chemicals for endocrine disrupting activity through binding to mammalian and fish oestrogen receptors is discussed in a study by Dang (Dang, 2010). An assessment of the risk posed to human health from environmental exposure to BPA is given by Patisaul (Patisaul, 2010) and emerging concerns regarding contaminants identified in the Upper Mississippi River in the USA and the regulatory measures put in place to reduce these, are discussed by Weiner and Sanheinrich (Wiener & Sanheinrich, 2010).

6. MISCELLANEOUS

Phytoestrogens, a class of endocrine disruptors found in plants, have been reviewed by Ball et al., Liu et al., and Taxvig et al., 2010. Other reviews published during this reporting period provide a discussion of human fertility (Joffe, 2010), the risks to the environment and human health from EDCs (Snyder & Benotti, 2010) and a general overview of endocrine disruptors (Pfaff, 2010). Finally a range of articles have also been published focusing on different/novel methodologies for investigation of EDC (Dåas et al., 2010; Flamegos et al., 2010; Huang et al., 2010; Majeau et al., 2010; Mita et al., 2010; Racz & Goel, 2010; Rivas et al., 2010).
References

1. HUMAN EXPOSURE MEASUREMENT AND MODELLING


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2. HEALTH EFFECTS

a. Reproductive/developmental health


b. Carcinogenic effects


c. Other effects

No relevant papers identified.
3. BIOLOGICAL MECHANISMS and TOXICITY TESTING


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4. ENVIRONMENTAL EFFECTS

a. Aquatic effects


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5. RISK ASSESSMENT, RISK MANAGEMENT and RISK COMMUNICATION


6. MISCELLANEOUS


