Hormone Disrupters, Environmental Estrogens, and Pesticides

Presented by
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INTRODUCTION

Headlines

Controversy Builds Over Threat of Common Chemicals

"A Group Of Eminent Scientists Summoned To Washington This Week Is Soberly Weighing The Panglobal Impact of Undersized Alligators Penises, 'Lesbian' Sea Gulls and Diminished Human Sperm Counts"

Wall Street Journal (March 7, 1996)

What's Wrong With Our Sperm?

"Men's Reproductive Cells Seem To Be In Serious Decline Worldwide. One Possible Cause: Chemical Pollution"

Time (March 18, 1996)

The "Endocrine Disrupter" Issue

Some "environmentalists" have asserted that numerous man-made chemicals can "mimic" or interfere with natural hormones, and thereby "disrupt" the normal functioning of the endocrine system in humans and wildlife.

Endocrine systems consist of complex feedback pathways that influence diverse bodily functions through the secretion of hormones by glands such as the pituitary, thymus, thyroid, adrenals and gonads.

The hormone given the most attention is estrogen, a sex hormone produced by both men and women (but in larger quantities by women).

Other hormones, such as growth / thyroid hormones, are also attracting attention.

"Jargon" in the Popular Press

Environmental Estrogens

Endocrine Disrupters

Endocrine Modulators

Hormone Mimics

Gender Benders

Hormone-Related Effects Described in the Popular Press on Humans and Wildlife

Abnormalities in Human Testes

Decreased Quantity / Quality of Human Sperm

Feminization of Wildlife Species

Possible Association Between Organochlorine Chemicals and Breast Cancer

Broad Generalization that All Pesticides Are Just Like DDT

Experimental Evidence of Interest

Precocious Metamorphosis of the Frog Tadpole (Rana pipiens) by Adding Thyroid Hormone (Thyroxin) to the Aquarium Water:

The mother frog was caused to ovulate in September, some seven months before the normal breeding period, by subcutaneously implanting six pituitary glands from adult female donors. The eggs were artificially inseminated. When the tadpoles developed hindlimb buds, minute amounts of thyroxin were added to the water. In about three weeks, the treated tadpoles reabsorbed their swimming tails, grew hindlimbs and then forelimbs, lost their horny teeth (used for plant feeding), shortened their intestinal tracts in preparation for carnivorous feeding, modified their respiratory and integumentary systems for terrestrial environments, and emerged as normal but miniature air-breathing adult frogs.

This experiment was first performed by Gudernatsch (1912), who fed bits of horse thyroid to young tadpoles. Those of you +/- "boomer" age may remember BSCS Biology in high school, and may have repeated this experiment.

Since thyroxin initiates the metamorphic changes long before they would normally occur, the resulting "froglets" are about one-third the size of those metamorphosing in nature. Thyroid-ectomized tadpoles never metamorphose, but grow to "giant" size.

Diagram of Human Endocrine System (showing location of glands)

Substances Implicated

A vast number of man-made chemicals and chemical by-products have been identified as potential hazards. Some environmental groups and scientists have focused attention on:

dioxins

PCBs

DDT and its metabolites

Other Pesticides (including other organochlorines, and the triazines)

alkylphenol ethoxylates

PVC products

bisphenol-A

phthalates

pulp and paper production effluents

spermicides and condom lubricants

Media Attention

The Society of Environmental Journalists promoted the issue at the October 1995 annual meeting.

Since then, major articles have appeared in *Scientific American*, *Esquire*, and the *New Yorker*; the BBC Television Production was shown on the *Discovery Channel*.

A new book, **Our Stolen Future**, was published in March 1996. Its publication was followed by a promotional tour;

reviews are starting to appear.

ENVIRONMENTAL ESTROGENS AND BREAST CANCER

Estrogenicity - Definition and Function

Estrogens are hormones produced by humans and other animals;

Secreted primarily by the ovaries (female);

Estrogens stimulate sexual maturation of females, and play an important role in reproductive function.

The Nature of the Physiological Response to Hormones

Hormones act through specific receptors.

A complex feedback mechanism provides balance.

Mammals have both "male" and "female" hormones.

Hormone effects are threshold-mediated.

Estrogen: Therapeutic Uses

Estrogen is used with other female hormones in:

Birth Control (for women of child-bearing age);

Hormone Replacement Therapy (for post-menopausal women)

- reduces the risk of osteoporosis, heart disease, and some GI cancers.

- some reports of increased risk of endometrial and possibly breast cancer*.

(*Studies indicate this effect is associated with use of estrogen alone. Current practice is to use estrogen with progesterone, and at lower rates.)

Xenobiotics Tested by the E-SCREEN Assay

Estrogenic Xenobiotics Non-estrogenic Xenobiotics

Herbicides Herbicides

none 2,4-D 2,4-DB

alachlor atrazine

butylate cyanazine

dacthal dinoseb

hexazinone metolachlor

picloram propazine

simazine trifluralin

Insecticides Insecticides

p,p,-DDT bendiocarb carbaryl

o,p,-DDT carbofuran chlordane

p,p,-DDE chlordimeform chlorpyrifos

o,p,-DDE diazinon heptachlor

DDT (technical grade isomer mixture) kelthane lindane

dieldrin malathion methoprene

chlordecone (kepone) mirex parathion

endosulfan pyrethrum rotenone

-endosulfan

-endosulfan

methoxychlor

toxaphene

Fungicides Fungicides

none chlorothalonil hexachlorobenzene

maneb metiram

thiram zineb

ziram

Estrogens are defined by their ability to induce the proliferation of cells of the female genital tract. The wide chemical diversity of estrogenic compounds precludes an accurate prediction of estrogenic activity on the basis of chemical structure alone. Rodent bioassays are not suited for the large-scale screening of chemicals before their release into the environment because of their cost, complexity, and ethical concerns. The E-SCREEN assay was developed to assess the estrogenicity of environmental chemicals using the proliferative effect of estrogens on their target cells as an end point. This quantitative assay compares the cell number achieved by similar inocula of MCF-7 cells in the absence of estrogens (negative control) and in the presence of 17-estradiol (positive control) and a range of concentrations of chemicals suspected to be estrogenic. Among the compounds tested, several "new" estrogens were found; alkylphenols (antioxidants, also used in the synthesis of detergents), phthalates (plasticizers), some PCB congeners and hydroxylated PCBs, and some insecticides (chlorinated hydrocarbons) were estrogenic by the E-SCREEN assay. In addition, these compounds competed with estradiol for binding to the estrogen receptor and increased the levels of the progesterone receptor and pS2 in MCF-7 cells, as expected from estrogen mimics.

E-SCREEN can be used to screen individual chemicals. Authors recommend using the E-SCREEN test to measure total xenoestrogen burden, once a protocol is developed to separate environmental estrogens from endogenous ones.

(A. M. Soto et. al., *The E-SCREEN Assay as a Tool to Identify Estrogens: An Update on Estrogenic Environmental Pollutants*, *Environmental Health Perspectives,* 103 (Suppl 7): 113-122, 1995.)

Estradiol

structure of estradiol

Structures of Selected Synthetic Chemicals Which Are Estrogenic

examples = structures of *o,p,*-DDT, methoxychlor, nonyl phenol tributyl tin, and bisphenol-A PCBs

Other Common Materials, Drugs and Natural Food Substances Can Have Estrogenic Effects

These include:

Oral contraceptives

Hormone Replacement Therapy Treatments

Breast Cancer Treatment Drugs (tamoxifen)

egg yolks and liver

grains and grain products (barley, corn, oats, rice, wheat) - including beer!

vegetable oils

apples, carrots, cherries, garlic, olives, parsley, peanuts, plums, potatoes, soybeans, yams

components of sewage effluent

Some scientists maintain that exposures to naturally-occurring hormone-like substances dwarf any likely ordinary exposures from industrial chemicals.

(**Food Safety**, J. M. Jones, Egan Press, St. Paul, MN, 1992.)

Zearalenone

Zearalenone is a natural toxin produced in foods by *Fusarium* infections.

Zearalenone is estrogenic, and a probable carcinogen ((NOEL 0.05 mg/kg/day).

1,800 mg in the diet of ewes produced 1/2 ppb in milk. However, this was sufficient to produce estrogenic effects in nursing lambs.

The Potency of the Physiological Response to Hormones

For comparative purposes, assign Estradiol a potency rating of 1.0.

Ethinyl estradiol potency is about 10.0;

Plant flavenoid potencies range from 0.01 to 0.0001;

DDT's potency is 0.0001 to 0.000001.

(Kupher & Bulger, *Estrogenic Actions of Chlorinated Hydrocarbons* in **Effects on Chronic Exposures to Pesticides on Animal Systems**, J. E. Chalmers and J. D. Yarbrough, Eds., Raven Press, New York, 1982.)

Estimated Human Exposure from Dietary Estrogens

Source Estrogen Equivalents

Birth Control Pills 16,675

Estrogen Replacement Therapy 3,350

Flavenoids in Foods 102

Pesticides 0.0000025

(S. H. Safe, *Environmental and Dietary Estrogens and Human Health: Is There a Problem?*, *Environmental Health Perspectives,* 103: 346-351, 1995.)

Estrogenic Effect of Pesticides Based on the E-SCREEN Assay

Compound Concentration RPE, % RPP, %

estradiol 10pM 100.00 100.0

DDT* 10M 79.61 0.0001

o,p,-DDT 10M 86.14 0.0001

p,p,-DDT 10M 71.00 0.0001

dieldrin 10M 54.89 0.0001

endosulfan* 10M 81.25 0.0001

-endosulfan 10M 77.17 0.0001

-endosulfan 10M 78.26 0.0001

1-hydroxychlordene 10M 40.00 0.0001

kepone 10M 84.00 0.0001

methoxychlor 10M 57.00 0.0001

toxaphene 10M 51.90 0.0001

Concentration describes the dose at which an estrogenic effect is detected; maximal cell yield is obtained at concentrations between 10 and 100 pM estradiol. Most xenobiotics are active at 10M. The RPE (Relative Proliferative Effect) measures the ratio between the maximal cell yield achieved by the xenobiotic and that of estradiol. The RPP (Relative Proliferative Potency) is the ratio between the minimal concentration of estradiol and the minimal dose of the xenoestrogen test compound needed to produce maximal cell yields x 100. Asterisks (*) denote that the compound tested was of technical grade.

(A. M. Soto et. al., *The E-SCREEN Assay as a Tool to Identify Estrogens: An Update on Estrogenic Environmental Pollutants*, *Environmental Health Perspectives,* 103 (Suppl 7): 113-122, 1995.)

Odds of Developing Breast Cancer (U.S. Female)

By Age 25: One in 19,608 By Age 60: One in 24

By Age 30: One in 2,525 By Age 65: One in 17

By Age 35: One in 622 By Age 70: One in 14

By Age 40: One in 217 By Age 75: One in 11

By Age 45: One in 93 By Age 80: One in 10

By Age 50: One in 50 By Age 85: One in 9

By Age 55: One in 33 In a Lifetime: One in 8

(J. H. Tanne, New York Magazine, October 11, 1993, pp 53-62.)

Breast Cancer Trends Among U.S. Women

Age (Caucasian women) Percent Change in Mortality Rate 1989-1992

30-39 - 8.7

40-49 - 8.1

50-59 - 9.3

60-69 - 4.8

70-79 - 3.4

80 + + 1.0

White Women (all ages) - 5.5

Black Women (all ages) + 2.6

All Women - 4.7

(Smigel, K., J. Natl. Cancer Inst., 87: (3), p. 173, February 1, 1995.)

Established Risk Factors for Breast Cancer in Females

Risk Factor High-Risk Group Low-Risk Group Relative Risk

Age old young ***

Country of Birth North America,

Northern Europe Asia, Africa ***

Socioeconomic

Status High Low **

Marital Status Never Married Ever Married *

Place of Residence Northern U.S. Southern U.S. *

Urban Rural *

Race 45 white black *

Race 40 black white *

Nulliparity yes no *

Age at First

Full-Term Pregnancy 30 years 20 years **

Oophorectomy

Premenopausally no yes **

Age at Menopause late early *

Age at Menarche early late *

Weight,

Postmenopause heavy thin *

History of Cancer

in One Breast yes no **

History of Benign

Proliferative Lesion yes no **

Any First-Degree

Relative with History yes no **

of Breast Cancer

Mother or Sister

with History of yes no ***

Breast Cancer

History of Primary

Cancer in Endo- yes no *

metrium or Ovary

Mammographic

Parenchymal dystplastic parenchyma normal parenchyma **

Patterns

Radiation to Chest large doses minimal exposure **

*** = relative risk greater than 4.0

** = relative risk of 2.0 to 4.0

* = relative risk of 1.1 to 1.9

(J. H. Tanne, New York Magazine, October 11, 1993, pp 53-62.)

Occupation as a Risk Identifier for Breast Cancer

Occupational Groups PMR OR (95% confidence interval)

Executive / Managerial 109* 1.27 (1.20, 1.35)

Professional 129* 1.55 (1.48. 1.61)

Technicians and related support 94 1.06 (0.97, 1.16)

Sales 99 1.20 (1.14, 1.27)

Clerical 113* 1.35 (1.30, 1.40)

Service 74* 0.81 (0.77, 0.84)

Farming, Forestry, and Fishing 75* 0.84 (0.66, 1.07)

Mechanics 117 1.72 (1.24, 2.40)

Construction Trades 64** 0.99 (0.64, 1.56)

Precision Production 97 1.12 (1.01, 1.25)

Machine Operators 84* 0.90 (0.88, 0.97)

Transportation 83** 0.80 (0.70, 1.07)

Laborers and Helpers 76* 0.82 (0.74, 0.90)

Armed Forces 66 0.75 (0.40, 1.38)

Housewives 91* 0.82 (0.80, 0.83)

* P 0.01

** P 0.01

(C. H. Rubin, et. al., *Occupation as a Risk Identifier for Breast Cancer*, American Journal of Public Health, 83: 1311-1315, 1993.)

Occupational Exposures and Female Breast Cancer Mortality in the U.S.

Cantor et. al. examined 31 possible workplace exposures, including to herbicides and insecticides.

Findings for styrene monomer, several solvents and some metal-related exposures suggest more investigation. No unusual results from workplaces with potential herbicide or insecticide exposures.

(Cantor, K. P. et. al., *Occupational Exposures and Female Breast Cancer Mortality in the United States*, Journal of Occupational and Environmental Medicine, 37: 336-348, 1995.)

Pesticides and Breast Cancer

Wolff et. al. observed increased circulatory DDE in breast cancer patients (1993).

Wolff, M.S., et al., Blood Levels of Organochlorine Residues and Risk of Breast Cancer, J. Natl. Cancer Inst. 85, 648-652 (1993).

Krieger and Wolff et. al. observed no correlation in a larger study (1994).

Krieger, N., Wolff, M.. et al., Breast Cancer and Serum Organochlorines: a Prospective Study Among White, Black and Asian Women, J. Natl. Cancer Inst. 86: 589-599 (1994).

Breast Cancer is NOT more prevalent where DDT was most heavily used:

- Southeast Asia

- Rural U.S.A.

- Asia

- Europe in the 1940's

Cessation of DDT use in the U.S. has not lead to a decrease in breast cancer.

Anti-chemical activists report several studies that correlate pesticide use, estrogens and breast cancer.

However, published reports show women with the highest exposure to pesticides have the lowest incidences of breast cancer mortality:

- published report show areas with the highest exposure to pesticides and in which pesticide use is the greatest do not have higher incidences of breast cancer mortality (Scientific American, October 1995; New York Magazine, 1993).

- studies show women who work with pesticides have lower than average breast cancer mortality (American Journal of Public Health, 1993).

- estrogen potency of pesticides is trivial compared to other sources (Environmental Health Perspective, 1995).

Comment

The notion of an environmental link to breast cancer has not been proved by scientific studies published in medical journals. While some researchers have explored the possibility of such a link for years, others worry that excessive attention to this area of inquiry could divert scientific expertise and resources from work more likely to yield definitive answers that can save women's lives.

(J. Pope, The Times-Picayune, New Orleans, LA, July 21, 1995.)

Lowering the Risk of Breast Cancer

Lower Alcohol Consumption

Reduce Dietary Fat

Increase Intake of Vitamins A, C, E; Carotenoids, Fiber

Increase Physical Activity

(Dr. Louise Brinton, Journal of the National Cancer Institute, 86: 1371-1372, 1994.)

Investigation of Environmental Effects Requires:

1. Determining the type of effect;

2. Identifying agents that could be responsible;

3. Establishing a dose-response relationship; and

4. Demonstrating that environmental concentrations match observed effect.

Studies Required for Pesticide Registration Which Detect Endocrine Effects

Mammalian

Developmental (rabbits, rats)

Multigeneration Reproduction (rats)

Subchronic (dogs, mice, rats)

Chronic (mice, rats)

Oncogenic (tumor production) (mice, rats)

Other

Reproduction Tests in Aquatic and Terrestrial Invertebrates

Chronic Fish Studies

Early and Full-Life Cycle Tests in Fish

Avian Reproductive Studies

Avian Teratology Studies

Examples of Current FIFRA Testing Requirements

Subchronic Dog and Rodent Studies

- three dose levels, one high enough to elicit toxicity.

- endocrine target organs are examined for macroscopic and microscopic changes in morphology and size:

pituitary gland testes

uterus thyroid

ovary parathyroid

adrenals epidiymis

mammary glands accessory sex organs

- two generation studies include observation of:

gonadal function lactation

estrous cycles weaning

mating behavior offspring's ability to reproduce successfully

conception

Food for Thought

A number of chemicals, including many natural food substances in normal diets, have hormone-like effects. Any or all of these have the potential to produce adverse health effects if the dose and potency are high enough. *Note: critical factors are DOSE and POTENCY.*

Although it is alleged that industrial "endocrine disrupters" appear to be causing problems on a global scale, there is lack of data documenting widespread problems in the human population.

In the book **Our Stolen Future**, a great deal of reliance is placed on the DES (diethystilbesrol) episode; however, the issue of dose and potency severely limit the application of this situation to all industrial chemicals and exposures. (DES is a synthetic estrogen given to women from the 1940's through the early 1970's to prevent miscarriages. It was found to cause reproductive anomalies, including structural abnormalities and cancer, in both male and female offspring of pregnant women taking the drug.)

Most of the wildlife data cited in **Our Stolen Future** comes from polluted "hot spots", such as Florida's Lake Apopka, a Superfund site. There, Lou Guillette found alligators with underdeveloped male sex organs. Other research heavily relied upon simply proposes "endocrine disruption" as a possible hypothesis for observed population effects.

Note: loss of habitat is the most frequently cited factor for the decline of wildlife populations, not reproductive inhibition.

Contrary to the claims of some, most chemicals introduced into commerce in the Unites States are, in fact, carefully tested for reproductive and developmental effects. This is especially true for pesticides.

Reregistration is requiring "older" pesticides meet the same standards in place for pesticides registered today. (Registrants must conduct and submit the additional studies, and they must be acceptable. Otherwise, the product registration is canceled.)

Governmental Activity - Endocrine Disrupters

The United States has begun a study of "Persistent Organic Pollutants" (POPs). This study includes the issue of endocrine disrupters.

EPA has held two scientific workshops and is developing a research agenda for the issue.

The National Science and Technology Council (chaired by the White House OSTP) considers the issue a

government-wide research priority.

The Department of Health and Human Services has numerous research projects underway.

The National Research Council began a two-year study on the issue with funding from EPA and the Department of the Interior.

The U.S, Senate attached an amendment to S. 1316, the Safe Drinking Water Act Amendments, that would require additional testing and provide EPA with new regulatory authorities.

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Additional information is available on the American Crop Protection Association Endocrine Issues Page

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