

The U.S. Federal Framework for Research on Endocrine Disruptors and an Analysis of Research Programs Supported during Fiscal Year 1996

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The potential health and ecological effects of endocrine disrupting chemicals has become a high visibility environmental issue. The 1990s have witnessed a growing concern, both on the part of the scientific community and the public, that environmental chemicals may be causing widespread effects in humans and in a variety of fish and wildlife species. This growing concern led the Committee on the Environment and Natural Resources (CENR) of the National Science and Technology Council to identify the endocrine disruptor issue as a major research initiative in early 1995 and subsequently establish an ad hoc Working Group on Endocrine Disruptors. The objectives of the working group are to 1) develop a planning framework for federal research related to human and ecological health effects of endocrine disrupting chemicals; 2) conduct an inventory of ongoing federal research programs; and 3) identify research gaps and develop a coordinated interagency plan to address priority research needs. This communication summarizes the activities of the federal government in defining a common framework for planning an endocrine disruptor research program and in assessing the status of the current effort. After developing the research framework and compiling an inventory of active research projects supported by the federal government in fiscal year 1996, the CENR working group evaluated the current federal effort by comparing the ongoing activities with the research needs identified in the framework. The analysis showed that the federal government supports considerable research on human health effects, ecological effects, and exposure assessment, with a predominance of activity occurring under human health effects. The analysis also indicates that studies on reproductive development and carcinogenesis are more prevalent than studies on neurotoxicity and immunotoxicity, that mammals (mostly laboratory animals) are the main species under study, and that chlorinated dibenzodioxins and polychlorinated biphenyls are the most commonly studied chemical classes. Comparison of the inventory with the research needs should allow identification of underrepresented research areas in need of attention. *Key words:* carcinogenicity, developmental toxicity, endocrine disruptor, immunotoxicity, neurotoxicity, risk assessment. *Environ Health Perspect* 106:105–113 (1998). [Online 28 January 1998] <http://ehpnet1.niehs.nih.gov/docs/1998/106p105-113reiter/abstract.html>

Over the last few years, there has been a growing concern that both synthetic and naturally derived chemicals may be causing a variety of unwanted health effects in both humans and wildlife populations due to their ability to impact the function, especially during developmental stages, of the endocrine system. In response to this concern, a federal interagency effort to coordinate research on endocrine disrupting chemicals was initiated through the President's National Science and Technology Council (NSTC). The NSTC is chaired by the president and its membership includes the vice president, the president's cabinet, and high-level advisors to the president, including John H. Gibbons, science advisor to the president. The NSTC is specifically charged with ensuring that science and technology are considered in the formulation

of federal policies and that the federal organizations have coordinated science and technology budgets and programs. The NSTC has several committees. One is the Committee on Environment and Natural Resources (CENR), which is responsible for coordinating environment and natural resources research and development across the federal agencies. The CENR has established five top priorities for the administration's environment and natural resources research and development investment.

- Global climate change
- National environmental monitoring and research
- Natural disaster reduction
- North American research strategy for troposphere ozone
- Endocrine disruptors.

Work on the endocrine disruptors initiative began in the fall of 1995 with the establishment of a Working Group on Endocrine Disruptors, chaired by the EPA and co-chaired by the U.S. Geological Survey and the Department of Health and Human Services. Participating departments and agencies are listed in Table 1. The objectives of this interagency working group are very specific: 1) develop a planning framework for federal research on human health and ecological effects of endocrine disrupting chemicals; 2) conduct an inventory to identify the nation's resources, current research projects, and the scientists conducting them; and 3) identify research gaps and facilitate a coordinated interagency research plan to address them.

As of November 1996, the first two goals have been accomplished, and a meeting was sponsored at the Smithsonian Institution to communicate efforts of the U.S. government to coordinate research activities on the general topic of endocrine disruptors. The primary purposes of the meeting were to share the framework for research needs agreed upon across the key federal organizations and to provide a preliminary analysis of the existent

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The compilation of the federal inventory would not have been possible without the efforts of many agency representatives to the CENR working group: Terri Rowles, Mike Bolger, Jane Robens, Sue Seiber, Cynthia Palmer, Ken Stills, James Koenig, George Vermont, and Steve Medford. In addition, the assistance of the following scientists was instrumental in analyzing the scope and content of the human health research projects: James Beall, Dori Germolec, Jean Harry, Dick Hill, Susan Seiber, and Michael Shelby. John Orr and Michael Choong provided invaluable assistance in setting up the computer database and Internet-accessible inventory of ongoing research projects. Finally, Joanne Rodman was instrumental in assisting in the formation and early meetings of the CENR Working Group on Endocrine Disruptors.

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risk assessment approach is lacking. Research is needed to understand the potential biological interactions of endocrine disruptors because there are such diverse chemical classes involved and because there are multiple mechanisms by which the endocrine system can be disrupted.

Measurements. The extent, magnitude, and trends of environmental exposures and effects of endocrine disruption must be documented to accurately identify and assess this problem. Exposure determinations/follow-up research is important to gauge the extent of endocrine disrupting chemical contamination in the environment, including the spatial and temporal trends of exposure in human and wildlife populations that show adverse effects. Existing exposure and effects data on endocrine disrupting chemicals should be compiled and evaluated to deduce local and national trends in population level effects. Evaluation of human health and ecological effects are most useful when information is consolidated from multidisciplinary research; consequently, well-planned and coordinated research on endocrine disruption is encouraged. Laboratory and field studies should be better integrated. Hypotheses generated by field studies should be followed up by controlled laboratory studies, and adverse effects identified in controlled laboratory studies should be validated in field studies. Sentinel species are those that are considered an indicator for the health of many components of ecosystems and connote the concept of early warning of potential problems. The identification and subsequent monitoring in the environment of sentinel species susceptible to the effects of endocrine disrupting chemicals is, therefore, an important way to provide an early warning sign of environmental contamination. Finally, database development is needed to provide a systematic way to organize information for use in problem formulation and retrospective risk assessment. Rigorous research efforts are needed to develop information systems that include 1) a compilation of the results of chemicals in various short-term screening tests; 2) prospective and retrospective analysis of health trends, i.e., status and trends; 3) field data on hormone levels and tissue burdens of endocrine disrupting chemicals; and 4) a global inventory of ongoing endocrine disruptor research to ensure that key uncertainties are being addressed and that redundancies are kept to a minimum.

The Inventory

The inventory consists of the voluntary contributions of the 14 member organizations of the working group and is the first such effort to compile relevant research projects across the federal government. An

early deliberation of the group focused on what criteria should be applied for inclusion into the inventory in terms of experimental designs and chemicals under study. In this initial version, the practice was left primarily to the contributing agency, with the general guidance that the inventory was to be used to evaluate the impact of endocrine disruption on biological processes, that we were concerned with side effects and not efficacy of pharmaceuticals, that there should be evidence that perturbations of the endocrine system are involved in the health effect of a chemical under study, and that for ecological studies, some documentation of actual exposure was considered important in determining the appropriateness for inclusion. The authors of this analysis provided oversight to ensure that the guidance was applied relatively evenly across organizations.

Nearly 400 projects were identified in constructing the initial inventory (Table 1), although it should be noted in analyzing the inventory that all projects are not of the same magnitude: one may be a large-scale field study with multiple subelements, while another may represent the work of a single investigator in a laboratory. No attempt was made to identify resources associated with the projects, so the analysis must be viewed in a semiquantitative sense. Furthermore, while efforts were made to ensure consistency in the use of categories, subcategories, endpoints, keywords, and chemicals under study, we recognize that improvements can be made in the future in this area. For example, it would be desirable to have all chemicals identified with CAS (Chemical Abstracts Service) registration numbers wherever possible. Additionally, it might also be useful to classify exposure studies by chemical classes, such as organochlorines, metals, polychlorinated biphenyls (PCBs), and chlorinated dibenzodioxins and dibenzofurans to identify indicator chemicals that are representative of key chemical classes. Hopefully the communication of this analysis will help develop that consistency in subsequent revisions and updates of the inventory. The bulk of the projects (63%) were submitted from four federal units: the NIEHS, the National Cancer Institute (NCI), the EPA, and the National Science Foundation (NSF). In retrospect, the efforts of several organizations were overlooked [e.g., National Institute for Occupational Safety and Health (NIOSH), the Occupational Safety and Health Administration (OSHA), the Consumer Product Safety Commission (CPSC), and the National Institute of Standards and Technology (NIST)], and these omissions (as well as updated contributions from current participants) will be rectified in the next

version of the inventory. Human health research dominated the inventory as a primary focal area (69%) followed by ecological research (18%) and exposure research (13%). It is noteworthy that more projects indicate exposure as a secondary ($n = 59$) rather than a primary focus ($n = 53$).

The analysis of the inventory focused on several central questions: 1) How do the research projects match the inventory in terms of categories and subcategories of research? 2) To what extent is there an appropriate balance in the range of organisms, biological endpoints, and chemicals under study? and 3) Are there specific areas in need of additional research, integration, or collaboration? The numerical analyses of project distribution by focal area, category, etc., were accomplished by fielded searches in an Oracle database. The project descriptions were subsequently transferred to the Internet site (<http://www.epa.gov/endocrine>).

Human Health Research

Of the 396 research projects included in the present CENR endocrine disruptor inventory, 273 (69%) listed human health effects as their primary focus. Approximately 21% of these fell under the Methods category, 57% under Models, and 21% under Measurements. The health effects projects were examined according to the toxicological endpoints of carcinogenicity, immunotoxicity, neurotoxicity, and reproductive and developmental toxicity. As shown in Table 2, the largest number of projects in the inventory was in reproductive and developmental toxicity, followed by carcinogenicity, neurotoxicity, and immunotoxicity.

The category "none specified" includes human health effects projects that did not specify one of the other toxicological endpoints. The breakdown of the health effects projects by endpoint yields a total that is larger than the total number of health effects projects because several projects address more than one experimental endpoint (e.g., both carcinogenicity and reproductive toxicity). A large number of projects listed for a given endpoint does not necessarily mean that the topic is being investigated sufficiently to support risk assessments for endocrine disruptors. To explore the utility of the present inventory of research projects for evaluating human risk, the health effects projects were examined by toxicological endpoint according to their distribution within the planning framework subcategories (Table 3), as well as by the nature of the research objectives of each group of projects.

Carcinogenicity. The focus of 75% of the projects on carcinogenic effects was in mechanistic/basic research or in exposure

determination and follow-up, and nearly half of these projects were on breast cancer. An intensive research effort on breast cancer is certainly needed because breast cancer rates have been increasing (1 out of 9 women in the United States will contract this disease in their lifetime), and there is concern of potential environmental contributions (e.g., synthetic organochlorine compounds acting via the estrogen receptor or by other mechanisms that modulate estrogen action). Research projects on possible relationships between endocrine disrupting chemicals and endocrine-mediated tumors at other sites (e.g., ovaries and prostate) are not well represented in the inventory. If this inventory has captured all the relevant ongoing federal research in these areas, there is then a need for additional research efforts on other organs where cancer outcomes may also be influenced by perturbations in natural hormonal activities. Two other areas that do not appear to be well represented in the inventory are projects on mixtures and on the development of predictive cancer risk models. Current research focusing on receptor-based quantitative models for dioxin-induced health effects may provide pioneering approaches for construction and validation of other predictive biological models of endocrine disrupting chemicals.

Although projects on database development appear to be few, progress in this area may ensue with data collected from projects

Table 2. Distribution of human health effect projects by toxicological endpoint

Toxicological endpoint	Number of projects
Carcinogenicity	89
Immunotoxicity	29
Neurotoxicity	59
Reproductive/developmental toxicity	107
None specified	59
Total ^a	343

^aValue exceeds total number of health projects, as some dealt with multiple endpoints.

on exposure determination and follow-up. The small number of multidisciplinary research projects may reflect underreporting rather than lack of multidisciplinary research activities. Of the four toxicological endpoints examined in this review, carcinogenicity contained the largest emphasis within human studies. Again, this was largely due to the numerous epidemiological projects addressing environmental contributions to breast cancer. In addition, several follow-up studies on diethylstilbestrol (DES) are included in the inventory. DES and halogenated synthetic compounds (organochlorine pesticides, PCBs, TCDD/dioxin) are by far the most frequently represented chemicals in the cancer related projects in this inventory.

Immunotoxicity. Even though laboratory studies indicate that endocrine disrupting chemicals can have significant immunomodulatory effects in animals, immunotoxicity projects were the least represented among the human health effects projects present in the inventory. Similar to the distribution of projects on carcinogenicity, the basic research subcategory contains the largest number of projects on immunotoxicity, and there is little apparent research activity on mixtures or on the development of predictive risk models. Several of the basic research projects examine biochemical and molecular effects that endocrine disrupting chemicals have on specific immune cell types, organ systems, or the inflammatory process in general.

Neurotoxicity. The basic research subcategory contained the largest number of projects on neurotoxicity, followed by the subcategory exposure determination and follow-up. Few projects in the inventory examined human neurological effects of individual chemicals or mixtures. There is an epidemiology study that assessed exposure to PCBs and effects on growth and behavioral functioning. Although most of the basic research projects are relevant to

the development of the nervous system and neurological effects of altered endocrine functioning, none of these addressed the effects of exposure to endocrine disrupting chemicals and human risk. Nevertheless, the basic research studies should be useful for assessing whether specific neurotoxins produce their effects by an endocrine disrupting mechanism.

Several projects are examining the endocrine system as a target of specific chemicals. In one project, there is an attempt to develop methods to assess subtle alterations in the developing brain, and another project involves the assessment of changes in neurobehavioral function following developmental exposure to pesticides. The inventory includes one coordinated multidisciplinary effort that addresses the ability of chemicals to disrupt the endocrine system during development.

Reproductive and developmental toxicity. Basic research is also the most heavily represented subcategory in the inventory of projects on reproductive and developmental toxicity. This subcategory includes many projects on the basic physiology and pathology of the reproductive and endocrine systems, but few projects on mechanisms and toxic consequences of endocrine disruption. The basic research projects should, however, provide useful information on how endocrine disrupting chemicals adversely affect reproduction or development. Several projects in the inventory are concerned with naturally occurring hormones. There are too few studies examining associations between exposure to exogenous substances and alterations in the human reproductive system to get an adequate sense of potential human risk. There are no projects addressing the existence of synergism or antagonism or mechanisms by which chemicals may interact. Efforts to develop and validate methods to detect and characterize endocrine disruptors, especially those that elicit activities other than estrogenic activity, are not well

Table 3. Distribution of human health effect projects by toxicological endpoint and subcategory listings

Category and subcategory	Toxicological endpoint (number of projects)				
	Carcinogenicity	Immunotoxicity	Neurotoxicity	Reproductive/developmental toxicity	None specified
Methods					
Hazard identification	9	3	11	14	3
Biomarkers	8	2	5	20	0
Models					
Risk models	3	1	4	14	4
Basic research	44	18	43	78	47
Mixtures	1	2	2	5	9
Measurements					
Exposure determination and follow-up	40	9	20	35	7
Multidisciplinary research	3	2	10	9	1
Sentinel species	2	3	2	4	0
Database development	2	2	1	8	0

represented in the inventory. There are no coordinated, multidisciplinary programs addressing endocrine disruption and human health risk. The development of mechanism-based predictive risk models will need greater input of human data.

Projects with no toxicological endpoint specified. Basic research is by far the most heavily represented subcategory of human health projects that do not specify one of the above toxicological endpoints. A major focus of the basic research projects is on regulation of receptor activation [aryl hydrocarbon (Ah) and estrogen receptors] and gene expression using *in vitro*-based cell culture systems. Few projects examine the activation and role of other hormonal receptors, investigate responses across species, examine antagonistic (e.g., anti-estrogenic) effects, or incorporate mechanistic data into evaluations of human risk.

General conclusions of human health research. Two recurring themes are evident from this analysis of the federal endocrine disruptor research inventory of human health effects projects. First, basic research is the subcategory that contains the largest number of projects for each of the toxicological endpoints examined. This is not too surprising, because a large number of the projects in the inventory are sponsored by the National Institutes of Health (NIH) and the NSF and because interest in endocrine disruption as a toxicological mechanism has expanded enormously in recent years. Many basic research projects are relevant to the role of the endocrine system during development (e.g., of the nervous system or reproductive system) or examine effects of altered endocrine functioning; however, few address effects of exposure to endocrine disrupting chemicals and human risk. These projects are included in the inventory because it is expected that they will contribute to our understanding of adverse effects mediated via the endocrine system and thereby be useful for assessing whether and how specific toxicological effects are produced by an endocrine disrupting mechanism. Although the basic research projects in the inventory are valuable for advancing the science on endocrine disruption, more effort will be needed to link these mechanistic data to assessments of human health risk. The second consistent finding is that studies on health effects of mixtures are not being addressed to any considerable extent. We expect the distribution of projects to change as the inventory expands and high priority research needs are identified.

Ecological Research

Nine federal organizations submitted projects with ecological effects as the primary focus area. Five organizations submitted a

total of nine projects, which leaves four organizations—the NSF, the Department of Interior, the EPA, and the National Oceanic and Atmospheric Administration—responsible for the majority of the ecological research.

The analysis is organized by the nine subcategories within the three major categories of Methods, Models, and Measurements. In a few cases, a category or subcategory other than that listed in the project was assigned to enhance consistency in the designation of categories across projects.

In the subcategory of hazard identification, nine projects are listed that provide development and/or validation of screening tests. There is a good mix of *in vitro* and *in vivo* tests. *In vivo* tests incorporate small fish, amphibians, and invertebrates. Both freshwater and saltwater fish are represented, as well as the clawed frog, the test species used in FETAX (Frog Embryo Teratology Assay-Xenopus). Most of the screening tests focus on the vitellogenin response with only two exceptions, tests that measure either growth or thyroid hormone responses. Also in this subcategory are two projects that focus on identification of active components in environmental mixtures.

The other subcategory in Methods is biomarkers. Some projects in the database may have been classified in this subcategory merely because a biomarker was used. However this subcategory was intended to be restricted to projects that develop new biomarkers or contribute toward validation of biomarkers. Seven projects were designed to develop or validate biomarkers, and again there was a strong focus on the vitellogenin response. In addition, one project examined the potential for retinal necrosis as a biomarker and two projects tested biomarkers using controlled multigenerational studies. The most common effort related to biomarkers is a group of 10 projects that evaluated effects in the field. Nearly all these projects were designed to measure ecological effects corollary to the vitellogenin response.

In the category of Models, only a few studies were directed at risk model development. However, there are additional projects that were designed to develop ecological risk models with exposure as the primary focus. Three projects examined receptor/effects linkages—two used fish and one used a reptile. A single effort to develop structure-activity relationships for endocrine disrupting chemicals and potential ecological effects is under way.

Basic research is prominently represented in the Model category, with 23 projects that examine basic endocrine regulation. While the projects in this group have broad taxonomic representation, only one mentions

chemical contaminants as a variable in the design. Most of these studies could be characterized as examining the relationship between hormone action and behavioral response. Also, five projects are dedicated to the development of animal models. Small fish models are best represented, but an amphibian model and a reptilian model are included as well. A single effort is under way linking hormonal events that control sex determination to toxic action.

Three projects could be classified in the mixtures subcategory of Models; all are testing environmental mixtures. Toxicity identification evaluation (TIE) procedures are being used in both *in vivo* and *in vitro* tests. None of the projects appear to be designed to provide information needed for additivity or synergy models. Projects dealing with environmental PCBs are not included in this subcategory although they could be considered mixtures. PCBs are often treated as a unit whether the exposure is expressed as a total PCB concentration or as total dioxin equivalents.

Projects classified as exposure determination and follow-up are well represented in the ecological database. Fourteen projects are conducting field monitoring of highly exposed or vulnerable populations, and these populations cover a broad taxonomic range. These projects make some measurement of exposure to the individuals and then assess some endpoint. The reproductive endpoint is most often targeted using an indicator of reproductive success such as fledgling or hatching success. Many of these projects also make additional measurements that are not strictly reproductive endpoints, such as morphology, behavior, and histopathology.

Two projects focus on developing exposure analysis tools. One uses a food web model to derive estimates of exposure to top predators. The other uses a semipermeable membrane device, which consists of an artificial membrane bag filled with a lipid material and is meant to mimic biological tissues absorbing lipophilic contaminants from water.

Under the subcategory of multidisciplinary research, six studies focus on integrating field and laboratory work. Despite the small number of projects, there is good taxonomic distribution, with birds, fish, and reptiles represented. There are no studies in the ecological health primary focus area that address the interaction between human and ecological health; however, this research inventory may initiate such studies.

Sentinel species are considered an indicator for the health of many components of ecosystems and connote the concept of being an early warning of potential problems.

Attributes associated with sentinel species include sensitivity to pollutants, widespread distribution, suitability for field and laboratory research, and a well-understood biology. Projects listed in the inventory develop information on four species that appear to meet enough of these criteria to be considered as sentinels: American kestrel (*Falco sparverius*), mink (*Mustela vison*), river otter (*Lutra canadensis*), and English sole (*Pleuronectes vetulus*). Another topic area identified under the subcategory of sentinel species is that of historically neglected species. Research is being directed on two animal groups that have been overlooked in ecotoxicological efforts: amphibians and dolphins. There are no efforts described in the inventory that focus on database development, the last subcategory.

While a broad range of chemicals are listed as agents under study in the inventory (Table 4), the majority of studies focus on PCBs, dioxins, and furans. There is also a significant amount of research underway on DDT and other chlorinated pesticides. Hormones are the chemical group next most

Table 4. Agents under study for ecological effects

Agent	Number of projects
Phytoestrogens	5
Polychlorinated biphenyls	28
Polycyclic aromatic hydrocarbons	5
Metals	8
Hormones	15
Environmental mixtures	9
Dioxins/furans	18
DDT	15
Chlorinated pesticides	15
Bleach kraft mill effluent	2
Atrazine	1
Total	121

Table 5. Distribution of health endpoints for ecological effect projects

Toxicological endpoint	Number of projects
Carcinogenicity	1
Immunotoxicity	4
Neurotoxicity	14
Reproductive/developmental toxicity	62
Growth/metamorphosis	8
Total	89

Table 6. Organisms under study for ecological effects

Organism	Number of projects
Insects	6
Other invertebrates	6
Fish	30
Amphibians	6
Reptiles	6
Birds	13
Mammals	7
Total	74

frequently studied, with most of these projects included in the basic research subcategory. Eight projects target metals, but only two mention tin; this is surprising given findings of imposex in bivalves, which may be due to tributyltin exposure. Two projects identified chlorotriazine herbicides, which have been proposed to have endocrine disrupting activities.

Four biological endpoints are defined in the original inventory design: reproductive (and developmental), neurologic, immunologic, and cancer. For the ecological research primary focus, a fifth category is perhaps warranted—growth and metamorphosis. Hormones that control metamorphosis would be restricted to ecological studies, and a number of species of fish and wildlife exhibit hormone-induced rapid growth phases that are critical in various life stages.

The overwhelming majority of projects in the ecological focus area use reproductive endpoints (Table 5). This is not surprising given that the regulatory premise for protection of wildlife has been based on population impacts rather than those at the individual level. This reality has directed research to the endpoint most likely to control population viability. Neurologic endpoints have the next highest frequency of occurrence in the database, but many of these studies are in the basic research subcategory and do not incorporate toxicant exposure. Eight projects identify a growth or metamorphosis endpoint, but few studies examine either immunologic or cancer endpoints. Extensive findings of cancer have been made in wild fish populations, and fish cancer models have been developed and used; however, research has not suggested a hormonally based mechanism for these environmental cancers.

For this analysis, projects in the ecological focus area are divided into seven taxonomic groupings (Table 6). Among these groupings, fish are the most frequently studied, with more than twice as many projects than birds. The other five groupings have much lower and approximately similar frequencies.

General conclusions for ecological effect studies. Developing a sound understanding of the ecological risk caused by endocrine disrupting compounds is greatly complicated by the large number of species that have to be considered. Despite the conservation of hormone structure across taxonomic groups, the importance of different hormones and the normal range of circulating concentrations vary considerably among species. Ecological health research is also limited by the resources available, which is demonstrated by the much lower number of projects when compared to human health.

Two areas of focus that were evident in the inventory stimulate further discussion. The first is the overwhelming number of ecological effects projects targeting the reproduction endpoint. The benefit of this emphasis is that reproduction has often been shown to be the most sensitive significant endpoint in wildlife that can result from xenobiotic exposure, thus offering the best chance of documenting injury. The question to be raised is whether this focus may compromise the ability to expand the knowledge base for the other endpoints, particularly those critical to ecological health such as immunotoxicity and growth/metamorphosis.

The second focus area that emerged is the significance of the abnormal vitellogenin response in males. The notoriety and utility of this response has generated a number of projects dedicated to the development of techniques that could eventually be used as screening bioassays for endocrine activity or even biomarkers of exposure in wild populations. The concern about this emphasis is that the relevance of vitellogenin in the blood of males to either individual health or to the viability of the population is not known.

Exposure Studies

The third focal area of the research inventory includes exposure studies that address the extent of exposure or the potential for exposure to endocrine disruptors. Exposure assessment is critical to assessing the potential for human and wildlife exposure to endocrine disruptors, establishing priorities for hazard identification, monitoring trends in contaminated media and body burdens, determining baseline levels to assess the impact of interventions, validating exposure and health effects associations, and reducing uncertainties in risk assessment. Exposure assessment is the least represented area of the research inventory with only 13% (53/396) of projects identifying exposure assessment as their primary focus (however, an additional 59 projects identified exposure as a secondary focal area of activity, which undoubtedly reflects the need to have exposure data to support any projected association between exposure and effects). For the purposes of this research inventory, exposure assessment has been defined to include the measurement or estimation of the magnitude, frequency, duration, and route of exposure to substances in the environment or in biological tissues. Also included under exposure assessment are studies that address demographics by linking the size and nature of the exposed populations to the parameters of chemical exposure. Not included in this focal area are studies that address the projected association between exposure and

effect or research addressing the development of biological markers for susceptibility and effect.

As with the other areas of the inventory, three categories were reviewed. These included Methods, Models, and Measurements (Table 7). Methods and models represent the tools used in conducting exposure assessments, and they are employed to identify levels of endocrine disruptors in both media and tissues. Research in these categories includes such projects as the development of gene probes for the diagnosis of exposure to steroids and recombinant detection methods for dioxinlike chemicals that can be used in dose reconstruction, and models used to estimate levels of endocrine disruptors in contaminated media and tissues. Examples include models to estimate organochlorine exposure and associated breast cancer risk and the development of molecular biomarkers of pesticide resistance in mosquito fish. In contrast to models and methods, measurement research represents the application of these tools to define actual levels of endocrine disruptors in environmental media and tissues. Examples include measurements of serum pesticides and PCBs in relation to breast cancer, as well as the measurement of PCBs and dieldrin burden among native Americans. As such, these measurements represent the systematic collection of information for subsequent analysis and potential linkage to health effects.

Table 8 provides a breakout of exposure assessment studies by subcategories. In this breakout it is clear that sentinel species, as well as mixtures research, may be underrepresented, at least from a numerical perspective. The same possibly would be true of database development. However, not all studies in the inventory reflect the same scope of effort. Thus, some projects listed under database development, such as the NHANES (National Health and Nutrition Examination Survey) study and other exposure inventories are clearly large in scope. This suggests that a greater emphasis is placed in the area of database development than would otherwise be indicated by the fact that only six studies identify database development as their primary focus.

In terms of the range of chemicals being assessed, PCBs, dioxins, and DDT are highly represented, with other compounds possibly underrepresented (Table 9). Small numbers of projects identified other potential endocrine disrupting chemicals such as alkylphenols (nine projects), phthalate esters (five projects), and nonpersistent pesticides (nine studies).

A further breakout of these studies by agent and endpoint is shown in Table 10 for those studies that provided relevant

keywords in the appropriate data fields. Table 10 shows a current emphasis on reproductive/developmental effects and a minor number of immunotoxicity and neurotoxicity studies. Some studies provide no endpoint, which indicates either a pure focus on exposure with no concomitant health assessment component or an inadvertent omission of a keyword in the biological endpoints field. Such issues should be resolved in future revisions to the inventory. Further, as reflected in Table 10, some studies have more than one endpoint under consideration. A catalog of exposure assessment studies by agent and organism (Table 11) indicates that the species breakout is highly skewed toward laboratory mammals and humans, with these two categories comprising over 50% of the studies in the inventory.

General conclusions of exposure projects. In general, there is balance of research activities across methods, models and mea-

surements, with 32, 28, and 52 studies being conducted with those categories as primary keywords for research categories and with exposure assessment as either the primary or secondary focus area. The emphasis on measurement studies reflects a focus on the actual identification of exposures where they are suspected. It appears that only limited work is ongoing on cost effective methods to identify hazard and contaminants, and relatively few studies are

Table 7. Research category distribution for exposure projects

Research category	Exposure (primary)	Exposure (secondary)	Total
Methods	16 (2) ^a	16 (4)	32 (6)
Models	16 (0)	12 (3)	28 (3)
Measurements	21 (6)	31 (2)	52 (8)
Total	53 (8)	59 (9)	112 (17)

^aNumbers presented as number of projects identified with a primary research category (secondary research category).

Table 8. Research subcategory distribution for exposure projects

Research subcategory	Exposure (primary)	Exposure (secondary)	Total
Methods			
Hazard identification	10 ^a	26	36
Biomarkers	7	22	29
Models			
Risk models	10	10	20
Basic research	18	13	31
Mixtures	3	3	6
Measurements			
Exposure determination/follow-up	16	14	30
Multidisciplinary research	3	12	15
Sentinel species	2	7	9
Database development	6	2	8
Total	75	109	184

^aNumbers presented as number of projects with corresponding primary or secondary research subcategory.

Table 9. Agents under study for exposure projects (a project may involve more than a single chemical)

Agent	Methods	Models	Measurements	Total
DDT/DDE	2	4	11	17
Estrogen	4	6	5	15
Oral contraceptive	0	0	5	5
Organochlorine	1	1	5	7
Polychlorinated biphenyls	16	9	30	55
Phytoestrogen	4	3	3	10
TCDD/dioxins	9	9	6	24
Total	36	32	65	133

Table 10. Cross-tabulation of exposure projects by agent and endpoint under study (a project may contain more than a single biological endpoint; some exposure projects did not list any of the four health endpoints as an endpoint)

Agent	Carcinogenicity	Immunotoxicity	Neurotoxicity	Reproductive/developmental	Total
DDT/DDE	6	1	2	5	8
Estrogen	4	0	1	3	8
Oral contraceptive	4	0	0	2	6
Organochlorine	0	0	0	1	1
Polychlorinated biphenyls	10	5	8	20	43
Phytoestrogen	1	0	1	2	4
TCDD/dioxins	2	3	1	8	14
Total	27	9	13	41	82

Table 11. Cross-tabulation of organisms and agents for exposure projects (a project may involve more than a single species)

Agent	<i>In vitro</i>	<i>In vivo</i>	Invertebrate	Fish	Amphibian	Avian	Reptile	Mammal	Human	Total
DDT/DDE	1	5	0	1	0	0	0	6	10	23
Estrogen	5	6	0	2	0	0	0	8	6	27
Oral contraceptive	0	0	0	0	0	0	0	0	5	5
Organochlorine	0	1	0	1	0	0	0	1	5	8
Polychlorinated biphenyls	8	18	1	8	2	1	1	24	27	90
Phytoestrogen	3	3	0	2	0	0	0	5	4	17
TCDD/dioxins	8	6	0	3	1	0	0	11	7	36
Total	25	39	1	17	3	1	1	55	64	206

directed at database development, mixtures, and sentinel species. However, the studies in the area of database development may be broader in scope than other studies and may, in fact, be addressed more comprehensively than the number of database development activities would suggest. As the scope of the endocrine disruptor research program is likely to continue to expand, it is essential that research priorities be developed to properly allocate resources to where the human health and ecologic risks are greatest. Outcomes of exposure assessment research should play a key role in this process as we learn more about the sources and long-term fate, transport, and bioavailability of endocrine disrupting chemicals.

As noted above, a major focus in the inventory appears to be on human studies, which is reported to be the focus of 64 of the total of 112 exposure assessment studies; however, closer examination of the individual projects with exposure assessment as the primary or secondary focus indicates that only 24 actually involve examination of relationships between exposure and outcomes in populations. The majority of these studies focused on issues related to breast cancer. Many of the projects identified as human oriented used human cell lines *in vitro* or examined various food sources for levels of endocrine disrupting chemicals. Only limited studies for invertebrates ($n = 1$), amphibians ($n = 3$), birds ($n = 1$), and reptiles ($n = 1$) were reported. The immune system ($n = 9$) appears to be the most underrepresented issue from a biological endpoint perspective. None of the four primary biological endpoints (reproductive, carcinogenic, neurologic, or immunologic) were indicated for a significant number of projects that identified exposure assessment as a primary or secondary focal area. With reference to the chemicals under study, there appears to be a reasonable distribution of chemicals being studied, although the emphasis has been on oral contraceptives and persistent organochlorines, with much less work in other areas (e.g., phthalates, nonpersistent pesticides, and phytoestrogens). With

respect to demographics, there is a general lack of exposure assessment information on vulnerable groups, both in terms of critical life stage and life style.

Summary and Conclusions

This is the first comprehensive effort to evaluate the involvement of the federal government in endocrine disruptor research. The analysis of the inventory provides a powerful tool to evaluate the strengths and weaknesses of current research efforts relative to the key uncertainties in the state of the science. The federal government is supporting a considerable amount of research on the topic, and several organizations have been actively working in the area over the past few years. By actively analyzing the current level of effort, the working group expects to assist the overall risk assessment process for endocrine disruptors by 1) ensuring that the federal investment is appropriately focused on the correct questions and uncertainties; 2) facilitating the coordination among the organizations to maximize the complementary nature of the research and to minimize duplications; and 3) providing the forum and means by which a coordinated federal program can have a reasonable probability of success.

The inventory analysis indicates that the overall effort is largest in human health research and in model development. Nearly 80% of the projects are contained within three subcategories: basic research, hazard identification, and exposure/risk models. Major research gaps identified in the analysis of the human health, ecological, and exposure research areas are presented in Table 12. Research targeted at developing predictive fate and transport models for specific endocrine disrupting chemicals and predictive dose-response models for potential endocrine disrupting chemical-related diseases are particularly needed in order to improve the foundation of future risk assessment and risk management activities. Within biological endpoints, there is a reasonable emphasis on effects on reproduction and development, but effects on other systems, particularly for wildlife, are lacking. As

laboratory and field studies increase in number, it is important that they consider multidisciplinary examinations so that the full ranges of biological responses can be identified and characterized. Additional biomarkers of response and the use of sentinel species for environmental studies should assist in the expanded characterization of hazard and risk. For wildlife studies, inclusion of growth and metamorphosis endpoints should be encouraged for relevant species; information on the normal ontogenetic endocrine patterns and their role in regulation of differentiation is needed; and the range of species covered should be expanded. As to chemicals under study, there is a clear dominance in studies on Ah and estrogen receptor ligands and related structural chemicals; much less attention is being placed on contemporary-use industrial chemicals and pesticides that may interact with the endocrine system via mechanisms independent of direct steroid-receptor binding (e.g., increased bioavailability due to displacement of hormones from serum-transport proteins or increased metabolic turnover through induction of liver-conjugating enzymes). Finally, work is needed to better understand the biological responses of exposure to multiple endocrine disrupting chemicals, both from the view of specific chemical mixtures and, more importantly, from the standpoint of developing generally applicable predictive models of joint action.

The member organizations of the CENR working group are committed to developing an integrated strategic plan to address the major gaps and uncertainties for endocrine disruptors. The establishment of the CENR framework, the development of the inventory, and the analysis of the match of the inventory to the framework provide the foundation for this process. Individual organizations must now take this information and develop research strategies based upon their respective missions. These must be widely circulated so that the various components of the federal effort remain informed of activities of sister organizations, thus allowing the total effort to become better coordinated and complementary. An

Table 12. Research gaps identified by matching current federal effort with the research framework

Human health effects	Ecological effects	Exposure assessment
More coordinated research on the development of predictive risk models based on results from mechanistic particular interspecies studies including greater input of human data	Characterization of effects on individuals and populations associated with the abnormal vitellogenin response	Research on mixtures, immunotoxicology, and transgenerational implications of exposure. Of utility in this regard would be the definition of accepted test protocols for each of these emerging areas of concern
Relationships between exposure to endocrine disrupting chemicals and endocrine-mediated cancers of the ovaries, testes, and prostate	Development of biomarkers of effect, particularly using noninvasive techniques, because many sensitive organisms are top predators with limited populations	More focused research to further establish priorities across organizations and thereby promote integration and collaboration among those in the federal sector and elsewhere pursuing research with respect to exposure to endocrine disruptors
Expansion of immunotoxicity studies in rodents to other species, including humans	Development of risk model information to address ecological risk assessment, particularly in addressing mixtures	Increased support for studies of fate and transport, particularly for new chemicals that may cause endocrine disruption
Assessing interindividual variability in exposure and response	Databases of baseline circulating levels of hormones for fish, wildlife, and invertebrates	
Evaluating potential synergisms and/or antagonisms in chemical mixtures	Increased emphasis on amphibians, reptiles, and invertebrates, which appear from limited work to be relatively sensitive to endocrine disruption	
	Basic research on the ability of xenobiotics to disrupt endocrine function in invertebrates	
	Broadening of the endpoints being tested, particularly immunologic endpoints, because disease is increasingly becoming a substantial factor limiting some animal populations	
	Increased attention to growth and metamorphosis as specific endpoints of ecological health	
	Broadening of the range test chemicals to include wider coverage of contemporary use chemicals	

iterative process is needed by which federal organizations supporting research on endocrine disruptors can evaluate the roles of their research programs in a larger context of national research needs on endocrine disruptors and contributions of sister organizations to these needs. The framework document and federal research inventory described in this paper provide the means to do this. Moreover, the CENR provides a coordinating mechanism for participating federal units to continue to evaluate research needs and exchange information.

A truly coordinated research strategy on endocrine disruptors should also include international information exchange and exchange of information with the private sector. The CENR anticipates working with the International Programme on Chemical Safety, the Organization for Economic Cooperation and Development, and the United Nations Environment Programme as they develop a global research inventory and an international assessment of the state of the science for endocrine disruptors.

REFERENCE

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