Linking Environmental Agents and Autoimmune Disease: An Agenda for Future Research

MaryJane K. Selgrade,[1] Glinda S. Cooper,[2] Dori R. Germolec,[3] and Jerrold J. Heindel[4]

[1]National Health and Environmental Effects Research Laboratory, U.S. Environmental Protection Agency, Research Triangle Park, North Carolina USA; [2]Epidemiology Branch, [3]Laboratory of Toxicology, [4]Division of Extramural Research and Training, National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina USA

Autoimmune diseases are influenced by multiple factors including genetics, age, gender, reproductive status, hormones, and potential environmental contaminants. A workshop, "Linking Environmental Agents and Autoimmune Diseases," was convened at the National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina, 1–3 September 1998, to review current knowledge about links between environmental exposures and autoimmune disease, to identify and prioritize research needs, and to develop an integrated, multidisciplinary research agenda. Participatants spent the last half-day of the workshop in small group discussions for the purpose of developing consensus on research needs. Research needs identified were *a*) develop research tools needed to explore links between environmental agents and autoimmune disease; *b*) establish a disease registry or surveillance system; *c*) develop and validate strategies for screening chemicals for the potential to induce or exacerbate autoimmune disease; *d*) develop an emergency response strategy to gain information from accidental exposures; and *e*) conduct hypothesis-driven research in occupationally exposed groups and/or in experimental animals. There was consensus that meetings like this workshop and projects that facilitate interactions between specialties should be encouraged. A multidisciplinary approach is needed to address this problem. *Key words*: autoimmune disease, epidemiology, hazard identification, research needs. — Environ Health Perspect 107(suppl 5):811–813 (1999).
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There are over 80 autoimmune diseases affecting 10 million Americans. These diseases are influenced by multiple factors including genetics, age, gender, reproductive status, hormones, and potential environmental contaminants. This monograph reports the proceedings of the workshop "Linking Environmental Agents and Autoimmune Diseases" held at the National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina, 1–3 September 1998. The workshop brought together immunologists, clinicians, epidemiologists, molecular biologists, and toxicologists to review current knowledge about environmental links to autoimmune disease and identify data gaps and future research needs in a multidisciplinary fashion. The articles in this monograph discuss this review (conducted during the first 2 days of the workshop). The final half-day of the workshop was devoted to developing a research agenda, which is the topic of this article. Speakers and participants met in small groups to brainstorm on the following topics: strategies for testing, common threads (among autoimmune diseases, chemicals, and other indicators of immunotoxicity), integrating epidemiology and animal research, organ-specific diseases, and risk factors (sensitive populations). A summary of each group's discussion was presented during the last session of the workshop, and a consensus on research needs and priorities (i.e., a research agenda) was developed (Table 1). Five broad research needs were identified and are described below. The foremost need (encompassed in the first four needs listed below) is to develop a preliminary database indicating a role for various environmental agents in the development or progression of various autoimmune diseases. The field cannot move rapidly into hypothesis-driven research (the fifth need listed below), particularly with respect to molecular mechanisms, without preliminary data on which agents and diseases are important.

Research Needs

Develop research tools needed to explore links between environmental agents and autoimmune disease. Because this field of study is fairly new, many of the tools needed to effectively study the links between environmental agents and autoimmune disease must be developed. Two much-needed tools are biomarkers of effect and validated questionnaires for use in epidemiology, field, and clinical studies. Superimposed on both of these is a need to understand the role that other factors such as genetic polymorphisms, gender, and infectious disease play in susceptibility to and expression of autoimmune disease.

Developing useful biomarkers inevitably requires an understanding of the mechanisms underlying autoimmune disease and potential interactions among environmental chemicals, the immune system, and target organs. Two possible types of chemical effects are of concern: initiation of autoimmune disease and exacerbation or acceleration of preexisting autoimmune disease. Biomarkers amenable to both human and animal studies would be ideal. A number of potential candidates were discussed, including the presence of autoantibodies or autoreactive T cells; alterations in T cells, B cells, and macrophages; increased total immunoglobulin G; human leukocyte antigen/major histocompatibility complex markers; and expression of cell surface proteins assessed by flow cytometry, serum enzymes, and cytokine profiles. There may be others as well. It was suggested that clinical trials (e.g., stage III drug trials) or groups exposed occupationally (e.g., to silica, pesticides, or anesthesia), as well as animal models of certain diseases may provide opportunities to address this issue. The most expedient solution would be the identification of biomarkers universal to all types of autoimmune disease; however, there are likely multiple mechanisms involved in the development of different autoimmune diseases. These different mechanisms will probably result in the need to develop different assays and biomarkers for certain autoimmune diseases. The complexity of autoimmune disease(s) is not unlike that associated with cancer. Certainly, the identification of mechanisms common to more than one type of autoimmune disease would yield the most useful biomarkers. For example, with the advent of cDNA microarray technology, it may be possible to identify patterns of gene expression that meet this criterion.

This article is based on a presentation at the Workshop on Linking Environmental Agents and Autoimmune Diseases held 1–3 September 1998 in Research Triangle Park, North Carolina.

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Address correspondence to J.J. Heindel, National Institute of Environmental Health Sciences, Division of Extramural Research and Testing, PO Box 12233 (EC-23), Research Triangle Park, NC 27709. Telephone: (919) 541-0781. Fax: (919) 541-5064. E-mail: heindelj@niehs.nih.gov

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Table 1. Research needs.[a]

Develop research tools needed to explore environmental agents/autoimmune disease links
Establish a disease registry or surveillance system.
Develop and validate strategies for screening chemicals
Develop an emergency response strategy to gain information from accidental exposures
Conduct hypothesis-driven research in exposed groups and/or in experimental animals

[a]The foremost need (encompassed in the first four needs listed above) is to develop a preliminary database. The field cannot move rapidly into hypothesis-driven research without preliminary data on which agents and diseases are important.

Validated questionnaires are needed for use in cohort and other types of epidemiology studies to assess exposure and both organ-specific and systemic effects that might be associated with chronic, low-dose (or occasionally acute, high-dose) exposures. Again, questionnaires need to consider two possibilities: initiation, or possible exacerbation/acceleration of autoimmune disease. In the first case the population at large may be at risk. In the second case a genetically sensitive subpopulation is at risk. Another concern is that autoimmune disease associated with chemical or drug exposure may differ somewhat from spontaneously occurring autoimmune disease; a related concern is that diagnostic criteria used for clinical research (prognosis and therapy) may not be appropriate or optimal for etiologic research.

Establish a nationwide surveillance network. Population-based registries for autoimmune diseases would help to identify clusters of cases potentially associated with environmental exposures as well as sensitive populations. Because individually most autoimmune diseases are relatively rare, surveillance of this type is needed to define the scope and magnitude of the problem. Essential to establishing such a surveillance network is a working definition (diagnostic criteria) of autoimmune disease. In developing a definition, the possibility should be considered that autoimmune diseases associated with chemical exposures may not be identical to spontaneous disease as mentioned above. There are a number of approaches that may be used to develop registries, including state-based or disease-specific registries. A variety of designs should be explored, as it is unclear which approaches will be most useful. Ultimately, strategies must be designed that include rural and minority populations. Registries are necessary to stimulate and facilitate research and reduce the likelihood that biases in selection of patients will produce inaccurate study results. A related activity, the development of cell (DNA) and sera banks on selected populations, would also facilitate research with respect to genetic predisposition, for instance.

Develop and validate strategies for screening chemicals for the potential to induce or exacerbate autoimmune disease. The goal is to develop a testing strategy, most likely in laboratory rodents, that could be used to assess chemicals for the potential to increase risks associated with autoimmune disease. The National Toxicology Program (NTP) (*1*) testing battery that is used to assess immune suppression is an example of such a strategy.

The first step is to develop tools for hazard identification. Few tests amenable to screening are currently available. During the workshop there was considerable discussion of the popliteal lymph node assay (PLNA) and the modified PLNA using reporter antigens (*2*). There was general agreement that an international, interlaboratory evaluation of these assays would be valuable and should include a broad range of chemical classes. Ideally, both known positives and negatives should be included in the validation of this assay, although identifying chemicals with these characteristics may be difficult. The possibility that a metabolite rather than the parent compound might be responsible for an effect has to be taken into account. Such a validation effort should provide information on reproducibility of the assay as well as an indication of the potential for false negatives and false positives. It is unknown at this point whether this assay can distinguish between initiators and potentiators. The workshop participants recognized that the PLNA, in its current form, identifies immunostimulatory compounds, not specific agents responsible for autoimmune effects. Further development of this assay to more specifically target autoimmune disease should be encouraged. There was also some interest in exploring the potential of other types of assays that detect immune stimulation as possible screening tests to begin to identify agents posing a risk of autoimmune disease. In particular, the recently revised Organization for Economic Co-operation and Development 407 guidelines (*3*) or the NTP-tiered testing approach (*1*) has produced considerable data on immune modulation. Because these tests were designed to assess immune suppression, the data have not been analyzed to determine their value in identifying agents that might cause or contribute to autoimmune disease. However, it may be that for some of these chemicals, stimulation of a response occurred and was probably ignored. Because data from these tests are already available for a number of chemicals and certain of these tests are now being required in some cases (*4*), an analysis of their value in identifying chemicals associated with autoimmune disease should be fairly straightforward and would yield valuable information. One concern is that these assays may need to be repeated in animals that are genetically predisposed to developing autoimmune disease (e.g., NZB/W mice) in order to address the possibility of exacerbation of disease versus initiation. Beyond screening for immunostimulatory effects, a number of the potential biomarkers listed earlier under developing research tools, as well as histopathology on selected organs, may also prove useful for screening purposes. In addition, there was a suggestion that it is possible to develop *in vitro* assays that provide a preliminary screening test.

Finally, it is necessary to continue to evaluate the use of currently available animal models of autoimmune disease and to develop additional animal models that may be used to identify environmental agents that potentiate systemic and organ-specific disease. Again, this requires a clear definition of autoimmune disease against which to assess the appropriateness of the animal models. Guidelines to be used in selecting appropriate animal models for testing suspect chemicals also must be developed. These guidelines should consider chemical structures, any observed clinical manifestations, types of immunostimulatory responses observed in screening studies, genetic predisposition, gender, and other issues. Animal models ultimately could be used to identify genetic targets and polymorphisms in these genes that could alter susceptibility. The presence of similar polymorphisms in homologous human genes would certainly support the appropriateness of an animal model.

Once testing strategies have been developed, an assessment is needed to determine whether rodents are good predictors of the potential to induce or potentiate autoimmune disease compared to clinical and epidemiologic data. Other issues more easily addressed after the above issues have been resolved include consideration of the most appropriate tests/methods to assess autoimmune effects that might result from prenatal exposures or from longer term chronic exposures, particularly in the elderly. Also, once a database is established, it is important to consider structure–activity relationships associated with autoimmune disease.

Develop an emergency response strategy. On occasion there have been large-scale accidental exposures (toxic oil) or unexpected effects from intended exposures (L-tryptophan) that may have yielded more information had someone been prepared to assess the situation in a timely fashion. A protocol for responding to such incidents must be in place

in advance, along with the means to rapidly obtain support, both financial and other (e.g., rapid institutional review board approval). Both clinical and epidemiologic approaches as well as exposure assessment must be included in such a strategy. This issue is not unique to autoimmune disease effects, and any emergency response strategy could be extended to include immune modulatory effects in general. Because of the resilience of the immune response, it is important to assess effects as soon as possible after exposure and to conduct exposure assessment. A result of such emergency response studies is the generation of hypotheses that could then be tested in controlled animal studies or perhaps in the exposed population.

Conduct hypothesis-driven research in occupationally exposed groups and/or in experimental animals. Hypotheses generated from case reports, clusters identified in surveillance studies, and the previously mentioned emergency response efforts must be tested in a strictly controlled fashion a. Identification of autoimmune effects after chemical exposure in animal studies should result in the development of hypothesis-driven human epidemiologic or clinical studies. Wherever possible, attempts must be made to link human and animal research. For example, considerable data describing the development of thyroiditis in animals exposed to dietary iodine were presented at this meeting (*5*), but no epidemiologic studies to assess the association between dietary iodine and thyroiditis in humans have been conducted. Similarly, animal data suggest an association between certain chemical exposures (e.g., mercury, trichloroethylene) and autoimmune disease, but there are no human data to substantiate these results (*6–8*). On the other hand, the association between certain chemical exposures (e.g., vinyl chloride; silica) and scleroderma has been studied in clinical settings (Parks et al., this monograph (*9*), but currently there are no data from animal models that test the hypothesis that exposure to these chemicals contributes to this disease. It is clear that some coordination of human and animal studies is needed to maximize our ability to use the data to assess the risk of autoimmune disease that might be associated with chemical exposure.

A number of major research questions were raised during the discussions that should be addressed in a hypothesis-driven manner: Are there gender differences in susceptibility to specific environmental agents? What genetic polymorphisms are critical in the development of autoimmune disease following chemical exposure? What is the role of the endocrine system? Are there distinct pathways leading to distinct systemic autoimmune diseases or do systemic autoimmune diseases share common etiologic risk factors? Are there clinical and/or mechanistic differences between idiopathic and chemically induced autoimmune diseases? Can exposure to an environmental agent exacerbate the natural history of an autoimmune disease? Do environmental factors (other than infectious agents) play any role in the development of type I diabetes? Does *in utero* exposure pose a risk of subsequent development of autoimmune disease? If so, what are the most sensitive periods for exposure? Does exposure during other developmental stages (neonatal, prepubertal, adult, aged) influence the risk of developing autoimmune disease? Is there synergy between viral infection and environmental chemicals in the development of autoimmune disease? There are certainly other questions that could be raised as well.

A Final Recommendation

There was consensus that meetings like this one and projects that facilitate interactions between specialties should be encouraged. Funding should be targeted to integrated studies that use multidisciplinary approaches to improve overall knowledge of the hazard, mechanism of action, and human health consequences associated with environmental agents and autoimmune disease. Meetings of this type should be held periodically as a means of exchanging information and focusing research to gain maximum benefit.

Addendum

The congressional budget for the National Institutes of Health (NIH) for 1999, which came out shortly after this conference, appropriated $30 million for research on autoimmune diseases. One project that will use this money is a Request for Applications (RFA), jointly sponsored by the National Institute of Environmental Health Sciences and several other NIH institutes, that is a direct result of the research ideas put forth at this conference and detailed above. The RFA "Environment-Infection-Gene Interactions in Autoimmune Diseases" will fund pilot/feasibility studies to determine the role of environmental agents (chemical, physical, infectious) and genetics in the initiation and progression of autoimmune diseases.

References and Notes

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