Sex differences in autoimmune disease

Article *in* Nature Immunology · October 2001

DOI: 10.1038/ni0901-777 · Source: PubMed

1 author:

Caroline C Whitacre
The Ohio State University
107 PUBLICATIONS **6,496** CITATIONS

Autoimmune diseases are more prevalent in women than men. A new interest in understanding the biology of this difference as well as funding opportunities have focused attention on research priorities in sex differences.

Sex differences in autoimmune disease

Caroline C. Whitacre

Department of Molecular Virology, Immunology and Medical Genetics, The Ohio State University College of Medicine and Public Health, Columbus, OH 43210-1239, USA. (Whitacre.3@osu.edu)

The autoimmune diseases include more than 70 chronic disorders that affect ~5% of the US population, a population in which these diseases have been intensively tracked and studied. Well over 100 years ago, when the earliest descriptions of systemic lupus erythematosus (SLE) and multiple sclerosis (MS) were recorded, it was noted that women are affected more often than men. A compilation of some of the more common autoimmune disorders with their sex distribution and incidence figures is shown (**Fig. 1**). The most striking sex differences are observed in Sjogren's syndrome, SLE, autoimmune thyroid disease (Hashimoto's thyroiditis and well as Graves' disease) and scleroderma, which represent a spectrum of diseases in which the patient population is >80% women[1]. There is a middle tier of relatively common diseases that includes rheumatoid arthritis (RA), multiple sclerosis (MS) and myasthenia gravis, in which the sex distribution is 60–75% women. A final group, which includes sarcoid, the more common inflammatory bowel diseases and immune-mediated (type 1) diabetes (also known as insulin-dependent diabetes mellitus or IDDM), are characterized by a female:male ratio that is approaching 1:1. In addition to a difference in prevalence between women and men, it is also recognized that the two sexes exhibit differences in disease presentation.

The study of autoimmune diseases has intensified within the past two decades, paralleling the virtual explosion of information and research conducted on the immune system. With an increased understanding of innate immunity, adaptive immune recognition, lymphocyte activation and the principles of immune tolerance, the tools are in place for creative approaches to the treatment and prevention of autoimmune diseases.

In any discussion of differences between men and women, one invariably runs into the debate over use of the words "sex" and "gender". In the biomedical literature, and that dealing with autoimmune diseases is no exception, these two terms are used interchangeably. In fact, the term "gender" is probably used more often, to get away from the reproductive connotation associated with the word "sex". This is almost certainly incorrect. The origins of these terms date to the mid-20th century when scholars spoke of a person's "sex" as being those characteristics attributed to biology, such as sex chromosomes, hormone concentrations and the physiology of sex organs[2]. This is in contrast to the term "gender", which refers more to a cultural and social framework that encompasses social interactions, social hierarchies and cultural practices, that is, how one is viewed. In a discussion of autoimmune diseases, a treatment of both sex differences and gender differences is warranted. Here, only "sex" differences will be discussed and will refer to biologically determined properties.

Increased focus on sex differences

Even though the female prevalence in autoimmune diseases has been recognized for over 100 years, much attention and research funding has only recently been focused on this area. What were the key events that led to such increased interest in this topic? A shift in philosophy and a series of events occurred that had an impact on the study of sex differences as they relate to autoimmune disease.

First, the way in which autoimmune diseases are viewed has subtly changed. In the past, each autoimmune disease was considered individually and had separate medical specialties and funding organizations (for example, the National Multiple Sclerosis Society (NMMS), Arthritis Foundation and Lupus Foundation). Researchers working on one autoimmune disease tended to focus on research questions pertaining only to that disease and target organ (with some notable exceptions). Because any single autoimmune disease is relatively rare, this approach resulted in isolated pockets of research activity, with some diseases intensively studied, whereas others were relatively neglected. During the 1990s it was realized that similar immune mechanisms were operative in more than one autoimmune disease. For example, activation of the $CD4^+$ type 1 helper T (T_H1) cells was shown to be important

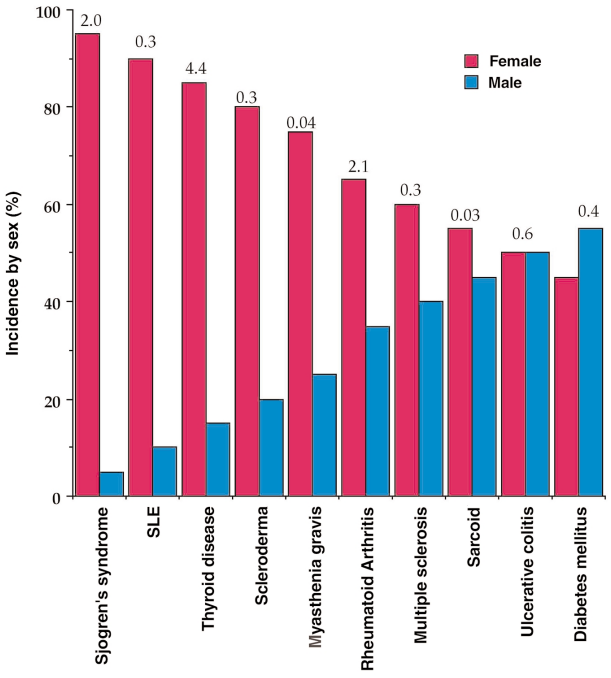

Figure 1. The sex distribution of the major autoimmune diseases. The numbers above the bars refer to the total number of disease cases (×1,000,000) in the USA[1,7].

in the pathogenesis of RA, MS and IDDM, although the antigenic specificities of the cells in the various diseases are quite different. The cells themselves, as well as their cytokine profiles and the overall cytokine milieu of the target organ, were shown to be important parameters in disease induction, with similarities between disease states. At the genetic level, additional similarities have been recognized. A genome-wide screen of RA sibling pairs identified several genetic regions for RA that also contribute to the overall genetic risk of SLE, inflammatory bowel disease, MS or ankylosing spondylitis[3].

Accompanying this shift in philosophy was the formation of the American Autoimmune-Related Diseases Association (AARDA), which promoted: "… bringing national focus to autoimmunity as a major health issue, and promoting a collaborative effort among researchers in order to find a cure for all autoimmune diseases". Learning from the cancer field, this organization advocated combining individual efforts to achieve a greater impact and visibility. In so doing, the number of individuals advocating collectively for increased research on autoimmune diseases approached 15–30 million.

Along with the change in philosophy that united research on autoimmune diseases, a series of independent events helped to further fuel the effort to expand research funding in the area of sex and gender differences in autoimmune diseases. The US National Institutes of Health (NIH) sponsored conferences devoted to autoimmune disease, specifically gender and autoimmunity, which brought together active researchers in the field. One particular conference was followed by an NIH Program Announcement in 1996 on "Gender in the pathogenesis of autoimmunity: mechanism", which was sponsored by six different NIH institutes under the leadership of the National Institute of Allergy and Infectious Diseases.

During 1996 and 1997, the NIH Office of Research on Women's Health held a series of regional and national meetings to review the NIH scientific agenda for research in the area of women's health. More than 1500 scientists, clinicians, social scientists, public policymakers, legislators and advocates came together to examine the achievements to date and directions for research in women's health for the future. The six-volume report from these meetings highlighted the importance of sex differences in normal and abnormal immune function and proposed determining the effects of sex steroid hormones on the immune response[4].

Another key initiative was the formation of a Task Force on Gender, Multiple Sclerosis and Autoimmunity by the NMSS in 1997. This task force brought together researchers and NIH program directors from diverse fields; these individuals specialized not only in MS but also in RA, SLE and scleroderma. The task force reviewed the sex-difference literature[5], developed a research agenda for the field[6] and made recommendations on funding initiatives that have led to NMSS expenditures of more than $2.2 million for research on sex and gender differences.

In fiscal year 1998, the United States House and Senate Appropriations Committee called for the Director of the NIH to convene a coordinating body for the study of autoimmune diseases, which would have the aim of synergizing research among the NIH institutes in this area. The Autoimmune Diseases Coordinating Committee, established in 1998, brought together representatives from the 22 NIH institutes offices and centers, the Food and Drug Administration, the Veterans Administration, the Centers for Disease Control and private organizations sponsoring research in the area of autoimmune diseases. The report of this committee[7] identified six areas of particular research opportunity, one of which was "gender and autoimmunity". In fiscal year 1999, Congress earmarked $30 million in new appropriations to expand support of autoimmunity research, bringing the NIH total for that year to $393 million. This money, particularly the new dollars, supported seven new NIH initiatives and expanded the scope of other ongoing activities. Prominent among the new programs were two new initiatives led by the NIAID: the Autoimmunity Centers of Excellence and the Immune Tolerance Network, a clinical research program that represents a consortium of institutions in the US and abroad for the purpose of the clinical evaluation of promising new tolerance therapies.

In November 1999, the Institute of Medicine formed a blue-ribbon panel named the Committee on Understanding the Biology of Sex and Gender Differences, which evaluated the current understanding of sex differences and its implications for disease and biology. This group worked for 1 year to determine the knowledge base, evaluate barriers to the conduct of research, and develop strategies to overcome those barriers. Their report was recently released and contains 14 recommendations for research priorities as well as addressing barriers to progress[8]. Prominent among those recommendations were "… to monitor sex differences and similarities in all human disease".

Sex differences and autoimmunity: the background

Basic immune responses differ between females and males, with most of the evidence gathered from work done in rodents. After immunization, female mice produce more antibody and show more vigorous T cell activation than male mice[9,10]. Similar approaches in humans, in which responses to vaccination were tested, have yielded mixed results, with either no differences shown or an increased antibody response in females[11,12]. It is notable that women have higher absolute numbers of $CD4^+$ lymphocytes relative to men[13], which likely contributes to their increased responses. Direct comparisons of cytokine production under conditions of immunization have shown higher production of T_H1 cytokines in females[14]. Cytokine secretion is generally enhanced *in vitro* in the presence of estrogen—observed most prominently with interferon-γ (IFN-γ), interleukin 1 (IL-1) and IL-10—and decreased in the presence of androgens (IFN-γ, IL-4 and IL-5)[15–17].

The increased prevalence of autoimmune disease in women, the sexual dimorphism of the immune response and the modulatory effects of sex steroids on immune function *in vitro* have focused attention on the role of these hormones—mainly estrogen, progesterone and testosterone—as primary mediators of the sex differences. Perhaps the most striking evidence comes from pregnancy, in which estrogen and progesterone increase greatly during the third trimester. In both MS and RA, disease activity decreases throughout pregnancy, but most profoundly during the third trimester, when estrogen and progesterone concentrations are highest[18,19]. This is often followed by a flare of disease activity during the *post-partum* period, when estrogen and progesterone concentrations fall. These observations are in contrast to SLE, which appears to either worsen or remain unchanged during pregnancy[20–22].

This fluctuation of disease activity during and after pregnancy has been explained by a hormonal environment during pregnancy that favors a T_H2 response. In MS and RA, this environment may suppress the ongoing T_H1 responses to central nervous system and joint antigens, whereas in SLE, a T_H2 environment would enhance antibody production and possibly exacerbate disease progression. Interestingly, men with RA have significantly lowered testosterone concentrations[23]. An alternative hypothesis to explain changes in disease during and after pregnancy has examined the genetic relationship between mother and offspring. Maternal cells remain present in the offspring and *vice versa*: offspring lymphoid cells have been identified in the maternal circulation years after birth, a situation referred to as microchimerism[24]. In situations of RA improvement during pregnancy, the children were more often disparate from their mothers at the HLA major histocompatibility complex

(MHC) class II loci, which suggested that immunological recognition of the paternal MHC class II caused modulation of the maternal autoimmune response[25].

Many of the findings in humans with autoimmune disease have been borne out in animal models. For example, animals with collagen-induced arthritis and experimental autoimmune encephalomyelitis (EAE) have decreased signs of disease during pregnancy with exacerbations of clinical disease activity *post-partum*[26]. Duplication of the hormonal environment of pregnancy with estriol pellets also suppresses EAE[27]. Manipulation of the hormonal environment by castration of mice and rats shifts the pattern of disease in IDDM, RA and EAE[26]. For example, castration of male NOD (nonobese diabetic) mice increases the frequency of IDDM, whereas oophorectomy of females decreases the incidence of disease. Injection of mice with the male hormone testosterone also suppresses EAE[28].

The modulatory effects of estrogen seem to be quite different between normal immune responses measured *in vitro* and autoimmune responses observed *in vivo*, with enhancement of the former and apparent suppression of autoimmunity. This dilemma was partly resolved with the realization that estrogen shows biphasic dose effects: lower doses facilitate immune responses and higher doses, as occur in pregnancy, suppress such responses[16].

Sex steroids may act directly on the immune system, modulating aspects of antigen presentation, lymphocyte activation, cytokine gene expression and/or homing of immune cells. The identification of estrogen and androgen receptors on immune cells provided a means for direct communication. Sex steroids also have indirect effects that must be considered. Sex hormones modulate the hypothalamic-pituitary-adrenal (HPA) axis and, thus, modulate the stress response, as oorphorectomy results in decreased corticosterone concentrations, whereas orchidectomy enhances the corticosterone response[29]. Females of many species, including humans, have higher corticosterone-cortisol concentrations than males do[29,30]; in addition, glucocorticoids suppress the production of sex hormones and the action of these hormones in tissues. The sharp spike of corticotropin-releasing hormone (CRH) and cortisol at parturition undoubtedly participates in the decline of estrogen postpartum. The discovery of an estrogen-response element in the promoter region of the gene encoding CRH indicated that these two hormone systems are inter-regulated. Therefore, the interactions between the sex hormones, HPA axis and immune system is complex: all these factors must be considered when studying the sex differences in autoimmunity (**Fig. 2**).

Genetics plays a key role in defining autoimmune disease susceptibility and determining the expression of sex hormones and neuroendocrine factors. In an individual with a susceptible genotype, exposure to environmental factors (such as sunlight, diet, allergens, infectious agents or environmental toxins) can act to initiate an autoimmune process. Critical modulating factors that can make the difference between disease expression or not include sex hormones, which can act reciprocally with hormones of the HPA axis or sympathetic nervous system. All these factors together affect the immune response to self and foreign antigens through modulation of cytokine production and effector cell function. The nature of the antigen and the character of the immune response—that is, T_H1 or T_H2—dictate the outcome of the immune response.

Research priorities

To understand sex differences in autoimmunity, it is first necessary to thoroughly understand sex differences in the basic immune response. Although some work has been done in this area, much more research is needed to understand the extent of the differences, from the earliest stages of antigen exposure to the final effector stages of immunity, in response to exogenous and self-antigens. Thus, differences between the stimulation of male and female innate immune responses by pathogens must also be studied. Within the adaptive immune system, processes such as lymphoid and myeloid cell development, antigen processing and presentation, cytokine production, natural killer cell function, tolerance induction and the regulatory influences on these processes need further clarification in males *versus* females.

High among research priorities in the area of sex differences is determination of the mechanisms by which the primary sex hormones

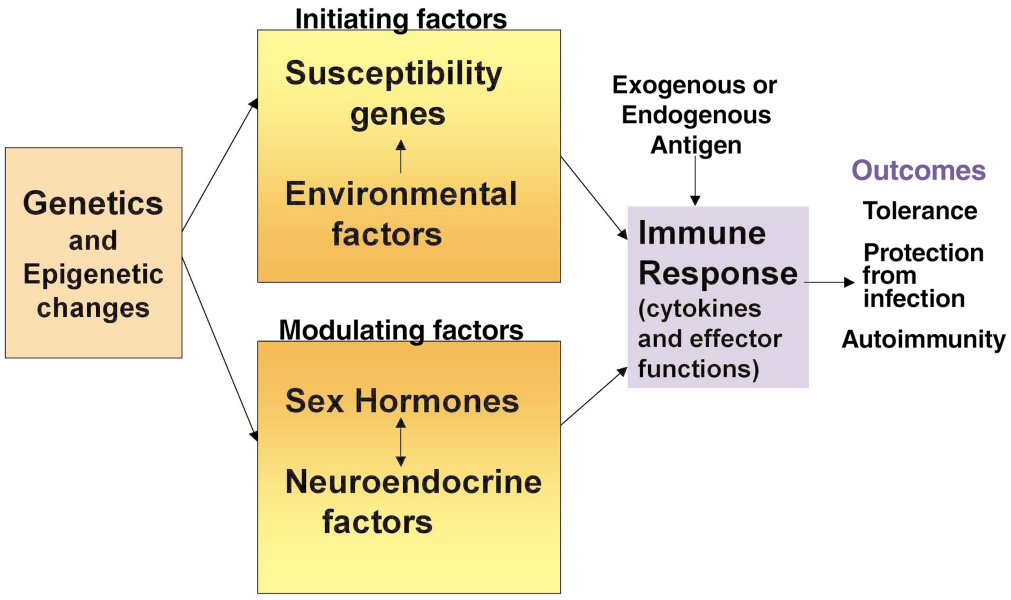

Figure 2. A model for the multifactorial nature of autoimmune disease. Sex hormones represent an important modulatory factor in the immune and autoimmune response. Sex hormones include the gonadal sex steroids as well as other hormones that indicate differences between men and women.

(estrogens including estriol, progesterone and testosterone) affect immune function. It is also important to extend mechanistic studies to other sexually dimorphic hormones, such as prolactin, growth hormone and insulin-like growth factor. Data on the effects of the sexually dimorphic hormones and hormone fluctuations on various phases of the immune response will be insightful.

Another area of high research priority is understanding the effects of naturally fluctuating hormone concentrations on the immune and autoimmune responses. The dramatic changes in disease activity during and after pregnancy that are documented for RA and MS provides an opportunity to follow disease-relevant immune responses over a relatively short time-frame. It is essential to determine the events that cause the *post-partum* flares of disease activity. Other opportunities for studying the effects of hormones on immune and autoimmune responses are during the menstrual cycle, oral contraceptive use and estrogen replacement therapy. It will be important to focus on the interplay of hormone systems, that is, sex steroids and stress steroids (CRH and cortisol), because both profoundly change during pregnancy and parturition and can affect the immune response.

The contribution of genetics to sex differences in autoimmune disease is currently unexplored. Genetic effects certainly operate at the level of the MHC in determining susceptibility to autoimmune disease, but the role of sex hormones in regulating these genes is not known. Now that the human genome sequence is known, specific hormone response elements can be localized and new hypotheses generated about the contribution of steroid hormones to genetic regulation. A new area in need of further research is the interaction between genetic and environmental factors.

Human studies of autoimmune disease, which will be crucial to the success of the priorities outlined above, will rely on data-sharing between centers. The incidence of many autoimmune diseases is low, so that any one center may not have a sufficient number of individuals meeting the criteria for entry into studies. In addition, it is hoped that the increased attention garnered by the sex difference area will promote inclusion of additional arms in clinical trials; for example, trials previously restricted to women may now include a male arm for comparison.

Never has there been greater interest and funding opportunities in the area of sex differences in autoimmune disease than now. At present, there is an NIH request for applications on “Sex-based differences in the immune response”, which is jointly funded by three NIH institutes, the Office of Research on Women’s Health and the NMSS[29]. This initiative calls for research that is broadly inclusive of all autoimmune diseases. Such funding initiatives, together with separate thrusts by specialized groups (in RA, SLE, IDDM and MS), provide an opportunity for significant research advances in the area of sex differences in autoimmunity.

1. Beeson, P. B. *Am. J. Med.* **96**, 457–462 (1994).
2. Fishman, J. R., Wick, J. G. & Koenig, B. A. *Agenda for Research on Women’s Health for the 21st Century: NIH Publication 99-4385*. Vol.2, 15–19 (NIH, Bethesda, MD, 1999).
3. Jawaheer, D. *et al. Am. J. Hum. Genet.* **68**, 927–936 (2001).
4. Collier, E., Diamond, B., Weinblatt, A. C., Whitacre, C. & Mitchell, K. C. *Agenda for Research on Women’s Health for the 21st Century: NIH Publication 99-4386*. Vol. 2, 93–100. (NIH, Bethesda, MD,1999).
5. Whitacre, C. C. *et al.* Sex Differences in Autoimmune Disease: Focus on Multiple Sclerosis. *Science* (1999). www.sciencemag.org/feature/data/983519.shl.
6. Whitacre, C. C., Reingold, S. C. & O’Looney, P. A. & Task Force on Gender, Multiple Sclerosis and Autoimmunity *Science* **283**, 1277–1278 (1999).
7. *National Institutes of Health Report of the Autoimmune Diseases Coordinating Committee*. (2000) http://www.niaid.nih.gov/publications/autoimmune.htm.
8. Wizemann, T. M. & and Pardue, M.-L. *Exploring the Biological Contributions to Human Health: Does sex matter?* (Board on Health Sciences Policy, Institute of Medicine, 2001).
9. Eidinger, D. & Garrett, T. *J. Exp. Med.* **136**, 1098–1116 (1972).
10. Weinstein, Y., Ran, S. & Segal, S. *J. Immunol.* **132**, 656–661 (1984).
11. Michaels, R. & Rogers, K. *Pediatrics* **47**, 120–123 (1971).
12. Vranckx, R., Muylle, L., Cole, J., Moldenhaser, R. & Peetermans, M. *Vox Sang.* **50**, 220–222 (1986).
13. Amadori, A. *et al. Nature Med.* **1**, 1279–1283 (1995).
14. Huygen, K. & Palfliet, K. *Cell. Immunol.* **85**, 75–81 (1984).
15. Wynn, D., Rodriguez, M., O’Fallon, W. M. & Kurland, L. T. *Neurology* **40**, 780–786 (1990).
16. Gilmore, W., Weiner, L. & Correale, J. *J. Immunol.* **158**, 446–451 (1997).
17. Araneo, B., Dowell, T., Diegel, M. & Daynes, R. *Blood* **78**, 688–699 (1991).
18. Nelson, J. & Ostensen, M. *Rheum. Dis. Clin. N. Am.* **23**, 195–212 (1997).
19. Confavreux, C., Hutchinson, M., Hours, M. M., Cortinovis-Tourniaire, P. & Moreau T. *N. Engl. J. Med.* **339**, 285–291 (1998).
20. Silver, R. & Branch, D. *Clin. Obstet. Gynaecol.* **6**, 565–600 (1992).
21. Lockshin, M. *Arthritis Rheum.* **32**, 665–670 (1989).
22. Petri, M. *Rheum. Dis. Clin. N. Am.* **20**, 87–118 (1994).
23. Spector, T. D. *et al. Clin Rheumatol.* **8**, 37–41 (1989).
24. Nelson, J. L. *Lupus* **8**, 370–374 (1999).
25. Nelson, J. L. *et al. N. Engl. J. Med.* **329**, 466–471 (1993).
26. Jansson, L. & Holmdahl, R. *Inflamm. Res.* **47**, 290–301 (1998).
27. Kim, S, Liva, S. M., Dalal, M. A., Verity, M. A. & Voskuhl, R.R. *Neurology* **52**, 1230–1238 (1999).
28. Dalal, M., Kim, S. & Voskuhl, R. R. *J. Immunol.* **159**, 3–6 (1997).
29. Wilder, R. *Annu. Rev. Immunol.* **13**, 307–338 (1995).
30. Griffin, A. C. & Whitacre, C. C. *J. Neuroimmunol.* **35**, 53–64 (1991).