

### Meeting Report

## The Emerging Epidemic of Non-Hodgkin's Lymphoma: Current Knowledge Regarding Etiological Factors

Paul H. Levine<sup>1</sup> and Robert Hoover

Environmental Epidemiology Branch, Division of Cancer Etiology,  
National Cancer Institute, Bethesda, Maryland 20892

A workshop on the increase in the incidence of non-Hodgkin's lymphoma was held at the NIH, Bethesda, Maryland, October 22-23, 1991. The objectives of the meeting included an evaluation of the magnitude of the increase and an assessment of how much was likely to be real *versus* artifact; an estimation of the amount of the increase that might be attributable to known or suspect NHL<sup>2</sup> risk factors; and an appraisal of information regarding molecular and immunoregulatory mechanisms involved in lymphomagenesis that could help explain the current trend.

The meeting was opened by cochairs Max Essex (Harvard University, Cambridge, MA) and Pelayo Correa (Louisiana State University, New Orleans, LA), who noted respectively the rapid growth of information on the role of viruses in the etiology of NHL, particularly retroviruses, and the difficulties in investigating the epidemiology of a disease such as NHL, which can no longer be categorized solely on a pathologic basis.

Richard Adamson (National Cancer Institute, Bethesda, MD) reported that between 1973 and 1978 the NHL incidence rate increased by more than 50%, this increase being one of the largest increases for any type of cancer in the United States. These increases have made NHL the sixth most common malignancy for both sexes, and the relationship between NHL and HIV infection will clearly augment the impact of this disease on the American population.

Session 1, on Time Trends and Pathologic Classification, was chaired by Iris O'Conor (NCI) and Gregory O'Connor (Loyola University, Chicago, IL). In the first presentation, Susan Devesa (NCI) summarized the data from the Surveillance, Epidemiology and End Results Program, noting that over the last 20 years the reported increase was almost 4% per year for males and 3% for females. The most striking increases were apparent among those born in the early 1900s. As part of this extensive review, Devesa noted that this increase was mirrored in a number of registries around the world and was paralleled by an increase in mortality, which was more prominent in the northern and midwestern United

States than in the South. Raymond Cartwright (University of Leeds, Leeds, England) reviewed the time trends and geographic patterns in the United Kingdom, noting a similar increase in incidence in that country and a geographic variation suggesting an environmental impact on low-grade histological subtypes of NHL that was not apparent for high-grade histological subtypes. Theodore Holford (Yale University, New Haven, CT) reviewed in detail the incidence rates from Connecticut for the period 1935-1989, looking for possible artifacts and confounders leading to an inappropriate interpretation of the data, concluding that the magnitude of the trend in Connecticut (10.3% every 5 years for females and 9.2% every 5 years for males over this time period) was of too great a magnitude to be entirely attributed to artifact.

Elaine Jaffe (NCI) initiated a discussion of the pathology aspects by reviewing the evolving classification of NHL, noting that the Rappaport classification, which was based solely on morphological features of the tumor, has given way to more sophisticated laboratory-oriented features initially involving immunophenotyping and now including virological and molecular markers. The need to reevaluate the previous lymphoma terminology was illustrated by her discussion of the recently described mantle cell lymphoma, which falls within several categories of the working formulation, and her observation that the small cell lymphoma group includes several different disease entities.

Peter Banks (University of Texas, San Antonio, TX) supplemented Dr. Jaffe's report emphasizing the impact of immunophenotyping, noting that a significant number of immunologically defined peripheral T-cell lymphomas would previously have been classified as Hodgkin's disease, raising the issue of investigating the possibility of misclassification between Hodgkin's disease and NHL as a contributing factor in the apparent increase in NHL incidence. Dennis Weisenburger (University of Nebraska, Omaha, NE) reported the application of more modern immunopathological classification to an epidemiological study of NHL in Nebraska relating specific histological subtypes to herbicide exposure. He also documented pathology review studies in Minnesota and Iowa which found misclassification of tumors by subtype as high as 60% for diffuse lymphomas. In the discussion, it was noted that not only were histopathological classifications changing but that etiologically determined categories (such as HTLV-I-associated leukemia/lymphoma) had more than one histopathological presentation. It was also noted that a comparison between the morphological variants included in the 1982 Working Formulation and the more recent classifications, which include newly recognized variants such as mantle cell lymphoma and MALT (B-cell lymphomas of mucosa-associated tissues), underscored the difficulty of identifying trends in specific

Received 7/10/92.

<sup>1</sup> To whom requests for reprints should be addressed.

<sup>2</sup> The abbreviations used are: NHL, non-Hodgkin's lymphoma; HTLV-I, human T-cell lymphotropic virus type I; EBV, Epstein-Barr virus; HIV, human immunodeficiency virus; NCI, National Cancer Institute; CFS, chronic fatigue syndrome.

NHL subgroups over time but were not likely to be responsible for the apparent increase of NHL in the past two decades.

Session II on Genetic and Environmental Determinants was chaired by Nancy Mueller (Harvard University) and Joseph Fraumeni (NCI). Alexandra Filipovich (University of Minnesota, Minneapolis, MN) reviewed the data from the Immunodeficiency Cancer Registry concerning lymphoma in genetically determined immunodeficiency diseases, and Martha Linet (NCI) approached the genetic aspects by reviewing reports of familial aggregations of lymphoma. More than three-quarters of the tumors in the Minnesota registry are lymphomas and leukemias, and while genetic predisposition and overt immunodeficiency are unlikely to be contributing to the marked recent increase in NHL incidence, the opportunity for studying mechanisms of lymphomagenesis in these patients may lead to a better understanding of lymphomagenesis in the general population. Leo Kinlen (Oxford University, Oxford, England) reviewed the available data on acquired diseases with altered immunity and therapeutic drug exposures, including a study of approximately 5000 organ transplantation patients followed in Britain, Australia, and New Zealand. In his evaluation of the data, Kinlen raised the possibility that mechanisms other than immunodeficiency could be responsible for the 25- to 50-fold increase in cancer associated with immunosuppressive therapy. Robert Hoover (NCI) noted that immune stimulation had been suggested as contributing to the increase in NHL in some of these patient groups, but he also pointed out that the risk of NHL in autoimmune disease was selective—substantially elevated in Sjogren's syndrome and rheumatoid arthritis but increased minimally or not at all in others, such as lupus erythematosus. The role of viruses in the etiology of NHL was discussed by Mueller, Alexandra Levine (University of Southern California, Los Angeles, CA), and William Blattner (NCI). Mueller first discussed HTLV-I, observing that the 1:1 male:female sex ratio of adult T-cell leukemia/lymphoma in view of the higher prevalence of infection in women *versus* men supports the importance of perinatal infection compared to the sexual transmission of the virus in the etiology of this malignancy. The role of EBV and the observation of elevated EBV titers as a predictor of lymphoma development was also discussed, but Mueller concluded that the cofactors impacting on the effect of viruses on the host were more likely to be involved in the increase in NHL incidence than any change in the pattern of virus infection *per se*. The impact of HIV on the recent increase in NHL was discussed by Levine, who noted that high-grade lymphomas were the major NHL type associated with the acquired immune deficiency syndrome epidemic and that she saw no evidence of a change in the pattern of low-grade lymphomas in California. William Blattner (NCI) discussed the integration of virological, immunological, and epidemiological studies, exemplified by a case-control study in Jamaica which noted that farming was an important risk factor for T-cell lymphoma, many of which were also HTLV-I associated. He also commented on the impact of HIV on the increasing incidence of NHL, noting data from the studies indicating that as many as 27% of the lymphomas in 1992 could be HIV related.

Shelia Zahm (NCI) and Neal Pearce (Wellington School of Medicine, Wellington, New Zealand) pre-

sented the accumulating data that pesticides may be playing a significant role in the etiology of NHL. Their studies in both countries suggest that pesticides may be contributing to the observed increase in NHL over time. The specific compounds associated with NHL have not been definitively identified thus far. Among the major concerns are the phenoxy herbicides, including 2,4-D, which is estimated by the Environmental Protection Agency to be applied (with related compounds) at an annual rate of 60 million pounds per year in the United States. In the reports investigating other possible environmental contributions to the increase in NHL, John Boice (NCI) reported that he could find no evidence that radiation was a significant etiological factor in NHL. Scott Davis (Fred Hutchinson Cancer Research Center, Seattle, WA) reviewed the available data on nutritional studies, and while no nutritional link to NHL has been reported, this could be due in part to a dearth of studies in this area. Davis noted, for example, the intriguing evidence in rodents linking changes in diet to abnormalities of the immune response and concluded that further studies in humans should be undertaken. Aaron Blair (NCI) reviewed a collaborative case-control study investigating occupational exposures to potential carcinogens and including a detailed histopathological review of the associated NHLs. Blair described some of the approaches to job classification relevant to exposures and emphasized the problems of exposure misclassification leading to potential misinterpretation of risk factors. Finally, preliminary reports on four ongoing studies were presented to close the session. Jonathan Buckley (University of Southern California, Los Angeles, CA) reviewed the data obtained by the Children's Cancer Study Group from approximately 100 hospitals in the United States, Canada, and Australia, examining a number of risk factors including frequency of infections and history of allergy. Thus far, no significant risk factors have emerged from the analysis other than data which support the previously reviewed studies that pesticides and herbicides could be associated with an increased risk of developing NHL. Paul Scherr (Centers for Disease Control, Atlanta, GA) reviewed a case-control study involving nine Boston hospitals between 1980 and 1982. The major occupational groups associated with an increased risk of developing NHL were agriculture (forestry), fishing, and construction. In this study, there appeared to be a correlation between specific exposures and histological subtypes of NHL: histiocytic lymphoma with pesticide exposure and diffuse large cell lymphoma with pesticide and dust exposure. Leslie Bernstein (University of Southern California, Los Angeles, CA) reviewed a study of NHL in Los Angeles County completed in 1984, and of the many risk factors analyzed, significant associations were found with macrodantin and furadantin and long-term use of steroids, while eczema, oral contraceptives, and pregnancy appeared to have a protective effect. Finally, Paul Levine (NCI) reviewed a study investigating CFS as a possible risk factor for NHL. Although anecdotal reports had suggested an association between outbreaks of CFS and the subsequent development of NHL, no significant statewide trends in NHL could be linked to the 1984-1986 outbreak of CFS reported in northern Nevada.

Session III on Molecular and Immunoregulatory Mechanisms was chaired by Michael Potter and Ian McGrath (NCI). Potter provided an overview on the devel-

opment of B-cells and lymphomagenesis, noting that the concepts regarding B-cell lineage are changing with a greater appreciation of the significant input from peritoneal B-cells. He stressed the importance of investigating the complex series of events in lymphoma pathogenesis, such as the role of chromosome translocations as a mutational process in lymphomagenesis. Michael Williams (University of Virginia, Charlottesville, VA) reported on his studies of chromosomal break points in centrocytic lymphomas and discussed the possible relationship between *bcl-1* and cyclin D1, noting the possibility of a cell cycle-related gene linked with lymphomagenesis. Ilan Kirsch (NCI) reviewed recent studies on chromosomal aberrations in NHL, reporting a significant increase in apparently specific chromosome translocations in the lymphocytes of farmers exposed to herbicides and insecticides. Ronald Levy (Stanford University, Palo Alto, CA) discussed the evidence that the development of follicular lymphomas is driven by the presentation of autoantigens, this process having its effect in the maintenance and progression of the tumor rather than necessarily being involved in initiation. John Tew and Andras Szakal (Medical College of Virginia, Richmond, VA) discussed aspects of the lymphoid follicle relevant to lymphomagenesis, including the structure and biology of the follicle in aging. Donald Mosier (Medical Biology Institute, La Jolla, CA) then described a mouse model for EBV oncogenesis, observing that EBV-infected cells from healthy human donors were oncogenic in mice with severe combined immunodeficiency disease but that there was extensive variation in the oncogenicity from donor to donor for reasons that remain unexplained. Alfred Evans (Yale University) and Paul Levine (NCI) discussed the increasing availability of laboratory assays using well-characterized serum banks and tumor tissues (including paraffin blocks) to perform retrospective laboratory-oriented epidemiological studies on the etiology of lymphoma. They emphasized the need to preserve biological materials with complete medical and epidemiological background information. The final laboratory-oriented presentation was by Magrath, who used Burkitt's lymphoma as a model for emphasizing the importance of using biological assays to understand the epidemiology of NHL. At least two forms have been identified which are morphologically indistinguishable, one form always being EBV associated and having molecular characteristics such as immunoglobulin gene rearrangements and *C-myc* oncogene activation with features differentiating this form (which is

the predominant form found in sub-Saharan Africa) from the form most commonly found in the United States.

Session IV, chaired by Robert Hoover (NCI), was utilized to review and discuss the information that had been reported in the first three sessions. In the first presentation, Patricia Hartge (NCI) examined the data presented on known risk factors for NHL and attempted to quantify the impact on the time trends. Using the estimates presented by the attendees as to the possible effects of histopathological misclassification, familial factors, occupation, immunosuppressive factors, and the recent impact of HIV infection, more than half of the increase among white men aged 0-64 could be explained. Nonetheless, an estimated 42% increase remained unexplained among males younger than 65 years of age. Exposures to pesticides in the environment, stress, diet, and hair dyes could be contributing to the increased incidence, but in order to account for the residual rise, the exposure(s) would need to have risen from 0% to 42% among white men aged 0-64 and carry an average relative risk of 2. The attendees discussed the possibility that the increase in NHL was in some way related to a general subclinical immunosuppression triggered by an as yet unidentified environmental agent, but thus far there was no evidence that such a phenomenon was occurring. Essex, however, did caution that information from animal models and emerging data relevant to human viruses could be used to support the possible importance of environmental factors, particularly viruses, producing such immunological perturbation on a wide scale. In addition to providing a forum for the development of specific studies in which new laboratory techniques could be linked to epidemiological studies investigating the etiology of NHL, there was a better understanding that the classification of NHL was continuing to evolve with the importance of laboratory characterization as an essential component in subdividing morphologically indistinguishable tumors. To further the effort on identifying the reasons for the disturbing trend in NHL incidence, the NCI is eliciting suggestions from the attendees and others in the scientific community for specific new approaches to addressing this intriguing and important issue.

The proceedings of the meeting, which include discussions of the presented papers and an outline of areas requiring specific attention, will appear in a *Supplement to Cancer Research* in October 1992.