Digestive Diseases Interagency Coordinating Committee (DDICC)
Research in Inflammatory Bowel Disease
Meeting Summary

April 30, 2003
1:00 p.m. - 4:15 p.m.
Natcher Building, Conference Room D
NIH Campus
Bethesda, MD

Participants

Chair:

Jay Hoofnagle, M.D.	National Institute of Diabetes and Digestive and Kidney Diseases

Executive Secretary:

Stephen James, M.D.	National Institute of Diabetes and Digestive and Kidney Diseases

Members:

Nell Armstrong, Ph.D., RN	National Institute of Nursing Research
Leslie Curtis	National Institute of Diabetes and Digestive and Kidney Diseases
Darla Danford, M.P.H, D.Sc.	National Heart, Lung, and Blood Institute
Frank Hamilton, M.D., M.P.H.	National Institute of Diabetes and Digestive and Kidney Diseases
Brian E. Harvey, M.D., Ph.D.	Food and Drug Administration
Mushtaq A. Khan, DVM, Ph.D.	Center for Scientific Review
Vishnudutt Purohit, Ph.D.	National Institute on Alcohol Abuse and Alcoholism
Patricia Robuck, Ph.D., M.P.H	National Institute of Diabetes and Digestive and Kidney Diseases
Jose Serrano, M.D., Ph.D.	National Institute of Diabetes and Digestive and Kidney Diseases
Deborah Willis-Fillinger, M.D.	Health Resources and Services Administration

Guests:

Roger DeRose	Crohn's & Colitis Foundation of America
Dale Dirks	Crohn's & Colitis Foundation of America
James Everhart	National Institute of Diabetes and Digestive and Kidney Diseases
Ivan Fuss	National Institute of Allergy and Infectious Diseases
Barbara Harrison	National Institute of Diabetes and Digestive and Kidney Diseases
Daniel Hollander, M.D.	The Eli and Edythe L. Broad Foundation
Robert Karp	National Institute of Diabetes and Digestive and Kidney Diseases
Gene Kestenbaum	Crohn's & Colitis Foundation of America
Li Liang, MD	Food and Drug Administration
Gavin Lindberg	Crohn's & Colitis Foundation of America
Peter Mannon	National Institute of Allergy and Infectious Diseases
Marjorie Merrick, M.A.	Crohn's & Colitis Foundation of America
Judith Podskalny	National Institute of Diabetes and Digestive and Kidney Diseases
Sharon Pope	National Institute of Diabetes and Digestive and Kidney Diseases
Suzanne Rosenthal	Crohn's & Colitis Foundation of America
Annette Rothermel, Ph.D.	National Institute of Allergy and Infectious Diseases
Clare K. Schmitt, Ph.D.	National Institute of Allergy and Infectious Diseases
Allen Spiegel, M.D.	National Institute of Diabetes and Digestive and Kidney Diseases
Aron "Ron" Yustein, M.D.	Food and Drug Administration

Speakers:

Richard Blumberg, MD	Brigham and Women's Hospital
Charles Elson, MD	University of Alabama School of Medicine
Daniel Podolsky, MD	Massachusetts General Hospital
John Rioux	Whitehead Institute
Robert Sandler, MD, MPH	University of North Carolina School of Medicine
Bruce Sands, MD, MS	Massachusetts General Hospital
Warren Strober	National Institute of Allergy and Infectious Diseases

Welcome and Introductions

Dr. Jay Hoofnagle, Director, Division of Digestive Diseases and Nutrition, National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institutes of Health (NIH)

Dr. Hoofnagle opened the meeting with a description of the Digestive Diseases Interagency Coordinating Committee (DDICC), a committee legislated by Congress to meet four times per year to discuss particular issues with regard to digestive diseases and to coordinate research activities between various institutes and organizations. Dr. Hoofnagle then introduced Dr. Allen Spiegel, Director of the NIDDK, who identified the current meeting's focus to be Inflammatory Bowel Disease (IBD), an area that has experienced tremendous progress in the face of enormous challenges. Participants were invited to introduce themselves and included representatives from government, industry, and non-profit organizations.

Dr. Spiegel stated that the NIH budget has doubled in the past 5 years, and the increase has brought with it a corresponding responsibility to provide justification to Congress that the funding is both reasonable and worthwhile. IBD is an area where such information is forthcoming. Dr. Spiegel introduced the organizer of the meeting, Dr. Stephen P. James, Deputy Director of the Division of Digestive Diseases and Nutrition, NIDDK, who invited the first speaker to the podium to begin the afternoon's presentations.

Overview of CCFA Strategic Plan "Challenges in IBD Research"

Dr. Richard Blumberg, Chief, Department of Medicine, Division of Gastroenterology, Brigham and Women's Hospital, Harvard Medical School

Dr. Blumberg presented an overview of the Crohn's & Colitis Foundation of America (CCFA), an organization with which he has been associated for 10 to 15 years at a variety of levels, and which, together with support from NIH, has nurtured his scientific career.

The CCFA is a national volunteer-driven, not-for-profit organization whose mission statement is "To cure and prevent Crohn's disease and ulcerative colitis through research, and to improve the quality of life of children and adults affected by these digestive diseases through education and patient and family support."

Dr. Blumberg stated that the CCFA's FY 2003 budget is approximately $25.8 million, raised annually by contributions from individual donors. More than 82 cents of every

dollar the Foundation spends goes directly to research and educational programs; approximately $9 million during FY 2003 will be allocated to the National Scientific Advisory Committee (NSAC) for projects related to the scientific mission of the CCFA. The CCFA consistently meets the standards set by organizations such as the National Charities Information Bureau and the Better Business Bureau, and is ranked very highly by a number of philanthropic study groups and The National Health Council.

The CCFA research mission, which is similar to the peer-review analysis pursued at NIH, has remained consistent over the last decade:

- To identify and fund the best peer-reviewed investigator-initiated research in IBD;
- To provide "seed money" to allow investigators to generate enough preliminary data to be competitive at the NIH level;
- To encourage outstanding young investigators toward a career in IBD research; and
- To identify and support emerging areas of research that could have an important impact on the understanding of the pathogenesis and etiology of IBD.

To date, the CCFA has committed over $80 million directly to research. The Foundation receives more than 150 grant applications per year, and currently funds more than 80 ongoing research grants. In this regard, and related to the meeting today, the CCFA has successfully partnered with NIH in the past, and anticipates future cooperative efforts with the Institutes as well as other organizations, including the Broad Medical Research Foundation. Dr. Blumberg pointed out that more than 60 percent of CCFA-sponsored researchers have obtained subsequent funding from the NIH for further IBD research, and greater than 75 percent of Research Training Award recipients are still involved in IBD research, which testifies to the success of both the programs and the peer-review process.

Dr. Blumberg outlined the leadership of the CCFA, and identified the committees within the NSAC that are involved in the determination of research spending direction and additional activities related to the pursuit of the Foundation's mission. Of particular relevance to today's meeting is the Research Initiatives Committee, which has been spearheading the information contained in the Challenges document.

CCFA funding includes the Senior Research Grant ($115,000/2 years), First Award ($69,000/3 years), and a series of research training vehicles designed to aid in career development. In addition, the Research Initiatives Committee is in the process of generating a new series of targeted IBD research initiatives based on the Challenges document.

The CCFA Challenges document is the product of a meeting held in Phoenix, Arizona in May 2002, which drew together section leaders and committees in the areas of both basic and clinical science to prepare a position paper identifying worthwhile aims for IBD research. Promising areas of research included:

- Identification of susceptibility genes in the general population as well as specific

subgroups of IBD (e.g., childhood onset, extraintestinal manifestations, and Mendelian disorders).

- Generation of surrogate markers to define clinical patterns, natural history, and prognosis of IBD.

- Achievement of a greater understanding of regulatory cells and their networks in experimental models and humans, and the biological delineation of the susceptibility genes (most importantly, NOD2).

- Generation of randomized clinical trials (RCTs) in important human disease models (e.g., post-surgical relapse or pouchitis), and to couple these RCTs with translational studies.

- Achievement of a greater understanding of the ecology of microbes that are driving the pathogenic and regulatory responses.

- Generation of rigorous clinical descriptors for molecular classification.

- Development of novel epidemiologic and environmental risk factors and further examination of already identified risk factors.

- Further understanding of biomarkers for dysplasia and cancer, as well as the interplay between microbiota, intestinal epithelial cells and the immune system.

- Consideration of the potential relevance of a consortium focused on mouse genetics to human IBD.

- Understanding the cellular and molecular mechanisms of tissue injury, repair, fibrosis, and fibrolysis.

Immunology and Microbiology of IBD

Dr. Charles O. Elson, University of Alabama, Birmingham

Dr. Elson began by stating that it has been estimated that 2 to 4 million genes are represented in the microbial ecology of the human gut, compared to 30,000 human genes, necessitating daily production of 3 to 4 grams of IgA. Citing data from animal studies, Dr. Elson demonstrated the enormous impact commensal bacteria has on the host, particularly with regard to the gut, including sizable increases in the maturation and turnover of the epithelial cell compartment, and development of the mucosal and systemic immune systems, as well as the enteric nervous system.

Pattern recognition or "toll-like" receptors situated on the surface of cells in the gut signal the presence of bacteria to the host epithelium and immune cells, indicating a dynamic interchange between these different types of cells. Advances in animal models during the past 10 years have provided data showing both effector and (more dominant) regulatory cells exist in the gut that are directed at bacteria. These models continue to examine effective regulation, excessive effector cell function, and epithelial cell function in the gut.

Dr. Elson identified several areas of research that warrant further attention:

- Regulatory and effector cells need to be more closely defined and better understood with regard to their origin, localization site, markers, recognition of antigens, and stimulation processes.

- Investigation of microbial antigens and adjuvants that activate not only pathogenic T cell response, but the generation of regulatory cells as well.

- Identification of successful human intestinal microbiota populations and their interactions with the immune system, currently being investigated using a new technique, gradient gel electrophoresis, a procedure through which the 16s RDNA gene is bacteria is amplified, sequenced and separated.

Dr. Elson stated that the creation of a human and mouse intestinal microbiota catalog will depend on developing appropriate quantitative technologies, and he suggested that NIH is the best suited entity for such an enormous undertaking. Probably more achievable is genome sequencing of the 10 to 20 bacterial species present in the intestine, since each strain requires approximately a day's work for mega-sequencing outfits, and this task could be contracted out to a number of high throughput gene sequence facilities.

Further insight into the mechanisms of tissue injury and repair could provide valuable information for the treatment of fibrosis, which is a major concern for patients with Crohn's Disease.

Dr. Elson emphasized that not only do researchers need to delineate the function of the genes that are identified as being associated with IBD, but the relationship of these genes to the triad of the microbiota, the epithelium, and the immune system must be better understood from within the IBD perspective. Recently developed technologies, such as genetically defined microbes, epithelial cell studies, induced mutant mice models, the availability of gnotobiotic facilities, and gene expression arrays, offer investigators the means for examining the mechanism and manipulation of gene interaction and its implications for IBD research.

Dr. Elson was asked to comment on the variation in microbiota composition between individuals, to which he responded that previous studies suggest that cause and effect in humans will be particularly difficult to determine, but that cataloging and the development of quantitative-like chips for major flora species would aid in understanding dysbiotic flora. Prioritizing sequencing of the intestinal microbes would be best determined by a consensus conference of investigators interested in IBD as well as microbial ecologists with expertise in the major categories of gut bacteria.

It was suggested by one participant that the feasibility of proteomic approaches also be considered as a novel technology or approach, and that the engagement of researchers not typically included in IBD conferences and studies would broaden the understanding and parameter of quantitative research methods.

Dr. Elson was asked to what extent gnotobiotic facilities could be shared regionally. Dr. Elson responded that a large, central facility often presents physical barriers to its use simply because researchers are unable to access it, and that placing a facility in an area where interested investigators would be likely to use it would make it a more effective resource.

The question was raised whether effort and funding put into microbial flora research would be better applied to understanding more about general host immune response rather than the actions of specific bacteria. Dr. Elson replied that response to flora is certainly central to IBD treatment; however, knowledge of particular antigens and mucosal immunology are inter-related fields.

In a final comment, the point was made that some sort of strategy ought to be developed to examine the dynamics between complex flora and the mucosa. It may be that taking a functional assay of the mucosa will provide leads that will ultimately result in a more concentrated understanding of specific genomes and their ecologies.

Genetics of IBD
Dr. John Rioux, Whitehead Institute, Cambridge

During the past 4 or 5 years, the use of a number of approaches demonstrating the very strong genetic component of IBD, Crohn's Disease, and ulcerative colitis has culminated in significant advances in the field of complex traits genetics, and in particular, IBD research. In order to truly capitalize on this new information, Dr. Rioux stated that the challenge is to not only identify genes which confer susceptibility to Crohn's Disease and ulcerative colitis, but to understand how genetic variants predispose individuals to the development of IBD, especially as this knowledge relates to drug treatment response.

Numerous studies using linkage analysis have been conducted with the goal of identifying specific genomic regions implicated in predisposition for complex common diseases. However, Dr. Rioux emphasized that the relatively small number of families involved in any single study, coupled with the modest gene effect, has resulted in sampling biases that have prevented the identification of a single common locus. This deficiency is not insurmountable, and recognizing that every study has limitations, Dr. Rioux proposed two methods for merging data from different studies: 1) meta-analysis, the combination of results from multiple studies (method used by the CCFA-sponsored International IBD Genetics Consortium), and 2) mega-analysis, the combination of genotype data across different studies (such as work currently being done by the NIDDK-sponsored IBD Genetics Consortium).

Dr. Rioux cited studies showing that linkage analysis is no longer considered the most powerful method for identifying IBD genes, and it has since been replaced by the association approach. Association has the strength to identify genes, even if they only have a very modest effect. In fact, the association approach has produced some surprising study results in that association studies conducted with regard to CARD15 susceptibility of Crohn's Disease in European-derived populations were positively replicated in all reported analyses. In addition, genotype-phenotype correlations have recently been undertaken which demonstrate that CARD15 variants are most strongly associated with ileal disease and with stricturing disease, and that of the known variants, none are associated with disease in African-Americans or Asians, which prompts research questions in this area that ought to be addressed. Association research analysis might also be applied to identify the genes underlying the linkage peaks which will become apparent following meta- and mega-analysis of study results.

Dr. Rioux offered an example of a study conducted by his group of hundreds of single nucleotide polymorphisms or genetic variants, which showed that the human genome, despite being 3 billion base pairs in length, travels as units or blocks of DNA. This is an important observation not only from

the perspective of genetic variation in a particular disease study, but also from a biological perspective, in that only a very few variants of any single gene exist in a population.

Dr. Rioux described an international haplotype map currently being sponsored by NIH within the U.S. together with the Japanese, Chinese, and Canadian governments, which will facilitate study of the human genome and how it relates to disease.

Dr. Rioux listed four priority areas of future research as reflected in the Challenges document. First, the identification of IBD genes warrants researchers' attention. Currently, only the CARD15 gene has been identified. Dr. Rioux proposed that research objectives ought to 1) include identifying the genes in the regions of association (IBD5, HLA); 2) identify genes in the region of linkage using the association approach, genome-wide scans and mega- and meta-analyses; and 3) initiation of a genome-wide association study of IBD using the internationally built haplotype map. In order to accomplish these goals, Dr. Rioux suggested a large, well-characterized collection of patients and controls must be made available. In addition, financial and scientific collaboration will be required due to the expense of a genome-wide association study and the need for a large number of families.

A second priority is identification of causal genetic variation in under-studied populations. Known variation in CARD15 and IBD5 plays a role in European-derived adult populations, and population differences in HLA alleles have been observed. However, these two loci have a very minimal effect in a number of populations. Dr. Rioux emphasized the importance of studies targeting African-American, Asian, Latino, and pediatric populations.

Thirdly, very little is known about the role of IBD genes in biology and pathophysiology. Dr. Rioux recommended that functional studies of variants, genetic studies of spontaneous and induced mouse models, and gene-environment studies in mouse models be undertaken, although answering these questions will ultimately depend on the development of novel methodologies.

Dr. Rioux stated that a fourth research objective is a greater understanding of the influence of genetic variation on disease. The recent finding of genotype-phenotype correlations between CARD15 and IBD5 and disease phenotypes has revealed interesting associations. However, Dr. Rioux remarked that their true influence on disease course and response to therapy is yet unknown, and suggested that researchers need to be able to molecularly classify IBD patients with regard to predisposition and biological state. Dr. Rioux suggested that researchers incorporate genetic screening, proteomics, serology, and expression analysis in both drug trials and in natural history studies, so that genes and gene variation predispose disease and lead to differential responses to drug therapy.

During discussion following Dr. Rioux's presentation, Dr. Strober suggested that perhaps the relationship between HLA and IBD has been over-examined, given its 30-year research focus. Dr. Rioux qualified Dr. Strober's comment, stating that, until now, research has centered on association between just a few variants, and has neglected to identify the genetic variation of the gene that predisposes the disease. Novel approaches to HLA research have only recently become possible with the advent and availability of newly developed technologies.

Dr. Rioux was asked what, if anything, NIH or other agencies should do differently in terms of providing resources for IBD research. Dr. Rioux answered that, insofar as well-characterized patients, families, and controls are concerned, establishment of collaboration opportunities among genetics researchers will be paramount to achieving a situation where it becomes natural for individuals to work together. Ms. Merrick (CCFA) suggested that a conference be considered to more clearly identify needs and coordinate plans to move forward in this area. Given the difficulty of funding abroad, one participant agreed that organization of collaborative opportunities would certainly foster international partnerships.

Dr. Rioux concluded with the comment that, given the expense of haplotype mapping the human genome, cobbling monies together from different agencies might be a means for making this rapidly evolving technology more accessible.

Epithelial Biology of IBD

Dr. Daniel Podolsky, Massachusetts General Hospital, Boston

Dr. Podolsky suggested that, as a benchmark of progress in IBD research, one should consider the change in perspective regarding the functional importance of the epithelium in understanding the pathophysiology of IBD. Researchers 20 or 30 years ago were concerned almost entirely with the dynamics of secretion and absorption of the digestive tract. In the 1980s, they began to appreciate that the epithelium, in addition to its absorptive and secretory properties, was essential as a continuous monolayer barrier. However, more recently, researchers have come to understand that the epithelium is an intrinsic and central participant in the nature of human interaction with the environment and our immune sensitivities to luminal flora.

Modern understanding of the anatomy of the epithelial barrier depicts a very highly evolved structure near the apical surface, which essentially forms a seal between one epithelial call and the cell adjacent to it that effectively prevents the ability of bacterial products, factors derived from the diet, to access the underlying lamina propria and classical elements of the mucosal immune system.

Dr. Podolsky spoke of the progress made in the last several years that has added considerable molecular detail to the morphologic structure of the epithelium, and he

presented a partial list of various proteins that collectively form the tight-junction structure and/or functionally modulate the tightness that it creates as a barrier. Certainly, a central objective in research over the next few years will be to acquire a more comprehensive understanding of how those proteins and others are dynamically regulated to sustain that barrier, and how the epithelial monolayer can be made to re-establish continuity after ulceration has occurred in patients with IBD.

Insight into the role of the epithelium as a dynamic component of the mucosal immune system has, in large part, been stimulated by thinking about the nature of the interaction with the luminal flora, both the commensal flora as well as known and specific pathogens. Dr. Podolsky said that it is increasingly clear that the epithelium does interact with the mucosal flora in a number of different paradigms. For example, researchers recognize that very small amounts of products derived from the luminal flora and perhaps from dietary intake are able to negotiate the epithelial barrier to gain access to the basolateral surface of the epithelium, as well as the underlying lamina propria elements. In addition, there is an increasing appreciation among researchers that the epithelial cell acts as a sensor and signal transduction unit, not only in stimulation of functional responses within epithelial cells themselves, but in that they also influence the functional state of the underlying mucosal lamina propria.

Dr. Podolsky described toll-like receptors, such as TLR2 and TLR6, which have been found to bind varying classes of patterned molecules, in essence allowing them to sense whole classes of microorganisms, rather than the specific recognition characteristic of the adaptive immune system. Dr. Podolsky stated that virtually all toll-like receptors are very highly expressed in epithelial cells, and therefore provide a dimension of its ability to serve in the innate immune system that had not heretofore been recognize. Dr. Podolsky commented that the question of why the presence of toll-like receptors does not result in a constant stimulation of an active immune response is an area that warrants further research. Studies with TLR4 suggest that clearly some mechanisms have evolved which allow the epithelium, while expressing receptors for the potential to respond to possible changes in the flora, to nonetheless control what would otherwise be a stimulus for ongoing inflammation.

NOD2, the protein encoded by the gene, mutations of which have been associated with Crohn's Disease in at least a minority of patients, contains a number of important functioning domains and can ameliorate some very key cell responses. Dr. Podolsky cited recent studies providing evidence that NOD2 expression is clearly present in epithelial cells.

Dr. Podolsky summarized the current conception of the epithelium as a very tightly adhered set of epithelial cells maintained by a complex, but probably dynamically regulated, set of tight junctions, in constant interface with a very dense set of microbes, and with the functional capability to produce a wide variety of regulatory peptides, cytokines, cemakines, and growth factors, which can not only modulate their own functional properties, but certainly coordinate functional responses in more classical

elements of the mucosal immune system.

Both direct and circumstantial evidence exist which implicate the epithelium in the pathophysiology of IBD, including:

- Altered structure within the tight junction, including alteration in the functional properties of that barrier, such as permeability;

- Mutant murine lines, where the functional defect has been reasonably demonstrated to be restricted to the epithelial compartment, and which have a phenotype indistinguishable from other murine forms of colitis;

- Expression of some key mediators of adaptive and innate immune receptors; and

- Critical dependence of the integrity of the barrier repair mechanism susceptibility with regard to the development of colitis and the ability to downregulate and control colitis once initiated.

Dr. Podolsky highlighted areas in both basic and applied science that may prove to be essential to further understanding of the mechanisms of IBD, particularly with regard to the role of epithelium. Lineage specification and the molecular basis of differentiation needs to be determined, which will require identification of the stem cell and regulation of the stem cell compartment. A more comprehensive understanding of the molecular basis of the barrier and its regulation will be essential to understanding its alteration in the context of IBD. Dr. Podolsky emphasized that a key goal should be to understand the mechanisms of the microbial-epithelial interaction, and what then becomes the functional definition of true commensalisms, symbiosis, and pathogenesis. More research is needed in the area of epithelial role in innate and adaptive immune mechanisms, as well as further definition of the mechanisms involved in epithelial repair.

Elucidating the role of the epithelium will be dependent upon a better understanding of the functional proteome within the normal epithelium, its relationship to the germ-free proteome, especially within the context of inflammation. Dr. Podolsky stated that one of the rate-limiting steps in much of the work focused on intestinal epithelium is the lack of researchers' ability to sustain primary cells in culture on an ongoing basis, resulting in a dependency on established cell lines. Expression profiles of epithelial cell populations, as well as imaging of their function in vivo would clearly accelerate both discovery of their role and characterization of their functional interaction with other mucosal cell populations.

Dr. Podolsky concluded by stating that a comprehensive understanding of the alterations in the epithelial compartment associated with or contributing to IBD would likely serve as a springboard for designing more effective therapeutic therapies, which is, after all, the ultimate goal of IBD research.

Following Dr. Podolsky's presentation, Dr. James pointed out that the NIH has recently funded two centers (one at Washington University and a second at Baylor), to define progenitor cells in a very comprehensive fashion, and to provide that information through the Internet as a resource tool for investigators.

Dr. Elson asked why NOD2 or CARD15, which are ancient genes, are now seemingly being resurrected as susceptibility factors for IBD, even though gastrointestinal infections were probably more common in centuries past than today. Dr. Podolsky speculated that it may only be as more genes and modifiers are identified that one will be able to explain the relatively recent appearance of Crohn's Disease. In terms of how NOD2 may or may not modify the background of enteric infections, it may turn out to be only one of the factors that define ultimate susceptibility to a particular pathogen. Another participant suggested that, from the perspective of evolutionary biology, NOD2 may have multiple functions and be involved in different diseases even in a historical timeframe.

Epidemiology of IBD

Dr. Robert Sandler, University of North Carolina, Chapel Hill

Dr. Sandler described the global prevalence of IBD, pointing out that while the disease tends to be common in Canada, the U.S., and in Europe, it is uncommon in Asia, Africa, and South America. In addition, a north-south gradient exists, so that people in Canada are more likely to have IBD than U.S. inhabitants, and within the U.S., those people in the northern parts of the country are more likely to get the disease than those living in the south. Importantly, when individuals migrate from low-incidence areas to a higher-incidence region, their IBD rates increase, indicating that environment plays a clear role in IBD occurrence.

Time trends for ulcerative colitis remain fairly flat. However, prior to 1932, Crohn's Disease was virtually unknown; since that time, incidence in nearly every registry has risen precipitously, often reaching a plateau in other regions, suggesting that some environmental change has been made manifest in the increase in the incidence of Crohn's Disease during the 20th Century. The disease also tends to be more prevalent in urban areas than in rural areas, and has a more frequent onset during the early adult years, although some studies suggest that a bimodal curve may exist. With respect to sex ratio, the incidence of IBD is approximately the same in both men and in women. The disease tends to be more common in individuals with higher socioeconomic status.

Dr. Sandler acknowledged the important role genes play in IBD, but stated that clearly, in order to understand IBD more fully, researchers need to understand the interplay between genes and environment.

Few analytic epidemiologic studies of IBD have been conducted; and Dr. Sandler stated that those that have been do not provide much information, since much of what they have

demonstrated has been the result of accidental discovery, rather than a product of good hypotheses. For example, smoking increases the incidence of Crohn's Disease and decreases ulcerative colitis, although the reasons for these correlations are as yet unknown. Studies have demonstrated that appendectomy appears to be protective against ulcerative colitis, and NSAIDs increase the risk for Crohn's Disease. Studies examining dietary factors that might be related to IBD have by and large been negative. Neither measles nor the measles vaccine has been shown to be related to Crohn's Disease, and breastfeeding decreases the incidence of Crohn's Disease.

Dr. Sandler summarized health care epidemiology of IBD, stating that the best estimates indicate that 1 million people in the U.S. have IBD, but that even those estimates, based on the only population-based studies that have occurred, came from an area where IBD rates are suspected to be higher than most other regions in the country. Neither do researchers know how much the disease costs, but it has been estimated that direct costs for Crohn's Disease and ulcerative colitis in 1998 reached $708 million and $387 million per year, respectively, with hospital in-patient costs representing the greatest portion of costs. Given the high numbers associated with in-patient hospital stays, out-patient hospital visits, emergency room visits, and physician office visits, Dr. Sandler emphasized that researchers must not lose site of the fact that not only does IBD affect a large number of individuals, but that quality-of-life studies must be conducted to keep a focus on the impact of the disease.

In Dr. Sandler's opinion, one of the major barriers to a better understanding of IBD is that, unlike cancer or hepatitis, the disease is not reportable. To a large extent, a lack of research-quality definitions of IBD and its phenotype hampers epidemiologic studies. With respect to ulcerative colitis, misclassification with infectious colitis often occurs. Very limited population-based data and little available information on costs and impact are further barriers, and the workforce of IBD epidemiologists is particularly small.

With those barriers in mind, Dr. Sandler identified population-based studies, including studies of health care costs and utilization, incidence and prevalence, and a combination of biological specimens, exposure information and clinical data, both on cases and on controls, as a primary need for furthering IBD epidemiological understanding. Data from these studies will provide a means for investigating the prevalence of genes in the general population, the gene-environment interaction, and gene-gene interaction. Greater support from both the CCFA and the NIH for gnotobiotic facilities is also necessary.

In terms of approaches, Dr. Sandler stated that new pathways to discovery will be dependent on population-based studies that have both biological and clinical repositories on both cases and on control. He described research teams of the future as being collaborative efforts between population scientists and laboratory scientists, involving public and private partnerships, from foundations to government to industry, and indicated that these types of partnerships will ultimately aid in sharing and alleviating the weight of funding in IBD research.

Dr. Sandler indicated a need to re-engineer the clinical research enterprise through translational research, integrated research systems focused on population-based studies, and recruitment of a larger workforce trained in all aspects of epidemiological research. Dr. Sandler has coined the phrase "biomedical informatics" to mean clinical data systems and repositories linking those systems together, and stated that IBD research needs a workforce able to make these resources available to investigators.

Dr. Sandler credited IBD researchers with making significant inroads into understanding IBD, progress which will surely be enhanced through collaborative efforts in the making and in the future.

At this point, a meeting participant remarked that public-private partnerships are an important notion, particularly with regard to industry. Considering the number of industry-sponsored studies already in place through NIDDK, this is a type of collaboration that may warrant expansion, although it may hinge on whether or not companies feel there is a competitive advantage in their participation.

The point was raised that, insofar as population-based IBD epidemiological studies are concerned, Iceland could provide a good source of study subjects, since they have been very well-studied and have benefited from a standard of medical care. Dr. Sandler responded with reference to a Pan-European study, and stated that although population-based studies in the U.S. are admittedly difficult, they can nonetheless be conducted.

Clinical Research in IBD

Dr. Bruce E. Sands, Massachusetts General Hospital, Boston

As he began his presentation, Dr. Sands remarked that patient-oriented clinical research is central to all the issues previously discussed during this meeting with regard to IBD, including patient characterization, laboratory research, human materials, and clinical and observational studies.

Key areas of study identified by the CCFA Clinical Challenges document include:

- Therapeutics (medical/surgical/other);
- Pharmacogenomics (genetic determinants of drug response and toxicity);
- Natural history/outcomes (prognosis);
- Epidemiologic/environmental risk factors;
- Phenomics (genotype-phenotype correlation);
- Interaction of genotype and epidemiologic/environmental risk factors in disease expression; and
- Biomarkers: Surrogate markers of clinical response, prognosis; dysplasia.

Dr. Sands described the current state-of-the-art patient care in randomized clinical control trials, stating that researchers are much better at inducing remission in both ulcerative colitis and Crohn's Disease patients, although maintaining remission over time is an area in need of improvement. RCTs may provide important insights into mechanisms of drug action, as well as disease pathogenesis. In addition, RCTs are specialized prospective cohorts, ideally suited for pharmacogenomic investigations.

RCTs also need to be used for the exploration of important human models, such as the maintenance of medically induced remission, the prevention of relapse following surgical resection, and early intervention studies which may provide information regarding alteration of the natural history of IBD. Dr. Sands pointed out that clinical trials are very much lacking in prospective data in children, and future research should focus on growth and developmental outcomes in this population. The benefits of complementary medicine, devices (such as aphoresis columns), and nutritional therapies should also be explored.

In order for RCTs to produce meaningful data that can be used to define prognosis prospectively and identify high-risk patients, Dr. Sands emphasized that outcome measures need to be better defined. Researchers need to move beyond short-term endpoints and even maintenance of remission to longer-term outcomes and natural history landmarks, such as the need for surgery or the development of complications. A critical area warranting closer attention is the determinant of the placebo response.

Dr. Sands named the identification of surrogate markers as the foremost objective before current IBD researchers. Objective findings from endoscopies are invasive, expensive, and often imperfect.

Observational studies are a second methodology for patient-oriented research. The CCFA Clinical Challenges group suggested the "moon shot" as a high priority for IBD research, so-called primary because of its expense, but still a relatively feasible project designed to identify potential high-risk IBD patients through the examination of new genes, gene and environment interactions, prognostic indicators, and outcomes. Perhaps a more feasible and certainly less expensive approach would be the development of a prospective incidence cohort followed serially for serologic markers, genotype, clinical

features, environmental factors, gut flora, and disease activity markers.

Dr. Sands commented that twin studies have a special role in IBD research, in that they are not strictly cohort studies, but are in a sense more similar to case-control investigations. In addition, Dr. Sands suggested that population-based cohorts are necessary to allow investigators to focus on environmental risk factors leading to discrepancy in disease expression. Currently, IBD research in special populations, particularly minorities, children, and women, needs to be addressed. To accomplish these goals, standard, validated definitions of a variety of endpoints, as well as possible risk factors, need to be established.

Dr. Sands outlined the priorities of the Clinical Science Research Agenda of the CCFA:

- Development of surrogate markers of disease activity, which would most readily translate into benefit for IBD patients;
- RCTs in important human disease models of the evolution of IBD (essentially, pouchitis and ileal resection);
- Definitions of the clinical description of patients for potential use in molecular classification of IBD;
- Definition of genotype-phenotype correlation with existing/novel genes of IBD (phenomics);
- Identification of novel epidemiologic and environmental risk factors for IBD; and
- Identification of biomarkers for dysplasia and cancer in patients with IBD.

Dr. Sands stressed that a crisis exists in the need for patient-oriented investigators in IBD, and that unless this deficit is corrected, many questions will go unanswered.

Dr. Strober offered a comment regarding partnerships between individual laboratories and pharmaceutical companies, saying that pharmaceutical companies may become reluctant to commit to funding better drugs for IBD when effective ones already exist. Dr. Sands agreed that, as more and more drugs become available, it is less likely that companies will want to invest money in what amounts to a relatively small patient population. Dr. Sands took the opportunity to mention an alliance between the CCFA and the CCFC (a group of investigators in Canada), working together to facilitate research that might not otherwise occur through an industry-approved mechanism.

The issue of the lack of trained investigators was revisited by several participants, including one individual who stated that the shortage is particularly acute among pediatricians. A second attendee offered information regarding loan repayment for medical school indebtedness available through NIH.

The comment was made that an academic experimental approach is currently being used in the field of neuroscience, and this might be a type of approach that would be applicable to IBD research, as well.

Intramural NIH IBD Research

Dr. Warren Strober, NIAID, NIH, Bethesda

Dr. Strober cited major areas of study in the Mucosal Immunity Section (MIS), part of the intramural program, including:

- Murine models of mucosal inflammation, such as TNBS-colitis, or colitis induced by the introduction of tri-nitrobenzene sulfonic acid into the rectum of mice. Using this model, the MIS has shown that Th-1 colitis is treatable with anti-IL-12.

- Initial description of murine TNBS-Colitis and demonstration that it is a Th1-mediated inflammation reversed by anti-IL-12.

- Demonstration that TNBS-Colitis can be regulated by induction of oral tolerance to TNP-substituted proteins and induction of TGF-β-producing cells.

- Use of TNBS-colitis model to examine mechanisms of anti-IL-12 therapy, to devise novel suppressor cytokine-based therapies and to probe mechanism of effector and regulatory cell generation in Th1 colitis.

- Genetic studies of TNBS-colitis, which have shown that it is not simply a chemically induced inflammation.

Dr. Strober pointed out the fact that innate immune factors are very important, in that the innate immune response involves the breaking or alteration of the mucosal barrier, followed by the entry of normally present materials in the gut, interaction with endrytic cells, and the reduction of IL-12. Within the context of the IL-12 production, specific adaptive immune responses lead to the reduction of interferon gamma and TNFα. This adaptive immune response is ultimately the cause of the disease.

The MIS is also very interested in the NOD2 defect, and is heavily engaged in studies of NOD2 deficiency and its impact on IBD, since the NOD2-deficient animal does not develop IBD.

Studies focusing on strain-specific susceptibility to TNBS-colitis have prompted researchers to conduct genome-wide searches in the SJL/J mouse, resulting in the identification of two susceptibility areas, one on chromosome 9 (tnbs1) and another on chromosome 11 (tnbs2), both of which have certain human homologues and linkages to other diseases. In addition to defining the areas of susceptibility, investigators have

identified what may be an important possible candidate gene that is currently being examined. Dr. Strober emphasized that collaborative efforts between scientists working on different aspects of this problem would be a very cost-effective research enterprise.

Dr. Strober described a second model, oxazolone-colitis, another skin-sensitizing factor, which, when injected into the rectum causes Th2-mediated colitis from the induction of IL-13 produced by NK-T cells. These study findings are significant in that oxazolone-colitis closely resembles ulcerative colitis.

Major studies of murine models of mucosal inflammation with oxazolone-colitis have included 1) the initial description of oxazolone-colitis and the discovery that it was a Th2-mediated colitis, and 2) the development of a more chronic model of oxazolone-colitis and the discovery that it is caused by IL-13 producing NK-T fells. Dr. Strober pointed out that these studies are a result of and focus of ongoing collaborative efforts.

Dr. Strober described a number of studies in which Dr. Mannon, Chief, Clinical Studies Unit, MIS, LCI, NIAID, is involved, including:

- Studies of the Clinical Efficacy and Immunologic impact of anti-IL-13 therapy in humans (done by multiple centers);

- Pilot study of the clinical efficacy and immunologic effect of G-CSF therapy in humans, important because it may be possible to maintain remission because of change in orientation of immune system; and

- Pilot study of the use of interferon-β in ulcerative colitis patients, (another area of possible collaboration, because if a consortium could be organized to study anti-IL-13 in patients, a new therapy for ulcerative colitis could be found).

Dr. Strober described novel therapies, including the creation of engineered regulatory cells that administer DNA that codes for TGF-β, effective in treating TNBS-colitis. TGF-β induces Smads (signals in cells that bind to the promoter of IL-10), demonstrating a close relationship between TGF-β and IL-10. This is significant because IL-10 is an antfibrotic protein that inhibits inflammation without causing fibrosis, which could lead to another possible area of collaboration: The development and utilization of regulatory cells to treat this form of inflammation.

Asked whether NOD2 knock-out mice have been crossed onto different genetic backgrounds, Dr. Strober replied that those studies are in the beginning stages. A meeting participant stated that IBD patients are not identical to NOD2 knock-outs, and that a reasonable question exists as to whether or not there is some residual functional capability of the Crohn's Disease-associated variations that is due to something other than a functional deficiency, a point with which Dr. Strober concurred.

Dr. Strober was asked to describe the commercialization process that would take place

should a partnership between scientific researchers and biotech industry take place. Dr. Strober answered that NIH has very well developed technology transfer branches. When a company develops a new treatment or substance, the technology transfer group will evaluate its commercial viability and, should it be determined to be commercial valuable, the group often pays to have the patent licensed to the company. However, despite the availability of this resource through NIH, companies are often reluctant to avail themselves of the service at least until their discovery has been registered, due in large part to the proprietary information and risk involved.

Regarding the issue of models, it was pointed out that while NIH has been able to partner with drug companies to provide additional data and analyses in many instances, this has not been the case with respect to osteoarthritis, where no drug exists and no randomized trials are being conducted. Development of a population-based cohort would advance understanding in the field of osteoarthritis as well as IBD by providing surrogate markers and reducing the costs of trials, and this aspect might entice drug companies to invest in a consortium that would be equipped to provide that resource. CCFA, to the extent that it is patient-based and would therefore facilitate patient recruitment, would be a valuable asset in this area.

NIDDK Portfolio in IBD Research

Dr. Frank Hamilton, Division of Digestive Diseases and Nutrition, NIH

Dr. Hamilton prefaced his summary of NIH-supported IBD research with the remark that this marks the 10th anniversary of the NIH Strategic Plan on IBD, and thanked Barbara Harrison for her instrumental effort in crafting the plan.

Research directions in IBD at NIH were the basis of a discussion at a meeting in 1989, from which a program announcement was developed that led to the addition of $2.5 million to the NIH budget specifically earmarked for IBD by Congress in 1993. Seven specific Themes were defined:

- Development of RFAs to encourage basic and clinical research in IBD. In 1993, 12 new awards were made amounting to $2 million. In 1995, the 12 RFA awards amounted to $2.5 million.
- Training of new investigators in IBD, an indication that an emphasis on manpower was identified as early as 1993, but which continues to be a critical limitation of IBD research.
- Formation of workshops focused on interdisciplinary areas.
- Use of interactive RO funding mechanisms, which was ultimately been found to be a very unsuccessful mechanism and has since been excluded from NIH efforts in this area of research.
- Enhancement of outreach activities through the Digestive Disease Centers. This has included co-sponsored programs on genetics of IBD, as well as immune/non-immune

interactions in intestinal inflammation, and the development of pilot and feasibility studies to encourage investigators to become involved in IBD research.
- Encouragement of and instruction in successful grant writing via national meetings, specifically aimed at young investigators.
- Development of biological resources and data collection, such as the co-sponsorship with CCFA of a meeting in 2003 to examine endpoints in clinical trials in Crohn's Disease.

A seventh Theme centered on the continued review of the Plan, and to that end, DDDN recruited Dr. Steve James from the University of Maryland to become the Deputy Director of the Division in 2001. Subsequently, Dr. Robert Karp was invited to lead the genetics of digestive diseases program, which has led to interesting and important interactions within the IBD community.

Dr. Hamilton outlined the success of IBD funding mechanisms within NIDDK, which has increased from $8.9 million in FY92 to over $36.0 million in FY02. Currently, NIDDK-supported IBD research includes the three Digestive Disease Centers, which have four program projects representing approximately 20 percent of the program product portfolio. One hundred twenty-one RO1 and RO3 IBD research grants are currently funded. A multi-center trial on IBD has been the focus of efforts by Pat Robuck. Judy Podskalny has encouraged Career Development Awards, which have bolstered the program and have contributed to an increase in the development of new investigators. The emphasis on the SBIR program as an area of high program relevance for the Institute has resulted in a growth in that area as well.

The growth in NIDDK-funded research over the last 10 years is a reflection of the entry of both new Principal Investigators and the commitment of experienced researchers in the field of IBD. Dr. Hamilton emphasized that the cooperation and risk-taking of NIH leadership has been a crucial element of the growth in IBD research and knowledge. He also extended appreciation for the partnership between NIH and CCFA, especially with regard to young investigators who have received seed money that allowed them to acquire preliminary data necessary for effective and important NIH grants.

Dr. James invited Karp to offer his remarks on the IBD Genetics Consortium. Dr. Karp described the 6-month Consortium, which is composed of six research centers and one data coordinating center, with a combined annual funding of $2 million. The Consortium is primarily concerned with identifying susceptibility genes for IBD. To that end, the Consortium is creating a patient repository, immortalized cell lines, and DNA samples, and will ultimately be forming an extensive database for analysis.

Discussion

Dr. James thanked the meeting's participants for their input regarding the exciting future research opportunities in IBD and stated that one of the most important aspects

researchers need to keep in mind is that maintaining continuous dialogue beyond meetings is imperative.

Dr. Spiegel posed a question for consideration: Has endoscopy and its availability blunted efforts at more effective, non-invasive imaging of the GI lining? He reminded participants that the NIH has a new imaging institute that ought to be utilized.

Dr. Sands commented that a variety of alterative efforts have been used, such as nuclear imaging, but even when they have been successful in quantifying disease activity, they have not been widely accepted by the community because they are relatively unavailable. Dr. Spiegel referenced the Institute of Imaging in Bioengineering, and suggested that institute is interested in fostering novel and innovative methods.

Dr. Spiegel raised the issue of the striking temporal trend in terms of Crohn's Disease as brought to the group's attention by Dr. Sandler, and wondered to what extent the combination of gene susceptibility and environmental factors has been crystallized so that it could be translated or applied. A meeting participant responded that, although the clinical assay technology has been licensed to a company, the technique has little value other than ascribing susceptibility to family members. Since, even for family members who are double-homozygous-recessive, the risk is quite low, the test is of relatively little benefit at this point.

Dr. James stated that the ability to effectively identify high-risk individuals using epidemiology is dependent upon prospective discovery, including the determination and development of appropriate markers.

Mrs. Rosenthal expressed appreciation for the availability of meetings and funding from the NIH, and suggested that a trans-NIH coordinating committee would be a valuable tool for the translation and integration of the Challenges Document with NIH resources. Recognizing what has historically been a difficult venture, Mrs. Rosenthal asked Dr. Spiegel to comment on the procedure for establishing joint funding across and within the NIH.

Dr. Spiegel responded by stating that the NIH leadership is extremely interested in working with IBD investigators to provide and support a myriad of scientific opportunities in an intramural fashion. Granted, the doubling in resources observed over the past few years will likely not continue, and limited resources will demand that issues be prioritized. Nonetheless, NIH clearly intends to foster and encourage the widest scope of intramural work as is possible. In order to remain cost-effective, NIH will examine and analyze issues one at a time, contact individuals, and determine the best means for networking to facilitate and achieve the necessary goals. While this may be a daunting task, it is the task at hand, and one that is not unique to IBD, since it pertains to many multi-disciplinary diseases, being addressed at many different levels.

Mrs. Rosenthal asked whether models such as the Juvenile Diabetes Foundation could

lend insight into methods for integrating information such as that contained in the Challenges document within the NIH in a timely fashion. Dr. Spiegel described coordinated efforts between the Heart Lung Institute and NIDDK currently in discussion regarding cardiovascular disease in Type 1 diabetes. The Allergy and Infectious Diseases Institute trans-NIH coordinating committee on automimmune diseases is another example of a concerted interdisciplinary effort at work. The NIAID funds the Autoimmunity Centers of Excellence, and welcomes applications with an IBD focus, since an infrastructure exists that would be available to aid in that type of investigation. Ultimately, Dr. Siegel concluded, the proof will lie in the progression of the science. Dr. Spiegel stressed the need for collaboration between CCFA and similar organizations, and stated that industry partnership is crucial for to research to be effectively translated to patients.

Drs. James and Spiegel concluded the meeting by thanking participants for attending and encouraging attendance at the next DDICC meeting, scheduled for June 16, 2003.