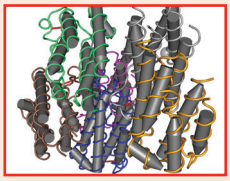
NewYork-Presbyterian

ADVANCES IN ADULT AND PEDIATRIC RHEUMATOLOGY

Affiliated with Columbia University College of Physicians and Surgeons and Weill Cornell Medical College

JANUARY 2014



Molecular structure of human interferon-alpha

Joan M. Bathon, MD
Chief
Division of Rheumatology
NewYork-Presbyterian/
Columbia University
Medical Center
jmb2311@columbia.edu

Mary K. Crow, MD, MACR
Physician-in-Chief and Chair
Division of Rheumatology
Hospital for Special Surgery
crowm@hss.edu

CONTINUING MEDICAL EDUCATION

For all upcoming education events through NewYork-Presbyterian Hospital, visit www.nyp.org/pro.

BEST HOSPITALS U.S.News & World Report NATIONAL RHEUMATOLOGY 2013-14

Rethinking the Paradigm for Lupus Nephritis

About 50 percent of patients who develop lupus nephritis do not adequately respond to current therapies, even the newest frontline agents, for the disease. With this information in hand, Robert J. Winchester, MD, an immunologist and rheumatologist and former Chief of the Division of Rheumatology at NewYork-Presbyterian/Columbia University Medical Center, set out to find out why, beginning with the question: Is the correct paradigm an immune complex disease mediated by autoantibodies?

Dr. Robert J. Winchester

"The treatment of lupus is currently driven by the concept of an antibody-mediated injury," says Dr. Winchester. "But the chronic injury in renal disease does not appear, for many patients, to fit into that paradigm."

Dr. Winchester, in conjunction with Vivette D. D'Agati, MD, Director of the Renal Pathology Laboratory at Columbia University College of Physicians and Surgeons, and her colleagues performed two sets of characterizations of renal biopsies to better define the immunologic character of the T cell infiltrate in lupus nephritis.

"We found that lupus kidneys have a variably patterned and often extensive infiltrate of predominantly clonally expanded T cells of CD4 and CD8 lineages," says Dr. Winchester. "They were fairly extensive, particularly in those patients whose disease had progressed. CD4+ T cells were prominent in nearly two-thirds of lupus biopsy samples and were distributed

(continued on page 2)

Interferon-Alpha in Systemic Autoimmune Disease: A Decade of Influential Research

Over the last 10 years, the studies of Mary K. Crow, MD, MACR, Physician-in-Chief and Chair of the Division of Rheumatology at Hospital for Special Surgery, have been influential in identifying interferon-alpha as a central mediator in systemic lupus erythematosus (SLE). "Our work in this area started in 2002, followed by a publication in 2003, early in the era of microarray gene expression studies," says Dr. Crow, who is also the Joseph P. Routh Professor of Rheumatic Diseases in Medicine at Weill Cornell Medical College.

As Dr. Crow and her colleagues in the Hospital's Mary Kirkland Center for Lupus Research reported in the December 2003 issue of *Autoimmunity*, studies using large-scale microarray technology to analyze global gene expression patterns in peripheral blood cells of lupus patients and control subjects demonstrated that genes regulated by interferon-alpha (IFNalpha) were among the most significantly overexpressed in SLE mononuclear cells.

"For years, there had been hints that interferon-alpha was elevated in lupus patients," notes Dr. Crow. "But it hadn't been a major focus of research. When we looked at the microarray data it was very striking that several hundred gene transcripts regulated by interferon were overexpressed in these patients. We considered that increased activity of IFNs may account for many of the immune system alterations that characterize SLE and contribute to autoimmunity. In fact, this IFN signature was associated with increased disease activity in lupus patients. From that point on, the nature of the major IFNs or other factors that drive the IFN-regulated gene expression signature seen in SLE became important

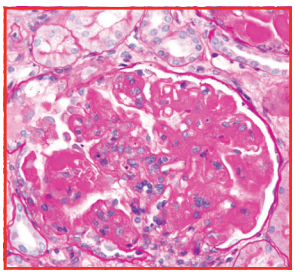
(continued on page 3)

Rethinking the Paradigm for Lupus Nephritis *(continued from page 1)*

as broad periglomerular aggregates or intermixed with CD8+ T cells forming periglomerular caps. While one might expect CD4+ T cells to be present because they help in the production of antibodies, we were surprised to find that there was a considerable proportion of CD8+ T cells. And nearly all of these were memory effector, which means that they were all set to have some effector function."

> **"The treatment of lupus is currently driven by the concept of an antibody-mediated injury. But the chronic injury in renal disease does not appear, for many patients, to fit into that paradigm."**
>
> **— Dr. Robert J. Winchester**

In addition, Dr. Winchester and his colleagues found that sometimes the CD8+ T cells were present in the periglomerular region around the glomerulus, and sometimes they were present within the tubule, and not just near it – a condition called tubulitis. "In fact, the vast majority of T cells found in the tubules turned out to be CD8+ T cells and exhibited features that suggest participation in an adaptive immune response," explains Dr. Winchester. "This indicated that they were presumably driven by some event. So our next question was are they clonally expanded? And the answer was yes. If you look at the T cell repertoire obtained through laser capture microdissection from these particular infiltrated areas, you can isolate the RNA of a small section to identify the nature of the T cell receptor. We found that all of the signatures of these being clonally expanded T cells were present, again arguing that they're driven by some antigen recognition event in the kidney that drives the T cell. What that something is, of course, is the main question. We don't know the answer to that."



Micrograph of diffuse proliferative lupus nephritis

The researchers' findings did not fit the paradigm that most of the injury in lupus is mediated by antibodies that are directed to autoantigens. "Susceptibility to lupus disease is associated primarily with class II MHC [major histocompatibility complex] molecules that present peptides to CD4+ T cells," says Dr. Winchester. "So what's a CD8 T cell doing there? When you go back and look at earlier events, again using Dr. D'Agati's treasure trove of old biopsies, in some instances the same T cell clone was found in the kidney up to six years earlier or in the blood of the patient initially and then it appeared in the kidney. So this clone persists and is always there. It may not always have been clinically associated with injury, but when we see the progression of the renal disease, and these clones expanding more and more, they appear to be driving a component of the injury. It would seem that both the CD4 T cells and CD8 T cells have the potential to play an important role in mediating injury, which may be relevant to development of progressive renal failure."

According to Dr. Winchester, this information should be used not as the therapeutic goal, but rather as a guide to therapy. "Certainly antibodies are responsible for acute flares and acute inflammation, but the chronic destruction and why therapy only works in some patients, but not in others, suggests that we just don't have the right target," says Dr. Winchester. "Rather than simply suppressing autoantibodies, which might not be responsible for the chronic manifestations of the disease, we might want to target these clonal expansions. Some of the medications used in the transplant field are related to those used in lupus, but they are used to suppress the T cells that would otherwise cause the rejection of the transplanted kidney. We should take a look at what our therapeutic endpoints are, how we use these agents, and we should use them much more in a way to affect the function of T cells. Assuming that this mechanism, which we think is a likely one, is confirmed, we may not need totally new agents, but rather we need to use them in more creative ways."

Reference Article

Winchester R, Wiesendanger M, Zhang HZ, Steshenko V, Peterson K, Geraldino-Pardilla L, Ruiz-Vazquez E, D'Agati V. Immunologic characteristics of intrarenal T cells: trafficking of expanded CD8+ T cell ß-chain clonotypes in progressive lupus nephritis. *Arthritis & Rheumatism.* 2012 May;64(5):1589-600.

For More Information

Dr. Robert J. Winchester • rjw8@columbia.edu

Dr. Winchester's Work Recognized with Prestigious Crafoord Prize

In May 2013, Dr. Robert Winchester was awarded the 2013 Crafoord Prize in Polyarthritis by the Royal Swedish Academy of Sciences. He shared the award with Peter K. Gregersen, MD, a graduate of Columbia University College of Physicians and Surgeons and now an investigator at the Feinstein Institute for Medical Research, and Lars Klareskog, MD, professor at Karolinska Institutet. The three were recognized for discovering how rheumatoid arthritis arises from the interplay of genes and the environment. Working together in the 1980s, Drs. Winchester and Gregersen made the first breakthrough when they found an explanation for why certain genes increase the risk of rheumatoid arthritis. The two scientists found that certain variants of HLA genes increase risk because the proteins they produce are more likely to attract the unwanted attention of the immune system. Dr. Winchester received the prize from the King of Sweden (photo) at a ceremony at the Royal Swedish Academy of Sciences.

Markus Marcetic

Interferon-Alpha in Systemic Autoimmune Disease *(continued from page 1)*

Dr. Mary K. Crow

areas of investigation for us. Researchers around the country were making the same recognition, and the field really took off."

Dr. Crow and her colleagues went on to study both the clinical and laboratory characteristics of the patients who had this high interferon signature, making a first-time observation that certain autoantibodies were associated with this interferon pathway and that the antibodies had specific characteristics. "They tended to bind to immune complexes containing RNA," says Dr. Crow. "As studies by us and by others continued, we recognized that those immune complexes containing RNA were actually playing a very important immunomodulatory and immunostimulatory role in driving the production of interferon-alpha and other pro-inflammatory mediators. It was suggested that interferon-alpha itself, or the receptors and signaling molecules that were responding to these immune complexes, might be therapeutic targets."

This school of thought emerged around the country and in Europe and was picked up by the pharmaceutical industry, which began thinking about how to target this new pathway that seemed to augment immune system activation in lupus. At the same time, Dr. Crow and her colleagues were pursuing genetic studies to try and understand why this was happening in lupus patients. "As data from the Genome-Wide Association Studies became available, we delved in detail into some of the genes that were statistically associated with lupus, making the further observation that several of those variable genes associated with lupus actually were associated with high Type 1 interferon in lupus patients and even in their families, suggesting a genetic association or genetic contribution to this high interferon production."

In time, Dr. Crow's laboratory studied several different lupus-associated genes, all of which seemed to relate to this high production of interferon or responsiveness to interferons. "Clearly we were seeing a genetic predisposition in many lupus patients to activating this Type 1 interferon or interferon-alpha pathway," says Dr. Crow. "We were defining a role for certain gene variants that made an individual more likely to make interferon and/or respond to interferon. At the other end of the spectrum, we were also looking at the clinical and serological manifestations to determine if patients with more severe disease were more likely to have this pathway activated and to understand at the molecular and cellular level what might be responsible for generating lupus flares."

More recently, the researchers have been looking at gene expression in blood cells and plasma proteins over time in a cohort of lupus patients in order to understand the role that the interferon pathway plays in the pathogenesis of the disease and its clinical manifestations, for example, making someone susceptible to a lupus flare. "As we've done these studies – longitudinal analysis of gene expression and protein expression – we've included additional molecular pathways that instigate or reflect activation of other cells," says Dr. Crow. "We are now in the process of analyzing very large data sets to characterize biomarkers, enabling us to establish a more biologic way of explaining what we see clinically."

Dr. Crow is particularly interested in understanding what is occurring at the underlying biologic level when the patient is relatively quiescent clinically, but that might lead to a flare a few months down the line. "If we can understand this, then maybe we can prevent that flare from happening," she says. At the same time, a better understanding of lupus at the cellular level can help to therapeutically target the disease and identify who might be appropriate patients to enroll in clinical trials that target certain pathways.

After a decade of work highly focused on the immunologic basis of lupus with the role of interferon-alpha as a primary focal point for that understanding, Dr. Crow believes there is now a much deeper understanding of the disease. "What we've learned about the genetic basis of lupus and how that plays out at the level of interferon pathway activation and how immune complexes can augment immune system activation have led to hypotheses about molecular targets for new therapeutics."

Many of these therapeutics are now in development, says Dr. Crow, and clinical trials of anti-IFN-alpha monoclonal antibodies are in progress.

Reference Articles

Niewold TB, Kelly JA, Kariuki SN, Franek BS, Kumar AA, Kaufman KM, Thomas K, Walker D, Kamp S, Frost JM, Wong AK, Merrill JT, Alarcón-Riquelme ME, Tikly M, Ramsey-Goldman R, Reveille JD, Petri MA, Edberg JC, Kimberly RP, Alarcón GS, Kamen DL, Gilkeson GS, Vyse TJ, James JA, Gaffney PM, Moser KL, Crow MK, Harley JB. IRF5 haplotypes demonstrate diverse serological associations which predict serum interferon alpha activity and explain the majority of the genetic association with systemic lupus erythematosus. *Annals of the Rheumatic Diseases.* 2012 Mar;71(3):463-68.

Crow MK. Interferon-alpha: a therapeutic target in systemic lupus erythematosus. *Rheumatic Disease Clinics of North America.* 2010 Feb;36(1): 173-86.

Niewold TB, Kelly JA, Flesch MH, Espinoza LR, Harley JB, Crow MK. Association of the IRF5 risk haplotype with high serum interferon-alpha activity in systemic lupus erythematosus patients. *Arthritis & Rheumatism.* 2008 Aug;58(8):2481-87.

Niewold TB, Hua J, Lehman TJA, Harley JB, Crow MK. High serum interferon alpha activity is a heritable risk factor for systemic lupus erythematosus. *Genes & Immunity.* 2007 Sep;8(6):492-502.

Kirou KA, Lee C, George S, Louca K, Peterson, Crow MK. Interferon-alpha pathway activation identifies a subgroup of systemic lupus erythematosus patients with distinct serologic features and active disease. *Arthritis & Rheumatism.* 2005 May;52:1491-503.

Crow MK, Kirou KA, Wohlgemuth J. Microarray analysis of interferon-regulated genes in SLE. *Autoimmunity.* 2003 Dec;36(8):481-90.

Crow MK. Interferon-α: a new target for therapy in systemic lupus erythematosus? *Arthritis & Rheumatism.* 2003 Sep;48(9):2396-401.

For More Information

Dr. Mary (Peggy) K. Crow • crowm@hss.edu

NewYork-Presbyterian

AMAZING THINGS ARE HAPPENING HERE

BEST HOSPITALS U.S.News HONOR ROLL 2013-14

Top Ranked Hospital in New York.
Thirteen Years Running.

Advances in Adult and Pediatric Rheumatology

NewYork-Presbyterian Hospital
525 East 68th Street
New York, NY 10065

www.nyp.org

Columbia University
College of Physicians and Surgeons

Weill Cornell Medical College