NATIONAL INSTITUTE OF DIABETES AND DIGESTIVE AND KIDNEY DISEASES (NIDDK)
NATIONAL INSTITUTES OF HEALTH (NIH)

Glomerular Disease: Pathophysiology, Biomarkers, and Registries for Facilitating Translational Research

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Bethesda, MD

Summary Report

TUESDAY, APRIL 17, 2012

INTRODUCTION

Gregory Germino, M.D., Deputy Director, National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institutes of Health (NIH), Bethesda, MD

Dr. Germino welcomed the meeting participants, who represent a full range of communities (e.g., academia, government, industry, and patient advocate organizations) engaged in a collective effort to facilitate translational research for glomerular disease. He highlighted the importance of the problem and the recent technological and scientific advances. Dr. Germino explained that he had witnessed slight progress in the field during his 20 years as a nephrologist, but he is optimistic that current research efforts will elicit large improvements in patient outcomes. Elucidation of the underlying pathologic mechanisms has been a large impediment, but persistence, coupled with new technologies, has led to recent advances in the understanding of glomerular disease. More research is needed to better understand disease physiology, identify biomarkers, and design effective therapeutics. Results from these studies will inform the entire field of glomerular disease.

OPENING REMARKS AND OBJECTIVES

Michael Flessner, M.D., Ph.D., Director of Inflammatory Renal Diseases, DKUHD, NIDDK, NIH, Bethesda, MD

Dr. Flessner welcomed the meeting attendees and expressed appreciation to the steering committee, which was co-chaired by Drs. Charles Alpers and Roger Wiggins. External steering committee members include Drs. Michael Braun, Ronald Falk, Agnes Fogo, David Salant, William Schnaper, Katalin Susztak, Aliza Thompson, and Shen Xiao. The NIDDK representatives to the steering committee were Drs. Michael Flessner, Paul Kimmel, Jeffrey Kopp, Kevin McBryde, Marva Moxey-Mims, Andrew Narva, Robert Star, and Yining Xie.
Dr. Flessner noted that the meeting would be videotaped, and he requested that speakers and participants asking questions use the microphones to ensure high audio quality. He reassured the presenters that confidential material would be removed prior to posting the conference videotape, presentations, and proceedings on the NIDDK website. Breakout sessions will not be videotaped, but science writers present in each room will summarize the discussions.

The goals of the conference are to: (1) discuss mechanisms that initiate and drive progression of glomerular diseases to End Stage Renal Disease (ESRD); (2) explore targets and pathways to therapeutic development for each glomerular disease; (3) assess existing biomarkers that define the diagnosis, initiation, progression, and/or relapse of glomerular disease; and (4) discuss approaches to cooperation among industry, academia, government, and non-nephrology researchers who deal with other organ systems. Experts in specific disease areas will present known mechanisms that initiate and drive the progression of glomerular disease. Defining novel biomarkers or surrogate markers of disease progression is necessary to replace the traditional outcomes of ESRD or death, which are too delayed to be useful in developing therapeutics or performing clinical trials. The fourth goal is the most important in developing therapies for glomerular diseases. Although glomerular diseases account for approximately 10 percent of ESRD, individually they are rare compared to diabetic nephropathy. The need exists to leverage collaborative efforts to develop international registries (e.g., NephCure), identify target populations, and foster research on biomarker development. Building relationships between industry, the academic medical community, government, and patient foundations will facilitate progress in the field.

STATE-OF-THE-ART LECTURES

Moderator: *Charles Alpers, M.D., Professor, Department of Pathology, University of Washington Medical Center, Seattle, WA*

Dr. Alpers highlighted the themes of the state-of-the-art lectures. He mentioned that this session was comprised of presentations describing perspectives that could be applied to glomerular disease to identify therapies and emphasize cooperation between industry, academia, non-governmental organizations (NGOs), and government.

Challenges in Therapy Development for Rare Diseases—Cystic Fibrosis

Frank Accurso, M.D., Section Head, , Pediatric Pulmonary Medicine, University of Colorado School of Medicine and Children's Hospital Colorado

As an example of how to approach the study of a complex disease, Dr. Accurso discussed the challenges faced by the Cystic Fibrosis (CF) field. He presented an overview of CF, noting that it is a genetic condition with a high morbidity that afflicts one in 3,500 births. The United States contains 35,000 individuals that are affected by this progressive lung disease that is characterized by a complex pathophysiology including infection, inflammation, fibrosis, and airway remodeling. Lung function suffers a precipitous decline during adolescence; the median age at death of a CF patient is 26 years. Dr. Accurso noted that CF is caused by defects in one gene that codes for the cystic fibrosis transmembrane conductance regulator (*CFTR*); many molecular mechanisms and unknown genetic susceptibilities underlie glomerular disease, thereby increasing the difficulty of study.

Dr. Accurso emphasized the crucial importance of disease registries in any clinical trial network to identify patients, set targets for therapies, and compare practice patterns. The Cystic Fibrosis Foundation (CFF) Therapeutics Development Network (TDN) was initiated in 1997 to accelerate new therapeutics and improve the treatment of CF. The TDN aspires to: facilitate the safety and

efficiency of Phase 1 and Phase 2 clinical trials, derive clues to CF pathogenesis, identify biomarkers, support young investigators, standardize approaches to compare outcome measures, promote synergy between sites, increase the patient pool, improve study design and statistical methods, and establish biorepositories.

The TDN, which operates out of a coordinating center in Seattle, is funded by the NIH, CFF, and industry. Other important components include the TDN clinical trial sites, National Resource Centers, the U.S. Food and Drug Administration (FDA), and other regulatory agencies. More than one-half of the studies originate in industry. The TDN coordinating center is comprised of biostatistics and data management personnel to analyze results, clinical trial operations staff to oversee the trials, a network operations department to assign studies to centers, and an administration department to determine how to pay for the studies. The TDN has grown from seven clinical trial sites in 1998 to 77 sites in 2009, and now supports Phase 3 trials. TDN oversight committees are responsible for scientific protocol review and publication of trial results. The core of each TDN site includes a Principal Investigator, Research Coordinator, Database Manager, and either a Laboratory Technician or paid service to provide laboratory support.

The TDN can claim successes in the form of 60 studies, dozens of publications, and grants awarded from the NIH and CFF. Approximately 2,000 patients per year participate in TDN studies. Significant TDN involvement resulted in the approval of the CFTR modulator VX-770 in January 2012. Dr. Accurso explained that the team science approach of the TDN has led to quality improvement in improving care for CF patients. Challenges include incorporating Electronic Medical Records (EMR) into the registry, prioritizing clinical studies, selecting and enfranchising trial sites, maintaining confidentiality, improving Patient Reported Outcomes (PRO), increasing patient enrollment, and collaborating with industry.

Discussion

Dr. Accurso clarified that most sites employ an intermediary person to input data from the EMR into the Cystic Fibrosis Foundation patient registry; the goal is to be able to directly translate data into the registry. The registry garners clinical data, pulmonary function, culture results, and treatment regiments. Site funding requires data submission. Dr. Accurso noted that the registry at his site is updated at least weekly. An attendee queried whether the TDN uses institutional or center Institutional Review Boards (IRB), and Dr. Accurso replied that institutional IRBs are utilized, but they are developing other methods. In response to another question, Dr. Accurso noted that 35 to 40 percent of centers are in the TDN.

Lessons From a Lupus Nephritis Study—ALMS: Successes, Failures, and Challenges
Neil Solomons, M.D., Vice President, Research and Development, Vifor Pharma, Victoria, Canada

Dr. Solomons thanked the meeting organizers and disclosed that he was an employee of the pharmaceutical company Vifor Pharma. He declared that the Aspreva Lupus Management Study (ALMS) was a large, challenging project that yielded numerous lessons, divided into categories of study design, operations, outcomes and interpretation of results, and safety issues. The ALMS

study was designed to evaluate whether mycophenolate mofetil (MMF) was as effective as, but safer than, intravenous cyclophosphamide (IVC) at treating lupus nephritis (LN).

Regarding challenges to study design, previous data were insufficient to quantify the treatment effect of IVC, requiring a superiority trial design to ensure regulatory success. Researchers considered whether the study should be open label or blind, ultimately deciding to forego sham injections in favor of an open-label study. They also deliberated the use of angiotensin converting enzyme (ACE) inhibitors and/or angiotensin receptor blockers (ARB) and the utility of a steroid taper. Operational challenges related to eligibility and recruitment included the reluctance of study sites to take repeated biopsies, the impact of prohibited prior treatments on recruitment, and consideration of the balance between trial speed and the desired sample size and demographics. Dr. Solomons noted that patient advocacy groups have a growing importance on clinical trial recruitment. The ALMS successfully relied on networks of physicians, coordination with pathology centers, and enthusiastic histopathologists to facilitate patient recruitment.

The ALMS, a 3.5-year study that required a large time commitment from patients, experienced poor subject retention. In addition to being costly, lack of retention affected the ability to detect statistically a clinically significant difference between treatment groups. The ALMS researchers explored methods to enhance subject retention, noting that most patients are motivated by receiving better treatment for themselves and advancing science. A patient appreciation program was initiated to incentivize consistent urine collection and cards were sent on subjects' birthdays.

Dr. Solomon explained the challenges associated with ALMS outcomes. He noted that the tendency is to include too many endpoints in the study, some of which lack utility. Interpretation of disease rating scales was inconsistent; expertise must be established within the study group. Other difficulties included local disease pattern variations (e.g., influenza), and differences in therapy protocols and urine-monitoring methods and consistency across the world. Dr. Solomon noted that rare disease studies often involve subjects from other countries, underscoring the importance of consistency in study procedures.

Results demonstrated that MMF was not superior to IVC when the primary endpoint was evaluated at 6 months. Statistical analysis uncovered racial and ethnic variations in IVC response: African Americans and Hispanics respond less well to IVC treatment, suggesting that genetic factors may be involved in the response mechanism.

In terms of post-treatment safety, subjects from Asia experienced the fewest infectious adverse events, but the infections were more likely to be severe and fatal. More of the deaths were associated with MMF treatment, but risk factors were not identified easily; one possibility is that four investigational sites in China mismanaged patients by initiating treatment when the patient had a pre-existing infection. Dr. Solomon summarized the ALMS results, noting that the international study was conducted with a diverse standard of care and outcomes. The design was challenging because of treatment effects that were difficult to quantify, and the safety issue might have resulted in part from differences in medical practice. As a consequence, the results were difficult to interpret. The critical importance of evidence-based trial management emerged from this study.

Discussion

One participant commented that it was surprising that minorities responded better to oral treatment rather than the intravenous treatment. Another attendee pointed out that it is important to store all data, even data that are not analyzed, because they may be useful in the future.

Glomerular Disease: Recent Travels and the Road Ahead
William Couser, M.D., Affiliate Professor, Department of Medicine, University of Washington, Woodinville, WA

Charged with giving an historical overview and future outlook for glomerulonephritis (GN), Dr. Couser reviewed the history of GN. The first demonstration of immune mechanisms occurred 107 years ago when scientists observed GN following injection of foreign serum and attributed the disease to "toxic bodies" formed in the circulation. In the 1960s, Frank Dixon, the father of renal immunopathology, defined the humoral immune mechanisms that underlie most forms of GN. He advanced the concept that the "toxic bodies" were circulating immune complexes that were passively trapped in the glomeruli, and the antibody component activated complement and damaged the glomerular basement membrane (GBM). Although some details of this hypothesis have since been revised, his research forms the basis for much of what is known today. In subsequent years, a direct role for antigen-specific T-cells in the development of GN has also been documented.

As glomerular disease research progressed through the late 20th century, further understanding of the multiple roles of the antigenic component of the "toxic bodies" evolved. Although antigens can indeed serve as components of preformed immune complexes, they can also localize independently in glomeruli, usually on a charge basis. These "planted" antigens, like normal glomerular proteins, can serve as a nidus for local, or in situ, immune complex formation. However, some were also shown to be directly toxic independent of antibody (e.g., via activation of the innate immune system); to function as molecular mimics resulting in autoimmunity; to initiate epitope spreading; and even to exert intracellular effects. Thus current paradigms depict a role for etiologic agents in activating both the innate and adaptive immune responses as well as in the subsequent development of both humoral and cellular immune mechanisms that mediate GN.

However, progress has been slow in identifying the etiologic events that initiate GN; this is a prime area to concentrate future research efforts. Infectious agents, including bacteria and viruses, have often been implicated. For example, mucosal infections by *Helicobacter pylori* may play a role in the development of IgA nephropathy (IgAN). Auto-antigen complementarity might link infectious pathogen-associated molecular patterns (PAMPs) and auto antigens in ANCA-associated GN, and several infectious PAMPs mimic auto antigens like the GBM antigen in Goodpasture's syndrome. Dr. Couser noted that evidence for both infectious etiologies and autoimmune features are now recognized in virtually all forms of GN. Better establishment of links between the two processes will further extend understanding of etiologic events in immune renal diseases and offer potential new therapeutic targets.

Dr. Couser outlined some pathogenesis goals for the field. Identification of target molecules, such as auto antigens, can now be accomplished by studying specificity of deposited antibodies and activated T-cells in human biopsy tissue. Studies focused on elucidating the role of antigen-specific T-cells in GN and genetic phenotyping are critical. Efforts to develop non-invasive technology to monitor intra-renal inflammatory events would be very useful. A "Systems biology" approach, using powerful integrative tools to link genomic and proteomic data with clinical and pathologic features is needed to select optimal therapy for individual patients.

Dr. Couser recounted his own 4 decades of involvement in treatment of patients with GN, which included the first use of both cytotoxic drugs and pulse steroids that remain standards of care today. Glomerular disease therapy has not kept up with progress in understanding pathogenesis because (a) researchers have yet to identify the best targets in man, (b) nephrology is too reliant on drugs developed to treat different processes in transplantation or rheumatology, (c) current therapeutics are toxic at optimally effective doses, and (d) drugs may have therapeutic effects that are unrelated to the rationale for using them. Because individual glomerular diseases are rare and usually chronic, large consortia and costly clinical trials are necessary to study therapy if ESRD is the only accepted outcome. Trials in progress do include some new therapeutics, but a major goal of the research enterprise has to be development of more targeted agents. Other goals include identification and agreement on biomarkers to reduce the length and cost of clinical trials; more attention to quality of life outcomes; and creation of glomerular disease registries.

New and exciting therapies may be targeted to both the sites of antigen exposure and antibody production as well as the site of tissue injury. Non-invasive real-time quantitation of specific inflammatory mediators in the kidney is becoming possible. Therapies to induce tolerance in patients and effect up regulation of protective molecules also are under development. A "kidney disease vaccine" to protect genetically predisposed individuals from nephritogenic infectious agents is even conceivable in the future.

Finally, Dr. Couser noted that most of the recent discoveries followed observations in animal models, and many of the important advances in the last 10-20 years emerged from relatively small laboratories led by physician-scientists. He cautioned that although the formation of large clinical and scientific consortia is necessary to accomplish many important goals, it is also essential to preserve a viable career path for the renal physician-scientist who understands both human disease and the technology necessary to translate future scientific advances in glomerular disease to the clinic.

DISEASE-SPECIFIC PATHOPHYSIOLOGIC MECHANISMS AND TRANSLATIONAL TOOLS

Moderator: *Roger Wiggins, M.D., Professor, Department of Internal Medicine, University of Michigan Health System, Ann Arbor, MI*

The purpose of this session is to answer the question: Where are we? Emphasis will be placed on therapeutic targets, potential diagnostic, prognostic, and alternative outcome biomarkers and diagnostic tests.

Minimal Change/FSGS: Genetics of Primary Podocytopathies
Friedhelm Hildebrandt, M.D., Professor, Department of Pediatrics, University of Michigan/Howard Hughes Medical Institute, Ann Arbor, MI

Dr. Hildebrandt described the status of research concerning minimal change disease (MCD) and focal segmental glomerulosclerosis (FSGS). He displayed a picture of a 3-year-old boy who presented with generalized edema and ascites because of steroid-resistant nephrotic syndrome (SRNS). Histological analysis confirmed FSGS, and over time the boy experienced characteristic renal function deterioration. FSGS in this case was caused by a homozygous point mutation in the *PODOCIN* gene, which results in protein dysfunction. This mutation is a full-penetrance biomarker conferring 100 percent risk of disease; it is one of the strongest cause-effect relationships in clinical medicine. Monogenic diseases, like FSGS, are defined by one disease-causing mutation in the entire genome, can be recessive or dominant, and different genes may cause similar disease patterns in different patients.

Dr. Hildebrandt explained that of the 6,000 monogenic diseases, the causative gene still is to be discovered in approximately one-half of the cases. Identification of causal alleles in nephrotic syndrome has been successful; the implication of *PODOCIN* and *NEPHRIN* genes, expressed in the podocyte foot process and slit membrane, guided the glomerular podocyte to the center of research efforts to understand disease pathogenesis. Subsequent identification of additional monogenic causes of SRNS facilitated understanding of essential components of glomerular function.

Monogenic causes are more frequent in early-onset SRNS than disease that presents later in childhood. Although mutations in *PODOCIN* are the most common, the myriad of rare monogenic disorders together result in a substantial number of disease phenotypes. To identify novel monogenic causes of childhood SRNS, Dr. Hildebrandt's laboratory recruited many afflicted families worldwide to provide free DNA sequencing of known causative genes. The researchers developed a new strategy for gene identification by mapping 1 million single nucleotide polymorphisms (SNPs) and identifying runs of homozygous regions in siblings. This procedure detected five candidate regions, none of which coincided with known disease loci, presenting an opportunity to identify a new nephrosis gene. The homozygosity mapping was combined with whole-exome capture and massively-parallel resequencing to refine further the relevant loci. One limitation to this approach is that capture and sequencing yields hundreds of variants that need to be parsed to the single causative mutation. After alignment with the reference sequence, evaluation of coding and splice variants, consideration of known polymorphisms, and manual inspection, Dr. Hildebrandt's research team identified the causative genes *ARHGDIA*, *KANK2*, and *MPDZ*.

All three gene products co-localize in glomeruli and co-immunoprecipitate, demonstrating molecular interactions. An *ARHGDIA* knockout mouse developed early onset nephrotic syndrome, further implicating the gene in SRNS. Subsequent research detected an afflicted child with a homozygous loss-of-function mutation in *ARHGDIA*, which abrogates physical interactions with three GTPases (i.e., Rhoa, Rac1, and Cdc42) that are important in glomerular function. The mutation also inhibits podocyte migration in a cell-based assay. The researchers knocked down *ARHGDIA* in zebrafish and detected generalized edema. This phenotype formed

the basis for an inhibitor screen to evaluate the efficacy of drugs. Preliminary data showed that Rac1 inhibitors are partially effective in preventing edema in the zebrafish *ARGHDIA* model.

Dr. Hildebrandt summarized the future outlook of SRNS, noting that it is feasible now to identify monogenic disease variants and every child should have that opportunity. Identification of causative mutations encourages the development of allele-specific animal models, improves the characterization of pathogenic pathways, enables allele-specific drug screening, and permits etiologic classification for therapeutic trials.

Discussion

Younger children are more likely to experience recessive monogenic disease. Recent evidence has shown that 20 percent of nephrotic syndrome results from dominant monogenic mutations. Polygenic cases are possible, but there are many single genes yet to be identified.

Minimal Change/FSGS: Mechanisms and Biomarkers for Primary Podocytopathies
Jochen Reiser M.D., Ph.D., Ralph C. Brown Professor, Chairman of Medicine, Rush University /,Chicago, IL / USA

Dr. Reiser explained that the kidney filtration barrier is a target for glomerular diseases such as MCD and FSGS. The foot process effacement of podocytes, the uniform signature of glomerular disease, is a reversible process. If effacement is not reverted, structural damage occurs, causing the podocytes to shrink, fail to proliferate, detach, and perish. This drives the loss of the glomerulus and ultimately the nephron. Kidney biopsies from patients with MCD or FSGS nephropathy always display some degree of foot process effacement. Previous data showed that cathepsin L expression, regulated by alternative translation, is another signature of proteinuric disease. Expressing the cytosolic form of cathepsin L induces foot process effacement.

MCD is a disease characterized by proteinuria, loss of glomerular basement membrane (GBM) charge, and steroid sensitivity. Angiopoietin-like 4 (ANGPTL4) can be expressed in podocytes and secreted into the GBM. In cases of MCD, the glucocorticoid sensitive protein is hyposialylated and lowers the GBM charge, driving severe proteinuria. ANGPTL4 is amenable to therapy; restoring sialylation through treatment with a sialic acid precursor reduces proteinuria in transgenic rats. Another interesting molecule is CD80, which is expressed persistently in cases of MCD and is secreted at high levels in the urine. CD80 expression decreases upon remission, demonstrating potential as a urinary biomarker.

FSGS is a progressive proteinuric glomerular disease. More than 20,000 people in the United States are affected by FSGS. Approximately 1,000 patients with FSGS receive a kidney transplant each year, yet FSGS often returns within days or weeks of the transplant for 30 to 40 percent of adults and 80 percent of children. A very specific role exists for the urokinase-type plasminogen activator receptor (uPAR) in FSGS etiology. This molecule is expressed ubiquitously on the cell surface of podocytes and other cells and requires interacting proteins to initiate signaling cascades. uPAR is induced in podocytes and other cells in cases of FSGS and diabetic nephropathy. Cleavage of domains on uPAR creates a soluble, circulating protein called soluble uPAR (suPAR). suPAR binds to beta-3 (β3) integrins, which catalyzes the active

configuration of the integrins and promotes podocyte motility along the glomerular basement mebrane (effacement). Injury-induced hypermotility of cultured podocytes is uPAR-dependent, and decreasing uPAR inhibits podocyte motility. The activity of the integrin can be measured with AP5, an activity-dependent antibody. Elevated blood suPAR levels create an active integrin configuration and significantly promoted effacement in recent studies. An antibody against β3 integrin also can inhibit podocyte-induced hypermotility and proteinuria in animal models. Inhibiting motility in the active phase of the disease is strongly anti-proteinuric. Repurposing anti-integrin antibodies for clinical trials of glomerular disease may be an efficient method to pursue new treatments.

The inhibition of suPAR might provide an ideal candidate approach—the proteins are soluble and might participate in injuring the podocyte under certain conditions. The molecule is elevated in the majority of FSGS patients but not those with other glomerular diseases. Patients with recurrent disease possess elevated suPAR levels relative to individuals in remission. In a study with 14 patients, strong suPAR expression correlated with recurrent FSGS and an elevated severity of podocyte effacement. Injecting recombinant suPAR is one therapeutic option, but animal studies have found that the molecule would need to be at a sufficient level to change the phenotype. Better ways of removing suPAR are needed; one possibility is improved plasmapheresis but better is a specific suPAR immunadsorption device. Expressing a mutant suPAR with 80 percent decreased binding to β3 integrin prevents proteinuria and effacement compared to expression of wild-type suPAR, demonstrating that the binding of β3 integrin to suPAR is sufficient to cause podocyte damage.

Researchers investigated whether suPAR could be blocked with antibodies, and discovered a dramatic effect of reduced proteinuria resulting from injecting antibodies twice weekly. A few patients in a FSGS trial experienced lower and stabilized suPAR levels, suggesting that a decrease in suPAR is helpful in establishing remission. Future studies will be important to understand why suPAR release is elevated in FSGS.

Discussion

Modulating sialylation might be useful for MCD because using sialic acid precursors modify protein expression, in contrast to steroids, which affect gene expression. Two research groups are developing sialylation-based therapeutics as an option for MCD.

A participant questioned what components affect suPAR levels. Dr. Reiser responded that studying infections has informed a lot of studies. MMF can lower suPAR levels by 15 to 20 percent. A MMF-like substance or longer treatment might increase efficacy. Integrin activation is lipid-dependent and Rituximab binds to podocytes directly affecting plasma membrane lipids. A current study, funded by the Nephrotic Syndrome Study Network (NEPTUNE), is investigating whether rituximab can inhibit podocyte integrin activation. Removing suPAR in a pre- and post-transplant setting and/or developing a biologic is the best option to progress treatment.

Membranous Nephropathy

David Salant, M.D., Professor, Department of Medicine/Nephrology, Boston University Medical Center, Boston, MA

Nephrotic syndrome results from massive albumin leakage into the urine, which causes hypoalbuminemia, edema, and a number of potential complications. Membranous nephropathy (MN) is an antibody-mediated cause of nephrotic syndrome and typically develops as a primary (or idiopathic) organ-specific autoimmune disease or less commonly it may be secondary to other conditions such as systemic lupus erythematosus, hepatitis B, certain drugs and cancer. Dr. Salant focused his discussion on primary MN because it is a common cause of nephrotic syndrome in adults.

The clinical course of MN is variable, and may include spontaneous remission, persistent proteinuria, progression to ESRD, and recurrence following kidney transplantation. Treatment involves potent immunosuppressive agents. MN morphology includes thickening of the glomerular capillary wall following basement membrane expansion around subepithelial immune deposits composed of IgG (predominantly IgG4) and complement.

Studies in the Heymann nephritis model of MN in rats demonstrated that the animals develop *in situ* immune deposits after mounting an antibody response against an intrinsic podocyte antigen. Podocyte injury, mediated by the IgG immune deposits and complement activation, causes proteinuric leakage and excessive matrix production resulting in glomerular basement membrane expansion.

In 2009, Dr. Salant's laboratory discovered the target antigen of primary MN in humans, M-type phospholipase A_2 receptor (PLA_2R), which is a member of the mannose receptor family. They identified autoantibodies (predominantly IgG4) against a conformation-dependent epitope of PLA_2R in the serum of primary MN patients, but not in secondary MN patient serum. Normal podocytes expressed surface PLA_2R, but the molecule relocated and colocalized with the anti-PLA2R antibodies in the immune deposits within MN glomeruli. In 2011, a large consortium study identified two risk allele associations in patients with primary MN—*HLA-DQA1* and *PLA_2R*. It is noteworthy that PLA_2R contains several SNPs with non-synonymous coding mutations in the anti-PLA_2R region.

In collaboration with investigators in the Netherlands, Sweden, China and the Mayo Clinic, the Salant group has shown that anti-PLA_2R is highly specific and about 75% sensitive for the diagnosis of primary MN and is a strong correlate of disease activity. Their studies showed that anti-PLA2R antibodies are present in the serum of patients while they are nephrotic, typically decline or disappear when they enter spontaneous or treatment-induced remission, and reappear if they relapse. Moreover, the decline in antibody levels generally precedes the decline in proteinuria by several weeks-months and is therefore a better marker of immunological disease activity than proteinuria, which may take time to resolve because of the basement membrane abnormalities.

Because prevention of MN is impractical, the ideal management strategy would be to restore tolerance to PLA2R, however this will require identification of the pathogenic epitope, determining the susceptibility-conferring genetic variations, and development of a suitable animal model for preclinical studies. Although immunosuppression is effective in many cases, ongoing antibody circulation does continue for several months and causes additional injury. This provides a window of opportunity to introduce complement inhibitors, apheresis (to remove circulating antibodies), immunoadsorbent therapy, decoy antigens, and/or blocking peptides to prevent further antibody-mediated damage until the anti-PLA2R antibodies disappear.

Pathophysiology of IgA Nephropathy

Jan Novak, Ph.D., Associate Professor, Department of Microbiology, University of Alabama at Birmingham, Birmingham, AL

IgAN is the most common primary GN, characterized by IgA1 deposits and activation of mesangial cells. The IgA1 found in glomerular deposits and circulating immune complexes is glycosylated aberrantly, and IgA1 in the deposits co-localizes with C3 and IgG, when present. IgAN is recurrent in more than 50% of transplant patients, suggesting the possibility of a causal IgAN of extra-renal origin. Synpharyngitic hematuria and proteinuria, ***i.e.,*** urinary abnormality associated with upper respiratory tract infections, accompany the disease.

Dr. Novak's studies and others have indicated a multi-hit mechanism for the development of IgAN. The first hit is an increase in IgA1 with galactose (Gal)-deficient *O*-glycans, followed by production of unique anti-glycan antibodies recognizing this aberrant glycoform of IgA1 (hit two). These two hits generate pathogenic IgA1-containing circulating immune complexes (hit 3), which ultimately deposit in the mesangium, causing the fourth and final hit, activation of mesangial cells and glomerular injury. Cytokines elicit a secondary effect on podocytes by inducing proteinuria. Genetic influences function in disease pathogenesis. Loci encoding genes involved in the major histocompatibility complex and regulation of glycosylation influence the first and second hits, and complement-related genes are involved in the third and fourth hits.

Human IgA1 possesses multiple sites for *O*-linked and two sites for *N*-linked glycans on each heavy chain. Some *O*-glycans can be Gal-deficient and sialylated; much complexity and heterogeneity exists in IgA1 glycosylation. Dr. Novak presented several critical questions and corresponding models and tools for research (***e.g.,*** immortalized IgA1-secreting cell lines and transgenic mice, cloning and production of anti-glycan antibodies, formation of immune complexes *in vitro*) that can be used to elucidate the mechanisms involved in aberrant glycosylation and formation of pathogenic immune complexes. Mass spectrometry, focused proteomics and glycoproteomics, kinome profiling, and gene- and protein-expression modulation tools are employed to answer questions in the field. For example, high-resolution mass spectrometry is used to analyze the sites of *O*-glycan attachment on IgA1 to determine whether Gal-deficient glycans are present at a specific attachment site(s) or as a mixture of variants glycoforms. Preliminary results indicate that 3-6 *O*-glycans per hinge region glycopeptide are the most common and that the attachment sites are not random.

Mesangial cell stimulation with IgA1-IgG immune complexes induces proliferation, protein kinase signaling, overexpression of cytokines and chemokines, and overproduction of matrix proteins. Notably, when inhibitors of protein kinases approved for use in cancer therapies were screened using this system, an inhibitor was identified that blocked the mesangial proliferation induced by IgA1-containing immune complexes.

Possible pathogenesis-related biomarkers and therapeutic targets include Gal-deficient IgA1, anti-glycan antibodies, Gal-deficient IgA1-IgG pathogenic immune complexes in the circulation. All four hits pose opportunities for interventions in the form of cytokine and signaling inhibition, blocking the Gal-deficient sites of IgA1 or antigen-binding sites of anti-glycan antibodies, and inhibiting the binding of immune complexes to mesangial cells or the cellular activation induced by these pathogenic complexes. Understanding IgAN pathogenesis will offer potential for disease-specific therapy testing and biomarker development. Notably, aberrant glycosylation occurs on different proteins in other diseases, such as breast cancer and other adenocarcinomas and chronic inflammatory diseases. Thus, significance of the studies of the abnormal *O*-glycosylation pathways in IgA nephropathy extends far beyond this disease.

Vasculitis

Ronald Falk, M.D., Professor, Department of Medicine, Kidney Center, University of North Carolina School of Medicine, Chapel Hill, NC

Dr. Falk discussed the lessons learned by the international vasculitis community about the disease. He noted that the vasculitis community has stimulated more progress as a group than any individual laboratory by sharing reagents, standardizing assays, and promoting randomized trials. Ant-ineutrophil cytoplasmic antibodies (ANCA) testing led to a number of phenomena, including a new classification system and naming convention. The vasculitis community recognized that disparate clinical symptoms actually were part of the same disease process. Dr. Falk discussed the names proposed by the Chapel Hill Nomenclature System, which codified the types of vasculitis and in doing so stimulated clinical trials. The clinicians systematically evaluated every iteration as a consequence of organizing the patients into larger groups with well-articulated endpoints. Two studies in particular evaluating the efficacy of rituximab and cyclophosphamide significantly changed the standard of care for ANCA vasculitis patients.

Dr. Falk suggested that ANCA disease might be best divided into proteinase-3 (PR3)-ANCA and myeloperoxidase (MPO)-ANCA disease because of the different natural histories, clinical phenotypes, and biology. A European genome-wide association study (GWAS) suggested that critical genes correlate not with the names, but rather with the ANCA serotype. Dr. Falk explained that antibodies interact with target antigens on the surface of cells. Neutrophils and monocytes are activated and cause damage. Animal studies elucidated the role of complement C5 in pathogenesis, provided information that anti-MPO IgG alone can cause disease while T-cells do not, indicated that neutrophils are required for pathogenesis, and suggested that neutrophil priming exacerbates disease. Abrogation of the alternative complement pathway and C5 receptor antagonists prevents disease in animal models; it will be important to determine whether the models correlate with human pathology.

An important consideration is whether all vasculitis is caused by antigen spreading or triggered by fibrinated proteins. Dr. Falk noted that the lack of assay standardization has been a barrier in resolving this issue. The role of the autoantibodies in promoting disease needs to be clarified. Autoantigen conformation might play a role; other possibilities include epigenetic expression, asymptomatic autoantibodies, and antigen regulation. Dr. Falk's laboratory developed an epitope mapping strategy to analyze the epitopes that occur in active disease, remission, or normal individuals and identified a single epitope specific to disease. Epitope-specific assays are important to understand disease progression and improve the utility of ANCA testing.

Research has indicated that in addition to monocytes and neutrophils, B-cells, T-cells, and plasma cells are important immune cell types in ANCA vasculitis. Scientists have learned that regulatory T-cells have defective suppression in ANCA vasculitis; additional research is necessary to determine the reason for the dysfunction and methods to reverse the process. Evidence indicates that regulatory B-cells might be protective of a relapse, which calls into question the wisdom of reducing their number with rituximab. Assays are needed to ascertain which B-cells are beneficial. Inhibitors of the complement cascade have been studied and present a target to abrogate acute vasculitis.

Overarching questions in the vasculitis field remain, including the reason why ANCA small vessel vasculitis is focal, and what molecular characteristics separate small, medium, and large vessel disease. The field needs a better understanding of the biology of remission and relapse, and biomarkers to detect remission and predict relapse would be useful. Current biomarkers, including creatinine and proteinuria, are not sufficiently sensitive. Numerous clinical therapeutics are being studied, with a focus on understanding how and when to use them; novel biomarkers are critical to target specific therapies and monitor disease activity and relapse.

C3 Glomerular Disease

Terry Cook, M.D., Professor, Department of Medicine, Imperial College, Hammersmith Hospital, London, U.K.

C3 glomerulopathy is a group of glomerular diseases characterized by uncontrolled activation of alternative complement pathways with C3 deposition. This disease is distinct from atypical hemolytic-uremic syndrome (HUS), which exhibits endothelial C3 activation without electron-dense deposits detected by electron microscopy. Dr. Cook discussed what is known about the pathogenesis of C3 glomerulopathy.

There are three complement pathways through which C3 can be activated (i.e., lectin, classical, and alternative pathways). The most important pathway relevant to C3 glomerulopathy is the alternative pathway, which is activated constitutively to detect cellular damage or pathogenic agents. The circulating protein Factor H is a circulating inhibitor of alternative pathway activation. It acts to accelerate the breakdown of the alternative pathway convertase and is a co-factor for factor I that metabolizes C3b to iC3b. C3 glomerulopathy results from an imbalance, which can be influenced by mutations or polymorphisms, between the regulators and activators of the alternative complement pathway.

Dense deposit disease (DDD) is defined by an electron-dense transformation of the glomerular basement membrane, occasionally with additional deposits on Bowman's capsule and the tubular basement membranes. This disease appears heterogeneous when viewed with a light microscope and can be membranoproliferative, mesangial proliferative, crescentic or endocapillary proliferative. Most DDD patients show low circulating levels of C3, indicating activation of C3 in the circulation. The disease is associated with C3 nephritic factor and Factor H deficiency or dysfunction.

C3 glomerulopathy without dense deposits has been called C3 GN, the morphology of which is variable by light microscopy and can include mesangial proliferation and membranoproliferative GN. There may be crescents and variable endocapillary inflammation. Dr. Cook presented examples of complement factor H-related protein 5 (CFHR5) nephropathy and MPGN type 3, demonstrating the distinguishing attributes of each. CFHR5 nephropathy has distinct subendothelial deposits, mesangial deposits and occasional subepithelial deposits. Biopsies that have previously been called MPGN type 3 have variable appearances with intramembranous deposits and may have prominent subepithelial humps.

Factor H-deficient mice deposit C3 on capillary walls and develop membranoproliferative glomerulonephritis over time. The mice do not develop glomerular disease in the absence of Factor B or capillary wall deposits in the absence of Factor I, implying that iC3b is deposited in the glomerulus from the circulation, not activated locally within the glomerulus. Therefore, therapeutics that sequester iC3b or inhibit Factor I might be effective in treating patients. Although Factor H-deficient mice crossed with C5-deficient mice still develop capillary wall deposits, they exhibit reduced glomerular inflammation, renal impairment, and mortality, implicating C5 in pathogenesis of C3 glomerular disease.

Genetic causes of C3 glomerulopathy have been defined. Some patients possess variations within the Factor H gene family. The Cypriot CFHR5 nephropathy is an autosomal dominant disease that results from a duplication of one allele of *CFHR5*. CFHR5 is detected in all complement-containing glomerular immune deposits, suggesting a role in complement activation or regulation in the kidney. Current studies suggest a role for CFHR5 in renal processing of C3. One form of familial MPGN type 3 results from a mutation that creates a CFHR3-CFHR1 hybrid protein.

Dr. Cook reiterated that causative mutations have been identified and polymorphisms are important for people who present with these diseases. Remaining questions to consider are: (1) why C3 targets the glomerulus, (2) why Bowman's capsule and the tubular basement membrane are not consistently affected, (3) which C3 metabolites target the glomerulus, and (4) why the deposit becomes dense. Understanding the pathogenesis also requires elucidating the roles of Factor H-related proteins and C5 and determining whether C5 is activated in the circulation or in the glomerulus. Scientists would like to better understand the significance and natural history of morphological variants and elucidate the cause of other phenotypic characteristics.

Therapeutic possibilities include replacement of missing factors (e.g., Factor H deficiency), removal of autoantibodies, sequestration of C3b in circulation, and prevention of iC3b generation. Reduction of glomerular inflammation and C5 inhibition are other potential treatments. A recent trial with eculizumab demonstrated efficacy in some cases; clinicians need to determine the best method to select patients for specific treatments.

Discussion

Dr. Cook clarified that finding small amounts of immunoglobulin does not exclude the fact that the disease is primarily caused by complement dysregulation. Allowing the presence of immunoglobin in the disease characterization might increase the incidence of C3 disease.

OUTCOMES FOR TRANSLATIONAL AND CLINICAL RESEARCH: PATIENT REPORTED OUTCOMES, BIOMARKERS, AND THE ROLE OF REGISTRY

Moderator: *Michael Flessner, M.D., Ph.D., Director of Inflammatory Renal Diseases, DKUHD, NIDDK, NIH, Bethesda, MD*

Clinical Trial Outcome Assessments: Measuring Treatment Benefit Across the Lifespan

Elektra Papadopoulos, M.D., M.P.H., Medical Officer, Office of New Drugs, Center for Drug Evaluation and Research, FDA, Silver Spring, MD

Treatment benefit is defined as the effect of treatment on how patients feel or function in their daily lives or on overall survival (FDA Guidance for Industry. *Patient-Reported Outcome Measures: Use in Medical Product Development to Support Labeling Claims.* Available at http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM193282.pdf). Well-defined and reliable assessments (Code of Federal Regulations 312.126) form the basis of FDA's conclusions of treatment benefit, which may then be described in labeling as claims using words that represent the concepts measured. Study endpoint measures can be classified into clinical outcome assessments (COAs), biomarkers and survival. Biomarkers (e.g., laboratory tests) are measurements that do not rely on factors such as rater judgment, training, motivation and effort. In contrast, these factors are inherent in the human reported measures, or COAs, which encompass patient-reported (PRO), clinician-reported (ClinRO) and observer-reported outcome measures (ObsRO).

Similar challenges exist when applying COAs for pediatric and rare diseases as for other uses, but with additional considerations that require early planning. Small sample sizes, poorly characterized natural history of disease, heterogeneous signs and symptoms, international and cultural variation in patient populations and clinical practice, and age variation in patients are often encountered with rare diseases. Disease manifestations can vary across the lifespan and, in the case of children, changing developmental characteristics (e.g., verbal ability) can influence patients' ability to provide self report.

The essential first steps of planning for endpoint selection are defining (1) the disease population, (2) the context of use and (3) the concept of measurement to define treatment benefit.

Disease definition should be explicit and targeted to the clinical trial population. The concept of measurement should be well defined and consistent with the study's objectives. These early planning steps can be challenging but are essential, because they drive the selection of the appropriate type of endpoint measure.

The final step is selecting the type of outcome assessment based on factors such as the need for clinical judgment (which necessitates a ClinRO), whether the patient can provide self report of symptoms or whether, as in the case of young children, observable indicators reflective of those symptoms must be identified. Self-report of symptoms and their impact on patients' daily lives provide *direct* evidence of treatment benefit and should be used when feasible and appropriate. Otherwise, observation-based reports based on verifiable observation (i.e., assessments detectable by the senses that do not rely on inference or interpretation) can provide indirect evidence of treatment benefit to support regulatory approval and labeling claims. Regardless of the type of outcome assessment selected, content validity (i.e., evidence that the instrument measures the targeted concept in the targeted context of use) is essential for a conclusion of treatment benefit. An instrument's other measurement properties (i.e., construct validity, reliability, or sensitivity to change) can only be evaluated once content validity has been established.

Well-defined and reliable COAs are urgently needed for documenting treatment benefits in many therapeutic areas. Early and strategic planning is critical, particularly for pediatric and rare disease populations. To increase efficiency in drug development, CDER has put forth a drug development tool qualification process (FDA Guidance for Industry *Qualification Process for Drug Development Tools* available at http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM230597.pdf) that provides a framework for development of publicly available scientific tools, including biomarkers and COAs, for application in multiple drug development programs over time.

Use of Data Standards and Disease Models in PKD. Why Do This Prospectively?
Ronald D. Perrone, M.D., Associate Chief, Division of Nephrology, Tufts Medical Center, Boston, MA

Autosomal dominant polycystic kidney disease (ADPKD) is characterized by a long period of stable kidney function during which extensive growth, irreversible scarring, and structural distortion occur. Despite significant advances in supportive care, the mean age of ESRD onset, 56 years, has not changed significantly in the past 20 years. Clinical research to identify new therapeutic agents that would block formation and/or growth of cysts at an early age would benefit greatly from identifying earlier biomarkers of kidney progression, such as total kidney volume (TKV), as a target endpoint for regulatory approval. A PKD outcomes consortium has been formed to create disease progression models to generate scientific consensus for adoption of the TKV as a biomarker and clinical endpoint for ADPKD, with the ultimate goal of making formal applications to the FDA and the European Medicines Agency.

The current paradigm for ADPKD progression includes a long (more than 40 years) period of normal kidney function, with early manifestations including hypertension and proteinuria followed by later symptoms of pain, hematuria, kidney stones, and infections. The present

regulatory endpoint, at which therapies are not likely to be effective, represents a substantial loss of kidney function, i.e., doubling of serum creatinine. Although expansion of TKV is more obvious in ADPKD than in glomerular disease, in both diseases underlying parenchymal injury is masked by glomerular filtration rates that are preserved by compensatory glomerular hypertension and hyperfiltration. For glomerular disease, the time scale over which normal kidney function is maintained (months to years) is relatively compressed.

The PKD outcomes consortium is comprised of the FDA, the NIDDK, academic institutions, philanthropic foundations, and pharmaceutical companies. The FDA recommended that the consortium construct a disease model to link TKV, the rate of size increase, and other secondary features of ADPKD, as well as collaborate with the Clinical Data Interchange Standards Consortium, Inc. (CDISC) and Critical Path Institute (C-Path) to standardize data in existing registries.

The goal of the consortium is to establish a registry of longitudinal data to be utilized in the creation of a disease model linking TKV with various endpoints in ADPKD. Dr. Perrone indicated that defining data elements for the registry retrospectively has been challenging, and a prospective approach—if it had been possible—would have been much better. Standardizing data on kidney size, liver cysts, and TKV has been difficult because of different measurement techniques, and it was not possible to map data on pain and quality of life due to heterogeneous collection methods.

The disease model to support qualification of TKV as an endpoint needs to determine the quantitative relationship between TKV and disease outcomes, including ESRD, mortality, declining renal function, and hypertension. Kidney disease has not been identified by the FDA as a priority disease for data element standardization. Dr. Perrone emphasized that standardization of data and research results is essential for conducting modern clinical research in today's complex, global, interconnected system.

Qualification of Diagnostic or Severity/Stratification Biomarkers
Courtney Lias, Ph.D., Director, Division of Chemistry and Toxicology Devices, Center for Devices and Radiological Health, Office of In Vitro *Diagnostic Device Evaluation and Safety, FDA, Silver Spring, MD*

Better diagnosis, treatment, and management of patients with kidney diseases requires the development of new tools, including biomarkers. Biomarker tests must be accurate, reliable, and validated for their intended use. Biomarker tests are employed in drug discovery, preclinical research, and clinical settings, including patient management and research. As diagnostic markers, they can identify and/or classify diseases; screen populations, often those that are asymptomatic; monitor for recurrence; and select responders to specific drugs. As prognostic markers, they are used to stratify disease by severity and or risk; predict disease development and progress; and identify which patients are at risk for an adverse drug reaction. Biomarker tests are sought as surrogate endpoints for diagnosis and prognosis, but their validation is difficult and requires special attention when determining decision points.

Evaluating effectiveness involves considering the sensitivity and specificity, and the positive and negative predictive value of the test. A test's positive predictive value indicates how likely it is for a patient with a positive test result to have the disease, whereas a test's negative predictive value indicates the likelihood that a patient with a negative test result does not have the disease. Predictive value depends on disease prevalence; if prevalence is very low, even a highly sensitive and specific test will result in many false positives. Biomarker tests therefore must be validated in the intended population of use.

In summary, new biomarker tests for diagnosis and prognosis of renal disease would benefit patients. Significant progress is being made in the development of biomarker tests, but they must be validated for particular purposes. Finally, Dr. Lias stressed that study design is important for the unbiased evaluation of effectiveness and reliability.

Possible Bases of Approval for Progression or Relapse of Glomerular Diseases
Aliza Thompson, M.D., Medical Officer, Center for Drug Evaluation and Research, FDA, Silver Spring, MD

Dr. Thompson offered a regulatory perspective, with the qualification that her views may not reflect the views of the FDA, on possible endpoints to support the approval of therapies for glomerular disease.

From a regulatory standpoint, drugs must be shown to be safe and effective for their intended use prior to marketing; evidence of their effectiveness must come from adequate and well controlled trials and must be substantial. The law does not define separate standards for approval for rare diseases; a regulation that does apply specifically to rare diseases is the Orphan Drug Act of 1983, which provides incentives for the development of drugs intended to treat small populations.

A drug's effectiveness can be shown by establishing effects on clinically meaningful endpoints (i.e., how a patient feels, functions or survives). Effectiveness can also be established using surrogate endpoints. A surrogate endpoint is a biomarker that is used in therapeutic trials as a substitute for a clinically meaningful endpoint; surrogate endpoints are expected to predict the clinical benefit of a therapy. Tolerance for the risks associated with a drug is influenced heavily by the benefits shown.

There is a high evidentiary standard for surrogate endpoints because there is substantial risk of adversely affecting the public health if a biomarker is falsely accepted as a surrogate endpoint. With regard to qualifying a biomarker as a surrogate endpoint, in general, it is not sufficient to show that the biomarker is correlated with a clinical outcome of interest; it is also important to consider if the biomarker is on the causal pathway of disease (i.e., mediating disease progression) and if there are data from intervention trials showing that treatment effects on the biomarker predict treatment effects on the clinical outcome of interest. Examples of accepted surrogates are blood pressure, which has supportive data from multiple intervention studies of agents working through distinct mechanisms showing that treatment effects on blood pressure predict treatment effects on CV outcomes, and certain electrolytes, which are thought to be directly causing a disease state.

Although it is reasonable to think that biomarkers that identify patients at risk of poor outcomes could also predict a treatment's benefit (i.e., serve as a surrogate endpoint), there are notable examples of biomarkers that performed well in identifying patients at risk for poor outcomes but failed to predict a treatment's effect on those outcomes. Lower hemoglobin levels are associated with cardiac disease risk in patients with chronic kidney disease, but trials showed that treatments that effectively raised hemoglobin levels also increased the risk of death, stroke, and other serious cardiovascular events. The failure of a surrogate could be due to an erroneous assumption about the relationship between the marker and clinical outcome. Dr. Thompson said, however, that many have argued that these failures are more likely due to unexpected adverse drug effects that minimize or outweigh treatment benefits. Because of this concern, there is less tolerance for risk when surrogate endpoints are used and for drugs with considerable toxicities, surrogate endpoints may not be a good path for establishing a drug's efficacy.

The term "glomerular disease" encompasses a collection of diseases, and suitable endpoints for establishing a drug's efficacy are likely to vary across these diseases, taking into consideration the pathophysiology, clinical course, and clinical manifestations of a given type of glomerular disease. Establishing an effect on progression to ESRD or on a marked and irreversible loss of renal function would provide convincing evidence of benefit across these diseases. Dr. Papadopoulos, in her talk, also addressed patient-reported outcomes as endpoints in clinical trials.

Proteinuria has been proposed as a potential surrogate endpoint in glomerular diseases. In approaching proteinuria as a surrogate endpoint, consideration should be given to the data supporting its use as a surrogate endpoint within the context of a specific disease. At present, it is unclear if proteinuria is causing/mediating progression to end-stage renal disease/ an irreversible loss of renal function, or if treatment effects on proteinuria will predict treatment effects on disease progression. It may be reasonable to use proteinuria as an endpoint under the accelerated approval pathway if certain conditions are met. Approval under this pathway is subject to the requirement to conduct postmarketing studies verifying the treatment's clinical benefit.

Dr. Thompson emphasized that the work being discussed at this conference will lay the groundwork for clinical trials. She urged researchers to consider the potential utility of their biomarkers as drug development tools, beyond their use as surrogate endpoints.

Discussion

A participant asked whether, in the context of the accelerated approval process, the FDA considers glomerular diseases to be serious, life-threatening illnesses; the mortality rates from many renal diseases are as high as some cancers, and they have serious cardiovascular effects. Dr. Thompson replied that although she could not make a blanket statement, she thought that many of the renal diseases discussed at this conference would be considered to be serious and/or life-threatening.

A participant pointed out that for some types of glomerular disease, proteinuria is considered to be a disease in and of itself, not just a biomarker, and treatment is directed to reduce proteinuria.

Dr. Thompson clarified that though proteinuria may be used to diagnose disease, patients don't complain of proteinuria per se. They may, however, complain of swelling and she noted that showing an effect on the symptoms that are important to patients could be a way to establish the benefits of a new therapy for glomerular disease. Another participant suggested that nephrologists should reconsider proteinuria as a candidate to be a surrogate indicator because no other biomarker has been proven to be better. Dr. Thompson responded that she was not arguing that proteinuria could never serve as a surrogate endpoint, but that more data, and disease-specific data, are needed to support its use in the context of specific glomerular diseases. Another participant pointed out that multiple clinical trials might be needed to qualify a given surrogate endpoint for a particular purpose.

Several participants called for better collaboration between the FDA, academia, and industry regarding glomerular disease. One participant, Dr. Falk, indicated that the American Society of Nephrology is drafting a memo of understanding, mission statement, and set of objectives for a partnership that will be called the Kidney Initiative for Innovation and Safety and that will involve all of these stakeholders.

CKD Biomarker Consortium: Sample Requirements for Discovery and Reproducibility of Renal Biomarkers

Jennifer Van Eyk, Ph.D., Professor, Department of Medicine, The Johns Hopkins University, Baltimore, MD

Dr. Van Eyk emphasized the magnitude of the effort required to develop new biomarkers for chronic kidney disease (CKD), citing expense, time considerations, technical challenges, and the complicated nature of experimental protocols. Possible types of biomarkers include imaging, metabolites, proteins, and genetic variants, but of these categories, proteomics offers potential biomarkers that are closest to acute events and therefore reflect the phenotypes for those events most directly. Biomarker development challenges include identifying appropriate technologies, cohorts, and analytes for each step of the process: discovery, verification, qualification, and implementation. Before embarking on biomarker development, investigators should consider whether a biomarker is clinically necessary, whether the required cohorts are available for discovery and validation given expected biological variability, what approach is best (*de novo* or targeted), and how to ensure sample quality. Recent advances have established a "pipeline" of sequential technologies to quantify thousands of proteins and their post-translational modified forms over a wide dynamic range. Better mass spectrometry technology has made possible quantitation with quality control measures, a broad dynamic range, targeted analysis of SNPs and peptides, good sensitivity, and multiplex assays.

Dr. Van Eyk presented an example of the types of standard curves, coefficients of variation, and recoveries achievable for peptides. She showed a sample discovery pipeline, which involves quality control at each analytic step. Establishing a specific sample collection protocol is very important. If protocols are established and appropriately followed, samples can be archived in a biorepository with confidence in the consistency of the results. Extensive method development has identified stable peptides for use as standards to assess sample quality, and techniques have been developed to remove high-abundance proteins from samples.

De novo biomarker discovery is not guaranteed success, but if all aspects are done correctly—including starting from appropriate clinical questions, evaluating the effects of other diseases, considering what the diagnostic will target, and assembling unique cohorts for each step—there is an increased probability of success. A recent example of successful *de novo* biomarker development is a suite of brain-specific proteins for brain injury. Dr. Van Eyk summarized the most important issues to consider before beginning biomarker development for diseases, rare or otherwise: disease specificity, viability of sample and technology protocols, and the labor and expense involved.

Discussion

An attendee asked whether the sample matrix of primary interest to nephrologists, urine, presents unique analytical challenges for biomarker development due to its high protein concentration. Dr. Van Eyck responded that despite analytical difficulties (e.g., a broad range in protein levels, high salt concentrations, protein precipitation, pH variation), extensive work has led to viable protocols for urine.

Pros, Cons, and Pitfalls of a Biological Repository
Elizabeth Wagner, M.P.H., Division of Blood Diseases and Resources, National Heart, Lung, and Blood Institute (NHLBI), NIH, Bethesda, MD

The NHLBI and the NIDDK have established collaborations between their biorepositories, which have overlapping focuses on patients with cardiac disease and obesity. Ideally, biorepositories operate by assisting with the design of new biospecimen collections. guiding biospecimen collection, and disseminating biospecimens for research protocols
The NHLBI Biologic Specimen Repository (Biorepository) has been managed for almost four decades with the aim of fostering blood safety, and contains almost 5 million specimens from more than 70 collections. Originally, the NHLBI Biorepository and Data repository were separate, but now more than 30 collections are linked to their phenotypic data. These linked collections and over 90 clincal study data sets are made available online through the Biologic Specimen and Data Repositories Information Coordinating Center (BioLINCC) at www.biolincc.nhlbi.nih.gov . In addition to the transfusion medicine collecioths, the NHLBI's Biorepository includes samples from multiple heart, lung and blood diseases and a variety of material types. Analyses of the samples have resulted in high-impact publications, and samples have been used in a range of studies related to blood safety. Building new collections and maintaining historical collections, however, requires significant financial and human resources. In the design phase, the involvement of a biorepository subject expert is essential. In addition, the processes by which biospecimens are acquired from the repository must be efficient to ensure that they are used. The BioLINCC program has successfully increased biospecimen use by providing online access and increasing visibility of the resource.

The NHLBI's Biorepository experience has shown that long-term planning is essential; experts in biorepository science and information technology are needed (particularly to link phenotypic data to their samples). Design for multiple purposes increases use. The NHLBI's long-term planning began in 2007 when it updated the Biorepository Operational Guidelines to follow best practices, and continued through 2009, with the implementation of a 5-year business plan. The

plan called for assessing the usage of samples and keeping only valuable collections. The NHLBI plans to build the Biorepository to maximize its value by critically evaluating new collections for possible inclusion; ensuring that sample collection, processing, and data storage are high-quality; and paying careful attention to ethical considerations, particularly those related to informed consent for historical collections.

Discussion

A participant called attention to the need to link the data to the phenotypic data and in addition to the investigators who make use of it, maximizing its usefulness for stimulating subsequent investigations.

An attendee asked how the NHLBI treats data identification and de-identification when consents for historical data often were granted for specific diseases or investigations. Ms. Wagner replied that the NHLBI honors the stipulations of consent agreements for historical data. A participant asked if the NHLBI reviews patient consent language before allowing a study to go forward. Ms. Wagner answered that the NHLBI is moving in that direction, but at present, its approach is to assign a biorepository expert to assist every study group in planning for data sharing.

DAY 2: WEDNESDAY, APRIL 18, 2012

PERSPECTIVE FROM INDUSTRY: DRUG DEVELOPMENT FOR GLOMERULAR DISEASES

Moderator: *Paul Kimmel, M.D., Senior Advisor, DKUHD, NIDDK, NIH*

Dr. Kimmel emphasized the importance of this session to the conference because of the major role that industry plays in research, product development, and regulation.

Phase III Planning for Novel Therapies in Autoimmune Glomerulonephritis: An Industry Perspective

Paul Brunetta, M.D., Principal Medical Director, Rare Disease Cluster, Genentech, Inc., South San Francisco, CA

Genentech and Roche study a number of renal conditions, including Lupus Nephropathy (LN) and primary Membranous Nephropathy (MN). Dr. Brunetta provided background on the involvement of these companies in multiple studies relevant to patients with autoimmune renal disease. Several issues complicate trial design. The FDA has not approved any new therapies for LN or primary MN; however, standard of care efficacy does not meet regulatory agency standards. Thus, non-inferiority trials against an unapproved active comparator drug (MMF versus Drug X) are unlikely to meet regulatory approval, despite the clinical significance of the information these trials would yield. However, many investigators believe that drug (MMF) versus placebo trials are unethical. Thus, LN guidance recommends an add-on trial design (e.g., MMF plus placebo versus MMF plus Drug X). The FDA generally accepts and understands this approach, but the approach increases the likelihood of safety consequences due to potential effects that may result from combined use of steroids, immunosuppressants, and biologics. In addition, the combinatorial approach is more expensive and does not align with monotherapy goals.

Another clinical trial issue is partial renal response (PRR), which represents stabilized renal function, inactive sediment, and significantly reduced proteinuria. The occurrence of PRR indicates positive prognostic benefits, but it is not an acceptable endpoint to achieve regulatory approval. The percentage of PRR improvement that is clinically meaningful remains unknown; however, PRR has yielded statistically significant long-term prognostic benefits compared to non-response. Natural history studies can influence the framework and data-gathering approach positively.

Histology grading warrants further exploration because it addresses the trial entry requirement for renal biopsy, which is problematic because it is not the standard of care and may reduce enrollment. Steroids are used inconsistently to induce and maintain therapeutic results in LN patients. Recent trials have suggested that the maximal serious infection rate is co-incident with the highest steroid doses, and there is a significant interest in the community to reduce steroids to the lowest effective dose. Two trials had included steroids at normal doses initially but tapered them to low doses over time, and significantly fewer serious infections were seen after the successful taper.

In summary, many issues remain for trial design, response definitions, and trial implementation; however, recent trials have been quite informative. Importantly, multi-company data sharing across trials will enable substantial advances for all sectors in hypothesis testing and analysis.

An Anti-macrophage Migration Inhibitory Factor Antibody for Treatment of Lupus Nephritis

Friedrich Scheiflinger, Ph.D., Vice President, Research and Development, Baxter BioTherapeutics, Vienna, Austria
Mahmoud Loghman-Adham, M.D., Medical Director, Baxter BioTherapeutics, Westlake Village, CA

Dr. Scheiflinger discussed the background and animal model findings for macrophage migration inhibitory factor (MIF). MIF is a multi-functional stress and growth factor that regulates cytokine and hormonal activity, as well as innate and adaptive immunity. MIF is a pleiotropic pro-inflammatory molecule expressed on a number of cell types, including immune, neurological, and skin cells. It negatively regulates MAP kinase phosphatase 1, an inhibitory regulator of inflammation, and thus overrides many local effects of steroids and promotes a local inflammatory response. With its many and varied systemic and local effects, MIF is a complex drug target.

MIF expression is upregulated in kidney disease, which induces macrophage accumulation and severe tissue damage, including glomerular crescent formation. The degree of upregulation is proportional to the degree of renal dysfunction, histological damage, and leukocyte infiltration in humans. Researchers use rats as a therapeutic model for testing the mechanism and efficacy of human anti-MIF antibodies and other drug candidates in renal autoimmune diseases. Anti-MIF treatment of rats with glomerular crescent damage effectively reduces proteinuria and prevents loss of renal function.

MIF is released in response to various stressors and it mediates injury by upregulating pro-inflammatory cytokines and chemokines and by antagonizing glucocorticoid activity. The general therapeutic mechanism of anti-MIF-mediated reduction of renal inflammation is that an antibody inhibits MIF, and thus inhibits MIF from secreting cytokines that recruit and activate leukocytes. Baxter BioTherapeutics is developing a clinical, fully humanized anti-MIF antibody to treat LN.

Dr. Loghman-Adham described Baxter BioTherapeutics' clinical plan for studying anti-MIF in various populations. The issue with traditional Phase I, II, and proof-of-concept trials is that the target is not expressed in healthy volunteers; thus, LN patients populate the entire study. Recruitment for LN trials has been slow for a number of reasons, including the rareness of LN, competing clinical trials, and requirements for histological diagnosis within 6 to 12 months. New and innovative strategies are needed for the completion of LN clinical trials and approval of new LN drugs.

Translating Targets into Therapies for Chronic Kidney Disease: Challenges and Opportunities

Glenn Reinhart, Ph.D., Director, Boehringer-Ingelheim Pharmaceuticals, Inc; Ridgefield, CT

Medical needs for renal disease are high and increasing. Recent advances in defining human disease mechanisms on a molecular level in both humans and preclinical models present multiple opportunities to identify targets with the potential to translate into improved patient care and clinical outcomes. The multi-step process to introduce a new drug to the market requires a number of years to complete:

1. Establish research objectives and validate the target.
2. Synthesize novel compounds.
3. Optimize and select drug candidate by testing compounds for efficacy and safety.
4. Scale-up synthesis and conduct rigorous safety tests in animals.
5. File an Investigational New Drug with the FDA.
6. Test tolerance and pharmacokinetics in humans (Phase I).
7. Test patient efficacy (Phase II).
8. Conduct large, clinical trials (Phase III).
9. File New Drug Application (NDA).
10. FDA review of the NDA.
11. FDA approval of the drug for marketing.

Major pitfalls include high failure rates for clinical Phase II and III studies, decreased NDA approvals, and the growing complexity of disease indications and clinical trials. Nonetheless, modern molecular and genetic science combined with the use of patient samples can be leveraged to strengthen the connection between molecular mechanisms of human disease and the preclinical models needed to optimize drug molecules. Challenges include high attrition rates, extensive regulation, and the historical need to rely on preclinical, proof-of-principle data to predict clinical outcomes in human disease. However, opportunity lies in the new insights being generated with regards to molecular mechanisms of human disease, which potentially can increase success rates in Phase II and III trials. Implementation of this process should include a strong focus in five areas: (1) developing targets that are drugable and disease-relevant by determining which signaling pathways drive the disease process in humans and enhancing access to relevant human data; (2) creating relevant and informative experimental models that express the targeted (human) pathway and use objective parameters to define clinical validation; (3) identify reproducible biomarkers to enable earlier decisions regarding efficacy in the clinic, improved dose selection, and stratified patient selection; (4) ensuring robust clinical efficacy and safety by evaluating current Phase II and III trials for validity and considering evolving patient selection practices and disease heterogeneity; and (5) pursuing ideal regulatory requirements by considering current and future registration endpoints (e.g., proteinuria) and the continually evolving clinical landscape.

Negotiating Renal End Points in Membranous Nephropathy
Caroline Savage, Ph.D., F.R.C.P., F.Med.Sci., Vice President and Head of Discovery Medicine, GlaxoSmithKline (GSK), Stevenage, U.K.

MN is a rare autoimmune disease diagnosed by renal biopsy. MN clinical presentation includes hypo-albuminemia and proteinuria, and its potential complications include hypertension, hyperlipidemia, increased risk for cardiovascular or thromboembolic events, decreased quality of life, and kidney failure. Natural history studies reveal a variable disease course—approximately one-third enter spontaneous remission, one-third experience persistent proteinuria, and one-third experience ESRD within 10 years.

Therapeutic approaches typically aim to control high blood pressure, proteinuria, and other symptoms. Patients destined for spontaneous remission should not be exposed to toxic immunosuppressant therapies; ideally, specific therapy is withheld in these patients unless renal conditions become severe. Unfortunately, robust biomarkers to predict likelihood of spontaneous remission are not available, so there is a tendency to withhold therapy in all patients for as long as possible. An ideal therapeutic solution would be used early in the disease to reduce renal function loss, administered until the patient achieves clinical remission indicated by reduced proteinuria, used to treat relapses, and not include steroids nor toxic alkylating agents or calcineurin inhibitors.

In renal diseases, a potential disease endpoint is proteinuria, a state that reflects loss of the glomerular filtration barrier and drives symptoms such as hypoalbuminemia and edema. Proteinuria is toxic because renal epithelial cells endocytose protein and release pro-inflammatory and pro-fibrotic mediators, which lead to chronic inflammation and renal failure. Reduced proteinuria is associated with improved long-term renal outcomes, and the nephrology community accepts it as an efficacy and activity biomarker. A 10-year cohort study in MN showed significantly higher rates of renal survival in patients who experienced complete (100%) or partial (90%) proteinuria remission compared to non-remission patients (45%). Nonetheless, some Regulatory agencies require additional studies on the long-term benefits of complete remission before they will consider proteinuria as an endpoint. Alternatively, agencies will consider serum creatinine production that is less than doubling as a renal function endpoint, but this is experimentally impractical. Several negative events that signify treatment failure are among other clinically meaningful endpoints under consideration. Potential secondary endpoints include the length of time for achievement of complete remission, patient questionnaires, and the 6-minute walk test (validated for CKD).

In summary, several potential under-explored endpoints effectively can measure the efficacy of drug candidates. The strongest candidates are: (1) partial or complete remission; and (2) a composite of clinically meaningful effects that represent treatment failure.

Addressing the Challenges to Develop New Drugs in CKD: Is Disease Stratification a Way Forward?
Maria Bobadilla, Ph.D., Dr.P.H., Head of Biomarkers and Experimental Medicine, F. Hoffmann-La Roche Ltd., Basel, Switzerland

CKD is a severe disease that presents a high unmet clinical need. Approximately 33 million people in the United States are afflicted, and sometimes patients die of cardiovascular disease before progressing to ESRD. Many common and orphan diseases in nephrology remain without adequate treatment guidelines, and the quantity and quality of randomized clinical trials is a major concern. The challenge is to stratify patients by their risk to progress to ESRD or perish from associated complications, and identify therapies that delay progression in patients that do not respond to the current standard of care.

Dr. Bobadilla explained that targeting fibrosis is the next frontier in the treatment of CKD. Tissue fibrosis is correlated with renal function, and regression of sclerosis has been observed under certain conditions. Normalizing the microenvironment restores glomerular architecture and might promote disease regression. Challenges exist in targeting fibrosis in CKD treatments. For one, fibrosis is not an approvable indication; an improvement in fibrosis associated with renal function is needed to qualify a clinical endpoint. Dr. Bobadilla noted that the disease paradigm needs to shift to a mechanism that is based on patient management by dissecting the mechanisms underlying clinical phenotypes and developing sensitive and non-invasive surrogate markers to quantify changes in disease progression. Cytokines associated with the fibrosis process probably are not sensitive or specific enough, but podocyte biology and genomic markers hold promise. To understand the etiology of disease, it is important to define the population that is to be treated.

Roche's kidney fibrosis strategy relies on collaboration with academia and contributions from the scientific community at large. The goal is to identify a bio-signature correlated with fast progression to ESRD, thereby shortening development timelines, reducing heterogeneity from the study population, and treating patients with a higher unmet medical need. Elevated cardiovascular disease risk in CKD patients needs to be addressed through trials designed with consideration of both dimensions. Notably, different conditions necessitate different approaches; FSGS and type 2 diabetic nephropathy (T2DN) require divergent patient stratification and translatability, which are key drivers of success.

Drug Development for Glomerular Diseases: Considerations in Ultra-orphan Settings
Camille Bedrosian, M.D., Senior Vice President and Chief Medical Officer, Alexion Pharmaceuticals, Inc., Cheshire, CT

Dr. Bedrosian discussed considerations for drug development for patients with ultra-rare disorders, which is Alexion's research focus. Ultra-rare disorders are those that affect fewer than 20 people per million. Furthermore, Alexion focuses on therapeutic candidates that have the potential for life-transforming impact on these patients and their families. One such drug is the complement inhibitor Soliris®, also known as eculizumab, which is approved for treating atypical HUS (aHUS) and paroxysmal nocturnal hemoglobinuria (PNH). PNH and aHUS are diseases caused by permanently uncontrolled and excessive complement activation.

Chronic complement activation results from loss of natural inhibitors due to genetic variation. The constitutively active complement pathway is designed to survey for abnormal internal processes or external intruders. The regulation of the pathway is highly controlled; dysregulation leads to anaphylaxis, inflammation, and thrombosis mediated by C5a and cell destruction, inflammation, and thrombosis mediated by C5b-9. Soliris® binds to C5 to inhibit the terminal portion of the cascade, leaving the proximal portion intact. Due to this mechanism of action, a side effect is an increased risk of meningococcal infections.

Alexion Pharmaceuticals is pursuing additional disease targets that have the potential to respond to eculizumab (e.g., Shiga toxin-producing *Escherichia coli* (STEC)-HUS, DDD, and C3 GN) and is in the process of developing four additional innovative therapeutic candidates. Alexion Pharmaceuticals is committed to clinical trials driven by highly motivated clinicians who have no suitable options to treat patients. In addition to prospective open-label studies, retrospective data have been collected from patients receiving the drug outside of a trial.

Ultra-rare diseases present many challenges. Target identification requires understanding of the pathogenesis of disorders that are not well studied. Trials often are designed with important input from experts in the field and without sufficient if any precedent. Inclusion and exclusion criteria must be narrow enough to exclude similar disorders, and patient recruitment requires significant effort from a global network to identify the rare disease patients. Regulators must evaluate data from trials in which a placebo control is impractical or unethical, and commercialization after approval requires long-term, international programs to educate physicians and patients. Registries are integral to the process.

Dr. Bedrosian commented that investigators can garner insights into rare diseases by looking beyond the conventional wisdom, realizing that each patient's experience is invaluable, deeply examining clinical trial data, and performing extensive followup. Partnerships, comprised of passionate investigators, industry innovators, dedicated regulators, practicing physicians, and patients and families are critical to progress the field of rare disease treatments for patients.

Discussion

Dr. Bedrosian clarified that the PNH study consisted of a randomized placebo-controlled trial. In contrast, aHUS trials were single-arm given the compelling nature of emerging available data, and the STEC-HUS trial also was single-arm because clinicians were requesting eculizumab in the face of a serious health crisis. The initial proof of concept trials in DDD have been investigator-initiated. Several design options would be considered for a subsequent DDD trial. Investigators should consider additional evaluations to fully understand the mechanism of Soliris® and identify patients who most benefit from treatment. Thoughtful stratification of patients is beneficial to drug development.

Industry Panel Discussion
Moderator and Panel of Speakers

Dr. Kimmel thanked the industry speakers for sharing their experiences and expressed appreciation to Dr. Flessner for planning the forum and inviting the speakers.

A participant noted that patient recruitment for clinical trials is a major problem for rare diseases. He solicited recommendations for the type of information that would be useful if a consortium established a registry of glomerular diseases. The panel responded that increased awareness through partnerships with appropriate consortia is important to increase the enrollment curve and improve recruitment efficiency. Currently, only 5 to 10 percent of patients with any given disease will enroll. Another idea is to define the population and acquire names and addresses to distribute survey information. Feedback is critical to understand whether groups of centers have access to a population that researchers are interested in studying.

The panel discussed priority activities that could be performed together with the goal of developing effective therapies to improve patients' quality of life. One suggestion is for pharmaceutical companies to collaborate in a pre-competitive space to improve PROs for glomerular disease. Many panelists agreed that collaborating to produce an effective PRO would be a useful endeavor in the pre-competitive space. Another opportunity is to collaborate to secure the limited funding from support programs. Performing natural history studies will help clarify usable outcomes to decrease the funding risk for these projects.

An attendee suggested that the GN field look to the model of the American College of Rheumatology criteria (ACR20) for drug approval to treat rheumatoid arthritis, emphasizing that the FDA should set the standard high for the first clinical trial and then accelerate the rate of approval for additional medical conditions. The panelists agreed that the ACR20 represents a partial response and underscores the importance of how a person feels in response to treatment. Likewise, partial proteinuria response might be clinically meaningful, especially in conjunction with improved PRO criteria.

Patients with severe disease tend to be highly motivated to participate in the development of new therapies, especially if there is a possibility of disease treatment. The panel considered methods to convince patients to become involved in long-term studies with complicated outcomes. Everyone agreed that patient enthusiasm and involvement is very important. Patient advocacy groups are critically important to recognize the disease and communicate options for treatment strategies and clinical trial opportunities.

The meeting attendees discussed the use of proteinuria as a primary or secondary endpoint. Some participants strongly believed that proteinuria is a valid primary endpoint. Understanding the pathophysiology of disease will facilitate the acceptance of indicators. Fibrosis is another example of an indicator that requires further study before it can be definitively labeled as such.

A participant queried which industry representatives would be willing to donate data from their clinical trials for the common good of patients. Some panelists noted that their companies had contributed standard of care data to large repositories. Each company must consider the legal

repercussions resulting from data contribution as well as other barriers, such as intellectual property rights. One option is for a biostatistician or clinician to approach an individual company with a specific analysis proposal to be granted access to the data. The Critical Path Institute is setting a precedent with their community-wide effort to improve the availability of neurobiology-related clinical trial data.

REGISTRY/COLLABORATION ROUNDTABLE

Moderator: *William Schnaper, M.D., Professor, Department of Pediatrics, Northwestern University Feinberg School of Medicine, Chicago, IL*

The registry and collaboration roundtable will stimulate discussion regarding the questions: What have we learned from other diseases and existing glomerular disease registries? What are the critical issues?

Collaboration in Rare Diseases Research

Stephen Groft, Pharm.D., Director, Office of Rare Diseases Research (ORDR), National Center for Advancing Translational Sciences (NCATS), NIH

A rare disease is defined in the United States as a disease with a prevalence of less than 200,000 individuals. Dr. Groft explained that rare diseases affect approximately 18 to 25 million people in the United States and 6 to 8 percent of the global population, although the exact prevalence is unknown. Collaborative efforts of the rare diseases community—including academic research investigators, medical specialists, federal research programs, patient advocacy groups, philanthropic foundations, and industry—are needed to best address the more than 7,000 genetic and acquired rare diseases. In December 2011, NCATS assumed responsibility for NIH's ORDR. Successful research and product development efforts for "orphan" and rare disorders occur primarily through a coordinating Disease-specific Steering Committee.

The ORDR is in the process of developing a web-based Global Rare Diseases Patient Registry and Data Repository (GRDR) to help aggregate existing patient registries. Another tool under development is the Registry of Biospecimen Repositories. In addition, the Research, Condition, Disease Categorization (RCDC) now includes a category for rare diseases and orphan drugs, reflecting 11.38 percent of the NIH research budget. A significant collaborative research activity is the Rare Diseases Clinical Research Network, which uses a data coordinating center with a strong emphasis on patient advocacy and community involvement, which has expanded to include 18 consortium members currently.

Dr. Groft reviewed the cycle from the biospecimen repository and natural history studies through better enrollment in clinical trials, Phase IV studies, and the patient contact registry. Patient registries face multiple challenges, particularly issues surrounding: informed consent; the globalization of rare diseases registries; the roles of government, service providers, private sector, and advocacy groups; and common data elements. The GRDR is working on these issues, and it provides information about rare diseases to clinicians and patients on its website. The Program currently is addressing data ownership and access concerns. Common data elements have been defined, and work is underway to develop specific data for individual diseases. Dr.

Groft said that a joint workshop between the NIH and FDA on May 16–17, 2012, will focus on natural history studies of rare diseases.

Discussion

A participant asked about efforts toward a centralized IRB for all of these diseases. Dr. Groft confirmed that a number of efforts are underway. The National Institute of Neurological Disorders and Stroke (NINDS), for example, has worked toward developing a centralized IRB for its programs. Localized IRBs remain a challenge for research networks, but resolution would ease many issues for investigators.

Dr. Groft explained that the novel approach in NCATS involves the Preclinical Innovation and Clinical Innovation Divisions. A trans-NIH Working Group regularly meets to review relevant applications and consider how to share resources among the Institutes and Centers. The CTSA Program is housed within NCATS' Clinical Innovation Program, and collaborative opportunities with an emphasis on translational research are welcomed.

Public-Private Partnerships/Collaborative Opportunities/An NIH View
Barbara Mittleman, M.D., Director, Public-Private Partnership Program, Office of Science Policy and Office of the Director, NIH

Dr. Mittleman began with a summary of key points made throughout the workshop. Efforts, resources, data about patient populations, and other foci of the community should be integrated to facilitate the greatest synergy and advance research. Better efficiency in extracting information from repositories can speed scientific progress and reduce costs. In addition, the cultures of sector stakeholders (e.g., academia, industry) vary widely, as do the reporting and reward systems in each sector, which often results in misunderstanding; communication is key.

The NIH can leverage connections to facilitate partnerships among external organizations. Success in partnerships relies on careful attention to details—it is important to determine who gets credit, who is responsible for resources, and what is the flow of work. A value proposition must be communicated persuasively and frequently to the leadership in each participating organization, and this necessitates a champion at each organization who is influential enough within the power structure to help leadership make decisions and commit resources.

Dr. Mittleman next provided an overview of the NIH's approach to public-private partnerships. Public-private partnerships are neither technology transfer agreements nor collaborations between individual scientists. A partnership agreement provides a formal structure (often with a memorandum of understanding) that facilitates relationships and often is located outside the government (e.g., Foundation for the NIH [FNIH], C-Path Institute). The government recognizes that partnerships offer a way to complement and leverage Federal resources. In some instances, the government cannot complete all the work itself; for example, patient advocacy groups have greater access to patients. In addition, resources always are limited, and established partnerships can provide agility and speed to scientific research. The principal sectors in biomedical public-private partnerships are: the NIH, industry, nonprofit organizations, academia, and patients/the public; each sector has a distinct mission and its own set of drivers and controllers. Partnerships

occur, for example, to advance science, promote public awareness, and support drug development. Dr. Mittleman noted that, with the Biomarker Consortium already established in the FNIH to promote biomarker science and qualification, there is no need to create new partnerships for the goals within NIDDK's glomerular research that fit within the existing Consortium framework.

NIH public-private partnerships can provide the NIDDK with a means to develop or obtain new tools, approaches, science, targets, compounds, trials, devices, and diagnostics. The NIH can partner with organizations on activities that are driven by rigorous science; with fair, inclusive, and transparent processes; compliant with Federal law, regulation, and policy; and focus on topics that are priorities to the Agency. To ensure success, shared goals and objectives must be defined, shared understanding of the task and requirements is needed, a common culture should be established, and communication is important. Product development partnerships offer a novel model, bringing venture philanthropy, drug development, and partnership together to overcome some of the barriers that industry has grappled with during the past 15 years.

Glomerular Disease Collaborative Network
Patrick Nachman, M.D., Professor, Kidney Center, University of North Carolina (UNC) School of Medicine, Chapel Hill, NC

Dr. Nachman provided an overview of the Glomerular Disease Collaborative Network (GDCN). The GDCN was established in 1985 at UNC to enhance communication between nephrologists and clinicians and compile clinical and outcome data on patients. The GDCN purpose is to investigate the etiology and pathogenesis of glomerular diseases and evaluate therapies.

GDCN patient registries follow patients from diagnosis to death, establish demographics and clinical characteristics, evaluate predictors of outcome, and observe current treatment trends. They also help in the design and recruitment for clinical trials as well as for laboratory, translational, and epidemiologic studies (e.g., through acquisition of biologic samples, questionnaires, interviews). Data about patients generally enter the registry when results of a renal biopsy conducted by a community nephrologist are uploaded to the GDCN database or following a referral to the UNC glomerular clinic for a renal biopsy; proper patient consent is required for inclusion into the database regardless of where the biopsy was completed. The GDCN process includes clinical and chart reviews, trial recruitment, newsletters for physicians about ongoing studies, surveys, and stored samples consent for laboratory tests.

GDCN patient registries currently encompass many disorders, including LN, FSGS, MN, and IgAN, among others. The database includes data concerning 16,500 patients, 1,000 of whom are in the pediatric group. Approximately 4,000 individuals have provided long-term consent, and many of these are enrolled in at least one study. The GDCN registries contribute to research and improved care through the collection of biologic samples in conjunction with clinical and pathological data; the provision of outcome predictors among patient cohorts of each disease; and the conduct of quality-of-life, health behavior, and epidemiologic studies. The registries also provide preliminary data for study design and sample size estimates (e.g., NEPTUNE), assist with recruitment for clinical trials, and produce and disseminate patient education materials. Eighty-five articles have been derived directly from GDCN registries and/or the bio-bank.

Strengths of the GDCN are that it is a collaborative effort involving passionate individuals that include patients, community nephrologists, and UNC faculty. It integrates the renal biopsy, extensive clinical data, and the bio-bank; it also is inclusive of incident and prevalent patients, and provides a demographic picture of the general population. Other strengths include a comprehensive data and medical records collection, representative of actual treatment trends, and long-term follow-up of patients.

Challenges include the lack of direct funding from granting agencies, absence of membership dues, and the lack of standardization of clinical information, laboratory tests, and data collection. In addition, recruitment and data collection can be difficult, and data collection and analysis, which are driven by specific studies, involve laborious data extraction from charts.

Future directions include discovering ways to simplify and expedite patient consent (e.g., through electronic contact) and improvements to the timeliness of data collection. The GDCN welcomes collaboration with other registries, and Dr. Nachman described a collaborative activity with the Toronto Glomerular Disease Network that resulted in nearly doubling a patient cohort.

Discussion

In response to a question about long-term consent from patients whose biopsy was conducted by a community nephrologist, Dr. Nachman indicated that the primary physician receives and reviews the form before providing it to the patient or sending it to UNC. He added that all patients are welcome to contact UNC for clarification about the form.

Toronto Glomerulonephritis Registry Group
Daniel Cattran, M.D., Professor, Department of Medicine/Nephrology, University of Toronto, Toronto, Canada

Dr. Cattran said that the Toronto Glomerulonephritis Registry (TGR) started in 1973 and was the template of the mission and functions of other registries described during this workshop. It also has benefited from collaborative opportunities with other registries during the past 30 years, that is, a way to accelerate the discovery and transfer of information. The TGR was designed for the patient, the nephrologist and pathologist, as well as the health care system. The number of people in North America with glomerular diseases currently is unknown ,a gap in knowledge that patient registries can help to define.

Dr. Cattran described TGR studies that indicate the utility of GN research. One study suggested the benefits of proteinuria reduction for membranous glomerulonephritis (MGN) patients in terms of disease remission. Clinical progress has been made as shown by studies that have revealed MGN remission data and patient survival across different periods of time has substantially improved . Other observational studies have been helpful in determining the value to the patient of achieving a partial remission in FSGS, as well as showing improvement in the rate of FSGS progression both in terms of outcome and remission rates over time. Studies of IgAN also demonstrate the benefits of proteinuria reduction for renal survival and help define partial remission in that disease as well. The TGR has aslso provided the infrastructure and

coordinating center for prospective randomized trial in MGN and ,FSGS The TGR is involved with basic science studies, including profiling idiopathic MPGN and HUS. In addition, TGR has been an active participant in NEPTUNE and is the largest recruiting centers for the network.

Future opportunities must take advantage of closer alignments between clinical and basic science. GN remains the most common cause of treatable ESRD. It is a rare disease, and conservative management is not sufficient to halt or reverse it. Immunosuppressive therapy continues to present challenges in terms of risks and benefit of therapy as well as costs . To address these challenges, TGR has worked with various groups throughout Toronto, including patient advocates, continued collaborative efforts with NEPTUNE and NephCure, and conducted surveys for needs assessments to help set priorities. To help with efforts across North America, TGR can help build a uniform renal biopsy classification and collection system, create a cost-effective glomerulonephritis research network of interested centers with high clinical volume for full data/biospecimen collections, and establish "centers of special expertise" to maximize science outputs for observational/clinical trials. Other activities could focus on better and more rapid communication among nephrologists, registries, and patients and collaborate in the creation of a" neutral" unbiased center for glomerulonephritis biopsies and data. These activities would significantly expand current scientific resources, provide opportunities for new investigators, support the creation of larger glomerulonephritis data bases , provide a unique environment for clinical investigation, and accelerate the timeline for the integration of basic science discoveries into the clinical domain.

Discussion

The moderator asked all roundtable presenters to consider the following question: Since different groups might have different interests, how realistic is it to assume that they will gather common data that will permit comparisons between groups or pooling of data for larger analysis?

The Nephrotic Syndrome Study Network (NEPTUNE)
Matthias Kretzler, M.D., Professor, Internal Medicine and Computational Medicine, University of Michigan, Ann Arbor, MI

NEPTUNE is a collaborative multidisciplinary effort of scientists from the US and Canada supported by NIH (ORDR, NIDDK) and the NephCure patient support group. It was initiated to establish a Nephrotic Syndrome multidisciplinary research and education platform in the Rare Diseases Clinical Research Network (RDCRN). NEPTUNE's overriding goal is to enable translational glomerular disease research, accomplished via five specific objectives. The first is to establish a collaborative, integrated, cost-effective investigational infrastructure that is amenable to conducting clinical and translational research on FSGS, MN, and MCD. NEPTUNE has 18 enrollment centers (17 in the United States and 1 in Canada) and follows a distributed governance structure that includes established working groups, protocols, pilot programs, and other entities.

NEPTUNE's second objective is to perform longitudinal and observational cohort studies on patients with incipient, biopsy-proven nephrotic syndrome. To conduct a prospective, non-blinded, and standardized evaluation of clinical outcomes, an observational cohort panel

comprises a consecutive sample of 450 eligible and consenting patients from the 18 enrollment centers over a period of 2.5 years, with minimum follow-up at 2.5 years. The patients are recruited as early as possible to increase the likelihood that early disease stages could be obtained for bio-banking and histological analyses. Patient information is used to define molecular, clinical, and histological disease subtypes as well as new disease predictors and therapeutic targets. A series of high-resolution clinical phenotypes are being determined based on molecular phenotyping and other parameters (473 total) from a broad range of areas (e.g., demographics, family history, medication history, adverse events, and histopathology). Biological samples and other parameters are collected during the clinical visits that occur every 4 to 6 months.

The third objective is to use unique resources, clinical data, and specimens from the NEPTUNE database to conduct pilot and ancillary projects to attain a holistic understanding of patients with nephrotic syndrome. In addition to conventional analyses (one category of molecular parameters is correlated with clinical outcome parameters) an integrated biology approach aims to combine multiple molecular and clinical data sources including (but not limited to) the genome, SNPs, transcriptome, epigenome, proteome, and morphology for a more complete definition of the underlying disease process. Overall, 18 ancillary projects have been received. In addition, four NEPTUNE pilot projects have been initiated on a variety of topics (e.g., sample collection, digital histopathology, and intervention trials).

NEPTUNE's fourth objective is to institute a training program that prepares post-doctoral and junior faculty candidates for clinical and translational research in glomerular disease. The fifth objective is to collaborate with the ORDR, the Data Management and Coordinating Center, and the Halpin and NephCure Foundations to implement a web-based exchange platform for use by lay people, physicians, and scientists.

NEPTUNE is also succeeding in effective outreach using a patient self-registry with total enrollment by April 2012 of 1,258 patients. In addition, NEPTUNE has broadened its integration by engaging international research networks that are involved in molecular glomerular disease. Efforts are underway to coordinate cohort studies on a global level with research networks representing more than 11,000 patients into cohorts who have collected more than 3,100 biopsies.

In summary, NEPTUNE serves as a resource for translational studies by providing an infrastructure that enables effective translational research.

European Registries for Glomerular Diseases
Charles Pusey, D.Sc., Professor of Medicine, Department of Medicine, Imperial College London, London, U.K.

The European Renal Association-European Dialysis and Transplant Association (ERA-EDTA) Immunonephrology Working Group was established in 2009 and contains members from various European countries. Rosanna Coppo currently is the chairman and conducted a National Registries of Glomerular Diseases 2012 Survey. The creation of this survey was done with the anticipation of attaining future collaborations between the ERA-EDTA and the American Society of Nephrology (ASN).

The survey includes several registries. The Spanish Registry of Glomerulonephritis started data collection in 1994 and has collected more than 20,000 native renal biopsies. The Italian Renal Biopsy Registry impressively has collected over 31,000 biopsies contained in two registries, since 1987. Other registries include the Scottish Renal Registry (2,000 biopsies), the Czech Registry of Renal Biopsies (10,000 biopsies), the Norwegian Kidney Biopsy Registry (8,000), and the Polish Registry (10,000 biopsies). All of the aforementioned registries include clinical data; however, these data may vary from registry to registry.

The registries have triggered several collaborative work projects. These include European studies to validate the Oxford classification of IgAN and to validate the European Vasculitis Study Group (EUVAS) classification of ANCA-associated GN. A collaboration from the United Kingdom (UK Registry for Rare Kidney Diseases [RADAR]) works to establish a registry of patients with membranoproliferative GN. Additionally, there is a proposal to develop a national registry of GN in England.

There are many glomerular disease registries in Europe accounting for about 95,000 European patients, and countries without registries are interested in developing them. There is an effort to coordinate and share these data across the continent.

Discussion

A participant questioned the impediments to data sharing across European countries. Dr. Pusey responded that the work is in the early stages of development, and that effort is currently being made to share access to all of the European datasets.

Lessons From the Neptune/Nephcure Contact Registry
Larry Holzman, M.D., Professor, Department of Medicine, Renal Electrolyte and Hypertension Division, University of Pennsylvania, Philadelphia, PA

Dr. Holzman described the currently operating NEPTUNE contact registry. Operated by the NEPTUNE consortium in collaboration with the NIH-sponsored RDCRN DMCC at the University of South Florida, this registry is a simple contact registry that targets patients with FSGS, Minimal Change Disease, Membranous Nephropathy, and undifferentiated Nephrotic Syndrome. This registry records contact and simple demographic information and a self-reported diagnosis and is aimed largely at enhancing patient education, facilitating clinical study

enrollment, and supporting other research initiatives. Subject recruitment occurs by web-based self-identification, care provider referral, and by very active recruitment by the NephCure Foundation. At present the NEPTUNE contact register contains more than 1300 subjects with a broad age distribution, of which the majority have reported a diagnosis of either FSGS or Nephrotic Syndrome.

While the contact registry in its present form has provided a useful mechanism for identifying glomerular disease subjects, it also has clear limitations that include sample bias and its data elements remain without validation. Reliance on self-referral via the internet selects for a more educated and affluent subject population that have internet access. As it is presently constructed, the NEPTUNE contact registry lacks glomerular disease-specific data elements. For this reason, it is presently being renovated to include detailed information on family history and glomerular disease-specific clinical course. This update also requests care provider contact information that should facilitate validation studies, provider education and study recruitment.

The NephCure mission supports research seeking the cause of primary FSGS and other diseases that cause idiopathic NS, improve treatments, and find cures. NephCure programs fund research (committed more than $10 million), as well as promote and advance outreach, engagement, education, and advocacy. The Foundation is presently preparing to create and maintain a new international glomerular disease registry. It hopes this will be useful to scientists in academia and industry in advancing glomerular disease science by facilitating our understanding of disease natural history and particularly by enhancing access to subjects by scientists seeking to enroll patients in research studies. As part of this effort, the Foundation envisions creating a clearinghouse of existing international registries. NephCure believes that it is best positioned to house and administer this project since it has established visibility and credibility among glomerular disease patients, it can provide efficient administration, it has no proprietary interest, and it brings financial resources and permanence to the effort.

Registries, EHRs, and the Cloud

John Sedor, M.D., Professor, Case Western Reserve University, Cleveland, OH

A registry is a book or volume containing important items that are regularly and accurately recorded. Patient registries have been effectively used for multiple purposes and produced findings that have elucidated causes of disease and impacted clinical practice. However, registries can be expensive to maintain and curate and are most informative when the registry design is for a specific goal. Economic austerity requires scientists to exhibit financial responsibility. With meaningful use of electronic health records (EHRs) and development of cloud computing platforms, there are greater opportunities for data access to assemble cohorts for research with agility and diminished cost.

Recent research demonstrates that EHR data can be effectively used for research. Disease phenotyping criteria have been developed, validated and used in GWAS. EHR data have been used in observational comparative effectiveness research, studies of disease prevalence and management in community settings, evaluating disease trends over time, and measuring compliance guidelines. The upfront cost is not insubstantial; however, when an EHR/cloud computing system is established, costs plummet. In fact, clinical data, which is already “paid

for," can be repurposed when institutions link EHRs to other clinical and research databases, for example biorepositories and pathology archives..

Cloud computing provides storage and analytical capabilities to a community of end-users and these platforms have been applied to EHR data. An example of a cloud computing platform system interfacing with EHR data is the Explorys EPM System. Through the auspices of the CWRU Clinical Translational Sciences Collaborative, Explorys provides CWRU investigators with a search engine for normalized EHR data. This interface permits a "Google-search-like" query of an institutional EHR to determine availability of patients with specific phenotypes required for research projects; allows capture of de-identified, population level data across health systems; and permits assembly of observational datasets, with a scalability not previously possible. Standard patient-oriented research models are expensive, engendering costly investments in personal and data collection. The new EHR/cloud computing paradigm promises decreased costs in needed FTEs and time required for project completion and promises to promote collaboration and data sharing.

Some investigators remain appropriately skeptical about uses of EHR data for research and certainly use of EHRs and cloud computing platforms will not be applicable for all research purposes. Implementation of EHR data for research purposes will necessitate addressing a number of issues. These barriers include validation of extracted data, development of data warehouses to link EHRs and research databases, need for interfaces between different EHR platforms, development of data sharing agreements between institutions, development of ad hoc querying tools, need to comply with human subject safety and privacy regulations and the need to harmonize ontologies of patient care versus research. Despite these challenges, use of HER data and cloud computing platforms provides an opportunity for investigators to address questions at scalability previously not possible and with greater efficiency and cost-effectiveness.

Discussion

A participant noted that diabetes was a common disease and questioned whether rare diseases, such as FSGS, had been evaluated for the benefits of EHR. Dr. Sedor responded that for glomerular diseases, specific EHR phenotypes must be defined and validated. Collaborations between researchers and clinicians will assist in this process. When accomplished, investigators will be able to assemble larger cohorts of these patients in order to better understand natural history and outcomes of treatments, for example.

An attendee asked about IRB approval to access EHR for patients with a disease of interest. IRBs might generally not allow for researchers to access EHR in a broad fashion (e.g., every patient with the word "kidney" in their file), and require more specifics. Dr. Sedor conceded that regulatory and IRB issues do exist. Some projects require the identification of individual patients, but partnerships between investigators at different institutions can be developed and IRB approvals can be attained with persistence. Genetic data present additional challenges since patients can be specifically identified with relatively small numbers of genetic variants.

A participant said that the presentations today highlighted many problems, including the globalization of research and the inherent coordination required. EHR, regulatory and consent

barriers have been addressed and overcome by different groups, and continue to be examined in research studies of patients with both rare and common diseases. Cooperation, collaboration, and time are required to further this effort.

Dr. Sedor indicated that they were working on using his institutions Epic EHR structure to automatically identify patients to enroll in studies based on and inclusion/exclusion criteria through the alert functionality. A participant noted that honest broker offices within organizations mitigate data accessibility issues and garner permission to conduct research. De-identifying data also can help to reduce the regulatory burden.

Registry/Collaboration Roundtable Discussion

A participant noted that the United Kingdom has initiated a program to solicit volunteers to provide DNA samples and simultaneously register for future research programs for which they could be recruited. DNA from more than 10,000 people in Cambridge has been sequenced already. This type of program could provide a platform to extend to patient populations. Society is very interested in helping advance medical progress.

"Meaningful use" is a significant term to clinicians and others involved with Medicare patients. In 2014, one of the meaningful use opportunities will be state-based registries. It would be useful to create a state-based glomerular registry in the same way that the oncology community has created their state-based cancer registry. This could present an opportunity to systematically generate high-quality data. Universities, hospitals, and insurance companies will be benefitting financially from "meaningful use" and those interests could be leveraged to improve the glomerular data collection and registry population.

SUMMARY

Robert A. Star, M.D., Director, DKUHD, NIDDK, NIH, Bethesda, MD
Michael Flessner, M.D., Ph.D., Director of Inflammatory Renal Diseases, DKUHD, NIDDK, NIH, Bethesda, MD

Dr. Star thanked Dr. Flessner and the organizing committee for their efforts and expressed appreciation for the productive meeting. The objectives were to bring together multiple parts of the glomerular disease community, including industry, patient advocacy groups, patients, and scientists. Dr. Star asserted that the meeting was successful and the groups developed wonderful ideas to move research forward.

Glomerular disease presents a large unmet clinical need, which has not improved for decades. Recent epidemiological and physiological studies have shed light into the "black box" of glomerular disease mechanisms, paving the way for rapid progress. It is imperative that the field move in an optimized and proactive direction. The community can collaborate in pre-competitive arenas to think broadly and discuss longer-term disease development plans. Collaboration also is necessary to qualify PROs, for use as a clinically relevant outcome measures. Working with the FDA to create industry guidance will be a useful next step. Another goal is to create common data definitions and data elements to inform future registries, along with developing standardized

disease models that better mimic human glomerular disease. Creating longitudinal registries and cohorts, perhaps populating with EMR data, will be critical. Glomerular disease research suffers from a lack of specific informative biomarkers, providing another opportunity for scientific advances. Characterizing and reducing the heterogeneity of patient subgroups will facilitate the evaluation of therapeutic interventions. The field first needs to validate pathophysiologic targets that can facilitate drug screens for hits, leads, and eventual drug identification. "Phase zero" studies could test various aspects of drugs early in the pathway to evaluate engaging and saturating the target.

Dr. Star detailed systematic changes that should be embraced by the glomerular field. Academics need to think long-term and broaden the scope of their work. Clinicians need to change their paradigm of treatment to involve all glomerular disease patients in clinical studies and trials. Industry needs to work together in the pre-competitive arena, share data for control and treatment groups, and share samples. Patients need to push the system to work faster and more efficiently in the right direction by becoming more engaged in research. The role of government will be to encourage these interactions and identify gaps where value can be provided. Dr. Star noted that this will not be an easy process, but is necessary to improve the quality of life for patients with glomerular disease.

Report—Breakout Group 1: Minimal Change/FSGS

Moderators: *William E. Smoyer, M.D., Professor/Vice President, Clinical and Translational Research, Department of Pediatrics/Clinical and Translational Research, The Ohio State University/Nationwide Children's Hospital, Columbus, OH*
Jochen Reiser M.D., Ph.D., Ralph C. Brown Professor, Chairman of Medicine Rush University , Chicago, IL USA

Breakout Group 1 discussed the many challenges surrounding rare diseases such as MCD and FSGS. As a result of these discussions, Breakout Group 1 suggested the following actions:

- **Conduct Studies to Better Define the Natural History of MCD/FSGS and Response to Therapy.** Because of low patient numbers, the usual approach of assembling patients and performing randomized clinical trials is not sufficient. Detailed studies need to be conducted to better define the natural history of MCD/FSGS and characterize the response to specific therapies. It is important to understand the primary and secondary pathways of disease pathogenesis (e.g., heterogeneity), and to identify and develop better biomarkers of both the disease stage (e.g., initiation, maintenance, or progression) and disease activity.

- **Include Plans for Meeting FDA Approval in Future Proposed Clinical Trials in MCD/FSGS.** To accelerate the development and approval of new therapies for MCD/FSGS the field needs to move rapidly toward the inclusion of plans for meeting FDA approval in all future proposed clinical trials in this disease. Investigators often realize too late that they have not done what will be needed to receive FDA approval for a drug. They need to demonstrate the ability to understand the mechanisms of action of these drugs and generate data that link quantitative reduction in proteinuria with clinical outcome, irrespective of the clinical agent. Patient Reported Outcomes (PROs) as well as objective outcomes are needed to correlate proteinuria values with outcomes.

- **Develop Multi-Center Clinical Trial Infrastructure for MCD/FSGS.** High priority should be given to the development of a robust multi-center clinical trial infrastructure for MCD/FSGS. This would both accelerate clinical trial enrollment and also enhance trial completion. Using this infrastructure to conduct a series of small proof-of-concept trials in treatable single-gene forms of MCD/FSGS would also better inform the best approaches for making subsequent larger investments.

- **Formalize Collaborative Relationships and Standardize Processes with Stakeholders**. Clinical trial enrollment and completion would also be greatly accelerated by the development of formalized collaborative relationships and standardized processes among all of the relevant stakeholders in the research arena (e.g., NIH, FDA, academia, industry).

- **Identify Markers of Disease Stage and Activity.** Investigators need to develop mechanisms to better delineate the entire phenotype at entry to clinical trials and beyond. Essential data should include: Clinical parameters, Histopathology, Genetics (e.g., biomarkers of monogenic causes and modifying/susceptibility genes), other etiologic biomarkers, and environmental exposures. A standardized minimal dataset that considers all of these issues

should be developed to facilitate registry and dataset harmonization. In addition, expert laboratories available to work with these registries should be identified.

- **Identify or Develop Biomarkers that are Relevant in Animal Models and Humans.** There has been frustration created with the prior identification of biomarkers that are relevant to animal models, but that are later found not to be useful in clinical trials of human MCD/FSGS. This disconnect could be reduced by developing high-throughput animal and cell culture models to screen small molecule libraries and biologics for MCD/FSGS treatment. In addition, screening available drugs for possible repurposing and partnering with industry regarding positive outcomes would also be a very auspicious approach. Breakout Group 1 identified numerous biomarkers of MCD/FSGS on which to focus research efforts. The best evidence of biomarker efficacy currently available is for proteinuria. This typically is measured as 24-hour collections and first-morning urine protein/creatinine ratios, but urine could also be analyzed for protein selectivity, as well as urinary albumin excretion (as a comparison to proteinuria). Additional biomarkers for MCD/FSGS for which evidence exists includes monogenic mutations and urinary podocytes. Finally, several putative biomarkers discussed included CD80, urine catalytic iron, suPAR, and cell-based functional assays.

- **Pursue Candidate Biologic Targets for Future Approaches.** Breakout Group 1 developed an extensive list of candidate biologic targets for future research, representing targets or pathways that were collectively identified by the group and had a good basis on which to focus future research efforts. Some of these targets included:

    - Cytosolic cathepsin L (general biomarker)
    - Angiopoietin-like 4 (MCD biomarker)
    - suPAR (FSGS causative molecule, biomarker)
    - The p38 MAPK / MK2 pathway
    - Reactive oxygen species,
    - Rho GTPases, large GTPases such as dynamin
    - The Jak/Stat pathway
    - Protective eicosanoids
    - Histone modifiers
    - The genes *SMPDL3b*, *APOL1*, and *COQ10*.

For each target chosen, the phase in which each of these targets are involved (e.g., initiator, mediator) should also be identified.

One participant reiterated the importance of ensuring that "patient advocacy groups" be included in the list of stakeholders.

Discussion—Breakout Group 1: Minimal Change/FSGS

Drs. Smoyer and Reiser described the group's charge to establish targets for future research and provide specific suggestions for moving the MCD/FSGS field in glomerular disease forward. The group focused its discussions on defining next steps and identifying candidate biomarkers and targets.

Because there are many subtypes of FSGS, including genetic subtypes (e.g., primary vs. secondary), more natural history studies are needed to help determine the most appropriate therapy for each subtype. Such studies should encompass genetics and physiology as well as pathology, which provide the basis for general phenotyping. Better delineation of the entire phenotype also should include clinical parameters and environmental exposures. In addition, studies of molecular diagnostics along with standard techniques could more clearly define or reclassify diseases.

To help advance the MCD and FSGS fields rapidly, single gene disorders should be identified within 5 years, and the pathogenesis studied thenceforward. In the meantime, a broad-based therapeutics approach should be developed to understand key pathways for both FSGS and MCD. Specific markers are needed to better distinguish between MCD and FSGS.

The pathway to regulatory approval should be clarified to expedite research and therapeutic approval. Specifically, focus should be placed on developing and gathering approaches, tools, and data to facilitate the FDA's review of potential therapeutic agents for FSGS and MCD. The FDA requires quantitative data that provide evidence that a given therapy is tied to the reduction of proteinuria. Nephrologists generally monitor change in kidney function and reduction of proteinuria as indicators of disease progression/regression. It has not been proven, however, that a 50% reduction in proteinuria is a benefit. The greatest issues are whether a 50% reduction in proteinuria could be used for all future drugs, and whether the reduction could be used both for nephrotic syndrome and CKD. Data that link proteinuria with ESRD are needed. Concerted efforts should address concerns about proteinuria in terms of nephrotoxicity. Studies are needed to support the reliance on proteinuria as a proxy metric.

A strategic framework should be developed that shows the clinical benefit of partial response versus full response and incorporates PROs. Outcomes are often based on how the patient feels, functions, or survives. PROs are needed in addition to clinical data, and these should be linked in a coordinated way.

Better mouse models should facilitate the translation of results from animal to human studies. One possibility is to generate data in human biospecimens that would indicate which mouse study outcomes would have the greatest relevance in humans. Translational biomarkers are difficult to obtain, and great skepticism exists in industry**.**

Scientists and clinicians should distinguish between initiation, maintenance, and progression of disease. MCD and FSGS involve complex processes, and better classification of where patients are in terms of these diseases would be valuable in enrollment of patients into the most relevant clinical trials for their disease stage.

Candidate targets include: cytosolic cathepsin L; angiopoetin like 4; suPAR; MK2, p38 MAPK pathway; reactive oxygen species (upregulation); RHO GTPases; activation/deactivation (e.g., integrins on podocytes); Jak/Stat pathway; glomerular epithelial cells. Participants recommended that for each target chosen, the phase in which each of these targets are involved (e.g., initiator, mediator) should be identified.

Specific biomarkers include: urinary albumin (to compare biomarkers against) and proteinuria for some cases; CD 80; monogenic mutations; urine catalytic iron; urinary podocyte markers, including urinary exosomes and podocyte RNA; proteins for selectivity; and suPAR.

Prenatal exposures could be considered in terms of risk factors, exposures, birth weight, and other possible contributors. Dietary exposures cannot be measured, and the effect of diet is unknown. Some patients, for example, have food allergies. There is a clear connection between food antigens and disease, IL-6, TNF-alpha, and others that should be explored. Small molecule libraries should be used to help identify druggable targets and define mechanisms (e.g., transgenic zebrafish to develop high throughput assays to span the three phases of initiation, maintenance, and progression).

Report—Breakout Group 2: Membranous Nephropathy

Moderators: *David Salant, M.D., Professor, Department of Medicine/Nephrology, Boston University Medical Center, Boston, MA*
Fernando Fervenza, M.D., Professor, Department of Nephrology and Hypertension, Mayo Clinic, Rochester, MN
Daniel Cattran, M.D., Professor, Department of Medicine/Nephrology, University of Toronto, Toronto, Canada

Breakout Group 2 emphasized that membranous nephropathy (MN) is a treatable and curable disease. The group noted that they based their recommendations on the assumption that a standardized reporting system for a data registry, linked to a biorepository, will be established. Specific recommendations from Breakout Group 2 include:

- **Identify key questions.** Can anti-PLA_2R be validated as a biomarker for active MN? Can other targets for secondary MN and anti-PLA_2R negative cases be identified? The group also discussed the necessity of defining a pathogenic and genetic basis for primary MN. Pathogenesis from animal models should promote progress to define pathogenesis in humans and the genetic basis of disease. Breakout Group 2 recommended forming a working group of patients, advocacy groups, nephrologists, and industry representatives to develop PROs and other efficacy endpoints. New therapies should be developed based on pathogenic mechanisms.

- **Validate anti-PLA_2R as a biomarker.** To validate anti-PLA_2R as a biomarker for MN, all future prospective MN studies should include baseline and follow-up measurements of anti-PLA_2R. Investigators should consult the FDA to define the study design necessary to validate anti-PLA_2R for diagnostic and therapeutic purposes. It will be relatively easy to establish

anti-PLA_2R as a diagnostic marker and more complex to use as a prognostic indicator for therapeutic studies.

- **Develop novel therapies.** MN is a therapeutically treatable disease; most patients who do not spontaneously enter remission respond to immunosuppressive therapy. Anti-PLA_2R is a reproducible biomarker with 75 percent sensitivity and 100 percent specificity. To modulate the target of anti-PLA_2R, investigators should be able to develop apheresis techniques, design small blocking molecules to inhibit binding to antigens, or develop affinity absorption to remove pathogenic molecules. A valid experimental model expressing anti-PLA_2R in podocytes will facilitate research in this field.

- **Better understand the pathogenesis of primary MN.** To accomplish this goal, it will be important to identify at-risk patients through epidemiologic studies (e.g., availability of a large dataset to identify patients who develop MN) and genetic investigations (e.g., Class II MHC or PLA_2R risk alleles conferring susceptibility). Defining the properties of anti-PLA_2R IgG_4 also is critical to better understand the pathogenesis of MN.

Discussion—Breakout Group 2: Membranous Nephropathy

The key questions to be answered for this disease pertain to etiology, pathogenesis, diagnostics, and therapeutics. Patients both negative and positive for PLA_2R need to be characterized carefully, especially because antibody-mediated disease (nephrotic or non-nephrotic) does not resolve spontaneously. In addition, the factors underlying disease progression or therapeutic nonresponse warrant consideration. For example, does the damage from the initial insult lead to the incorporation of other epitopes that subsequently trigger rapid response and disease progression?

Rather than focus on disease cures and symptom alleviation in an effort to establish areas of funding significance, one participant considered the utility of combining narratives from the nephrotic disease spectrum into a focused objective that cuts across all funding objectives to advance the acceleration of disease interpretation and solutions. The development of therapeutic agents based on patient-derived data has failed because of a lack in common resources. The nephrology community has not presented the problem and potential management solutions effectively to the NIH and public. The CF community, however, has succeeded by soliciting organizations with specialized research capacity and incorporating their efforts into a uniform process, which tends to garner higher public and funding approval. The resulting product would overlap with NEPTUNE, which is based on registry projects over many years, but it would go in greater depth. Retrospectively, a vast amount of information exists for MN, but it is not cohesive in comparison to other glomerular disease states. Prospective studies are needed to answer the important questions in an accelerated timeframe.

Having the FDA and NIH at the same table is critical; generating large amounts of data is useless if the FDA will deem it unacceptable. Worldwide, researchers are studying proteinuria in nephrotic disease, but U.S. regulatory agencies (FDA) will not accept proteinuria as an endpoint because of insufficient interventional trials. Industry needs an acceptable endpoint to conduct clinical trials in a reasonable amount of time. In terms of efficacy and clinical studies,

incomplete or partial remission is not acceptable to the FDA. Complete remission is an acceptable endpoint if it is accompanied by a long-term outcome study. Because proteinuria is inconsistent as a surrogate marker, the FDA prefers the use of clinical manifestations of the disease that are relevant to the patient. A patient exhibiting deteriorating proteinuria needs rescue therapy, which causes surrogate markers to become clinical manifestations of the disease and relevant to the patient. FDA's role at this early stage was questioned—NIH should fund research, collect the evidence, and provide the successful disease-treatment measures with the FDA. Does early FDA consideration skew the process?

A grant on patient-centered outcome research has been established to examine patient preferences, providing an opportunity to use a center-based approach similar to the CF approach, but including other elements, such as quality of life. However, the development of patient reported outcomes (PROs) is difficult and requires validation and datasets comprised of 1,000 patients, which is impractical and not sustainable at this time. Biopsy was suggested as a critical point of data collection because it is a unique feature of MN standard care.

The participants recommended that a working group comprised of representatives from both the nephrologic drug industry and patient advocacy groups in North America and the United Kingdom be established to develop meaningful efficacy parameters for MN, including patient-centric measures, renal function biomarkers (proteinuria included), and rescue therapy.

Regarding biomarkers, the group agreed that anti-PLA_2R is a drug-treatable target; most patients who do not enter remission spontaneously do respond to immunosuppressive therapy. Anti-PLA_2R is a reproducible biomarker (about 75% sensitive and 100% specific), but small studies are needed to test and validate anti-PLA_2R. The immunological relevance should be confirmed in preclinical surrogate models and presented as causal evidence to the FDA. A prohibitively large amount of antibody precludes the use of monkey studies. Another suggestion was to develop an animal model via transgenic antigen expression in the glomeruli. Finally, although anti-PLA_2R is associated with disease in approximately 80 to 85 percent of patients, excess proteinuria and creatinine occur in all patients and warrant extensive testing and validation.

With the assumption that a standardized reporting system for a data registry linked to a biorepository would be established, the group recommended the following for MN:

- Validate anti-PLA_2R as a biomarker for active MN.
- Identify biological targets for secondary MN and anti-PLA_2R-negative patients.
- Define underlying pathogenic and genetic mechanisms for primary MN.
- Develop new therapies based on pathogenic mechanisms.
- Form a working group comprised of nephrology industry representatives and patients in North America and coordinate with the United Kingdom to develop meaningful efficacy measures for MN.
- Include anti-PLA_2R baseline and follow-up measurements in future prospective studies on MN.
- Consult with the FDA on acceptable study designs for validating anti-PLA_2R for diagnostic and therapeutic purposes.
- Develop an informative experimental model that demonstrates anti-PLA_2R's utility as a reproducible biomarker.

- Use epidemiologic and genetic studies to identify patients at risk for primary MN.
- Develop a human PLA_2R transgenic model.
- Define the properties of anti-PLA_2R IgG_4.

Report—Breakout Group 3: IgA Nephropathy

Moderators: *Heather Reich, M.D., Ph.D., Clinician Scientist, Department of Nephrology, University of Toronto and University Health Network, Toronto, Canada*
Jan Novak, Ph.D., Associate Professor, Department of Microbiology, University of Alabama at Birmingham, Birmingham, AL

The most common cause of primary glomerulonephritis is IgA nephropathy (IgAN). One third of patients with IgAN in North America will progress to end-stage kidney failure. Due to a lack of understanding regarding the pathogenesis of IgAN, there is no targeted, *i.e.*, disease-specific, therapy for this disease. Furthermore, physicians' ability to identify patients at highest risk of progressive disease is limited; as a consequence, individuals at low risk of progression may be unnecessarily exposed to highly toxic chemotherapies, and patients at high risk of kidney failure may not receive sufficiently early and potent therapy.

The current environment is ideal to move forward with international collaborative network-based approaches to improve understanding of the pathogenesis of IgAN and develop clinically relevant non-invasive biomarkers of disease activity and progression. Breakout Group 3 noted that the environment is ideal because of the recognition of the importance of global collaborative efforts to study rare diseases, enthusiasm and energy from patient advocacy groups and the pharmaceutical industry, and an emerging framework of understanding of the pathogenesis of IgAN. There are several existing successful models and resources to leverage to help build collaborative frameworks. Breakout Group 3 presented the following specific recommendations and priorities:

- **Understand disease immunopathogenesis and develop disease-specific targeted treatments.** Defining pathogenic immune response patterns and upstream pathways involved in the production of aberrantly glycosylated IgA1 will facilitate the identification of therapeutic targets and development of novel pharmacologic interventions. A better understanding of the heterogeneity, origin, and levels of anti-glycan antibodies is needed. Defining the inciting triggers and mechanisms responsible for the formation of immune complexes, and the composition of pathogenic immune complexes that stimulate mesangial cell proliferation, is an important need. Discoveries from genetic studies will identify new targets that can be tested using *in vitro* and animal models. This information could lead to clinical trials using novel and existing compounds directed at these targets.

- **Identify new biomarkers of disease activity and progression.** The identification of biomarkers of disease activity, severity, and progression is an important research need. Better understanding of the mechanisms responsible for immunopathogenesis and progression of IgAN will facilitate identification of such biomarkers. While markers specific for IgAN are desirable, identification of non-IgAN-specific markers of early disease

progression will also enhance prognostication and monitoring of all patients with glomerular-based disease.

- **Develop new diagnostic tests for IgAN.** Emerging technologic advances should be explored to develop non-invasive diagnostic and monitoring tests. These may include high-resolution mass spectrometry for analyses of aberrant glycosylation of IgA1 and characterization of pathogenic immune complexes as well as novel imaging procedures, including target-specific molecular imaging and functional MRI approaches.

- **Establish a global collaborative network.** An international collaborative network should be comprised of committed investigators and advocacy groups, including NIH investigators. An international perspective is necessary as IgAN is a clinically and geographically heterogeneous disease with both genetic and environmental contributions to pathogenesis and prognosis. The group identified a need for large-scale, interdisciplinary collaboration to facilitate a better definition of morbidity and mortality in patients with IgAN, and fully characterize the multiple steps in the pathogenesis of the disease. It is critical to view the disease from an interdisciplinary perspective, including basic and clinical research. A reasonable hope exists that these studies will identify new targets that will be disease-specific and associated with markers that allow investigators to follow the progression of the disease and treatment response.

- **Relate therapeutic interventions with definitive outcomes.** IgAN is the most common primary GN and is potentially treatable. Challenges to establishing the efficacy of therapeutic interventions to prevent disease progression include the facts that IgAN is relatively rare, compared to cancer or cardiovascular diseases, and asymptomatic, progression of IgAN frequently occurs over the course of several years, and few surrogate markers of response have been definitely correlated with definitive outcomes (*i.e.,* organ and patient survival). The impact of interventions on definitive outcomes such as kidney and patient survival must be clarified. Demonstration of histologic improvement following treatment also remains unexplored as a measure of response, and a reproducible and clinically validated scoring system, the Oxford classification, will now permit better quantification of histologic changes in response to interventions.

Discussion—Breakout Group 3: IgA Nephropathy

The scope of IgAN is global, although the lack of biopsy-based registries precludes reliable estimates of the true incidence and prevalence of the disease. Consequently, the extent of disease burden in the United States remains poorly defined. Dialysis-based registries are not an accurate reflection of disease burden as at the current time, most people who are placed on dialysis have not had a biopsy to determine the cause of kidney failure. This suggests that a substantial proportion of patients with IgAN never receive biopsy-confirmed diagnosis prior to starting dialysis; as a result the impact of IgAN may be substantially underestimated. In the absence of a kidney biopsy, there must be a sound diagnostic test in order to support a clinical trial of people with IgAN.

IgAN is an important issue in Canada because it is the second leading cause of ESRD, and its proportion of ESRD has been increasing each decade since the 1970s. The population of people with IgAN in major cities in Canada includes a substantial proportion of individuals of Asian descent (approximately one-third), which is different from that in the United States. This difference offers unique opportunities for collaboration with U.S. researchers. Because of a lack of biopsy-based registries in the United States, it will be difficult to compare incidence and prevalence rates between the countries. One of the only population-based studies of IgAN in the United States was conducted in Kentucky and spans a two-decade interval 1975-1994.

The following are areas that were discussed as priorities for research.

A priority is to establish a biopsy-based registry to better understand disease incidence and prevalence. It is possible that a collaborative network would be needed to address the registry issue. This network should be pathology-based with biopsy-proven IgAN as the point-of-entry for a study. Because IgAN is considered an orphan disease in the United States, conducting studies will be a challenge. It is unlikely that electronic health record databases could be used until there is a wider application of biopsy-proven diagnosis.

Improving targeted treatments for IgAN is a challenge because of the rare disease designation; pharmaceutical companies show little interest in this area. Advocacy groups, such as the IGAN Foundation of America, are raising awareness and providing education, but more is needed to raise the profile to those who fund research.
The "multiple hit" pathogenesis was proposed as a reasonable model for pathogenesis of IgAN. This was the model presented by Dr. Novak, and is described in detail in his presentation. Practically all "hits" have significance in producing IgAN and are events that may provide opportunities for intervention. Much remains to understand about the model, but it is mature enough that additional research should be considered.

There are mouse models that also could be useful for the study of at least some specific aspects of the pathogenesis of IgAN. Sufficient information regarding mechanisms is available for a clear understanding of the steps needed to initiate IgAN. This should lead to identification of risk factors as an area of potential research. Other areas of research include finding a biomarker better than proteinuria, and a search for auto-antibodies involved in the process, such as the level of IgG antibodies that have an impact on IgAN.

A simple biomarker that has been considered is proteinuria, which is correlated with a more rapid rate of renal function decline in IgAN at far lower levels than in FSGS and MGN. However, reduction in proteinuria has not been related in randomized studies to prevention of end-stage kidney disease in IgAN.

The search for biomarkers depends on having a source of clinical biologic samples linked with detailed longitudinal clinical phenotype data; such a resource is generally deficient at this time. Novel biomarkers of disease response and activity must demonstrate a closer correlation with outcome than proteinuria to be of clinical relevance. The search for biomarkers not only helps to improve prognostic and monitoring capabilities, but offers new insights regarding the biologic mechanisms responsible for the development and progression of IgAN.

Input from FDA participants emphasized that regulatory requirements should be considered in the design of clinical and mechanistic studies of IgAN. Of great importance is developing specific IgAN patient reported outcomes (PROs), which are largely unexplored endpoints in studies of patients with IgAN. While this may be more challenging in a largely asymptomatic disease, research is required to better quantify the impact of IgAN on patient well-being and qualitative measures of patient health. Data need to be collected to show that an intervention influences PROs. Another potential measure of clinical improvement that remains largely unexplored in patients with IgAN is change in histologic appearance of kidney biopsy. While biopsy is an invasive procedure, improvement in histology may be a clinically relevant endpoint from a regulatory perspective.

In general, there is a need for research to identify the genetic components of IgAN. This may be the way to develop a relationship between histological outcomes and treatment responses. Ultimately, the goal is to design individualized treatments based on genetic profile considered in combination with other biomarkers of disease activity and risk.

Research in IgAN will benefit from using cross-cutting, interdisciplinary investigations. For example, in the pathogenesis model described by Dr. Novak, many of the hits described have relevance to many diseases and conditions, including FSGS. The use of functional MRI is improving the identification of kidney damage and inflammation in the kidney. Application for the study of IgAN should strongly be considered as a possible alternative to repeat biopsies before validated biomarkers are introduced in the clinical practice.

The nephrology community must engage with everyone with a stake in IgAN, and that approach will require large-scale, interdisciplinary collaborations. Data must be shared to establish biomarkers, especially in the search to show that treatment is connected to outcomes. This is vital to creating an information and research base to work with the FDA to gain approval for clinical trials and treatments. Along these lines, the letter of understanding being developed between ASN and FDA will be helpful in developing these new approaches and moving them to clinical and laboratory practice.

Report—Breakout Group 4: Vasculitis

Moderators: *Peter A. Merkel, M.D.,M.P.H, Chief, Division of Rheumatology, Professor of Medicine and Epidemiology University of Pennsylvania School of Medicine, Philadelphia, PA*
Patrick Nachman, M.D., Professor, Kidney Center, UNC School of Medicine, Chapel Hill, NC

Vasculitis research and treatment is at a more advanced stage than other glomerular diseases. Breakout Group 4 discussed opportunities to move the science to the next level. The group considered as vasculitis all vasculitides that include glomerular disease as a potential manifestation. They placed particular emphasis on ANCA-associated vasculitis (AAV), including granulomatosis with polyangiitis (GPA, Wegener's), microscopic polyangiitis (MPA), eosinophilic granulomatosis with polyangiitis (EGPA, Churg-Strauss), as well as the less well characterized diseases of cryoglobinemic vasculitis (CV), anti-GBM disease, IgA vasculitis (Henoch-Schoenlein), and drug-induced vasculitis. It was noted that hepatitis C virus-associated

CV poses different challenges than non-viral-associated CV. Specific recommendations by Breakout Group 4 include:

- **Attain long-term remission off treatment and improve outcomes.** In terms of treatment needs and goals, it is not unreasonable to begin thinking about curing the disease. Clinicians have accomplished long-term remission on treatment, but there can be complications and toxicity is a problem. Attaining long-term remission off treatment is an important goal, in addition to being able to identify patients who do not need long-term therapy. Reducing treatment-related toxicity, especially glucocorticoid-related toxicities and infections, and improving long-term outcomes by preventing damage and improving quality of life is important. Physician education to identify and diagnose patients earlier will improve long-term outcomes by having patients receive therapy more rapidly and facilitate referral to centers of excellence

- **Advance personalized treatment strategies in AAV.** Stratification variables, including ANCA type (e.g., anti-PR3, anti-MPO); clinical phenotypes (e.g., GPA, MPA); disease pattern (e.g., initial presentation, relapse); genetics and epigenetics; circulating markers in serum, cells, or urine; and histology or tissue markers need to be tested and validated longitudinally to incorporate as part of the disease paradigm and patient strategy. Vasculitis phenotypes are clinically different; signatures to identify the severity of the response will be useful. Treatments can target immunity or inflammation, may be different depending the phase of the illness, be disease-specific designer drugs or repurposed generic therapeutics (e.g., immunomodulatory agents used for other diseases such as rheumatoid arthritis).

- **Identify prognostic and diagnostic biomarkers.** ANCA has not achieved its promise as a marker of disease activity, response to treatment, or predictor of relapse. Candidate and discovery approaches should be employed to identify prognostic biomarkers (e.g., predictors of relapse, cured patients, markers of response potential, markers of treatment toxicity susceptibility) and diagnostic biomarkers (e.g., identify the 10% of ANCA-negative cases and the extent of disease severity).

- **Utilize available resources.** Current successful resources include research networks such as the Vasculitis Clinical Research Consortium (VCRC), European Vasculitis Study Group, French Vasculitis Study Group, Glomerular Disease Collaborative Network, and Italian Cryoglobulinemia Group. The vasculitis field has claimed numerous successes in the form of performing large, international randomized clinical trials, building cohorts, developing assays, partnering with industry and patient advocacy groups, and securing funding from government, industry, and private foundations. The VCRC, a member of the NIH Rare Diseases Clinical Research Network (RDCRN) has a strong partnership with industry and patients, and has an online patient contact registry with more than 3,000 patients from around the world. Long-term longitudinal cohorts identified through the VCRC have participated in clinical trials for six diseases (some drugs have gone on to labeling) and have supplied comprehensive data, DNA samples, and other biospecimens. The VCRC Data and Biospecimen Repository contains longitudinal data and samples from more than 1,000 patients. The repository has samples from recent clinical trials, a large DNA collection, and

an extensive collection of sera, plasma, and urine specimens linked to longitudinal clinical data.

- **Advance clinical and translational research in vasculitis.** Regarding funding efforts to advance studies in vasculitis, Breakout Group 4 suggested four priorities:
    1. Support research to develop personalized approaches to treatment (e.g., biomarkers, new drugs, trials).
    2. Maintain, expand, and leverage existing registries and cohorts. This can be accomplished by increasing the number of study centers, avoiding reinvention or parallel developments, and encouraging global collaboration by supporting an ongoing centralized organization rather than an *ad hoc* system.
    3. Focus on vasculitis as the overarching theme. Separate initiatives by research agenda rather than by proteinuric disease characteristics. Focus on systemic diseases in which glomerulonephritis is highly prevalent. Facilitate and encourage industry, NIH, and FDA attention to vasculitides.
    4. Continue to advance research in non-AAV vasculitides by connecting to AAV research, as well as performing specific studies and expanding the registry to other diseases.

Discussion—Breakout Group 4: Vasculitis

To refine specific ideas instead of generalities, Breakout Group 4 decided to focus on key vasculitis-specific issues, instead of universal disease concepts such as increasing quality and quantity of life, and discovering less toxic drugs. The breakout group determined that scientific opportunities of high value should be pursued. A cure for vasculitis is the optimal goal and should not be dismissed as a possibility.

Vasculitis is unique because it is a disease in which there already are international collaborations and trials that have found great success. Due to the moderately mature nature of the vasculitis research community, more precise outcomes are required for vasculitis because patients are doing better now than 20 years ago. The definition of a long-lasting "cure" is required because new therapies show an 80 to 85 percent acute success rate (remission-induction) and have significantly lowered mortality. Dealing with the acute disease is relatively easier than maintaining remission. Relapses and the cumulative burdens of disease and therapy substantially impact patients' quality of life.

Understanding which patients will undergo relapse is imperative. Additionally, to be considered is the definition of relapse because some patients appear to be in immunological remission, but routinely progress to relapse.

Another aspect of vasculitis that is somewhat different from several other glomerular diseases is that vasculitis is often a multi-organ system disease that often necessitates that patients are seen and cared for by doctors in various specialties.

Although ANCA titers do not correlate with disease reliably enough to directly affect patient care and treatment decisions, combining ANCA testing with patient-reported symptoms, direct

immune tests (e.g., T-cells), genetic profiling, and circulating markers (e.g., cytokines) might provide new opportunities for disease stratification and prognostication. Finding a single biomarker in vasculitis is likely a naïve goal. A diagnostic biomarker should be novel and powerful alone, or complementary to what already is available.

A more personalized medicine in each stage of the disease (before diagnosis and during initial treatment, relapse, and remission) should be realized in 10 years. Such approaches would likely incorporate various factors stemming from research in genetic profiling (pharmacogenomics on drug response, epigenetics, transcriptomics, and proteomics), circulating serologic markers, circulating cellular markers, and clinical phenotypic data.

Other treatment needs and goals include a cure, knowing when to treat a patient (and when to stop), prevention of damage, and early identification of disease and referral. Treatment targets are immunity and inflammation. Disease-specific and validated measures of quality of life are needed in vasculitis to advance treatments and ensure we are directing therapy to target what matters to patients.

Expanding and maintaining current vasculitis registries, repositories, and networks will be beneficial. This should include patients who are in remission so researchers can better understand the circumstances that create a vasculitis flare. Patient-level data from standardized questionnaires, including quality of life assessments, should be obtained more frequently than annually. This requires international partnerships and for work to go beyond one grant cycle. The NIH should help support these needs and support pathways for conduct of ancillary studies. Leveraging already-available NIDDK- or NIAMS-supported biorepositories is an option.

While shared data and specimens in a repository is a good idea, issues regarding data "ownership" and sharing are issues to be considered. Most researchers want to have time to analyze their data and specimens, often collected based on years of work, before they are shared with the entire vasculitis community.

Other areas of interest are in treating vasculitis differently to shorten treatment and remove the disease entirely, instead of having remission. Most patients are given long treatments as the default; however, they may not all require that practice. Patients with monocyclic vasculitis are distinct from those that have relapses; however, the pathogenesis is not understood well enough to predict a patient's future course. Non-AAV should not be forgotten, as these forms can be severe and life-threatening, although they are rare.

Educating the general public and primary care physicians (PCPs) would be beneficial in the avoidance of delays in referrals to specialists.

Many pharmaceutical companies now focus some resources to develop medications for rare diseases. Partnering with industry provides new options for government funding for vasculitis. The NIDDK can leverage their monetary resources with other government agencies to achieve such funding avenues.

Final suggested funding priorities in vasculitis research are: (1) support research to develop personalized approaches to treatment (e.g., biomarkers, new drugs, trials); (2) maintain/expand/leverage existing registries/cohorts; (3) focus on vasculitis as the overarching theme; and (4) continue to advance research in non-AAV vasculitides.

Report—Breakout Group 5: C3 Glomerular Disease

Moderators: *Terry Cook, M.D., Professor, Department of Medicine, Imperial College, Hammersmith Hospital, London, U.K*
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Breakout Group 5 declared that it is an exciting time to progress forward the state of C3 glomerular disease. There is a rapidly developing recognition of the spectrum of C3 glomerulopathy and major changes in the methods of classifying GN, particularly MPGN. A range of new drugs directed at the complement pathway are in preclinical or clinical (e.g., eculizumab) use. Excellent animal models are available, and others are in development. There is a high likelihood of targeted therapies becoming available in the following decade of research. Research for C3 glomerulopathy might have implications for other immune complex GN diseases, such as IgAN. Breakout Group 5 suggested the following specific actions:

- **Define and establish diagnostic criteria for C3 glomerulopathy.** The real challenge is to understand which part of the complement pathway is involved in pathogenesis in individual patients to allow appropriate therapeutic interventions. Progress in C3 glomerulopathy would benefit from recognition of the disease by pathologists and clinicians, definition of the range of histological appearances, and correlation of histological appearances with functional complement studies, genetics, clinical course, and outcomes. Increased capacity and timeliness of laboratory assessment of complement activation and complement gene variations is essential. Breakout Group 5 also plans to introduce measures to better identify cases of C3 glomerulopathy, including recurrent disease in transplanted kidneys. Clinicopathological correlation would be facilitated by the establishment of a registry with renal biopsies sent to a central site that can perform digitization of light microscope slides; digital electron microscopy and immunofluorescence images would also be acquired. The pathology would be classified by a panel of pathologists to establish criteria for diagnosis and allow the correlation of histological with clinical features.

- **Raise awareness.** To increase recognition, the field will hold an international consensus meeting to define C3 glomerulopathy and criteria for diagnosis in Hinxton, U.K., August 2012. The first C3 Glomerulopathy Focus meeting will be sponsored by Alexion Pharmaceuticals. A consensus statement produced by the meeting will be published to increase recognition and diagnosis of the condition by pathologists and nephrologists.

- **Establish a registry of cases.** A registry is needed to collect renal biopsies, clinical phenotypes, serum for complement analyses, and DNA from patients. The registry should focus on the correlation of pathologically defined features with clinical course and biomarkers. Funding and other resources from government and pharmaceutical sources

should be identified and leveraged. Standardization of complement assays, including alternative pathway activation and C3 nephritic factor (C3 NeF), and reagent and sample sharing between investigators will help progress research forward.

- **Develop specific biomarkers.** Biomarker analysis is another important research component identified by Breakout Group 5. In addition to generic glomerular disease markers biomarkers are needed specific to C3 glomerulopathy (e.g., C3 NeF, serum membrane attack complex [MAC], glomerular MAC, and genetic analysis of mutations and polymorphisms). Other possible biomarkers for assessment include urine MAC, urine C5a, and identification of C3 fragments in a biopsy.

- **Conduct clinical trials of therapies and preclinical studies.** To improve therapies, it will be important to develop a network of centers to manage clinical trials, design carefully controlled clinical trials, and populate a registry with data to allow the selection of appropriate patients for clinical trials. Selection of patients for trials based on knowledge of pathology, pathogenesis, and likelihood of progression will facilitate effective clinical trials. The parallel development and use of animal models to understand fundamental pathophysiology, generation of humanized mice, and development of non-invasive monitoring procedures (e.g., MRI using particles targeting iC3b) will be useful in the preclinical arena.

Discussion—Breakout Group 5: C3 Glomerular Disease

The breakout group emphasized the timeliness of research on C3 glomerulopathy, a newly designated disease with a spectrum of genotypes and phenotypes that are characterized poorly. Treatment options have expanded recently with the approval of new drugs targeting the complement pathway. Excellent animal models are available, and new ones are in development. The breakout group participants suggested that there is a high likelihood that targeted therapies can be developed rapidly—within the next decade—as was done in the case of HUS. In addition, participants suggested that progress in treating and understanding C3 glomerulopathy might have implications for other immune complex glomerular diseases, such as IgA nephropathy and immune complex-mediated MPGN. C3 glomerulopathy needs better recognition by pathologists and clinicians through education; definition of its range of histological appearances (e.g., C3 glomerular nephritis vs. DDD, presence of low levels of immunoglobulins); correlation of its histology with functional complement studies, genetics, clinical course, and outcome; increased capacity for and timeliness of diagnostic laboratory assessments; and a rationale for clinical trial design. The breakout group discussed several priorities to progress C3 glomerular disease:

The first actionable area for furthering the field is to define C3 glomerulopathy and establish criteria for its diagnosis. To this end, an international C3 glomerulopathy focus meeting, sponsored by Alexion Pharmaceuticals, is planned for August 2012 in Hinxton, U.K. Participants will include pathologists, complement system biologists, and clinicians. To increase recognition and diagnosis of the condition by pathologists and nephrologists, a product of the meeting will be a draft consensus statement for publication that defines the disease and its diagnostic criteria.

The breakout group participants suggested that subsequent meetings in the United States and other countries might be beneficial to increase recognition of the disease.

Identifying cases of C3 glomerulopathy and establishing a registry, which should include cases of recurrent disease in transplant kidneys, was a proposed second step for moving the field forward. The registry will allow correlation of pathologically defined features with clinical courses and biomarkers. Participants discussed issues that will need consideration when establishing the registry, including its size, classification criteria, recruitment, content, standardization, and support. Desired size and classification criteria are linked. The registry will collect clinical details (including long-term follow-up); serum for complement analysis; DNA samples (requiring IRB oversight); and digitized renal biopsy images (i.e., light and electron microscopy, immunofluorescence). Biopsy image standardization was discussed, and centralized digitization was determined as a possible approach. All pathology will be classified by a panel of pathologists. For recruitment, participants suggested a multinational effort that focused on large centers to accrue cases quickly and small centers for inclusiveness, especially of pediatric cases. Participants doubted that archived samples could meet inclusion criteria. A better understanding of the natural history of the disease through the study of registry cases is essential for designing clinical trials.

The breakout session participants recognized that standardization of complement assays, including those for alternative pathway activation and C3 Nef, would help advance the field. In general, measuring alternative pathway activation is more difficult than serum levels. Participants noted that although some assays are reliable, such as testing for serum Factor H, others appear to yield inconsistent results. Reagent and sample sharing were highlighted as strategies to foster standardization. Participants also pointed out that cost is a factor in the clinical setting, especially for genetic testing. They agreed that increased capacity to perform complement assays will provide clinicians with results more quickly.

Beyond existing generic glomerular disease biomarkers, it is important to develop C3 glomerulopathy-specific biomarkers for clinical use. This was suggested as the fourth actionable item in the breakout session. Biomarker development will be feasible due to improved testing capabilities. Biomarker candidates include C3 Nef, serum MAC, glomerular MAC, and genetic analyses for mutations and polymorphisms. Participants stressed that clinicians need information about selecting tests for diagnosis, and genetic testing will be crucial. Other possible biomarkers, suggested in part by results from animal models, are urine MAC, urine C5a, and the characteristics of C3 fragments from biopsies.

Designing clinical trials involves establishing endpoints and selection criteria. Clinical trial design will be informed by results from the C3 glomerulopathy registry. Clinical trials of targeted therapies will require a network of centers. Therapeutic approaches proposed by the participants included replacement of missing complement pathway factors, auto-antibody removal, and inhibition of alternative pathway activation. Some of these approaches have shown promise in animal models.

Continued progress creating animal models and development of non-invasive monitoring are two crucial preclinical tasks. Animal models, particularly humanized ones, will improve scientists'

understanding of the fundamental pathophysiology of C3 glomerulopathy. Participants agreed that development of non-invasive monitoring (e.g., MRI using particles targeting iC3b) was important to allow physicians to monitor patients over time without the need for repeat biopsies.

Report—Breakout Group 6: Cross-cutting Data Standards and Alternative Outcome Biomarkers

Moderators: *Larry Holzman, M.D., Professor, Department of Medicine, Renal Electrolyte and Hypertension Division, University of Pennsylvania, Philadelphia, PA*
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Breakout Group 6 discussed data standards and biomarkers that are needed by all glomerular diseases. Cross-cutting action items include the following:

- **Build registries to capture data from glomerular disease patients.** This was the recommendation with the highest priority. The registry needs defined and harmonized common essential data elements to build on available data. Developing and utilizing a common platform for the registry, with participation from all interested parties, will ease the exchange of information. The registry should contain longitudinal entry of clinical descriptors to understand natural history, molecular elements, and pathologic characteristics crucial to disease identification. Convening a meeting to continue direct dialogue with all interested parties would be useful to help define the minimal essential dataset to achieve goals and build on experience with existing registries. The specific aims of the registry are to facilitate patient education, perform natural history studies, understand treatment responses, and encourage comparative effectiveness research.

- **Define common biological mechanisms of initiation and progression across all glomerular diseases.** This will be accomplished by identifying appropriate biomarkers, creating biomarker panels, and evaluating fibrosis as an endpoint. Notably, all of the proposed methods for defining biological mechanisms would include repeat renal biopsies for comparison, which would be tremendously useful and increase the power of discovery for markers, mechanisms, and treatable events.

- **Develop and validate PRO tools, useful for multiple glomerular diseases.** This is a key priority. Quantifying how patients feel during disease progression or in response to therapy will facilitate the development of additional qualified outcomes. Another recommendation is to investigate factors (rare or common) that predispose individuals to develop glomerular diseases. Investigators should search for commonalities while understanding that disease-specific factors are informative.

- **Investigate common predispositions underlying both vascular and kidney disease.** Breakout Group 6 noted that patients with glomerular disease are susceptible to vascular disease. Investigating common predispositions underlying both vascular and kidney disease

is a useful endeavor. Elements common to both diseases worthy of future research include lipids, lipid metabolism, ApoL1, chronic inflammation, and oxidative stress. Excluding CKD patients from investigations of cardiovascular disease is not useful for CKD.

- **Identify factors that distinguish progressors from non-progressors for defined at-risk populations.** Preventing disease progression is important, and it is critical to understand why certain patients' progress and others do not. Evaluating a broad panel of factors, such as biomarkers for progression, lipid abnormalities, and pharmacogenomics will enrich clinical trials.

- **Encourage collaboration between industry and the FDA to establish clinically meaningful valid endpoints under specific contexts of use.** Producing a guidance document that details a road map for the evaluation and approval of potential markers for use in clinical trials will be a useful outcome of the multidisciplinary discussion.

Discussion

The second biopsy is important for approval of new therapeutics if it is utilized in conjunction with proteinuria or other biomarkers. This should be emphasized in future proposals to the FDA. Proteinuria has not been defined as a surrogate biomarker or real biomarker. Investigators need to have very careful and comprehensive conversations with the FDA to progress toward approval.

Although glomerular disease progression has a defined signature of final events culminating in kidney failure, it is important to retain the specificity of initial events.

Discussion—Breakout Group 6: Cross-cutting Data Standards and Alternative Outcome Biomarkers

Glomerular diseases have common pathways for progression that can serve as biomarkers. These biomarkers include optimization of traditional measures of structure and function, as well as novel biopsy-derived and urine markers. It is an urgent priority to make progress in this area that has the potential to fundamentally alter the management of glomerular diseases and prevent progression, the cause of greater than approximately 10 percent of ESRD costing more than $7 billion to treat each year in the United States.

The group participants decided that from a research perspective it would be useful to define a clinically meaningful pathologic classification for each disease, consisting of common data elements, degree of chronicity, and the presence or absence of normal glomeruli. Some participants noted that although useful information can be gained by morphologic observation, it may be obscuring the realities of the underlying biology. FSGS is an example; researchers know that multiple biologic causes produce common morphological presentations. The level of expertise of the renal pathologist is critical to identify the subtle differences in disease classifications. One participant noted that the glomerular disease advisory group (GDAG) initiated a study in which renal pathologists evaluate biopsies and collect demographic, clinical, diagnostic, and descriptive data to better understand the incidence of glomerular diseases.

Most patients with clinical signs of glomerular disease undergo a renal biopsy; developing kidney biopsy markers would be helpful to progress the field. Repeat biopsies might find additional information (e.g., podocyte number, normal or damaged glomeruli, transcriptomic and proteomic analyses). Clinicians' reluctance to perform repeat biopsies needs to be addressed, and the repeat biopsy needs to be evaluated as a potential endpoint that meets FDA criteria as a marker for clinical trials.

The proteinuria and urine protein:creatinine ratio are relatively sensitive but they are non-specific markers of kidney injury that can potentially be made more specific by proteomic optimization. The breakout group attendees discussed whether the field could do better than proteinuria as a urinary measurement. Additional urinary biomarkers can provide a global picture of glomerular function to predict outcome. Urine mRNA analysis and mass spectrometry analysis of proteins derived from urine can provide semi-quantitative information describing all renal epithelia. Urine markers can provide frequent non-invasive information at a low cost to guide therapeutic decision making.

Identifying signatures for predictors of disease progression is important. Interstitial fibrosis cuts across all glomerular diseases and should be available as an outcome. Better and more quantitative fibrosis measurements would promote its approval as an outcome indicator. Investigators should use established trials and new trials to determine the relationship between progressive loss of kidney function and renal fibrosis.

Glomerular filtration rate (eGFR) estimates have served as major decision-making markers. They inevitably suffer from inaccuracies, particularly in the critical relatively normal range where it is inherently difficult to account for renal reserve. Ethnic and racial variation exists in eGFR measurements and needs to be considered when planning a trial or analyzing outcomes.

Structural markers, such as renal ultrasound, can provide a semi-quantitative estimate of size and echogenicity; this can be a cost-effective low-risk screening tool. Counting glomeruli may be useful; an MRI scan provides non-radiation high resolution images to evaluate kidney volume and potentially could provide an estimate of glomerular (nephron) number and mass per kidney. Additional imaging techniques in the kidney are under development in animal models but need to be translated to humans.

The participants agreed on the importance of developing a route map for the evaluation of potential markers with the FDA and industry. It is important to perform separate studies to discover and validate biomarkers; doing both in one study diminishes the ability to interpret results. It would be useful for the FDA to determine what criteria will be used to evaluate potential biomarkers.

Registries can help solve the problem of patient education, finding rare patients, and trial enrollment. Industry appreciates registries as a method to alert people to interventional trials and recruit subjects. The breakout group discussed the preferred content of a universal registry and the questions it would answer (e.g., biomarker development, validation, and discovery; disease prevalence; response to conventional therapy; safety and efficacy; natural history of disease). At

a minimum, the registry needs to provide contact lists containing patient names, contact information, and diagnoses. One useful option would be an informed consent for patients to consider when they register for the database. Group members discussed whether it would be better to keep specialized registries for each disease or have one large registry, which might be logistically challenging to coordinate.

Biospecimens should be collected rigorously, be uniform, and be available to the research community. Collection of data standards and protocols must be consistent across studies, and data captured and stored in registries need to be standardized. Data on patient care, outcomes, and medical validation need to be harmonized to meet the criteria of the glomerular community.

A long-term observational cohort will help to understand better the glomerular disease pathology. A challenge for a prospective cohort is time and heterogeneity. Additionally, patient cohorts should be developed for testing biomarkers in the setting of clinical trials. It is useful to have representation across multiple countries and have access to pure populations to ensure sufficient power to identify variability.