



The University of Michigan
Medical Center
Ann Arbor, Michigan, USA

Friedhelm Hildebrandt, M.D.

Professor of Pediatrics and of Human Genetics
Investigator, Howard Hughes Medical Institute
Frederick G.L. Huetwell Professor for the
Cure and Prevention of Birth Defects
University of Michigan Health System
Pediatric Nephrology
1150 W. Medical Center Drive, 8220C MSRB3
Ann Arbor, MI 48109-5646, USA
Fax: +1 734-615-1386
Email: fhilde@umich.edu

HUM000044173 2003-0906

January 21, 2012

Gene Identification in Childhood Kidney Disease

Dear Colleagues,

Thank you for your interest in the mutational screening of patients with nephrotic syndrome (NS), congenital anomalies of the kidney or urinary tract (CAKUT), nephronophthisis (NPHP), or patients that have another rare kidney disease.

Nephrotic Syndrome

We are performing mutational analysis in the *NPHS2*-gene (podocin) and *WT-1*-gene. Our aim is to find out whether there is any correlation between the occurrence of mutations in the *NPHS2*-gene and the clinical outcome of these patients (e.g. response to steroids and cytotoxic drugs, relapse after transplantation) (Karle et al. *J Am Soc Nephrol* 13:388, 2002). This genetic analysis is investigational and is performed in the setting of a research laboratory and there are no universal standards for the performance of these studies. The investigators endeavor to attain the highest standards in their analysis, but these analyses should not be considered diagnostic tests, rather investigational genetic tests, not intended to replace other clinical or laboratory evaluations or treatments that would otherwise be considered the standard of care.

CAKUT

Identification of new genes causing CAKUT will offer new insights into the pathomechanisms of urinary tract malformations, as well as kidney development. CAKUT accounts for a significant degree of morbidity seen in children possessing such lesions. Clinically these abnormalities comprise the most common causes of infant and childhood chronic renal insufficiency and ultimately renal transplantation. The purpose of this proposal is to provide critical data needed to elucidate the genetic causes that underlie these various syndromes and provide a potential screening tool for families at high risk. Additionally, insights gained from this study will provide us and the research community with new information involving the abnormal and normal development of the genitourinary tract, which will have a potentially larger patient application in the future.

NPHP

Nephronophthisis (NPHP), an autosomal-recessive cystic kidney disease, is the most frequent genetic cause of chronic renal failure in the first two decades of life (Hildebrandt 2009). It is characterized clinically by a defect in urinary concentrating ability and progression into terminal renal failure in adolescence. Renal histology exhibits renal tubular basement membrane disruption, tubular atrophy with interstitial fibrosis, and cyst formation. NPHP can occur in association with extra-renal defects. These include retinal degeneration, ocular motor apraxia, cerebellar vermis aplasia, congenital hepatic fibrosis, and developmental defects of bone (Hildebrandt & Zhou 2007). By positional cloning of 10 different recessive genes (*NPHP1-10*) as causing NPHP, if mutated, we have contributed to the discovery of a new group of diseases, "ciliopathies," which are caused by mutations in genes that are expressed in primary cilia and centrosomes.

We will use state of the art molecular genetic diagnostics to identify the exact genetic cause of the renal disease that a child is suffering from (or a family member is suffering from, if that patient requests molecular genetic diagnostics for their own kidney disease). To find this cause we may use techniques that look at changes in very many genes at the same time, some of which may be known to cause other diseases. Because we are not experts on other diseases, we will alert participants to the fact that we will not evaluate or report changes in any other genes that are not the direct cause of the child's (or family member's) kidney disease. This means that if the participant is interested in gene identification or risk

identification in any other disease, they will have to request an independent molecular genetic test for those.

These genetic tests are presently considered investigational and are part of a research protocol. There is no cost for the blood draw, shipping or processing of the samples to the patients or family members of the patients who agree to participate in the study. Office visits for physicians or genetic counselors are not paid for by this study, nor are any other laboratory tests. Results of genetic analyses are generally available 3-6 months following the receipt of a sample. Results are transmitted directly to the corresponding physician and not to individual participants. Participants will therefore need to depend upon their local physician to communicate and explain the results of the genetic tests. The investigators would be happy to discuss the results of the genetic testing with any local physician who wishes to do so. **No results will be reported for individual participants who do not have a diagnosis of NS, CAKUT or NPHP at the time of enrollment.**

If an individual is found to have NS, CAKUT or NPHP after enrolling in the study a local physician may contact the investigators, at which time results of any genetic testing which has been performed can then be released to the local physician. Local physicians, or their representatives, are expected to review the consent document with prospective participants and indicate that they feel the participant understands the nature of the study by signing the consent document before the participant signs the consent document. In addition to the copy that is returned to the investigators, the participant and the local physician should also keep a signed copy of the consent.

We also kindly ask you to fill out a clinical questionnaire which includes not only important information on the family history, the clinical picture, the response to treatment, and extrarenal associations, but also on the ethnicity of your patient. Recent studies and our own data suggest that ethnic groups are affected differently by mutations in genes causing nephrotic syndrome, such as podocin and nephrin. Our group is interested in elucidating genotype/phenotype correlations in this disease. We, therefore, want to encourage you to describe your patient's ethnicity in as detailed a way as possible. Please feel free to check more than one box and/or use the "other" checkbox with a more detailed description.

Please return the following items to the investigators:

1. Signed consent document.
2. Health questionnaire.
3. Blood sample: 3-10ml EDTA or Na-Heparin blood for each participant.
4. Outside the U.S.: Customs Invoice (see end of document)

Blood samples without a signed consent document cannot be processed or analyzed.

As in the past, we are happy to provide free shipping of your blood samples. Therefore, we would like to kindly ask you to contact our laboratory for information on free shipping. Virginia Vega-Warner will be happy to help you and can be contacted by e-mail at vvegaw@umich.edu. DNA samples can be shipped by regular mail.

Please e-mail us at the time of shipping with the shipping number, so that we can track the package and ensure safe delivery. Thank you again for your participation. Please do not hesitate to contact us with any questions or concerns.

Best regards,



Friedhelm Hildebrandt, M.D.

Professor of Pediatrics and of Human Genetics
Investigator, Howard Hughes Medical Institute
Frederick G. L. Huetwell Professor for Cure and Prevention of Birth Defects

Patient's Name: _____

II. Treatment

1. Corticosteroids Yes No

Steroid sensitive

Complete response

Relapse Yes No

Partial response

Relapse Yes No

Steroid resistant

2. Cytotoxic drugs Yes No

Cyclosporine Yes No

Name of drug: _____

Clinical response: _____

Clinical response: _____

3. Dialysis / Renal Transplantation MM / YYYY

Date of end stage renal failure: ____ / ____

1st transplantation: ____ / ____

2nd transplantation: ____ / ____

Unsuccessful transplantation because of:

Recurrence

Graft loss because of:

Recurrence

Rejection

Date of transplant failure: ____MM/____YYYY

III. Extrarenal Association

The patient suffers / suffered from one of the following diseases:

Deafness

Short stature

Urinary/genital tract anomalies

Blindness

Facial dysmorphism

Heart anomalies

Microcephaly

Hexadactylia

Allergies

Mental retardation

Spondyloepiphyseal dysplasia
(Schminke-disease)

Other: _____

IV. Remarks

Thank you very much for your assistance.

Please provide us with the following information in order to facilitate further correspondence.

Name: _____

Phone: _____

Address: _____

Fax: _____

Address: _____

eMail: _____

BLOOD SAMPLE COLLECTION FOR MUTATIONAL ANALYSIS

1. Email: Before shipping samples please send an email to the below mentioned email address to alert us to your incoming shipment; or email at the time of shipping the samples, please include the tracking number, so we can be certain to receive them within 2 days or otherwise track them.

2. Venipuncture: Draw 1-3ml (neonate), 10ml (child), 20ml (adult) EDTA-blood or Na-Heparin under sterile conditions (wear gloves, do not touch rim of tubes); immediately invert tubes several times to prevent coagulation. If syringes and tubes are being used rinse syringe with Na-Heparin.

3. Storage: Always keep blood samples at room temperature! (Never chill, never freeze!)

4. Transport: Protect samples from the cold by wrapping them in gauze or packaging them in Styrofoam. Do not forget to contact us! Send samples and filled-out forms (informed consent and clinical questionnaire) inside the shipping package. Place customs forms outside the shipping package. Address the package to the person listed below, and ship by the fastest route possible (2-day Express Air Mail, Federal Express, DHL Worldwide Express, or UPS). Get a guarantee and tracking number from the carrier to deliver samples to our destination within 1-2 days (regular air mail is much too slow for blood samples).

If you like, you can use our personal courier account. For information on the account number please contact Virginia Vega-Warner at v_vegaw@umich.edu or Professor Friedhelm Hildebrandt at fhilde@umich.edu.

Thank you for your cooperation!

Send samples to:

Prof. Dr. med. F. Hildebrandt
University of Michigan, Department of Pediatrics
1150 W Medical Center Dr, 8220C MSRB3
Ann Arbor, Michigan 48109-5646, USA
Fax: 734-615-1386
eMail: fhilde@umich.edu

Friedhelm Hildebrandt, M.D.
Professor of Pediatrics and of Human Genetics
Investigator, Howard Hughes Medical Institute
Frederick G.L. Huetwell Professor for the
Cure and Prevention of Birth Defects

*University of Michigan Health System
Pediatric Nephrology
8220C MSRB III, Box 5646
1150 W. Medical Center Drive
Ann Arbor, MI 48109-5646, USA*

*Fax: +1 734-615-1386
Email: fhilde@umich.edu*

Invoice

Shipper:

Consignee:

Prof. Dr. med. F. Hildebrandt
University of Michigan, Department of Pediatrics
1150 West Medical Center Dr, 8220C MSRB 3
Ann Arbor, Michigan 48109-5646, USA

Content:

1 Parcel containing:
Documents and human blood or DNA, **non-hazardous, non-toxic, non-infectious**, sample for laboratory research use only, no commercial value.

\$ 1 value for customs purposes only.

Date / Signature