Special Note: Genetic Testing Registry (GTR) Public Forum
9 a.m. to 12 p.m., Nov. 2
Walter E. Washington Convention Center, Room 147
801 Mount Vernon Place, N.W., Washington, D.C.

Online Pre-registration at http://oba.od.nih.gov/gtr/gtr_meetings.html.
Free on-site registration is also available.
For more information on the Genetics Testing Registry,
visit http://www.ncbi.nlm.nih.gov/gtr/.

The GTR project will be overseen by the NIH Office of the Director. The National Center for Biotechnology Information, part of the National Library of Medicine at NIH, will be responsible for developing the registry, which is expected to be available in 2011. The GTR public forum is one way that NIH is engaging stakeholders — such as genetic test developers, test kit manufacturers, health care providers, patients, and researchers — for their insights on the best way to collect and display test information.

NHGRI at the ASHG 60th Annual Meeting
November 2-6, 2010

Day 2: Wednesday, November 3, 2010

INVITED PRESENTATIONS

- **SESSION TITLE: 2. COMPLEX DISEASE GENETICS RESEARCH IN ADMIXED POPULATIONS**
  Invited Scientific
  Ballroom A, Level 3, Convention Center
  Wed Nov 3, 2010 08:00AM -10:00AM

Session Descriptions:
Admixed populations are of special interest in human genetics because of the potential to exploit admixture linkage disequilibrium to map disease genes, especially with regard to diseases with different frequencies across populations. Furthermore, other types of genetic studies, including linkage, GWAS and candidate gene studies are often conducted in admixed populations. This session will describe methods used to estimate ancestry proportions, infer local ancestry of chromosomal segments, and conduct admixture mapping of complex diseases and traits. Successes and failures of admixture mapping and implications of admixture for GWAS, including replication, fine-mapping, and imputation will be evaluated. Examples such as kidney diseases, diabetes, dyslipidemia, asthma, and prostate cancer will be considered. Opportunities presented by admixed populations in the mapping of complex diseases will be discussed. As the field enters the post-GWAS era, it is essential to identify situations where the uniqueness of admixed populations can be exploited to identify susceptibility genes.

Presentation Information

- 08:00AM-08:05AM
  Introduction. *C. N. Rotimi* NHGRI/NIH.

- 09:25AM-09:45AM
  GWAS in African Americans: Findings, replication, fine-mapping, and implications of admixture for imputation. *D. Shriner* NHGRI/NIH.

- 09:45AM-10:00AM
  Questions and answers. *C. N. Rotimi* NHGRI/NIH.

- **SESSION TITLE: 9. IDENTIFIABILITY IN THE ERA OF GENOME-SCALE RESEARCH**
  Social Issues
  Room 147, Level 1, Convention Center
  Wed Nov 3, 2010 08:00AM -10:00AM

Session Descriptions:
Maintaining the privacy of individually identifiable health information is an important consideration in the protection of human research participants. Historically, researchers have coded biological specimens and study data to allow for analysis while protecting the privacy of participants. The de-identification of study data likewise has been proposed as a privacy safeguard in the development of large-scale repositories for data-sharing. However, genomic research raises new questions because genomic data in itself allows for individual identification, if an identifiable sample is available for matching. Survey data suggest that researchers and Institutional Review Board (IRB) officials have mixed views about the risks of re-identification and related harms associated with the collection and sharing of genomic data, and that appropriate procedures for participant protection often require lengthy discussion between IRBs and researchers. This session will explore the implications of identifiability in genomic research from diverse perspectives.

Presentation Information

- 08:20AM-08:40AM
  NIH policy developments. *L. L. Rodriguez* NHGRI/NIH.

PLATFORM PRESENTATIONS

- **SESSION TITLE: 11. INTERVENTION AND TREATMENT OF GENETIC DISORDERS**
  Program Number 16
  Ballroom C, Level 3, Convention Center
  Wed Nov 3, 2010 10:30AM-01:00PM
  Presentation Time: 11:45AM - 2:00NOON

Abstract Content
N-Acetyl Cysteine (NAC) Reverses Early- Stage Hepatic Phenotype of an Antisense Oligonucleotide Mouse Model of Niemann Pick Disease, Type C. *R. Fu[1,2], A. Incao[3], C. A. Wassif[1], W. J. Pavan[3], F. D. Porter[1]* 1) Program in Developmental Endocrinology and Genetics, NICHD, NIH, DHHS, Bethesda, MD, USA 20892; 2) Health Science Center, Peking University, Beijing, China 100191; 3) Genetic Disease Research Branch, NHGRI, NIH, DHHS, Bethesda, MD, USA 20892.

Niemann-Pick Disease, Type C (NPC) is an autosomal recessive disease characterized by excessive cholesterol and glycosphingolipids storage, and progressive neurological deterioration. Deficiency of either NPC1 or NPC2 leads to failure to efflux unesterified cholesterol and lipids from the late endosome/lysosome compartment. This primary defect in intracellular lipid transport initiates a pathological cascade that includes a deficiency of 27-hydroxycholesterol and neuroactive steroid, perturbed sphingosine metabolism, neuroinflammation, induction of apoptosis, and oxidative stress. Increased oxidative stress in NPC is supported by a number of *in vitro*, *in vivo*, and clinical studies. Based on these data, we hypothesized that N-acetylcysteine (NAC), a prodrug for glutathione supplementation, would reduce oxidative stress and thus potentially provide therapeutic benefit in NPC. To test this hypothesis we used a *NPC1* antisense oligonucleotide (AS oilgo) mouse model. This mouse model replicates many aspects of NPC liver disease and has the potential to be used for rapid *in vivo* testing of candidate drugs. Mice were treated with 1% NAC added to the drinking water. In comparison to untreated mice, we observed a significantly ($p < 0.0001$) decreased liver to body weight ratio in NAC treated mice, and serum transaminase levels (ALT and AST) were reduced to control values. Consistent with the bioavailable cholesterol deficiency found in NPC, many SREBP2 target genes (SREBP2, HMGCS1, NSDHL, SQLE) were significantly elevated (more than 1.5 fold > control, $p < 0.05$) in *NPC1* AS oligo mice compared to controls. NAC treatment reduced expression of these cholesterol homeostatic genes. Although total cholesterol levels were unchanged, initial results suggested that the fraction of unesterified cholesterol was significantly decreased ($p < 0.05$). This series of experiments demonstrates the utility of the *NPC1* AS oligo mouse model for rapid screening of candidate drugs, and suggests that NAC treatment addresses one aspect of the NPC pathological cascade, and thus is a candidate drug for use in combinatorial therapy in the treatment of NPC.

- **SESSION TITLE: 11. INTERVENTION AND TREATMENT OF GENETIC DISORDERS**
  Program Number 17
  Ballroom C, Level 3, Convention Center
  Wed Nov 3, 2010 10:30AM-01:00PM
  Presentation Time: 12:00NOON-12:15PM

Abstract Content

Successful use of lipoplex for delivery of the small molecule ManNAc and the *GNE* gene to rescue a mouse model of Hereditary Inclusion Body Myopathy (HIBM). *T. Yardeni[1,4], C. Ciccone[1], S. Hoogstraten-Miller[2], D. Darvish[3], Y. Anikster[4], J. Nemunaitis[5,6], P. Maples[5], C. M. Jay[5], W. A. Gahl[1], M. Huizing[1]* 1) Med Gen Branch, NHGRI/ NIH, Bethesda, MD; 2) OLAM, NHGRI, NIH, Bethesda, MD; 3) HIBM Research group, Encino, CA; 4) Tel - Aviv University, Sackler Faculty of Medicine, ISRAEL; 5) Gradalis Inc., Dallas, TX; 6) Mary Crowley Cancer Research Centers, Dallas, TX.

HIBM is an adult-onset, progressive neuromuscular disorder, caused by *GNE* mutations. *GNE* encodes the ubiquitously expressed, key enzyme in sialic acid (SA) synthesis, UDP-GlcNAc 2-epimerase/ManNAc kinase. We created an HIBM mouse model, mimicking the Persian-Jewish founder mutation M712T. Mutant mice (-/-) unexpectedly died before day 3 of life (P3) from severe glomerulopathy due to hyposialylation, which could be partially rescued by oral supplementation of the SA precursor N-Acetyl-D-mannosamine (ManNAc). We assessed the efficiency and efficacy of ManNAc delivery in liposomes (ManNAc-Lipoplex), and human *GNE* gene delivery in liposomes (*hGNE*-Lipoplex) in our HIBM mouse model. Newborn pups (P1) were retro-orbitally injected with ManNAc-Lipoplex or *hGNE*-Lipoplex. Mice were watched for clinical signs and survival beyond P3, and tissues were tested at P5 for sialylation, histology, glomerular disease and *hGNE* expression. *hGNE*-Lipoplex injections yielded no surviving -/- pups beyond P3, however, wild type survivors showed *hGNE* expression in tested tissues at P5, indicating no toxicity of *hGNE*-Lipoplex and efficient gene delivery to tissues. Interestingly, -/- pups that died before P3 also showed *hGNE* expression in their tissues (as early as P2); treatment at P1 may not allow enough time for sufficient protein translation and SA production. In contrast, ManNAc-Lipoplex injections at P1 yielded survival beyond P3 in >90% of -/- pups. These pups showed improved sialylation of glomerular sialoproteins at P5. ManNAc-Lipoplex treated -/- mice continued to live beyond weaning; the oldest mice are now 4 months. The development of a muscular phenotype in these mice, similar to the symptoms of HIBM, and the effects of ManNAc treatment on these symptoms can now be assessed. Our studies demonstrate: 1) retro-orbital injection in newborn mice is an efficient method for systemic delivery of compounds; 2) small molecules can be efficiently delivered in Lipoplex; 3) systemic delivery of a gene in Lipoplex yields gene expression in tissues after one day; 4) ManNAc-Lipoplex can be applied to increase sialic acid levels and systemic or intramuscular ManNAc-Lipoplex therapy should be considered for the treatment of patients with HIBM, and may also be considered for other disorders of hyposialylation (i.e., certain cancers, certain renal disorders).

- **SESSION TITLE: 17. GENE IDENTIFICATION IN NEUROPSYCHIATRIC DISEASE**
  Program Number 80
  Room 147, Level 1, Convention Center
  Wed Nov 3, 2010 10:30AM-01:00PM
  Presentation Time: 12:45PM-01:00PM

Abstract Content

Exome sequencing identifies sequence variants in two genes at the SPG43 locus. *G. Landoure[1,2,3], J. O. Johnson[4], D. Hernandez[4], K. B. Meilleur[5], A. Britton[4], M. Sangare[1,3], C. Rinaldi[1], M. Traoré[3], B. Traynor[4], K. H. Fischebeck[1]* 1) Neurogenetics Branch, NINDS/NIH, Bethesda, MD; 2) Department of Medicine, University College London, London, UK; 3) Service de Neurologie, Hôpital du Point G, Bamako, Mali; 4) Laboratory of Neurogenetics, NIA/NIH, Bethesda, MD; 5) Center for Research on Genomics and Global Health, NHGRI/NIH, Bethesda, MD.

Hereditary spastic paraplegias are inherited neurological disorders characterized by progressive spasticity and weakness, sometimes associated with muscle atrophy. We previously reported a family from Mali (West Africa) with two sisters affected by spastic paraplegia with weakness of the lower limbs and marked atrophy of the distal upper extremity muscles. There was no known consanguinity, but the proportion of identity by descent by SNP analysis was higher than expected, consistent with parental inbreeding. We performed homozygosity mapping and identified a region of extended homozygosity at chromosome 19p13.11-q12 shared by the affected sisters and not by the other unaffected family members or unrelated controls (Meilleur et al, 2010). The region of interest spans 17.7 Mb, and contains about 150 annotated genes. Sequencing of candidate genes in the region, including RAB3A, GDF1, FKBP8, and εCOP, was negative. We then performed genome-wide exome sequencing in one affected individual, and found 3 homozygous single nucleotide variants in 2 genes within the previously identified locus. Two of the sequence variants are located in the same codon (first and second nucleotides). Sequencing of the rest of the family members showed that these changes segregated with the disease in the family. All the sequence variants affect residues that are conserved across species from human to fruit fly, and they are predicted to be deleterious. In addition these sequence variants were not found in 50 ethnically matched controls. Sequencing of more controls is under way to exclude possible polymorphism. Our study highlights the efficiency of exome sequencing by identifying sequence variants in 2 genes at the SPG43 locus, of which one or more may be disease-causing in the family studied here. Additional investigations including functional studies of the candidate gene products are now being done to support our findings.

POSTERS

- **POSTER SESSION: CANCER CYTOGENETICS**
  Program Number 431
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Abstract Content

Extensive whole-chromosome aberrations detected by SNP array in a neurofibromatosis type 1-associated glomus tumor. *D. Stewart[1], A. Pemov[1], E. Beert[2], H. Brems[2], E. Legius[2]* 1) Genetic Disease Research Branch, NHGRI/NIH, Bethesda, MD; 2) Department of Human Genetics, Catholic University Leuven, Leuven, Belgium.

Introduction. Glomus tumors are painful benign tumors of the glomus body, a thermoregulatory shunt located in the fingertips. We recently reported that NF1-associated glomus tumors arise from bi-allelic inactivation of the gene *NF1*. We performed analysis of the genomic architecture in two sporadic and four NF1-associated glomus neoplasms using high-resolution Illumina SNP arrays. **Methods**. DNA was collected from primary cell cultures (except for one case, when DNA was also extracted directly from tumor tissue) that were established from dissected glomus tumors. Comprehensive *NF1* sequencing was performed on DNA from NF1-associated tumors. DNA samples were processed and hybridized to the Illumina HumanOmni1-Quad microarrays. White blood cell DNA was used as germline control in NF1-associated glomus tumors (germline DNA for the patients with sporadic glomus tumors was unavailable). Analyses of copy number variants (CNV) and loss of heterozygosity (LOH) regions was performed using GenomeStudio (Illumina). Every putative CNV or LOH region from NF1-associated tumors was compared to its matching germline DNA sample. For the sporadic tumors, where control germline DNA was unavailable, normal copy number for each region was assumed to be 2 and deviation from that was considered as copy number loss or gain. **Results**. We hybridized DNA from the genomes of four NF1-associated and two sporadic glomus tumors. We found germline mutations of *NF1* in all 4 tumors and a somatic mutation in *NF1* in three of the four NF1-associated tumors. In the fourth tumor there were multiple large-scale chromosomal aberrations, including evidence of likely mitotic recombination of chromosome 17q. Across the entirety of chromosomes 2, 3, 4, 5, 6, 8, 9, 13, 18, 19 and 21, we observed B allele frequencies of 0%, ~33%, ~67% and 100% in conjunction with a log R ratio of ~0. We also found evidence of a large number of smaller CNVs shared by multiple tumors. **Conclusions**. This is the first report of bi-allelic inactivation of *NF1* arising from mitotic recombination of chromosome 17q in an NF1-associated glomus tumor. The multiple large-scale chromosomal aberrations in 1/4 NF1-associated glomus tumors may be due to chromosomal trisomy or, less likely, mosaic whole-chromosome uniparental disomy (UPD). Somatic aneuploidy (and/or UPD) may be more common than expected in NF1-associated glomus tumors.

- **POSTER SESSION: . CARDIOVASCULAR GENETICS**
  Program Number 711
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Abstract Content

Population diversity and GWAS-based genetic variants for BMI: The PAGE Study. *K. E. North[1, 12], M. Fesinmeyer[2], M. D. Ritchie[3], N. Francescini[1], P. Buzkova[4], U. Lim[5], M. Quibrera[1], M. Gross[6], K. Glenn[3], S. Buyske[7], C. Kooperberg[2], C. S. Carlson[2], R. Li[8], C. A. Haiman[9], L. R. Wilkens[5], L. L. Marchand[5], R. L. Prentice[2], L. H. Kuller[10], L. Hindorff[8], J. Manson[11], C. Chen[2], U. Peters[2]* 1) Dept Epidemiology, Univ North Carolina, Chapel Hill, NC; 2) Fred Hutchinson Cancer Research Center, Seattle, WA; 3) Center for Human Genetics Research, Vanderbilt University, Nashville, TN; 4) Department of Biostatistics, University of Washington, Seattle, WA; 5) Cancer Research Center of Hawaii, Honolulu, HI; 6) Department of Laboratory Medicine and Pathology, University of Minnesota, Minneapolis, MN; 7) Department of Statistics & Biostatistics, Rutgers University, Piscataway, NJ; 8) Office of Population Genomics, National Human Genome Research Institute, National Institutes of Health, Bethesda, MD; 9) Department of Preventive Medicine, Keck School of Medicine, University of Southern California, Los Angeles, CA; 10) Department of Epidemiology, University of Pittsburgh, Pittsburgh, PA; 11) Department of Medicine, Harvard Medical School, Boston, MA; 12) Carolina Center for Genome Sciences, Univ North Carolina, Chapel Hill, NC.

The successes of genome-wide association studies (GWAS) in mapping loci that influence complex traits advance our understanding of genomic influences on common diseases. Translation of such knowledge into clinical and public health applications requires exploration of these associations in different ancestral groups. The NHGRI-supported 'Population Architecture using Genomics and Epidemiology (PAGE) ' consortium of population-based studies accomplishes this by investigating the epidemiologic architecture of well-replicated genetic variants associated with complex traits. PAGE is comprised of CALiCo (Causal Variants Across the Life Course, a consortium of ARIC, CARDIA, CHS, and SHFS), EAGLE (Epidemiologic Architecture for Genes Linked to Environment, with participants from National Health and

Nutrition Examination Surveys), Multiethnic Cohort, and Women 's Health Initiative studies. We selected twenty putative body mass index (BMI) related SNPs identified by prior GWAS in European and European American populations, and genotyped each SNP in a diverse sample of up to ~70,000 subjects representing European American (N=39,421), African American (N=14,573), Mexican American (N=7,317), Asian and Pacific Islanders (N=3,236), and American Indian (N=7,317) individuals. We compared allele frequencies between ancestry groups and examined each SNP for departure from Hardy-Weinberg equilibrium within each subpopulation. The association between each SNP and BMI was analyzed stratified by PAGE site and self-identified race using additive genetic models and linear regression (mixed models for family relatedness), and adjusting for age and current smoking status specific for each sex strata. Results were combined across study but within ancestral groups using fixed effects meta-analysis techniques. Interestingly, some well established genetic variants did not replicate in any population we considered (for example, rs2815752 in NEGR1). However, several SNPs did replicate in most of the populations, for example rs7498665 in SH2B1 demonstrated a significant effect ($P < 0.05$) in all but the Mexican American subpopulation. Given known differences in allele frequencies and linkage disequilibrium patterns across ancestral groups, our findings suggest a complex architecture of BMI and the need for considering genetic, cultural, and environmental population differences in interpreting the increasing body of genetic associations emerging from well-replicated GWAS.

- **POSTER SESSION: COMPLEX TRAITS: THEORY AND METHODS**
  Program Number 1017
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Abstract Content

The evaluation of a loss-of-function GBA variant found in patients with Parkinson disease. *N. Tayebi, Y. Blech-Hermoni, JH. Choi, W. Westbroek, BK. Stubblefield, E. Sidransky* Section on Molecular Neurogenetics, Medical Genetics Branch/NHGRI, NIH, Bethesda, MD.

Recent studies demonstrate an increased frequency of mutations in the gene encoding glucocerebrosidase (GBA), the enzyme deficient in Gaucher disease, among patients with Parkinson disease. One particular GBA mutation, c.84dupG, a founder mutation in the Ashkenazi Jewish population, was found with a frequency of 2.1% among Ashkenazi patients with Parkinson disease as compared to 0.27% in controls. In this mutant allele, the insertion of a G introduces a frameshift in the signal peptide sequence of glucocerebrosidase, causing a complete deficiency of the enzyme and generating a STOP codon ,TAA, 25 amino acids downstream from the insertion. The mutation has never been encountered in the homozygosus state and is presumed to be lethal. We evaluated the consequences and potential toxicity resulting from this alteration. RNA was extracted from fibroblasts from patients heterozygous for c.84dupG and controls. An RNA protection assay, RT-PCR using different primer sets, and subcloning followed by sequencing of each allele were performed, excluding an alternative exonic splice site. In vitro translation assays indicated no translation of the predicted 25 amino acid peptide. The small peptide was synthesized and an antibody was generated. Using the conjugated peptide as a positive control, the small peptide as not detected in Western blots of fibroblasts from patients with the c.84dupG allele. Moreover, the peptide was not seen in these fibroblasts on confocal microscopy. No morphological changes were observed in Cos-7 and CHO cell lines transfected with a pcDNA3.1 GBA construct expressing c.84dupG, excluding a toxic effect of the peptide. These studies indicate that c.84dupG is a loss-of- function mutation, implicating the deficiency of enzymatic activity in Parkinsonian patients carrying this mutation.

- **POSTER SESSION: . MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2139
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Abstract Content

Animal modeling of TARP syndrome: Knocking down and knocking out *Rbm10*. *J. J. Johnston[1], K. S. Bishop[3], E. J. Spaulding[1], J. K. Teer[1, 2], P. F. Cherukuri[1, 2], N. F. Hansen[2], S. K. Loftus[1], K. Chong[4], J. C. Mullikin[2], L. G. Biesecker[1, 2]* 1) Genetic Disease Research Branch, NHGRI, Bethesda, MD; 2) NIH Intramural Sequencing Center, Bethesda, MD; 3) Zebrafish Core Facility, Genetics and Molecular Biology Branch, NHGRI, Bethesda, MD; 4) Prenatal Diagnosis and Medical Genetics Program, Mount Sinai Hospital, Toronto, ON, Canada.

Micrognathia, glossoptosis, and cleft palate comprise a common malformation, Robin sequence. It is a component of the TARP syndrome, which comprises Talipes equinovarus, Atrial septal defect, Robin sequence, and Persistent left superior vena cava. This disorder is X-linked with apparently 100% pre- or postnatal lethality in affected males. We have previously characterized two families with TARP, performed massively parallel sequencing of X chromosome exons, filtered the results, used a unique algorithm to characterize variants, and showed that TARP is caused by truncating mutations in *RBM10*, which encodes RNA binding motif 10. These are the only known human mutations in this gene and there are no recognized animal models of this disorder. We hypothesize that *RBM10* dysregulates other developmental genes that cause other forms of Robin sequence or clefting. To better define the role of *RBM10* in development, we

proposed to use zebrafish as a model organism. We designed a total of 3 splice morpholinos targeting zebrafish *rbm10*. Fish were collected at 6-8 dpf for alcian blue staining of cartilage to identify abnormal jaw development. The first morpholino yielded a phenotype of mandible underdevelopment, consistent with the human phenotype. The second and third morpholinos did not yield a recognizable phenotype. Only 1 of the 3 morpholinos blocked splicing as shown with RT-PCR analysis from 3 dpf fish. We hypothesized that maternal transcripts may rescue the embryos and worked toward a translation blocking morpholino. As the 5 ' end of the zebrafish transcript was incomplete, we used 5 '-RACE to identify alternative splicing with 2 alternative translation start sites (equally represented in cloned products). Due to the complexity of the 5 ' end of the transcript and the negative result from the splice morpholinos we concluded that the fish is not a useful model for TARP syndrome. Concurrently, we have identified two mouse *Rbm10* knockout ES cell lines (*Rbm10*$^{tm1a(KOMP)Wtsi}$, *Rbm10*$^{GT(CSI176)Byg}$), which we obtained and are working towards creating transgenic lines.

- **POSTER SESSION: . THERAPY FOR GENETIC DISORDERS**
  Program Number 3075
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Abstract Content

Long-term phenotypic correction of a lethal mouse model of methylmalonic acidemia using rAAV9-mediated gene therapy and metabolic improvement after re-boosting at 1 year. *J. S. Sénac, R. J. Chandler, C. P. Venditti*
Genetics and Molecular Biology Branch, NHGRI, NIH, Bethesda, MD.

Methylmalonic acidemia (MMA) is an inherited metabolic disorder most commonly caused by the deficient activity of methylmalonyl-CoA mutase (MUT). The disorder carries a poor prognosis for long-term survival. We have recently demonstrated that gene therapy with an adeno-associated virus serotype 8 (rAAV8) vector can both rescue Mut$^{-/-}$ mice from neonatal lethality and provide long-term phenotypic correction. Over time, hepatic Mut RNA levels and Mut protein significantly decreased in the rAAV8 treated Mut$^{-/-}$ mice, suggesting that translation to human subjects may require multiple rounds of gene delivery throughout life. Our goal was to develop a new gene therapy vector using an alternative AAV serotype. We engineered an rAAV9 vector to express the murine Mut cDNA under the control of an enhanced chicken beta actin promoter (rAAV9-CBA-mMut). rAAV9-CBA-mMut was delivered directly into the liver of newborn Mut$^{-/-}$ mice at a dose of 1x10^{10} GC (N=9). A small group (N=5) of older treated Mut$^{-/-}$ mice were re-injected via the intraorbital route after 1 year with 2x10^{9} GC per gram. Effects on survival, growth, metabolites, in vivo propionate oxidative capacity were used to determine the efficacy of rAAV9 gene therapy. Untreated Mut$^{-/-}$ mice (N=58) uniformly perish, with most dying during the newborn period. However, rAAV9 treated Mut$^{-/-}$ mice survived for more than 1 year. On an unrestricted mouse diet, the treated Mut$^{-/-}$ mice displayed elevated methylmalonic acid levels in the plasma yet were vigorous and have maintained 80% of the weight of age, sex and diet matched controls. Expression of the Mut transgene, as early as 24 hours post injection, appears to correlate with early phenotypic correction. rAAV9 treated mice that received a second dose at one year showed metabolic improvements as early as 72 hours post injection and experienced a full correction of propionate oxidative capacity 2 weeks post injection. These experiments demonstrate that rAAV9 is an efficient gene therapy vector for the treatment of MMA in a mouse model that faithfully replicates a severe form of the disorder, even at lower dose than that used in previous studies with rAAV8. Re-treating older Mut$^{-/-}$ mice with a dose of rAAV9 similar to that used in human clinical trials produced a measurable effect, rapidly. Our data support the use of rAAV9 as a vector with potential human translation in the treatment of MMA.

- **POSTER SESSION: THERAPY FOR GENETIC DISORDERS**
  Program Number 3081
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 5:00PM-6:00PM

Discovery and Evaluation of a Non-Iminosugar Glucocerebrosidase Inhibitory Series with Chaperone Activity. *O. Motabar[1,2], W. Zheng[2], J. Marugan[2], E. Goldin[1], W. Westbroek[1], N. Southall[2], E. Sidransky[1]* 1) Medical Genetics Branch, NHGRI, NIH, Bethesda, MD; 2) NIH Chemical Genomics Center, NHGRI, NIH, Rockville, MD.

Gaucher disease is a lysosomal storage disorder (LSD) caused by a deficiency in the enzyme glucocerebrosidase (GC). Small molecule chaperones have been proposed as a promising therapeutic approach for the treatment of LSDs. Most of the small molecule chaperones described in the literature have iminosugar scaffolds. Here, we present the discovery and evaluation of a new series of GC inhibitors with quinazoline cores. This series was discovered through the quantitative high throughput screening (qHTS) of 326,770 compounds, in which several quinazoline analogues were identified. Structure-activity relationship (SAR) optimization yielded compounds with an IC50 close to 300 nM. These compounds were able to inhibit the hydrolysis of the natural substrate, and were selective for GC when tested against other lysosomal enzymes. Thermal denaturation experiments demonstrated the capacity of this series to stabilize GC. Immunocytochemistry on control and GC deficient fibroblasts with anti-GC and anti-LAMP2 antibodies was performed followed by laser scanning confocal microscopy. Upregulation and colocalization of GC to the lysosomes was seen with some of the compounds from this series. Systematic synthetic modifications enhancing the usefulness of these

compounds are presented as part of the SAR. This new class of GC inhibitors may open new avenues for the development of alternative therapies for patients with Gaucher disease.

- **POSTER SESSION: COMPLEX TRAITS: THEORY AND METHODS**
  Program Number 938
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Familial Idiopathic Scoliosis in Males: Localization to Chromosome. *C. M. Justice[1], K. Swindle[2], S. Cook[2], J. Dunn[2], N. H. Miller[2]* 1) Genometrics Section, IDRB, NHGRI, NIH, Baltimore, MD; 2) University of Colorado, The Children 's Hospital, Denver, Colorado.

Idiopathic scoliosis is present in 2 to 3% of children or adolescents and is defined by a lateral curvature of the spine ≥ 10 ° for which the cause is unknown. For curvatures of 10 degrees, the proportion of affected females to affected males is approximately 1:1, but the proportion of affected females to affected males increases dramatically as the magnitude of the scoliotic curve increases [Roach 1999]. In our initial population of 202 families, there were approximately 7 affected females for every affected male at curvatures ≥ 30 °. Lateral curvatures of 30 degrees are less prevalent in the pediatric population, with a prevalence of approximately 0.2 percent [Miller 1999]. The goal of this study was the identification of genetic determinants in families that include males with severe lateral curvatures. The males with severe curve subset was comprised of 25 families (207 individuals) in which at least one male was diagnosed in adolescence with a ≥30 ° lateral curvature. There were 123 scoliotic individuals (48 male; 75 female), and 85 unaffected individuals (45 male; 40 female) in this subset. A genomic screen was performed with a modified CHLC v.9 marker set. Fine mapping was done with a custom SNP panel and ABI Taqman methodology on an ABI 377 platform. The initial genome-wide screen and subsequent analyses were analyzed by model-independent linkage analysis using SIBPAL (S.A.G.E. v5.1). The genome-wide linkage analysis for the qualitative and quantitative traits resulted in significant p-values (2 adjacent markers with p-values < 0.01) on chromosomes 2, 16 and 22. The most significant p-value was obtained for the qualitative analysis (threshold set at ≥ 10 °) for d22s689 (p-value= 4.2 x10-8), for which an adjacent marker, d22s685 (p-value = 3.8 x x10-4) was also significant. Fine mapping with SNPs confirmed this region as being linked to FIS in our subset of families with adolescent males with curvatures ≥30 °. Significant SNPs lie primarily in the introns of the LARGE gene, integral to the development and maintenance of skeletal muscle, and SFI1, responsible for the integrity of the chromosomal centromere complex. Future goals include association and sequencing analyses of this region. FIS is a complex genetic disorder, and utilizing clinical criteria may aid in decreasing the heterogeneity of our large familial idiopathic study population, and enhance the successful identification of specific genes responsible for this disorder.

- **POSTER SESSION: COMPLEX TRAITS: THEORY AND METHODS**
  Program Number 982
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Fine-mapping linkage and family-based association of idiopathic scoliosis and chromosome 1. *D. Behneman[1,2], C. Justice[1], T. Beaty[2], K. Y. Liang[2], N. H. Miller[3], A. F. Wilson[1]* 1) Genometrics Section, IDRB/NHGRI/NIH, Baltimore, MD; 2) Johns Hopkins University, Bloomberg School of Public Health, Baltimore, MD; 3) University of Colorado Health Science Center at Aurora, Aurora, CO.

Idiopathic scoliosis is a lateral curvature of the spine with unknown etiology. Previous studies have reported linkage to several regions across the genome, as well as linkage and association with several candidate genes. In this study, we tested for association in two regions on chromosome 1 that were previously identified and replicated with genome-wide linkage analysis. Region 1 is 8 Mb on the p arm; Region 2 is 19 Mb on the q arm. Linkage analysis and family-based tests of association were performed in two samples: 95 families most-likely to have autosomal-dominant mode of inheritance (Sample 1) and 187 families selected for parent-child trios (Sample 2). Scoliosis was defined as two qualitative traits (lateral curvature ≥10 ° and ≥30 °) and as a quantitative trait (the degree of lateral curvature). Model-independent sib-pair linkage analysis, as well as allelic, genotypic and haplotypic tests of association, were performed on 110 SNPs in Sample 1 and 18 SNPs in Sample 2 within Region 1, and among 202 SNPs in Sample 1 and 214 SNPs in Sample 2 from Region 2.

For Region 1, 13 SNPs showed evidence of linkage and another 9 SNPs showed evidence of association in Sample 1, and in Sample 2 linkage was found with 2 SNPs and association was found to another 5 SNPs, for at least one scoliosis phenotype. At the 30 ° threshold, linkage and association were observed for rs4512614 and rs4654549 in Sample 1 and rs10915548 in Sample 2.

For Region 2, 16 SNPs showed evidence of linkage and another 13 SNPs showed evidence of association in Sample 1, and in Sample 2 linkage was found with 45 SNPs and association was found to another 20 SNPs, for at least one scoliosis phenotype. In Sample 2, linkage and association were observed for rs11586173, rs7519036, rs12117045 and rs6428417 at the 10 ° threshold. However, in Sample 1 none of the SNPs were significant for both linkage and association.

Candidate genes were identified within 1 Mb of these linked SNPs and within 0.1 Mb of associated SNPs then prioritized by statistical evidence (p-values) and biologic plausibility (gene function). These findings suggest gene(s) in these regions may contribute to scoliosis, an important step in determining genetic etiology for this disorder.

- **POSTER SESSION: GENOMICS**
  Program Number 1708
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Methods for analyzing next-gen whole exome sequence variation in small pedigrees with unknown diseases. *M. Sincan[1,2], TC. Markello[1,2], DA. Adams[1,2], K. Fuentes-Fajardo[2], H. Carlson-Donohoe[1,2], C. Toro[2], C. Tifft[2], PF. Cherukuri[3], JK. Teer[3], P. Cruz[3], N. Hansen[3], JC. Mullikin[3,4], WA. Gahl[1,2]* 1) Medical Genetics Branch, NIH/NHGRI, Bethesda, MD; 2) NIH Undiagnosed Diseases Program, NIH, Bethesda, MD; 3) Genome Technology Branch, NHGRI, NIH Bethesda MD; 4) NIH Intramural Sequencing Center, NIH, Bethesda MD.

Next generation sequencing technologies are becoming more available as a tool to diagnose genetic causes of diseases. We have sequenced 28 individuals from 8 families with undiagnosed diseases. Our families include both affected and unaffected individuals. Each family has three or four individuals with one or two affected individuals. The power of next gen sequencing gives us a very long list of potentially disease causing variations that are shared among various members of the families. For our families, the number of variants is between 79,710 and 107,817. Using conserved domain based analysis to determine the potential deleteriousness of these variants can reduce the number of variants to between 6,051 and 7,613. This is still a very big number to analyze. To this end, we have applied Mendelian filters to check if genotypes of family members are concordant with affected and unaffected status and various Mendelian inheritance models that are not in conflict with the current pedigree. The nature of our problem, i.e. dealing with undiagnosed diseases, forces us to apply different possible inheritance models. After we remove the positions that are not compatible with our filter, we still end up with a list of variants numbering in the hundreds for a single family. We also applied filters based on other families and SNPs that have frequency information. We assume that our cases are most likely rare diseases, if we find the same variants in other families and/or in dbSNP with a reasonable frequency, we flag them as potentially less likely to cause the disease in question. We found that it is a complex task to search for disease causing changes in the human genome, but data available to meta tag the variome is becoming increasingly available and tools we develop let us cross match our patients ' data to this growing body of information. We illustrate the tools we have developed to make these analyses.

- **POSTER SESSION: GENOMICS**
  Program Number 1740
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract

NCBI database for genetic associations with high-throughput molecular phenotypes. *J. Paschall[1], D. Hoffman[1], N. Sharopova[1], W. Gan[2], M. Feolo[1], S. Sherry[1], J. Ostell[1], JP. Struewing[2]* 1) National Center for Biotechnology Information, Bethesda, MD; 2) National Human Genome Research Institute, Bethesda MD.

The success of genome wide association studies (GWAS) has identified hundreds of robust genetic associations across a range of disease phenotypes. In most instances, however, the causative variants and thus the molecular pathways linking a genomic variant to disease susceptibility remain unknown when the associated SNPs do not map to exons. It is critical for post-GWAS studies to identify which of these may influence gene regulation. An important approach to bridging this gap is testing for association between genomic variants and molecular level phenotypes such as gene expression (eQTL), epigenomic, and proteomic data. The NCBI has developed a searchable database focused on archiving and visualizing genetic associations with high throughput molecular phenotype data: (http://www.ncbi.nlm.nih.gov/gtex). This effort originated with the eQTL data from the NIH Common Fund Genotype-Tissue Expression (GTEx) project http://nihroadmap.nih.gov/GTEx/ which will assess the genetic component of gene expression variation across many different human tissues. The scope of this database extends to all existing and future high-throughput molecular association datasets. Current datasets include those derived from both microarray and RNA-seq technology. Association results are searchable by expressed gene, genomic variant, and tissue-type, and are visualized graphically as histograms and as tracks within a genome browser. Links will be computed and automatically

updated between SNPs found to be associated with molecular phenotypes, and SNPs found to have disease phenotype associations as recorded in dbGaP and the NHGRI GWAS catalog (www.genome.gov/gwastudies). The goal is to allow the results of a new disease phenotype study to be quickly queried against all available molecular phenotype association data. This will provide additional biological support in interpreting those results, and allow for background processes to re-scan both disease and molecular phenotype associations over time in a continuous data-mining effort

- **POSTER SESSION: GENOMICS**
  Program Number 1784
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Identification of DNA-associated Proteins by Sequence-Specific Capture and Mass Spectrometry. *H. Guillen Ahlers[1], S. Mirza[1], S. Zhang[1], M. Zickus[1], R. Cole[1], M. Zelembaba[1], M. Chesnik[1], C.-H. Wu[2], S. Chen[2], Y. Yuan[2], G. Kreitinger[2], M. Scalf[2], M. R. Shortreed[2], L. A. Cirillo[1], L. M. Smith[2], M. Olivier[1]* 1) Wisconsin Center of Excellence in Genomics Science, Medical College of Wisconsin, Milwaukee, WI; 2) Wisconsin Center of Excellence in Genomics Science, University of Wisconsin, Madison, WI.

Recent advances in genomics and proteomics have brought us closer to reaching a detailed and comprehensive understanding of our genome and how it is regulated. Numerous proteins mediate DNA stability, control its activity, and regulate transcription of the genetic information. However, currently no technologies exist that allow the dissection of these protein-DNA interactions in a comprehensive global manner, and examine alterations in disease. To overcome this challenge, we report on the development of an entirely novel technology. Unlike ChIP-chip methodology, where DNA sequences that interact with individual known proteins are characterized, the Wisconsin Center for Excellence in Genomics Science (CEGS) utilizes an oligonucleotide capture technology to isolate targets of interest in a sequence-specific manner in order to analyze protein complexes attached to these regions. In an initial study, the mouse insulin-like growth factor-binding protein 1 (IGFBP1) promoter region was used as an in vitro model system. Specific capture oligonucleotides were designed and attached to gold surfaces using linker chemistry and amino-terminated oligonucleotides. Hybridization was optimized to sequester PCR-products containing an exposed single-stranded overhang. After on-chip protease digestion, FoxO1 binding to the DNA sequence was detected by tandem mass spectrometry using an LTQ XL mass spectrometer. Due to its high binding affinity, the reaction was carried out without a cross-linking step between FoxO1 and the DNA. Our analysis demonstrates efficient capture of FoxO1-DNA complexes in a sequence-specific manner. Capture technology and mass spectrometry allowed the detection of 1 pMol of captured FoxO1 protein using a PCR product with a FoxO1 binding site while an alternative PCR product with a mutated binding site did not lead to the detection of FoxO1. The on-chip digestion and sample preparation can be performed on small array surfaces (<5 mm2). Additional efforts are under way to allow the selective capture of restriction fragments directly from genomic DNA, rather than specific PCR products. This work was funded by the Wisconsin Center for Excellence in Genomics Science through NIH/NHGRI grant 1P50HG004952.

- **POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2206
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Initial analysis of whole exome and whole genome sequencing in the NIH Undiagnosed Disease Program. *T. C. Markello[1], D. A. Adams[1], K. Fuentes Fajardo[1], M. Sincan[1], H. Carlson-Donohoe[1], C. J. Tifft[1], T. M. Pierson[1,4], C. Toro[1], J. K. Teer[3], P. F. Cherukuri[3], N. F. Hansen[3], S. S. Ajay[3], H. Ozel Abaan[3], E. H. Margulies[3], P. Cruz[3], J. C. Mullikin[2], W. A. Gahl[1], NISC Comparative Sequencing Program* 1) Undiagnosed Diseases Program, NIH/NHGRI, Bethesda, MD; 2) NIH Intramural Sequencing Center, NIH, Bethesda, MD; 3) Genome Technology Branch, NHGRI, NIH, Bethesda, MD; 4) Neurogenetics Branch, NINDS, NIH, Bethesda, MD.

The NIH Undiagnosed Diseases Program began analyzing single families with whole exome and whole genome sequencing in October 2009. We have completed sequencing 28 whole exomes and 1 whole genome. The current plan is to complete 60 exomes and 7 genomes by September 2010. The first 8 families chosen for sequencing included 3-four member families with two affected siblings and both parents, 3- three member families with a single affected along with both parents, and one four member family with one affected and one unaffected sibling with both parents. The final family had 4 individuals, with an autosomal dominant parent -child inheritance. One family had both parents and one affected child analyzed by whole exome, and the other affected child by whole genome sequencing. One family involved a consanguineous first cousin mating. 4 families had skeletal phenotypes and 7 of the 8 had significant neuro-developmental phenotypes. Exome capture and sequencing produced a total of 674Mb high quality sequence and an average coverage of 78.4% of the UCSC " known genes" sequence. The single whole genome data was truncated to the rest of the family's exome data. To analyze the sequences, high-density SNP array data previously obtained on

these same individuals were used to construct linkage regions for recessive, dominant, and conjoint new mutations; this excluded up to 80% of the genome for recessive cases with two affected children. dbSNP130 data were used as an optional filter. For each variation from the reference sequence every family trio or quartet was examined for loci that followed homozygous, compound heterozygous recessive, or autosomal dominant (germ line mosaic) new mutation inheritance patterns. The final selection criterion used conservation information by the CDPred algorithm to prioritize candidates by the potential deleteriousness of each variation. For the single case of consanguinity, there were 113 variants that met criteria, 27 that were not in dbSNP130, and only 2 that had significant CDPred scores. One variation was identified and verified to be a homozygous mutation in AFG3L2, the first recessive diagnosis involving this gene. For the other families we have between 68 and 201 viable candidates that pass all exclusion constraints. These candidates are being evaluated using additional bioinformatics and laboratory assays. Analysis of these data and the remaining whole exome and whole genome evaluations are ongoing.

- **POSTER SESSION: PSYCHIATRIC GENETICS, NEUROGENETICS AND NEURODEGENERATION**
  Program Number 2582
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Do glucocerebrosidase mutations affect the neurobiology of PD? : Dopamine synthesis and regional brain activity in *GBA*-associated parkinsonism. *O. Goker-Alpan[1, 3], J. Masdeu[2], A. Ianni[2], G. Lopez[1], D. Eisenberg[2], P. Kohn[2], C. Groden[1], M. Chalfin[2], K. F. Berman[2], E. Sidransky[1]* 1) Section on Molecular Neurogenetics, I MGB/NHGRI, NIH, Bethesda, MD; 2) Section on Integrative Neuroimaging, Clinical Brain Disorders Branch, NIMH, NIH, Bethesda, MD; 3) Center for Clinical Trials, Springfield VA.

Mutations in the gene encoding glucocerebrosidase (GBA), the enzyme deficient in Gaucher disease (GD), are a common genetic risk factor for parkinsonism. Subjects with Parkinson disease (PD) are five times more likely to carry *GBA* mutations. Although the clinical phenotype of patients carrying *GBA* mutations may resemble sporadic PD, the age-of-onset of parkinsonian manifestations is earlier, and cognitive impairment is more prevalent. We investigated whether *GBA* mutations alter the neurobiology of PD, studying *in vivo* brain dopamine synthesis as a marker of neurodegeneration, and resting regional Cerebral Blood Flow (rCBF) as an index of brain activity in 103 subjects (38F/65M). Positron emission tomography (PET) was performed using ^{18}F-Fluorodopa (FDOPA) and $H_2{}^{15}O$ to evaluate regional brain dopamine synthesis and resting rCBF respectively. Eight subjects had sporadic PD [mean age 60 ±6 years]; 8 had GD and parkinsonism [54 ±10 years]; 10 had GD but no parkinsonism [50 ±13 years]; and 7 were Gaucher carriers, but did not exhibit parkinsonism [50 ±18 years]. Subjects in the last two groups had a family history of PD. All PD subjects had similar UPDRS scores and disease duration. As the age-of-onset was earlier among *GBA* mutation carriers, each study group was compared to its own age and sex matched healthy control group, made up by twice the number of study subjects. Data were assessed with both region of interest and voxel-based methods. Striatal dopamine synthesis was markedly decreased in PD subjects regardless of *GBA* mutation status, with a greater loss in the caudal striatum (51% loss in putamen Ki), and relative sparing of the caudate (29% loss). Striatal dopamine was decreased ($p < 0.05$) in the putamen of somesubjects with GD and a family history of parkinsonism, without overt signs of parkinsonism. rCBF was decreased only in the patients with GD and parkinsonism, in a pattern resembling that in diffuse Lewy body disease. This, the first *in vivo* study evaluating the pattern of neurodegeneration in both *GBA* hetero- and homozygotes, demonstrates that although the pattern of dopamine loss in *GBA*-associated parkinsonism is similar to sporadic PD, the resting brain activity is decreased in areas affected in diffuse Lewy body disease, potentially explaining the increased cognitive impairment. The absence of a gene dosage effect may support the gain-of function-hypothesis as a mechanism for the development *GBA*-associated parkinsonian pathology.

- **POSTER SESSION: PSYCHIATRIC GENETICS, NEUROGENETICS AND NEURODEGENERATION**
  Program Number 2584
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Expanding the phenotype of Autosomal Dominant Leukodystrophy associated with LMNB1 duplication. *C. Toro[1], SG. Ziegler[2], C. Groden[1], CD. Blair[3], K. Cao[3], H. Carlson-Donohoe[2], DR. Simeonov[2], FS. Collins[3], WA. Gahl[1,2]* 1) Undiagnosed Diseases Program, NHGRI, National Institutes of Health, Bethesda, MD; 2) Medical Genetics Branch, NHGRI, National Institutes of Health, Bethesda, MD; 3) Genome Technology Branch, NHGRI, National Institutes of Health, Bethesda, MD.

Autosomal Dominant Leukodystrophy (ADLD) is a rare disease due to duplications of LMNB1, encoding lamin B1. Patients typically present in the third or fourth decade of life, with progressive dysautonomia, spasticity and cerebellar dysfunction. Patients with ADLD are often misdiagnosed early on in their disease with multiple sclerosis. Dominant inheritance, distinct MRI features of widespread subcortical demyelination with sparing of subcortical U fibers,

involvement of corticospinal tracts and middle cerebellar peduncles, and absence of inflammatory CSF changes, support the diagnosis. Histopathological abnormalities available in a few cases are restricted to glial cells. Over-expression of LMNB1 has been suggested as the mechanism leading to ADLD. We expand the phenotype of ADLD by describing an otherwise previously healthy 47 year-old man with a 5-year history of dysautonomia, spasticity, ataxia and lower extremity pain. An Illumina Omni-Quad Chip identified a ~360 Kb duplication encompassing the LMNB1 gene on chromosome 5q23.2, confirmed by multiplex PCR. The absence of family history suggested a *de-novo* duplication. PAS positive bodies in eccrine sweat glands were present in a forearm skin biopsy. Sural nerve biopsy had no axon or Schwann cell abnormalities. Immunofluorescence staining for nuclear envelop proteins revealed abnormal nuclear morphology. The patient 's cultured fibroblasts had increased nuclear blebbing at passage 7 (30% compared to 11% in controls); cellular senescence was also enhanced, as measured by senescence associated β-galactosidase assay. Melanocytes grew slowly, with ballooned cytoplasm and misshapen nuclei (76% blebbed vs 47% in controls; 11% binucleated vs 0% in controls). However, qRT_PCR and Western blotting revealed that lamin B1 mRNA and protein were not over-produced in our patient 's cultured fibroblasts. The role of lamin B1 as an integral structural component of the nuclear envelope explains the morphological changes expressed in cultured cell lines from our patient. In addition, lamin B1 exerts transcription regulatory roles through its dynamic interactions with chromatin. A disturbance in these regulatory processes might be central to the emergence of demyelination in AOLD. Our patient 's cells provide a model system to study the LMNB1-dependent events, including those required for myelin maintenance in the adult human brain. The cells also provide a venue in which to investigate therapeutic interventions.

- **POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY**
  Program Number 2830
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

A Sequential Test Algorithm for DNA Pooling/Bootstrap-Based Studies. *J. I. Velez Valbuena, M. Arcos-Burgos*
Med Genetics Branch, NHGRI/NIH, Bethesda, MD.

DNA pooling is a practical way to reduce the cost of large-scale association studies to identify susceptibility loci. In contrast to individual genotyping, in DNA pooling we can combine samples from *N* cases and *M* controls into two single pooled sample tests and estimate the allele frequency of hundred of thousand SNPs using high-throughput genotyping technologies. Selection of disease-associated SNPs is performed based on their *P*-value after a statistical test has been run. For the subset of those SNPs determined to be significant, all individuals are then genotyped to corroborate results from the pooled samples.

The strategy described above has been successfully used for years. However, there are situations in which either it is not possible to recruit the number of patients (cases) needed based on power estimations or replication of DNA pools is important. In both situations, the limitation is the availability of DNA samples.

When using sequential testing, formally presented in 1945 as sequential probability ratio test (SPRT), units of interest are sequentially included as they are generated, and a statistical test is run at every stage. In the context of DNA pooling, units would be represented by the pooled samples coming from each group of cases and controls while using a bootstrapping re-sampling strategy.

We have developed a SPRT algorithm for identifying disease-associated SNPs when comparing cases and controls via DNA pooling that is at least as powerful as the strategy previously described, but that needs less DNA samples than the strategy described above. Along with our algorithm, we also provide a way to estimate the number of stages needed, i.e., the number of SNP-chip pairs to stop the algorithm achieving a desired significance and power levels. We illustrate how our approach works using GWAS on Oppositional Conduct Disorder (OCD) and Attention Deficit Hyperactivity Disorder (ADHD).

- **POSTER SESSION: THERAPY FOR GENETIC DISORDERS**
  Program Number 3094
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Wed 10:00AM-7:00PM
  Presentation Time: Wed, Nov 3, 2010, 6:00PM-7:00PM

Abstract Content

Cholesterol And The GM_2 Ganglioside Accumulate In Chédiak-Higashi Syndrome Cells And Are Released By A Small Lysosome Corrector Molecule. *AR. Cullinane[1], W. Introne[1], M. Huizing[1], WA. Gahl[1], W. Zheng[2], W. Westbroek[1]*
1) Medical Genetics Branch, NHGRI (NIH), Bethesda, MD; 2) NIH Chemical Genomics Center (NHGRI), National Institutes of Health, Bethesda MD.

Chédiak-Higashi syndrome (CHS; MIM #214500) is a rare autosomal recessive disorder characterized by partial

oculocutaneous albinism, immunodeficiency, late onset neurological features and a mild bleeding tendency. Mutations are identified in the LYST/CHS1 gene in approximately 90% of patients, the protein product of which has an unknown function but is thought to be involved in the regulation of lysosomal size and trafficking. Approximately 85% of affected individuals with nonsense or frameshift mutations develop the classic early-onset accelerated phase of lymphoproliferative infiltration of the bone marrow and reticuloendothelial system. Atypical late onset CHS patients who generally have missense mutations, and classic patients who undergo stem cell transplantation develop neurological features in adolescence and early adulthood. These features include low cognitive abilities, balance abnormalities, ataxia, tremor, absent deep-tendon reflexes and motor and sensory neuropathies. The cellular hallmark of CHS is the presence of enlarged lysosomes and lysosome-related organelles in many cell types. Confocal immunofluorescence microscopy of two atypical CHS patients ' fibroblasts and melanocytes showed an accumulation of GM_2 gangliosides and cholesterol in enlarged lysosomal structures. GM_1 did not accumulate in these cells. When a small lysosome corrector compound was added to the growth medium of the affected fibroblasts for five days of culture, both the GM_2 and cholesterol accumulation dispersed and the cells appeared to be more like control cells. Using dot blot analysis, the actual amount of GM_2 also decreased after drug treatment in the patient cells similar to that of control cells. The lysosomal size in CHS cells was not corrected by compound treatment. Both GM_2 and cholesterol accumulation has previously been shown to cause failure of neuronal dendrite outgrowth, impaired oligodendrocyte development and myelin biogenesis and neurodegeneration. The identification of these two molecules accumulating in CHS cells correlates with neurological dysfunction and could explain the neurological findings in CHS. Furthermore, the discovery of a small compound that reduces the accumulation of these molecules could potentially be used in the clinical management of the condition.

Day 3: Thursday, November 4, 2010

INVITED PRESENTATIONS

- **SESSION TITLE: 25. RESOURCES AND METHODS FOR ANALYSIS OF GENE FUNCTION IN MOUSE MODELS**
  Invited Scientific
  Room 207, Level 2, Convention Center
  Thu Nov 4, 2010 08:00AM-10:00AM

Session Descriptions:
Recent progress in efforts such as the NIH-supported Knock-out Mouse Project (KOMP) as well as EUCOMM (European Conditional Mouse Mutagenesis Program), NorCOMM (North American Conditional Mouse Mutagenesis project) and Texas A&M Institute for Genomic Medicine (TIGM) has resulted in the generation of large numbers of ES cells carrying defined mutations. The aims of this session will be to inform the general community of human geneticists about these reagents and how they can be employed to investigate mammalian development and human disease. Colin Fletcher will present an overview of these resources and will discuss how they can be accessed. Monica Justice, Andy Peterson and David Beier will provide illustrations of methods for their utilization and examples of medically-relevant results.

Presentation Information

- 08:05AM-08:50AM
  The KOMP and other targeted mouse mutant resources. *C. Fletcher* NHGRI/NIH.

- **SESSION TITLE: 20. FINDING HIGH-RISK SUSCEPTIBILITY GENE VARIANTS USING NEWER ANALYTICAL AND GENOMIC TOOLS**
  Invited Scientific
  Ballroom A, Level 3, Convention Center
  Thu Nov 4, 2010 08:00AM-10:00AM

Session Descriptions:
Although much attention is currently focused on genome-wide association studies and the detection of common, low-risk susceptibility gene variants, findings from diverse studies ranging from linkage and heritability evaluations to loss of heterozygosity (LOH)/comparative genomic hybridization (CGH) and functional genomics examinations suggest that many high-risk susceptibility gene variants remain to be identified. Discovering these high-risk genes, however, has become more complicated. The advent of new technologic methods such as arrayCGH and next generation sequencing and the incorporation of epidemiologic/clinical data and/or molecular genetic/functional genomics data plus enhancement of analytic strategies will help to identify additional high-risk susceptibility gene variants. In particular, combining these clinical/epidemiologic, analytic, and laboratory strategies should enhance the prospects for finding more high-risk susceptibility gene variants. This session will examine the current state of high-risk susceptibility gene variant identification and the challenges and opportunities to improve gene discovery.

Presentation Information

- 08:20AM-08:45AM
  Linkage was successful, so why can't we find a gene? Examples from cancer and other diseases.
  J. Bailey-Wilson NHGRI/NIH.

POSTERS

- **POSTER SESSION: CANCER GENETICS**
  Program Number 445
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content
Canine Gastric Cancer GWAS Identifies a Single Locus in the Chow Chow. *D. M. Karyadi[1], E. A. McNiel[2], E. Karlins[1], N. Madrill[2], E. A. Ostrander[1]* 1) Cancer Genetics Branch, NHGRI/NIH, Bethesda, MD; 2) Dept. of Small Animal Clinical Services, Michigan State University, East Lancing, MI.
The role of predisposition genes in human gastric cancer (GC) is not well understood. However, strong associations between specific dog breeds and GC risk have been reported, suggesting a mechanism for identifying genes important in GC susceptibility. Breeds at increased risk of GC include the Chow Chow, Belgian Sheepdog, Belgian Tervuren, and Keeshond. Of note, Chow Chows have striking 10-20-fold increased risk of developing GC compared to other breeds. GC in Chow Chows is clinically and morphologically similar to familial forms of the disease described in humans and, like human GC, has a grave prognosis. Diagnosis is usually made late in the course of the disease when effective treatment is rarely feasible. In humans mutations in limited number of genes have been suggested to increase risk, such as the *CDH1* gene and genes in the DNA mismatch repair pathway. However, these genes explain only a portion of familial human disease, little if any sporadic disease, and none of the canine disease. We hypothesize that a more global evaluation of the canine genome is likely to reveal genes of interest for both human and canine GC susceptibility. We conducted a genome-wide association study with Chow Chow cases, and unrelated and related controls using the Illumina CanineHD BeadChip with 170,000 SNPs. The final data set consisted of 125,713 SNPs informative in Chow Chows. Using the single-locus chi-squared test of significance, we calculated the allelic association of each SNP with the disease phenotype. In the analysis of cases and unrelated controls, the top 4 most significant SNPs were all at the same genomic locus (P_{raw} = $6.83x10^{-5}$ - $1.09x10^{-5}$), creating a double peak spaced 20 Mb apart. Chromosome-wide permutations (n=100,000) were performed to test for significance of this locus. For the SNP with the best disease association, the chromosome-wide empirical P value is significant at P = 0.021. Analysis of the Chow Chow cases and entire set of controls (unrelated plus related) identified the same locus. Chromosome-wide permutations (n=100,000) were also significant for the top SNP (P_{emp} = 0.033). None of the peaks in other genomic regions are significant at the chromosome-wide level in either analysis. We are in the process of fine-mapping this region and using haplotype analysis in order to identify the causal variant. Our findings would likely highlight genes and biological pathways important for future studies of both human and canine GC.

- **POSTER SESSION: CANCER GENETICS**
  Program Number 503
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

Linkage study of prostate cancer in high-risk African American families from Louisiana. *E. M. Ledet[1], J. E. Bailey-Wilson[2], D. M. Mandal[1]* 1) Dept Genetics, Louisiana State Univ HSC, New Orleans, LA; 2) NHGRI, NIH, Baltimore, MD.

Prostate cancer is a complex multi-allelic disease and the most common malignancy in men throughout the world. In the United States, a lifetime risk of mortality from prostate cancer is 3% for white men and 4% for African-American men. Thus far, disease susceptibility loci have been identified for this cancer but definite locus-specific information is not established due to the tremendous amount of genetic heterogeneity. Previously, we performed a genome-wide linkage scan on three families using microsatellite markers and identified a region on chromosome 22q13. Given the small sample size of three families, this lod-score value does not reach the genome wide significance level. However, we have since continued recruitment and accrued 20 large high-risk African-American families with at least 3 affected individuals; additionally, 28 large high-risk Caucasian families have been recruited. Demographic information and relevant clinical information has been documented from the hospital pathological report on the affected. Recently an Infinium II HumanLinkage-12 panel (Illumina, Inc.) with 6,090 SNP markers was performed on 180 DNA samples, including 15 African American families and 4 Caucasian families. This panel is optimized for linkage detection for both monogenic and polygenic disorders; SNPs are distributed on every chromosome with an average gap of 441 Kb and 0.58 cM. Three samples were discarded from analysis due to poor array performance, for a total of 177 samples imported into BeadStudio version 3.3.7; a proprietary calling algorithm was used and SNPs which failed quality control or calling were removed from the data set. Quality control and pruning of released SNPs was performed using PLINK. Linkage analysis is ongoing on the African-American cohort with Merlin and Genehunter-Plus. In this study we intend to identify any markers associated with prostate cancer in high-risk African American families and document any correlation between clinical features, such as prostate specific antigen (PSA), 'age of onset ' and/or Gleason score.

- **POSTER SESSION: DEVELOPMENT**
  Program Number 1329
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

A *Sox10* sensitized mouse ENU mutagenesis screen: uncovering pathways in development and disease. *D. E. Watkins-Chow[1], K. E. Leeds[1], R. Mullen[1], D. L. Silver[1], K. Buac[1], H. W. Hwang[1], I. Matera[2], S. K. Loftus[1], D. M. Larson[1], A. Incao[1], W. J. Pavan[1]* 1) Genetic Disease Research Branch, NHGRI, NIH, Bethesda, MD; 2) Laboratorio di Genetica Molecolare, Istituto G. Gaslini, Genova, Italy.

Melanocytes are specialized, neural crest-derived cells responsible for pigment production in the skin. Genetic interference of neural crest development can present as altered pigmentation in skin and/or hair and can be associated with debilitating diseases (neurocristopathies) including deafness, blindness, cleft lip, congenital megacolon, and albinism. Disruption of genes that regulate pigmentation can also affect a diverse array of tissues, due to inherent pleiotropic roles of genes that have been co-opted for function in non-neural crest-derived structures. We have used a *Sox10* sensitized mouse ENU mutagenesis screen to uncover previously uncharacterized pathways in melanocyte development, to provide insights into development of additional organ systems, and to generate models relevant for dissecting human disease etiology. This screen identifies mutations that increase the phenotypic severity of *Sox10* haploinsufficient mice ($Sox10^{Lacz/+}$) that carry a mutation in a transcription factor essential for melanocyte development. From analysis of 600 pedigrees, we have identified five dominant modifiers of the *Sox10* phenotype (*Mos1-5*) and four recessive modifiers of the embryonic $Sox10^{LacZ}$ expression pattern (*msp1-4*). The causative mutations associated with these modifiers affect genes involved in a variety of functions including hedgehog, neuregulin, and semaphorin signaling as well as ribosomal and RNA binding proteins. Comparative analysis of melanoblast development confirms that these mutations affect different time points in development and that different mechanisms lead to the observed melanoblast phenotypes. Further characterization of genes identified in the screen will contribute to our understanding of human genome function, provide additional disease models for human neurocristopathies and identify additional candidate pathways for melanoma progression.

- **POSTER SESSION: DEVELOPMENT**
  Program Number 1331
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

Patterns of sensory processing and their relationship to the neurobehavioral phenotype of children with Smith-Magenis syndrome (SMS). *R. S. Morse[1], H. Hildenbrand[2], A. C. M. Smith[1]* 1) NHGRI/Office of the Clinical Director, NHGRI, NIHOCD, NHGRI/NIH, Bethesda, MD; 2) Rehabilitative Medicine Dept, NIH, Bethesda, MD.

Smith- Magenis Syndrome (SMS) is a rare (1/15,000) microdeletion syndrome of chromosome 17 p11.2 associated with a specific pattern of physical, developmental and behavioral characteristics. The neurobehavioral phenotype is distinct and complex, characterized by high rates of outbursts/tantrums, attention-seeking, impulsivity, aggression, hyperactivity, distractibility, toileting difficulties, stereotypies (repetitive or self-stimulatory behaviors), sleep disturbance and self-injurious behaviors (Dykens & Smith, 1998). We suspect that underlying sensory processing deficits contribute to previously observed maladaptive behaviors, social difficulties and functional deficits that characterize the syndrome. This cross-disciplinary study investigates the relationship between sensory modulation and maladaptive behaviors characteristic of SMS using two validated instruments derived from the disciplines of occupational therapy and behavioral psychology. The Sensory Profile (SP) Caregiver Questionnaire and Child Behavior Checklist (CBCL) were collected from 25 parents of individuals (13F/12M; ages 3-25yrs; mean 7.16 yrs) with a confirmed SMS diagnosis. Results: Clinically meaningful low scores for one or more of the SP quadrants were documented in 24/25 (96%) of SMS subjects, indicative of sensory processing/modulation problems (Registration = 22; Seeking = 18; Sensitivity = 17; Avoiding = 21). Only one individual scored within the normal limits for all four SP quadrants (male; 3 years of age). On the CBCL, 20/25 (80%) SMS subjects had clinically significant high total scores for maladaptive behavior. Spearman Correlation Coefficients revealed statistically significant inverse relationships ($p=<0.05$) between the SP quadrant scores and the total scores on the CBCL. These results support the hypothesis that in individuals with SMS sensory modulation difficulties are associated with maladaptive behavior and functioning. Further examination of the relationship between patterns of sensory processing and maladaptive behaviors may expand the behavior management approach and contribute to improved social participation and daily function in individuals with SMS.

- **POSTER SESSION: . EVOLUTIONARY AND POPULATION GENETICS**
  Program Number 1453
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

The LPHN3 Common Haplotype Variant Predisposing to Attention-Deficit/Hyperactivity Disorder (ADHD) is Ancestral to the Protective Variant. *M. Muenke, M. Arcos-Burgos* Med Gen Branch, NHGRI/NIH, Bethesda, MD.

Recently we identified (Latrophilin 3) LPHN3, a novel Attention Deficit/Hyperactivity Disorder (ADHD) susceptibility gene and showed that a LPHN3 common variant confers susceptibility to ADHD and predicts effectiveness of stimulant medication. LPHN3, a brain-specific member of the LPHN subfamily of G-protein coupled receptors, is expressed in ADHD-related regions, such as amygdale, caudate nucleus and cerebral cortex. Further, this ADHD susceptibility variant affect metabolism in neural circuits implicated in ADHD. Here by genotyping more than 300 SNP variants, including some spanning the LPHN3 genomic region associated to ADHD, in different species of monkeys (sanguinus (n=1), macaca mulatta (n=2)), and primates (gorilla gorilla (3), pan troglodytes (8), and pongo pygmaeus (2)) (a phylogeny covering ~70 millions years of evolution), we were able of reconstructing a phylogenetic tree showing that the LPHN3 variant of susceptibility is ancestral to the LPHN3 variant conferring protection against ADHD. Furthermore, the reconstructed phylogenetic tree suggests that the evolutionary splitting between these variants happens before modern humans separated from great apes. This phylogenetic scenario is in strong agreement with the suggested selective beneficial advantage of genetic variants conferring susceptibility to ADHD and consequently with the fact that the cluster of behaviors outlined by the ADHD syndrome are and/or were indeed very old and normal.

- **POSTER SESSION: EVOLUTIONARY AND POPULATION GENETICS**
  Program Number 1459
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

Genetic Variation and Positive Selection at the WFDC locus in Hominids. *Z. Ferreira[1,3], A. Andres[1], W. Kretzschmar[1], J. Mullikin[1,2], W. Swanson[4], K. Gonder[6], S. Tishkoff[5], A. Stone[7], E. Green[1], B. Hurle[1], NISC Comparative Sequencing Program* 1) NHGRI, NIH, Bethesda, MD; 2) NIH Intramural Sequencing Center, Bethesda, MD; 3) IPATIMUP Rua Dr. Roberto Frias, s/n 4200-465 Porto, PORTUGAL; 4) University of Washington Seattle, WA; 5) Departments of Genetics and Biology, University of Pennsylvania, Philadelphia, PA; 6) Department of Biological Sciences University at Albany, State University of New York, Albany NY; 7) Department of Anthropology, Arizona State University, Tempe, AZ.

The whey acidic protein (WAP) four-disulfide core domain (*WFDC*) genes encode protease inhibitors with roles in innate immunity and regulation of the endogenous protease kallikreins (*KLK*). Through a comparative genomics strategy that involved the re-sequencing of the *WFDC* centromeric sub-locus in twelve primates, it was previously shown that a striking number of contiguous *WFDC* genes and the neighboring seminal genes (semenogelin I and II - *SEMGI* and *SEMGII*) show strong patterns of positive selection in primates. It is unusual to find a series of tightly linked genes that all show robust patterns of positive selection, as hitchhiking effects make it difficult for successive selective sweeps of tightly linked loci. Despite this co-localization of positively selected genes, the *WFDC* locus does not stand out as particularly unusual in humans with respect to known genetic variation databases. We tested whether the levels and patterns of genetic variation within the *WFDC* locus differ among hominid species. We sequenced 19 genes of the *WFDC* locus, plus 54 evenly spaced non-coding regions in 71 humans from three HapMap populations and 68 western equatorial African chimpanzees from three *Pan* subspecies. A set of 47 unlinked and neutrally evolving loci was also surveyed to assess the general patterns of diversity. Overall, we generated ~23 and ~13 Mb of high-quality sequence data from humans and chimpanzees, respectively, enabling the identification of 541 human and 847 chimpanzee single-nucleotide polymorphisms (SNPs) and 487 human-chimp fixed differences. Ongoing research includes detecting recent positive selection events using classical neutrality tests and identifying both incomplete and complete sweeps as well as detecting differences at inter and intra-species level. Test significance is being assessed through coalescent simulations under different demographic scenarios. Unique features of this study include examining the dynamic nature of the *WFDC* locus in primates, capitalizing on the sheer number and verified demographic origin of the chimpanzee samples, and noting the lack of ascertainment bias in SNP collection. Our efforts offer insights on the evolutionary forces driving the rapid diversification of *WFDC* and *SEMG* genes in hominoids, and improve our knowledge about the biological dynamics of rapidly evolving genomic regions in primates.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2029
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

Alpha-synuclein expression and localization in cultured neurons from glucocerebrosidase-deficient mouse models. *W. Westbroek[1], W. Xiao[1], S. W. Klontz[1], Y. N. Blech-Hermoni[1], M. R. Cookson[2], E. Sidransky[1]* 1) Medical Genetics Branch, NHGRI, NIH, Bethesda, MD; 2) Laboratory of Neurogenetics, NIA, NIH, Bethesda, MD.

Gaucher disease is an autosomal recessive lysosomal storage disorder caused by mutations in the glucocerebrosidase gene (GBA). In patients with Gaucher disease, deficiency of the enzyme glucocerebrosidase (GCase) leads to accumulation of the glycolipid glucosylceramide in reticulo-endothelial cells in the spleen, liver, bones, and, in neuronopathic forms, the brain. A subset of patients with Gaucher disease develop Parkinson disease, an adult-onset neurodegenerative disease characterized by motor dysfunction due to loss of dopaminergic neurons in the substantia nigra and alpha-synuclein protein aggregation into Lewy body structures in the brain. Recent studies have shown that subjects with Parkinson disease are over five times more likely to carry GBA mutations than controls, but the molecular mechanisms by which the two diseases are related remain unknown. We utilized mouse models of Gaucher disease including both the null allele knock-out and point mutation models to investigate the effect of GCase deficiency on alpha-synuclein expression and cellular localization. Primary embryonic hippocampal cells were cultured from E-18 mice homozygous and heterozygous for the null-allele, as well as embryonic mice with other common mutant GBA alleles. We performed immunocytochemistry on distinct primary neuronal cultures with several anti-alpha-synuclein antibodies, followed by laser scanning confocal microscopy. We found that neuronal cells with deficient or absent GCase activity had increased expression and altered localization of alpha-synuclein.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2093
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

Methylmalonic acidemia and optic nerve atrophy: reversal of sub-acute loss of visual function with anti-oxidant therapy. *J. L. Sloan[1], N. S. Hauser[1], I. Manoli[1], W. M. Zein[2], K. Bowles[2], K. O'Brien[3], B. P. Brooks[2], C. P. Venditti[1]* 1) Genetics and Molecular Biology Branch, NHGRI, Bethesda, MD; 2) Ophthalmic Genetics and Visual Function Branch, NEI, Bethesda, MD; 3) Medical Genetics Branch, NHGRI, Bethesda, MD.

Isolated methylmalonic acidemia (MMA) results primarily from a defect in the mitochondrial enzyme methylmalonyl-CoA mutase (MUT). Patients present with recurrent metabolic crises and multisystemic complications including growth retardation, chronic renal failure, pancreatitis and developmental delay despite optimal management. Studies in MMA knock out mice and patient tissues have suggested that mitochondrial dysfunction plays a significant role in the pathophysiology of MMA. Optic nerve atrophy (ONA) has been described as a rare complication of the disorder, and the pathophysiology is unknown. Patients with isolated MMA evaluated through NIH study 04-HG-0127 (clinicaltrials.gov identifier: NCT00078078) “Clinical and Basic Investigations of Methylmalonic Acidemia and Related Disorders ” underwent comprehensive ophthalmologic evaluation. We report four *mut^0* patients with ONA out of the 56 with isolated MMA followed in our protocol. All four patients were males, and presented with a decreased visual acuity at the ages of 7, 22, 23 and 24 years. Vision loss was bilateral but asymmetric and the progression was variable. Best-corrected vision ranged from 20/40 to 20/800 and in two patients visual evoked potentials were severely diminished and delayed. There were no apparent biochemical or environmental triggers shared by the patients and none had an acute metabolic decompensation during the onset of the symptoms. Each carried two mutations in the MUT gene. Two patients were negative for the Leber Hereditary Optic Neuropathy mitochondrial mutations. Patient 4 was a 24 year-old male, 4 years status-post cadaveric kidney transplantation, whose vision progressively worsened from 20/20 to 20/40 OS and 20/125 OD over a 4-week period. Daily oral coenzyme Q10, vitamin E, ascorbic acid, thiamine, and intravenous infusions followed by oral N-acetylcysteine were employed. The patient ’s visual acuity improved to 20/25 OD and 20/20 OS over one month, although the nerve fiber layer thickness remained decreased. Our experience suggests that 1) ONA is a late onset complication of isolated MMA and therefore all patients should have periodic thorough ophthalmologic evaluation and counseling about the symptoms of ONA. 2) Anti-oxidant therapy can be used to at least partially reverse the functional visual effects of optic nerve pathology in MMA patients in the acute setting.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2037
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content

The Genetics of Hermansky-Pudlak Syndrome. *R. Hess, R. Fischer, W. A. Gahl, M. Huizing* Medical Genetics Branch, NHGRI/NIH, Bethesda, MD.

Hermansky-Pudlak syndrome (HPS) is a disorder of lysosome-related organelle (LRO) biogenesis, resulting in oculocutaneous albinism, a bleeding diathesis, and occasional colitis or pulmonary fibrosis. Eight human HPS subtypes are identified (HPS1-8). Since an accurate diagnosis of each subtype has important prognostic and therapeutic implications and also provides insights into the cell biology of LROs, we extensively characterized each HPS subtype. We have studied 260 HPS patients at the NIH Clinical Center. Our molecular analyses indicated that HPS-1 (188 patients, 13 HPS1 mutations) comprises the largest group due to a founder mutation in NW Puerto Rico. HPS-2 results from mutations in AP3B1, encoding the beta3A subunit of adaptor complex-3, a coat protein that mediates vesicle formation. We identified 3 HPS-2 patients, harboring 4 different AP3B1 mutations. We also identified 13 HPS-4 patients (10 HPS4 mutations), 22 HPS-3 patients (10 HPS3 mutations; with founder mutations in Central Puerto Rico and in Ashkenazi Jews), 10 HPS-5 patients (13 HPS5 mutations), and 5 patients with HPS-6 (8 HPS6 mutations). We have not identified any HPS-7 or HPS-8 patients and only one patient/family of each subtype has been reported. Our remaining 12 unclassified HPS patients provide opportunities to identify new HPS-causing genes. Several genes, some corresponding to HPS mouse models that manifest both hypopigmentation and a platelet storage pool deficiency, are good candidates. Any new genetic causes of HPS will aid in elucidating the mechanism by which melanosomes, dense bodies, and lysosomes are created. Our extensive molecular studies allowed for genotype-phenotype analysis. We found that that HPS-1 and HPS-4 patients are at increased risk for developing pulmonary fibrosis and granulomatous colitis. HPS-2 patients have persistent neutropenia and had recurrent childhood infections, and may develop pulmonary fibrosis. HPS-3, HPS-5, and HPS-6 patients are clinically milder, with no apparent pulmonary involvement. These findings reiterate that an accurate diagnosis of each HPS subtype has important prognostic implications.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2119
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content
Urinary Exosomes: Biomarkers for Kidney Disease. *D. Maynard[1], W. Westbroek[1], W. Gahl[1], M. Gunay-Aygun[1,2]* 1) Section on Human Biochemical Genetics, Medical Genetics Branch, NHGRI/NIH, Bethesda, MD; 2) Office of Rare Disease Research, Office of the Director, NIH, Bethesda, MD.

Oral-facial-digital-syndrome type 1 (OFD-1) is an X-linked, male lethal ciliopathy, characterized by malformations of the face, oral cavity and digits and polycystic kidney disease (PKD). Renal cysts in OFD-1 generally originate from the glomeruli; cysts in autosomal dominant polycystic kidney disease (ADPKD) develop from any part of the nephron. Polarity in kidney epithelial cells is essential to the integrity and function of the kidney, since insertion of specific transporters and other proteins into apical membranes lining the renal tubule lumen or basal domains adjacent to the interstitium is critical. Studies of ADPKD renal tubule epithilia demonstrate that defects in membrane polarity affect specific sets of proteins involved in sodium transport and in EGF signal transduction. OFD-1 renal cells have not been studied.

Exosomes are 50-90 nm vesicles containing discrete packets of cytosol and are released by many cell types. Exosome membranes have the same orientation as the plasma membrane. Study of urinary exosomes provides a noninvasive means for detection and analysis of protein-expression/trafficking changes in renal tubule cells. Exosomes from OFD-1 and other PKD patients may contain a different contingent of membrane and/or cytosolic proteins due to protein mis-targeting/polarity defects in renal epithelial cells. We isolated exosomes from the urines of OFD-1 and other ciliopathy patients with PKD and from controls. We removed Tamm-Horsfall protein from the exosome samples and demonstrated by electron microscopy that this technique does not alter their morphology. We used proteomics combined with mass spectrometry to analyze the protein content of the exosomes. Preliminary proteomic results from OFD-1 and control urine samples found known exosome marker proteins (CD63, CD81, and CD9) as well as VPS (TSG101, Alix, and VPS28) proteins involved in multivesicular body (MVB) targeting and biogenesis. Proteins associated with hypertension and PKD or other kidney diseases (aquaporin-2, polycystin-1, -2, podocin, and neprilysin) were also found. These findings establish urinary exosomes as a system that reflects the contingent of proteins present in OFD-1 cells, and that can be investigated in other kidney disorders.

- **POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2389
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 5:00PM-6:00PM

Abstract Content
Characterizing expression of Proteus marker proteins in skin using immunohistochemistry. *M. J. Lindhurst[1], J. C. Sapp[1], C. R. Lee[2], L. G. Biesecker[1]* 1) GDRB, NHGRI/NIH, Bethesda, MD; 2) NCI/NIH, Bethesda, MD.

Proteus syndrome (PS) is a rare disorder that is hypothesized to be caused by a mosaic gene alteration lethal in the non-mosaic state. The phenotype of PS is highly variable. Objective diagnostic criteria have been published that has allowed for a more precise definition of the disorder. These criteria require: a mosaic distribution of lesions, a sporadic occurrence, and a progressive course of the disease plus a combination of characteristic manifestations. Characteristic manifestations include cerebriform connective tissue nevi (CCTN), linear epidermal nevi (LEN), vascular malformations, disproportionate overgrowth, and others. While it is straightforward to recognize a PS lesion in an affected patient or to recognize a histological section from a patient with PS as abnormal, a cellular phenotype had not been established. Recently, we reported the results of a micorarray experiment comparing cultured skin fibroblasts from affected PS lesions to skin fibroblasts cultured from non-PS skin. We found seven genes, *CD9*, *COL14A1*, *COL15A1*, *COL21A1*, *COL6A3*, *EML1*, and *FBN2*, that were up-regulated in the PS-affected cultures. When qRTPCR was used with a larger sample set to validate the micorarray results, *CD9*, *COL14A1*, *COL21A1*, and *EML1* showed significant differential expression. Immunohistochemistry using antibodies to CD9, COL15A1, and FBN2 showed increased staining of dermal fibroblasts and the surrounding extracellular matrix in five plantar CCTN sections from four PS patients when compared to plantar skin controls from five non-PS individuals. This staining was non-uniform within a section and varied among the PS and control sections. Presently, we are characterizing the staining in more detail to see if additional structures within the skin show any differential expression and to look for correlations between the areas with increased staining and other characteristics such as morphology or staining of other types of skin markers. In addition, we are testing antibodies to COL21A1 and COL6A3 to see if they are up-regulated in vivo. We are also examining skin biopsies taken from other areas, both affected and unaffected, of PS patients and are planning to compare the staining to other types of collagenomas and lesions found in other overgrowth syndromes. This survey will help not only in understanding the

biology of skin lesions in PS, but may also lead to the development of markers that can be used to distinguish different overgrowth syndromes.

▪ POSTER SESSION: CANCER GENETICS

Program Number 444
Exhibit Hall A, Lower Concourse Level, Convention Center
Thu 7:00AM-7:00PM
Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

Mapping Transitional Cell Carcinoma of the Bladder in the Dog. *H. G. Parker[1], E. M. Kwon[1,4], D. W. Knapp[2], P. Bonney[2], E. McNiel[3], E. A. Ostrander[1]* 1) Comparative Genomics Section, Cancer Genetics Branch, NHGRI, NIH, Bethesda, MD; 2) Purdue Comparative Oncology Program, Dept. of Veterinary Clinical Sciences, Purdue University, West Lafayette, IN; 3) Dept. of Veterinary Clinical Sciences, University of Minnesota, Minneapolis, MN; 4) Program in Human Genetics and Molecular Biology, Johns Hopkins University School of Medicine, Baltimore, MD.

Transitional cell carcinoma (TCC) of the bladder in the domestic dog is an ideal model of invasive bladder cancer in humans, which takes the lives of more than 14,000 people each year. Both diseases develop spontaneously, respond similarly to drug treatments, and have nearly identical histopathology. TCC is the most common cancer of the urinary bladder in pet dogs. It is an aggressive disease resulting in metastasis in approximately 50% of all dogs diagnosed. Currently little is known about the genetic basis of human bladder cancer. Recent GWAS studies identified two risk loci in both European and Asian populations, however, they account for only a small fraction of the overall risk of developing the disease. In comparison, our group has identified a small number of dog breeds that show a greatly increased risk of bladder cancer, as much as 20 fold higher than average, indicating a strong inherited component to the disease. In order to find mutations that increase susceptibility to TCC of the bladder, we have conducted a genome-wide association study using the Affymetrix canine v2 SNP chip on 122 Scottish terriers (ST), West Highland White terriers (WHWT), and Shetland Sheepdogs (SSD) with confirmed diagnoses of TCC compared to 135 dogs of the same breeds that are nine years or older and have never been diagnosed with cancer of any form. After correcting for both population structure and kinship, we have identified two primary susceptibility loci within the three breeds with $p < 1x10^{-6}$. The association at locus1 is driven primarily by the ST and is increased with the addition of WHWT but not with SSD. Locus2 is associated with the TCC in the WHWT and SSD when analyzed apart from the ST and does not show association with the disease in the ST. We hypothesize based on allele frequencies within the region that the ST are nearing fixation at locus2, explaining the 20-fold increased risk in this breed. We have designed and run a Goldengate assay of 768 SNPs covering both loci at ~15kb density in order to fine map the region and assess gross copy number within the germline and within tumor DNA at both loci. We expect to find overlapping regions between the three breeds that contain causal mutations that increase overall risk of bladder cancer. The identification of genetic risk factors for TCC will allow selection of molecular targets for early detection and treatment in both humans and dogs.

▪ POSTER SESSION: COMPLEX TRAITS: THEORY AND METHODS

Program Number 1110
Exhibit Hall A, Lower Concourse Level, Convention Center
Thu 7:00AM-7:00PM
Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

Replication of GWAS loci for fasting plasma glucose in African Americans. *E. Ramos[1], G. Chen[1], A. Doumatey[1], D. Shriner[1], N. P. Gerry[2], A. Herbert[3], H. Huang[1], J. Zhou[1], M. F. Christman[2], A. Adeyemo[1], C. Rotimi[1]* 1) Center for Research on Genomics and Global Health, NIH/NHGRI, Bethesda, MD; 2) Coriell Institute for Medical Research, Camden, NJ; 3) Department of Genetics and Genomics, Boston University School of Medicine, Boston, MA.

Chronically elevated blood glucose (hyperglycemia) is the primary indicator of type 2 diabetes, which has a prevalence that varies considerably by ethnicity in the United States with African Americans disproportionately affected. Genome-wide association studies (GWAS) have significantly enhanced our understanding of the genetic basis of diabetes and related traits including fasting plasma glucose (FPG). However, the majority of GWAS have been conducted in populations of European ancestry (EA), including a recent meta-analysis of multiple cohorts. Thus, it is important to conduct replication analyses in non-EA populations to both verify and identify shared loci associated with FPG across populations. We used data collected from nondiabetic individuals (n = 927) that participated in the Howard University Family Study, a cohort of African Americans, to replicate previously published GWAS of FPG. In addition to comparing SNPs directly, we queried a 500-kb window centered on each reported SNP for additional markers in linkage disequilibrium (LD). Using direct SNP and LD-based comparisons, we replicated multiple SNPs previously associated with FPG and strongly associated with type 2 diabetes in recently published meta-analyses and related GWAS in EA populations. The replicated SNPs included those in or near TCF7L2, SLC30A8, G6PC2, MTNR1B, DGKB-TMEM195, and GCKR. We also replicated additional variants in LD with the reported SNP in ZMAT4 and adjacent to IRS1. We replicated multiple GWAS variants for FPG in our cohort of African Americans. Using LD-based strategy, we also identified SNPs not previously reported demonstrating the utility of using diverse populations for replication analysis.

- **POSTER SESSION: GENETICS EDUCATION**
  Program Number 1688
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

Results from a survey investigating the background and training of pre- and post-doctoral fellows in statistical genetics and genetic epidemiology. *A. Wilson, M. Krishnan, The NIH Working Group on Statistical Geneticists Training Needs* Genometrics Section, NIH/NHGRI, Baltimore, MD.

With increasing numbers of genome-wide association and sequencing studies being performed, the scientific community has been deluged with data that require scientists who can develop and perform statistical genetic analyses. There is concern that there are not enough scientists to perform these analyses. A survey was conducted in 2008-2009 to determine the number of pre- and post-doctoral trainees and their type of training and potential expertise. The survey also queried the background and expertise of their faculty level trainers, whether the number of training slots available was adequate and whether these positions were filled. Members of the International Genetic Epidemiology (IGES) were invited to participate through the IGES newsletters prior to the 2008 IGES annual meeting. A second notice went to the IGES membership at the end of January and to selected members of the American Society of Human Genetics. In February 2009, there were 391 responses, with 197 from faculty level trainers, and 96 and 100 responses from pre- and post-doctoral trainees, respectively. Sixty-one percent (105/172) of the Faculty level trainers had NIH funding, and 32% (55/171) had pre-doctoral students supported on NIH T32 training or F fellowship grants. The number of current pre-doctoral trainees per trainer was 2.4 (132 trainers) and the total number of pre-doctoral trainees “in the pipeline ” was 318. Similarly, the average number of post-doctoral trainees per trainer was 1.2 (130 trainers) and the total number of post-doctoral trainees “in the pipeline ” was 150. The most frequent response for the reason that training slots were unfilled was that there were not enough qualified domestic applicants. The distribution of pre-doctoral fellows (96) was roughly normally distributed with the largest number of pre-docs in their third year of training. At least 40% of the pre-doctoral trainees had not had a single formal course in the following disciplines: computational biology, computer science, genetics, mathematics, medical genetics, molecular biology, population genetics or quantitative genetics. The distribution of years of training for post-doctoral fellows was more uniform than that of pre-doctoral fellows, with 64% (51/80) having two or less years of training. Based on the distribution of length of post-doctoral fellowships, roughly 11 - 17 post-doctoral fellows complete training each year.

- **POSTER SESSION: GENOMICS**
  Program Number 1888
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

Comparison and application of whole exome and genome sequencing on an individual with high risk for atherosclerosis. *J. K. Teer, N. F. Hansen, P. F. Cherukuri, L. L. Bonnycastle, P. Cruz, P. S. Chines, H. Ozel Abaan, E. H. Margulies, E. D. Green, J. C. Mullikin, L. G. Biesecker* NHGRI, NIH, Bethesda, MD.

Massively parallel sequencing has allowed broader interrogation of genomes for variants that cause disease. Continuing improvements now allow whole human genome sequencing in a relatively short period of time. However, targeted sequencing requires fewer bases than a whole genome, and therefore allows more targeted samples than whole genome samples. We have compared coverage of the CCDS exome in NA18507 (HapMap Yoruba) using three different exome capture kits. We find that genotype sensitivity (% of the CCDS regions covered with high-quality genotype calls) is similar between all methods: 86%-88%. In comparison, previously reported 30x whole genome coverage of the CCDS regions in NA18507 was ~73%. We have also compared a more recent 60x whole genome sequence of a ClinSeq ™ individual with exome capture. 60x whole genome sequence covered 86% of the CCDS using 192Gb total sequence, whereas exome capture covered 89% with 6.7Gb total. Both methods showed >99.9% overall concordance with genotype chip calls.

We have implemented a secondary analysis pipeline to realign reads using a gapped aligner, cross_match, and to call genotypes using a Bayesian based program, Most Probable Genotype (MPG). Genotypes are then annotated for coding status and potential detriment using CD_Pred. We have also developed a graphical java tool, VarSifter, to view, sort, and filter the resulting data, allowing investigators to focus on interesting variants. Using these tools we have sequenced and analyzed more than 70 exomes as part of the ClinSeq ™ program.

We have performed both whole genome shotgun and exome sequencing on a ClinSeq ™ individual at high risk for atherosclerosis. To get the highest coverage, we have merged the two data sets, resulting in 94.9% genotype coverage of the CCDS. Using our analysis tools, we identified 3,719,419 total variants, 22,264 of which were coding. We have

examined the variants, and have limited the list by removing variants previously observed in 8 HapMap samples, by including non-synonymous single-nucleotide variants and frame-shifting deletion/insertion variants, and by examining variants within a linkage region. Many variants fit these criteria, and we are currently evaluating which are most likely to be causative.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2014
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content
Further characterization of Congenital Disorder of Glycosylation IIb in siblings. *G. A. Golas[1], L. A. Wolfe[1], M. He[2], B. Xia[2], W. Zhang[2], X. Song[3], R. Cummings[3], D. R. Adams[1], S. Yang[1], A. Gropman[1, 4], C. J. Tifft[1]* 1) Undiagnosed Diseases Program, NHGRI Bethesda, MD; 2) Department of Human Genetics, Emory University, School of Medicine, Decatur, GA; 3) Department of Biochemistry, Emory University, School of Medicine, Decatur, GA; 4) Department of Neurology, Children 's National Medical Center, Washington, D.C.

Purpose: Describe clinical and biochemical features of two siblings presenting to the Undiagnosed Diseases Program (UDP). Case review: Two siblings born to non-consanguineous, Northern European parents with negative family history and maternal hypothyroidism and gestational diabetes were evaluated. The older sibling was delivered at 40 weeks gestation by emergency C-section. Birth measurements were normal. Postnatal course was complicated by hypotonia with poor latch and suck requiring bottle feeding. By 8 months of age, he had global developmental delay. Comprehensive molecular and biochemical evaluations were unrevealing. Evaluation at age 11 by the UDP, revealed mild dysmorphia, moderate hypotonia, joint laxity, normal reflexes, and no organomegaly. Weight was 90th centile and height 10th centile. He was non-verbal and able to sit independently and ambulate with assistance. His sister was delivered at 40 weeks gestation by repeat C-section. Birth measurements were normal. She developed generalized seizures within the first 24 hours. The CK was 592 U/L and ammonia 79 umol/dL. Brain ultrasounds and CT were normal. When evaluated at age 6 by the UDP, she was mildly dysmorphic with nystagmus, severe hypotonia, joint laxity, adducted thumbs bilaterally, normal reflexes, and no organomegaly. Height and weight were both below the 3rd centile, at the 50% centile for a 3-½ year old. She was non-verbal and unable to sit independently. Methods used: Prospective evaluation and clinical biochemical testing. Evaluations revealed generalized cerebral atrophy, delayed myelination and low NAA by brain MRI/MRS in both sibs, cortical visual impairment with optic nerve atrophy. CSF studies identified cerebral folate deficiency in the younger sibling. Urine oligosaccharides on both sibs demonstrated a tetrasaccharide band. Further investigation, identified it as Hex4 with 3 glucose and 1 mannose. The mannose is at the reducing end. DNA studies on the identified three mutations in the GCS1 gene and single mutations in the mother and unaffected sibling. This gene encodes alpha-glucosidase Ia, a protein described in the Congenital Disorder of Glycosylation type IIB in a single case report in 2000. Results: This case underscores the difficulties of diagnosing complex multi-system disease especially when affected siblings have slightly different phenotypes. It also expands the phenotype of CDGIIb.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2026
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content
Whole Exome Sequencing identifies AFG3L2 homozygous mutations resulting in a novel autosomal-recessive progressive myoclonic epilepsy-ataxia-neuropathy syndrome. *T. M. Pierson[1,2], D. A. Adams[1, 3], F. Bonn[4], P. F. Cheruki[5], J. K. Teer[6], N. F. Hansen[5], P. Cruz[5], N. I. S. C. Comparitive Sequencing Program[7], J. C. Mullikin[6,7], R. W. Blakesley[6], G. Golas[1,3], J. Kwan[8], T. Markello[1,3], C. Blackstone[2,9], A. Sandler[10], K. Fuentes Fajardo[1], C. Tifft[1,3], E. Rugarli[11], W. A. Gahl[1,3], T. Langer[12,13], C. Toro[1]* 1) NIH Undiagnosed Diseases Program, NIH Office of Rare Diseases and NHGRI, NIH, Bethesda, MD; 2) Neurogenetics Branch, NINDS, NIH, Bethesda, MD; 3) Office of the Clinical Director, NHGRI, NIH, Bethesda, MD; 4) Institute for Genetics, University of Cologne, Cologne, Germany; 5) Genome Technology Branch, NHGRI, NIH, Bethesda MD; 6) Genetic Disease Research Branch, NHGRI, NIH, Bethesda MD; 7) National Intramural Sequencing Center, NIH, Bethesda MD; 8) EMG Section, NINDS, NIH, Bethesda, MD, USA; 9) Cellular Neurology Unit, NINDS, NIH, Bethesda, MD; 10) Division of Surgery, Children's National Medical Center, Washington, DC; 11) Biocenter, University of Cologne, Cologne, Germany; 12) Institute for Genetics, Centre for Molecular Medicine (CMMC), Cologne Excellence Cluster on Cellular Stress Responses in Aging-Associated Diseases (CECAD), University of Cologne, Cologne, Germany; 13) Max-Planck-Institute for Biology of Aging, Cologne, Germany.

In neurogenetics, a single gene may be involved in several distinct disorders. Examples of this phenomenon include genes for signaling molecules, cellular membrane trafficking, and energy metabolism. These genes, initially identified with one disease, were “rediscovered ” in the context of another. Whole exome sequencing (WES) is used to search for new pathogenic mutations, but it also identifies variants in known genes that lead to variant phenotypes. We used WES

to identify new homozygous mutations in the AFG3L2 gene resulting in a novel progressive myoclonic epilepsy-ataxia-neuropathy syndrome. Two brothers presented in late infancy with progressive ataxia, dysarthria, spasticity, and myoclonic epilepsy. Associated features included ptosis, dystonia, and action myoclonus. Cognition was intact. Testing revealed cerebellar atrophy and polyneuropathies. Muscle biopsy uncovered mitochondrial respiratory dysfunction and abnormal structure. mtDNA was mildly depleted. The parents (first cousins) were normal neurologically, except the mother had asymptomatic mild cerebellar atrophy on MRI imaging. The AFG3L2 gene causes autosomal-dominant spinocerebellar ataxia, type 28 (SCA 28), which is phenotypically similar to other progressive late-onset SCAs with dysarthria, eye movement abnormalities, and ataxia. AFG3L2 is a nuclear encoded mitochondrial protein that forms oligomeric m-AAA protease complexes, which play a major role in mitochondria ribosomal assembly and proteome quality control. AFG3L2 forms homo-oligomeric m-AAA complexes, it also forms hetero-oligomeric m-AAA complexes with paraplegin, the protein mutated in autosomal recessive spastic paraplegia, type 7 (SPG7). Paraplegin is unable to form homo-oligomers and requires AFG3L2 to function; lack of activity results in lower extremity spasticity. Our patients ' mutation, resulting in a missense Y616C substitution, reduces AFG3L2 enzymatic activity in yeast expression studies, without a dominant negative effect. This mutation also inhibits the functional interaction between AFG3L2 and paraplegin in yeast expression studies. These results indicate the brothers ' complex phenotype is likely the combination of abnormal activity of AFG3L2 in cerebellar cells and paraplegin in motor neurons. In summary, WES was used to identify novel homozygous mutations resulting in a new disorder primarily consisting of a combination of the SCA28/SPG7 phenotypes in association with other mitochondrial signs and symptoms.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2074
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content
Clinical phenotype in a mother and her son with a novel mutation for Fabry disease. *S. Yang, K. O'Brien, C. Tifft* NHGRI, National Institute of Health, Bethesda, MD.

Fabry Disease (FD) is an X-linked lysosomal storage disease caused by mutations of the alpha-galactosidase A (GLA) gene resulting in a deficiency of the alpha-galactosidase A (alpha-GAL-A) enzyme. Male hemizygotes with classical Fabry disease often show the complete spectrum of symptoms beginning in mid childhood while female heterozygotes demonstrate a variable phenotype ranging from that seen in male patients to asymptomatic. We describe the case of a 45-year-old African American female with 12 year history of hypertension who developed rapid elevation in serum creatinine with insignificant proteinuria leading to renal biopsy for a diagnosis. The biopsy showed distended cytoplasm, myelinoid fibures, and zebra bodies in the podocytes consist with Fabry disease. At the time of her initial genetic evaluation, she denied any clinical symptoms of Fabry disease and there were no findings on physical exam to support the diagnosis. The family history was likewise negative. However, the molecular analysis of the GLA gene revealed a novel missense mutation in exon 6 (c806 T>A; p.Val269Glu) of unknown clinical significance. A detailed workup at the NIH Clinical Center was initiated which revealed multi-organ system involvement including left ventricular hypertrophy, increased Gb3 (globtriaosylceramide) excretion, whorl-like corneal opacities (cornea verticillata), hypohidrosis, and possible mild acroparesthesias (peripheral neuropathy). The patient 's 23-year-old asymptomatic son was also tested and found to carry the V269E mutation. His AGA activity of 5.6 pmol/punch/hr was well within the range for patients with Fabry disease (2.0-12.0). Our findings conclude that the V269E mutation may be associated with a milder clinical course; however, close clinical monitoring remains important. Enzyme replacement therapy (ERT) has been discussed and offered to both patients.

- **POSTER SESSION: METABOLIC DISORDERS**
  Program Number 2106
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content
Creation of a Mouse Model for Lowe Syndrome and Dent Disease 2 by Humanizing a Paralogous Modifier Gene. *R. Nussbaum[1,2], E. Chan[1], I. Bernardini[3], Y.-M. Kuo[1], W. Gahl[3], S. Bothwell[1]* 1) Medicine, UCSF, San Francisco, CA; 2) Institute for Human Genetics, UCSF, San Francisco, CA; 3) Medical Genetics Branch, NHGRI/NIH, Bethesda, MD.

The Lowe OculoCerebroRenal syndrome (OCRL) is a pleiotropic X-linked human disorder characterized by congenital cataracts, cognitive disability, and proximal renal tubular dysfunction, particularly low molecular proteinuria and often aminoaciduria, phosphaturia and bicarbonaturia. OCRL is caused by loss-of-function mutations in the *OCRL* gene encoding Ocrl, a type II phosphatidylinositol bisphosphate 5-phosphatase. Mutations in *OCRL* can also cause a proximal tubular disorder known as Dent Disease type 2, in which the disease is limited to the proximal renal tubules. A first attempt to create a mouse model for OCRL/Dent 2 failed when we found that *Ocrl*- mice are unaffected. We reasoned that the disparate phenotype between humans and mice with loss-of-function mutations in *Ocrl*/*OCRL* resulted

from differences in how the two organisms cope with loss of the enzyme rather than in differences between the two species in the function of the enzyme itself. We hypothesized that *Inpp5b* and *INPP5B*, which also encode a type II phosphoinositide 5-phosphatase in mice and humans, respectively, might underlie the disparate phenotype in the two species because (1) they are the closest paralogs to *Ocrl* and *OCRL* in the respective genomes of mice and humans, (2) *Inpp5b* has overlapping function in vivo with *Ocrl*, and (3) the two species differ in a number of important ways in how *INPP5B* and *Inpp5b* are expressed. We used a bacterial artificial chromosome containing *INPP5B* to create transgenic *Ocrl*-;*Inpp5b*-/-mice expressing *INPP5B*. All showed reduced post-natal growth, low molecular weight proteinuria, and aminoaciduria when hemizygous for the *INPP5B* insertion but not when homozygous for the insertion. Ophthalmological abnormalities were not present. We have created the first animal model for OCRL/Dent Disease 2 by humanizing a modifier paralog in mice carrying the mutant disease gene.

▪ POSTER SESSION: METABOLIC DISORDERS

Program Number 2110
Exhibit Hall A, Lower Concourse Level, Convention Center
Thu 7:00AM-7:00PM
Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

RRM2B mtDNA Depletion Syndrome presenting with Pyruvate Dehydrogenase Deficiency. *L. A. Wolfe[1,2], J. J. McGrath[2], L. J. Wong[3], G. D. Vladutui[4]* 1) Undiagnosed Diseases Program, NHGRI, Bethesda, MD; 2) Department of Genetics, Yale School of Medicine, New Haven, CT; 3) Baylor College of Medicine, Houston, TX; 4) Robert Guthrie Biochemical & Molecular Genetics Laboratory, Kaleida Health Laboratories & the University, Buffalo, NY.

Purpose: Describe clinical and biochemical features of a male infant presenting with failure to thrive and severe diarrhea in the neonatal period. Case review: A 2-week-old male born to consanguineous Hispanic parents after an uneventful pregnancy and delivery, presented at 2 weeks of age with failure to thrive and severe diarrhea was admitted to the hospital for further evaluation. Laboratory evaluation revealed normal acylcarnitine profile, urine organic acids, and CK. Plasma lactic acid (15 mmol/L, normal <2.2), and pyruvate (0.48 mmol/L, normal 0.03-0.10) were elevated with a mild increase of alanine (435.4 uM, normal 142-421). Due to a high L: P ratio (33), a mitochondrial electron transport chain (ETC) disorder was suspected. Common mtDNA point mutations and deletions were negative. Sequencing analysis of 5 genes (SUCLG1, DGUOK, POLG1, SUCLA2, TK2) responsible for mtDNA depletion, complex IV assembly genes (SURF1, SCO1, SCO2, COX 10), SDH subunits A-D, and complex I assembly genes (C6ORF66, NDUFA1) were all negative. Methods used: prospective evaluation and clinical biochemical testing. A novel hemizygous unclassified missense variant, c.677G>A (p. R226H) in the PDHA1 gene was detected. On muscle biopsy, abnormal mitochondria with proliferation were noted. Profound deficiencies in complexes IV and II-III were detected with reduced complexes I and I-III activities. Meanwhile, deficiency in pyruvate dehydrogenase complex (PDC) activity was detected in blood lymphocytes. The patient was started on carnitine and ubiquinol, which improved his diarrhea, and he was discharged home. Subsequently, the patient 's clinical condition continued to decline and he became ventilator dependent. MtDNA depletion syndrome was suspected and sequence analysis of the RRM2B gene, encoding a newly discovered p53-inducible ribonucleoside reductase subunit was performed. A homozygous c.109_110delAA (p. K37EfsX14) mutation was revealed. Results: Although an ETC deficiency may be secondary to the PDC defect, it is not known if mtDNA depletion would cause PDC deficiency. Thus, the clinical significance of p. R226H in PDHA1 remains unclear. This case underscores the complications in diagnosing mitochondrial ETC disorders.

▪ POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS

Program Number 2356
Exhibit Hall A, Lower Concourse Level, Convention Center
Thu 7:00AM-7:00PM
Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

Molecular & clinical analysis of the *retinoic acid induced 1* gene (*RAI1*) in patients with suspected Smith-Magenis Syndrome without the 17p11.2 deletion. *A. C. M. Smith[1], T. Vilboux[2], C. Ciccone-Stevens[2], J. Blancato[3], G. Cox[4,5], W. Introne[1], W. A. Gahl[1,2], M. Huizing[2]* 1) Office Clinical Dir, NHGRI/NIH, Bethesda, MD; 2) Medical Genetics Branch, NHGRI/NIH, Bethesda, MD; 3) Dept. of Oncology, Georgetown University Medical Center, Washington, DC; 4) Div. of Genetics, Children 's Hospital Boston and Dept Pediatrics, Harvard Medical School, Boston, MA; 5) Genzyme Corporation, Cambridge, MA.

Smith-Magenis syndrome (SMS) is a complex developmental disorder characterized by multiple congenital anomalies. The syndrome is primarily ascribed to a ~3.7 Mb de novo deletion on chromosome 17p11.2. Haploinsufficiency of multiple genes likely underlies the complex clinical phenotype. *RAI1 (Retinoic Acid Induced 1)* is recognized as a major gene involved in the SMS phenotype. Extensive genetic and clinical analyses of 36 patients with SMS-like features, but without the 17p11.2 microdeletion, yielded 5 patients with *de novo* and 5 patients with novel *familial RAI1* variants. Haplotype analysis showed two major *RAI1* haplotypes in our primarily Caucasian cohort; the novel *RAI1* variants did not occur in a preferred haplotype. RNA analysis revealed for the first time that *RAI1* mRNA expression was significantly

decreased in cells of patients with the common 17p11.2 deletion, as well as in those with *de novo RAI1* variants. Expression levels varied in patients with familial *RAI1* variants and in non-17p11.2 deleted patients without identified *RAI1* defects. No correlation between SNP haplotype and *RAI1* expression was found. Two clinical features, ocular abnormalities and polyembolokoilomania (object insertion), were significantly correlated with decreased *RAI1* expression. While not significantly correlated, the presence of hearing loss, seizures, hoarse voice, childhood onset of obesity and specific behavioral aspects and the absence of immunologic abnormalities and cardiovascular or renal structural anomalies, appeared to be specific for the *de novo RAI1* subgroup. Recognition of the combination of these features may assist in referral for *RAI1* analysis of patients with SMS-like features without detectable microdeletion of 17p11.2.

- **POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY**
  Program Number 2866
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Thu 7:00AM-7:00PM
  Presentation Time: Thu, Nov 4, 2010, 6:00PM-7:00PM

Abstract Content

The PhenX Toolkit: Facilitating the use of common measures in genomics research. *H. Pan[1], D. Jackman[1], V. Bakalov[1], K. Chang[1], A. Flynn[1], W. Huggins[1], J. Levy[1], D. Nettles[1], Y. Qin[1], H. Ray[1], P. Schad[1], N. Whitehead[1], M. Zmuda[1], H. Junkins[2], E. Ramos[2], L. Strader[1], C. Hamilton[1]* 1) Research Computing Division, RTI International, Research Triangle Park, NC; 2) National Human Genome Research Institute, Bethesda, MD.

To facilitate cross-study comparisons, PhenX (consensus measures for Phenotypes and eXposures) created a Toolkit of common measures for researchers to use when designing genomics-based studies. The PhenX Toolkit provides the user with a web-based interface for searching, browsing and selecting PhenX Measures and protocols. For each PhenX Measure, the Toolkit provides the user with a brief description of the measure, the rationale for selecting the measure, protocol(s) for collecting the measure, and supporting documentation. The Toolkit contains over 200 measures (15 research domains). Measures for the six remaining domains will be included in the Toolkit by the end of 2010. To expand its utility, the PhenX Toolkit has extended its browse and search capabilities and its collaborative efforts. The “Smart Query Tool ” provides the option to use either a high-specificity search through measure and protocol names, synonyms and keywords or a full-text search. The “Data Collection Worksheet ” will enable Toolkit users to integrate PhenX measures into their studies more easily while the Data Dictionary provides users with variable names, identifiers and many attributes in several formats. Mapping PhenX measures and variables to related research efforts will facilitate data interoperability, thus helping Toolkit users combine and/or harmonize data. To demonstrate its utility, PhenX has mapped PhenX measures, protocols and/or variables to studies in dbGaP, the Public Population Project in Genomics ’s (P3G) Data Schema and Harmonization Platform for Epidemiological Research (DataSHaPER), and electronic Medical Records and Genomics (eMERGE), and the results of mapping will be presented. PhenX measures are accessible using the caBIG CDE browser. Logical Observation Identifiers Names and Codes (LOINC) is developing LOINC codes for PhenX measures. These codes will make enable identification of PhenX variables in electronic medical records (EMRs), clinical data repositories and other resources, thus facilitating cross-study analysis. The PhenX Toolkit provides the research community with freely available, well-established, low-burden, high quality measures, and the bioinformatics support to use them effectively. Broad acceptance and use of PhenX Measures can facilitate identification of genes associated with common diseases, as well as gene-gene and gene-environment interactions. Supported by: NHGRI, Award No. U01 HG004597.

PLATFORM PRESENTATIONS

- **SESSION TITLE: 45. MECHANISMS AND TREATMENT OF METABOLIC DISEASE**
  Program Number 213
  Room 202, Level 2, Convention Center
  Fri Nov 5, 2010 08:00AM-10:30AM
  Presentation Time: 09:30AM-09:45AM

Abstract Content

Metabolic sink therapy in methylmalonic acidemia using a novel muscle-specific transgenic mouse model. *J. R. Sysol[1], I. Manoli[1], L. Li[2], J. Senac[1], R. J. Chandler[1], V. Hoffmann[3], P. Zerfas[3], J. Schnermann[2], C. P. Venditti[1]* 1) Genetics and Molecular Biology Branch, NHGRI, NIH, Bethesda, MD; 2) Kidney Disease Branch, NIDDK, NIH, Bethesda, MD; 3) Division of Veterinary Resources, ORS, NIH, Bethesda, MD.

Methylmalonic acidemia (MMA) is caused by deficiency of the mitochondrial enzyme methylmalonyl-CoA mutase (Mut) and results in massive elevations of methylmalonic acid in tissues and body fluids. Studies in knockout mice (*Mut*$^{-/-}$) and transplanted MMA patients have suggested that a large portion of circulating methylmalonic acid derives from extrahepatic organs, mainly the skeletal muscle. To examine the effects of restoring skeletal muscle expression of the Mut enzyme on the *Mut*$^{-/-}$ phenotype and gain insight into the efficacy of targeting skeletal muscle for metabolic “sink ” therapy for MMA, we generated mice that express the *Mut* gene under the control of an insulated, muscle-specific promoter (*Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$). *Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$ mice were born in Mendelian proportions, showed greater than 83% survival past day of life 60 (N=40), and achieved 40-50% of their heterozygous littermates weight through the first year of life. Muscle-specific *Mut* RNA expression in *Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$ mice was 103 ±4.6% compared to *Mut*$^{+/-}$ and was accompanied by abundant immunoreactive enzyme in muscle. To further assess transgene function, we measured the oxidation of 1-^{13}C propionate into $^{13}CO_2$. The *Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$ mice metabolized 18.4 ±3.6% of the label in 25 min, compared to 76.5 ±4.5% in *Mut*$^{+/-}$ and 10 ±2% in *Mut*$^{-/-}$. Baseline plasma MMA levels (μM) were 1107.9 ±66 in transgenic mice, compared to <5 in controls. The *Mut*$^{-/-}$;Tg$^{\text{INS-MCK-}Mut}$ animals develop significant liver pathology, characterized by giant eosinophilic vacuoles and megamitochondria formation, which was associated with decreased respiratory chain complex IV activity (18.2 ±7.4% relative to controls), similar to the *Mut*$^{-/-}$ mice. More variable changes were noted in the tubular epithelial cells and were associated with a decreased glomerular filtration rate as measured by inulin clearance. Selective muscle expression of the Mut enzyme by transgenesis at levels matching or exceeding the heterozygous controls resulted in near uniform rescue of the neonatal lethal phenotype of the *Mut*$^{-/-}$ mice, but was unable to prevent liver and kidney damage. This novel murine model demonstrates that enzymatic correction of skeletal muscle can augment metabolism in MMA, but cell-autonomous and/or toxic effects from circulating metabolites may mediate hepatic and renal tubular pathology. It also provides a new platform for testing of liver and/or kidney-directed gene, cell and other therapies for this disease.

- **SESSION TITLE: 55. INFLUENCE OF POLYMORPHISMS ON DISEASE RISKS AND TRAITS**
  Room 145, Level 1, Convention Center
  Fri Nov 5, 2010 01:30PM-04:00PM
  Presentation Time: Fri, Nov 5, 2010, 03:00PM-03:15PM

Abstract Content

Solute Carrier 2 (SLC2A9) on Chromosome 4 is associated with Uric Acid in African Americans. *B. Charles, D. Shriner, A. Doumatey, J. Zhou, A. Adeyemo, C. Rotimi* CRGGH, NHGRI/NIH, Bethesda, MD.

Purpose: Uric acid is the primary byproduct of purine metabolism. Hyperuricemia is associated with body mass index (BMI), sex, hypertension (HTN), renal disease, and other complex diseases, including the metabolic syndrome and type 2 diabetes (T2D). Multiple genome-wide association studies (GWAS) conducted in individuals of European ancestry (EA) have reported associations between uric acid and specific genomic loci. The purpose of this study was to identify novel susceptibility loci for serum uric acid levels in African Americans and to replicate previous GWAS finding for uric acid in European ancestry populations. Methods: A total of 1,017 African Americans who participated in the Howard University Family Study conducted in Washington, DC where included in this study. Genotyping was conducted using Affymetrix ® Genome-wide Human SNP Array 6.0 with genotyping calls determined by Birdseed, v2. Imputation was conducted using MACH and the HapMap reference panels for CEU and YRI. A total of 2,400,542 SNPs were assessed for association with uric acid using PLINK under the additive model with adjustment for age, sex, BMI, glomerular filtration rate, HTN, T2D status, and the 2 principal components identified in the assessment of population stratification. Results: Three variants in the gene SLC2A9 achieved genome-wide significance for association with serum uric acid (p-values ranging from 3.66x10-9 to 1.79x10-10). Conclusions: The most strongly associated locus for serum uric acid levels in individuals of European ancestry was also the most strongly associated locus in this African American sample.

This finding provides significant evidence for the potential role of SCL2A9 in uric acid homeostasis across human populations.

- **SESSION TITLE: 60. NEW FINDINGS IN KNOWN CLINICAL DISORDERS**
  Program Number 364
  Room 146, Level 1, Convention Center
  Fri Nov 5, 2010 04:30PM-07:00PM
  Presentation Time: Fri, Nov 5, 2010, 06:15PM-06:30PM

Abstract Content

Evaluation of alpha-synuclein aggregation in brain samples from patients carrying *GBA* mutations. *J. Choi[1], M. Cookson[2], G. Lopez[1], E. Goldin[1], O. Goker-Alpan[1], B. Stubblefield[1], E. Sidransky[1]* 1) Section on Molecular Neurogenetics, Medical Genetics Branch, NHGRI, National Institutes of Health, Bethesda, MD; 2) Cell Biology and Gene Expression Unit, Laboratory of Neurogenetics, NIA, National Institutes of Health, Bethesda, MD.

Recent findings demonstrate an increased frequency of mutations in glucocerebrosidase (*GBA*), the enzyme deficient in the lysosomal storage disorder, Gaucher disease (GD), among patients with synucleinopathies. Neuropathologic findings in some patients who developed both GD and parkinsonism revealed Lewy bodies and synuclein-positive inclusions in vulnerable brain regions. The association of *GBA* mutations and alpha-synuclein aggregation was examined by evaluating pathologic specimen. In this study, proteins were extracted from cerebral cortex from subjects carrying *GBA* mutations with and without a clinical history of parkinsonism. These include samples from patients with Lewy body dementia, Lewy body variant Alzheimer 's disease, Parkinson disease and patients with different types of Gaucher disease. A total of 26 brain tissue samples were analyzed. 11 were from patients without Lewy body disorders (eight had *GBA* mutations and three were controls), and 15 had Lewy body disorders (nine carried *GBA* mutations). The samples were homogenized and fractionated into TBS-soluble, SDS-soluble and urea-soluble fractions. Most patients with synucleinopathies were shown to exhibit oligomeric forms of alpha-synuclein in the insoluble fraction, including the subjects with *GBA* mutations. However, patients with Gaucher disease and no clinical evidence of parkinsonism did not display oligomeric forms. The amount of the oligomeric alpha-synuclein correlated best with the degree of Lewy body pathology. These studies indicate that patients with syncleinopathies carrying *GBA* mutations show biochemical characteristics typical of Lewy body disorders, but these changes are not always seen with glucocerebrosidase deficiency.

- **SESSION TITLE: 63. ETHICAL, LEGAL, EDUCATION AND POLICY ISSUES**
  Program Number 393
  Room 145, Level 1, Convention Center
  Fri Nov 5, 2010 04:30PM-07:00PM
  Presentation Time: Fri, Nov 5, 2010, 06:00PM-06:15PM

Abstract Content

Reactions of Smokers to an Information Pamphlet About Genetic Testing for a Common Lung Cancer-Associated Gene Variant. *S. C. Sanderson1, J. Shepperd2, C. M. McBride3, S. Docherty4, S. C. O'Neill5, I. M. Lipkus4* 1) Department of Genetics and Genomic Sciences, Mount Sinai School of Medicine, New York, NY; 2) Department of Psychology, University of Florida, Gainsville, FL; 3) Social and Behavioral Branch, National Human Genome Research Institute, NIH, Bethesda, MD; 4) School of Nursing, Duke University, Durham, NC; 5) Cancer Control Program, Georgetown University, Washington, DC.

Most common gene variants are only weakly associated with complex diseases such as lung cancer. Little is known about how smokers respond to learning about genetic testing for such variants. Our aim was to assess smokers ' reactions to an information pamphlet describing genetic testing for a common lung cancer-associated gene variant (GSTM1-null) . We assessed whether smokers (a) understood that the gene variant slightly influenced lung cancer risk, (b) felt the information to be useful in decision-making about genetic testing for lung cancer risk, and (c) were interested in being tested for the variant. Participants were 131 students who smoked cigarettes. They read and than answered questions about a 20-page information pamphlet, "Genetic Testing for Lung Cancer Risk ", which included information that people missing the GSTM1 gene (GSTM1-null) may have a 20% higher lung cancer risk than those who are GSTM1-present, and that lifetime risks are estimated to be 11% and 9% respectively. Most (78%) interpreted the GSTM1-present result to indicate that lung cancer risk is "slightly lower than average ", whilst 18% interpreted it as meaning "average " risk, and 2% as "much lower than average ". Similarly, most (89%) interpreted the GSTM1-null result as "slightly higher than average "; 3% interpreted it as "average " risk, 6% as "much higher than average ". When asked whether the information would help people make a decision about genetic testing for lung cancer risk, 68% said "yes ", 29% "somewhat ", 3% "no ". When asked to rate how understandable the information pamphlet was overall, the mean score was 6.60+0.71 (where 1=not at all to 7=completely understandable). The mean scores for importance and interest in getting tested for GSTM1 were 5.79+1.17 (where 1=unimportant to 7=important) and 5.21+1.66 (where 1=not at all to 7=extremely interested). The results suggest that smokers may perceive genetic information as important even when they understand a gene variant is only slightly related to disease risk, and express interest in genetic testing using a marker not highly predictive of disease. Further research is needed on how best to convey information to the public

about small effects on risk conferred by common genetic variation, and to explore why people are interested in receiving personal genetic information about genetic variants of low penetrance.

POSTERS

- **POSTER SESSION: . COMPLEX TRAITS: THEORY AND METHODS**
  Program Number 1127
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

PhenX Measures: Enhancing Research Studies of Gene-Environment Interactions. *W. Huggins[1], L. C. Strader[1], D. C. Whitcomb[2], B. Entwisle[3], B. Pescosolido[4], L. Goldman[5], J. A. Hammond[1], T. Hendershot[1], R. K. Kwok[1], H. Junkins[6], E. Ramos[6]* 1) RTI International, Research Triangle Park, NC; 2) Gastroenterology, University of Pittsburgh School of Medicine, Pittsburgh, PA; 3) University of North Carolina at Chapel Hill Carolina Population Center, Chapel Hill, NC; 4) Indiana University at Bloomington, Bloomington, IN; 5) Johns Hopkins University Bloomberg School of Public Health, Baltimore, MD; 6) National Human Genome Research Institute, Bethesda, MD.

Few Genome-Wide Association Studies (GWAS) have included robust environmental exposure measures even though gene-environment interactions modulate the severity and presentation of virtually all human disease. The PhenX (consensus measures for Phenotypes and eXposures) Toolkit offers high quality, well-established, standard measures of phenotypes and exposures from 21 domains for use in GWAS and other large-scale genomic research efforts. For each research domain, a Working Group (WG) comprised of academic and clinical researchers from diverse institutions and disciplines is assembled to prioritize a set of low burden, well established measures. Here, we demonstrate a possible scenario to examine gene-environment interactions by highlighting measures from four PhenX Domains: Gastrointestinal, Environmental Exposures, Social Environments, and Psychosocial. The Gastrointestinal WG identified a range of disorders that can be influenced by an individual 's environment including abdominal pain, recurrent constipation, and gastrointestinal cancers. These modulating factors can be assessed by other standard measures in the Toolkit. For example, the Environmental Exposures measures include water source, occupation history, and contact with common chemicals and solvents. The Social Environments WG identified a range of relevant exposures that address aspects of work, family, and neighborhood/community. The Psychosocial WG selected measures related to stress, coping, wellbeing, and social connectedness. Researchers can visit the web-based PhenX Toolkit (https://www.phenxtoolkit.org/) to review and select measures. For each measure, the Toolkit provides a description, selection rationale, protocol(s) for collecting the measure, and references. Users can browse, select and download the protocols for measures that are relevant for their research. The Toolkit provides a common currency for investigators who wish to add measures that are outside their primary research focus or to extend their research through the inclusion of salient PhenX measures to go beyond to explore the physical and social environments to determine the etiology of complex diseases. As investigators incorporate PhenX measures into their studies, there will be new opportunities for cross-study analyses to identify loci with small effect sizes and to discover gene-gene and gene-environment associations. Supported by: NHGRI, Award No. 1U01 HG004597-01.

- **POSTER SESSION: COMPLEX TRAITS: THEORY AND METHODS**
  Program Number 1187
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

Metabolic effects of common and rare variants in the Glucose Kinase Regulatory Protein. *D. Ng[1], M. G. Rees[2], S. L. Ruppert[1], J. C. Mullikin[3], M. C. Skarulis[4], L. G. Biesecker[1], NISC Comparative Sequencing Program* 1) Genetic Disease Research Branch, NHGRI, NIH, Bethesda, MD; 2) National Center for Human Genome Research, NHGRI, NIH, Bethesda, MD; 3) Genome Technology Branch, NHGRI, NIH, Bethesda, MD; 4) Clinical Endocrine Section, NIDDK, NIH, Bethesda, MD.

ClinSeq ™ is a large-scale medical sequencing (LSMS) project at the National Institutes of Health (NIH). The goal of ClinSeq ™ is to study the technical, medical, and genetic counseling issues associated with LSMS and its application to personalized medicine. The research aims are two-fold: 1) identify clinically relevant gene variants that impact on health and return these results to study participants to guide medical management; 2) generate hypothesis-driven clinical and molecular research to discover new genetic variants that contribute to disease susceptibility. Glucose Kinase Regulatory Protein (GCKR) was selected as a candidate gene for study in ClinSeq ™ because a common coding SNP rs1260326 (p.Pro446Leu) was found to be associated with hypertriglyceridemia in a replication study of quantitative trait loci in type II diabetes mellitus. Rodent studies showed that GCKR regulates glucose kinase (GCK) activity in the liver. GCKR sequesters and stabilizes GCK in the hepatic nucleus in the fasting state. GCKR inhibition of GCK is enhanced by

fructose 6-phosphate and reduced by fructose 1-phosphate. We hypothesized that GCKR variants with reduction or loss of GCK inhibition would result in abnormal cellular localization of GCK, altered glucose metabolism, and development of hepatic steatosis. To test this hypothesis, we analyzed over 700 ClinSeq ™ participants with Sanger Sequencing to identify GCKR variants. Seventeen rare (n=42 individuals) and one common ((rs1260326), n=355 heterozygotes, n=141 homozygotes) GCKR coding variants were identified. Cellular effects of these genetic variants were studied by co-transfecting CFP-tagged mutant GCKR and YFP-tagged wild-type GCK constructs into Hela cells. Microscopy revealed a spectrum of GCK cellular localization ranging from entirely cytoplasmic (p.Val103Met) indicating a total loss of GCKR inhibition, to preservation of GCK nuclear to cytoplasmic shuttling (p.Arg540Gln). Select ClinSeq ™participants with GCKR variants were invited to the NIH for metabolic phenotyping. Oral glucose tolerance tests with and without fructose showed a spectrum of glucose metabolism ranging from accelerated to delayed glucose clearance. Preliminary data from hepatic MRI scans showed an increased triglyceride content in 3 out of 4 phenotyped individuals. Studies are ongoing to compare individuals who carry the same variant with each other and with wild-type controls to determine if there is a genotype-phenotype correlation.

- **POSTER SESSION: . MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2285
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

Novel mutations in the HPS1 gene among Puerto Rican patients. *C. Carmona-Rivera[1], R. A. Hess[1], K. O'Brien[2], G. Golas[2], E. Tsilou[3], J. G. White[4], W. A. Gahl[1,2], M. Huizing[1]* 1) Med Gen Branch, NIH/NHGRI, Bethesda, MD; 2) Office of Rare diseases,NIH/NHGRI, Bethesda, MD; 3) Oftal Gen Vis Func Branch, NIH/NEI, Bethsda, MD; 4) Dept of Laboratory Medicine, U. Minnesota, Minneapolis, USA.

Hermansky-Pudlak Syndrome is a disorder of oculocutaneous albinism and platelet storage pool deficiency. Eight different disease causing genes have been identified, whose gene products are thought to be involved in the biogenesis of lysosome-related organelles. HPS type 1 (HPS-1) is the most common HPS subtype in Puerto Rico, with a frequency of 1:1,800 in the northwest of the island due to a founder mutation, i.e., a 16-base pair duplication in exon 15 of the HPS1 gene (c.1472_1487dup16; p.H497QfsX90). We identified three Puerto Rican HPS-1 patients who carried compound heterozygous HPS1 mutations. One patient was heterozygous for c.937G>A, causing a missense mutation (p.G313S) at the 3 ' splice junction of exon 10. This mutation resulted in activation of a cryptic intronic splice site causing an aberrantly spliced HPS1 mRNA that included 144-bp of intronic sequence, producing 11 novel amino acids followed by a stop codon. The other two patients were heterozygous for the previously reported c.972delC in HPS1, resulting in a frameshift and a premature stop codon (p.M325WfsX6). These findings indicate that, among Puerto Ricans, other HPS1 mutations apart from the 16-base pair duplication should be considered in the analysis of this population.

- **POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2299
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

Identification of recombinant alleles using Quantitative Real-time PCR: Implication for Gaucher disease. *A. Velayati, M. A. Knight, B. K. Stubblefield, E. Sidransky, N. Tayebi* Section on Molecular Neurogenetics, Medical Genetics Branch/ NHGRI, NIH, Bethesda, MD.

Background: Gaucher disease is an autosomal recessive disorder caused by the deficiency of glucocerebrosidase. The glucocerebrosidase gene (GBA) is located in a very gene-rich region on chromosome 1q 21. The presence of contiguous, highly homologous pseudogenes for both GBA and metaxin 1 at this locus increases the likelihood of DNA rearrangement. We describe an easy method to identify and analyze recombinant alleles in patients with Gaucher disease. Methods: Genomic DNA from twenty patients with Gaucher disease known to carry recombinant GBA alleles and five controls were studied. Six different probes for either the GBA gene or pseudogene were designed to identify DNA rearrangements, as well as copy number variation within the GBA locus. Quantitative real-time PCR using TaqMan probes was performed on genomic DNA, and β-globin was co-amplified as an internal control. Southern blot analyses using the restriction enzyme HincII and direct sequencing were performed to confirm the real-time results. Results: GBA fusions and duplications could be detected in all the cases, corresponding to the Southern blot results. Different sites of recombination could also be distinguished. Conclusion: Quantitative real-time PCR is a sensitive and rapid method to detect fusions and duplications in patients with recombinant GBA alleles. Since this technique is faster and cheaper than Southern blotting, it can be applied as a suitable method in diagnostic laboratories. Keywords: Gaucher disease, glucocerebrosidase, recombinant alleles

- **POSTER SESSION: PSYCHIATRIC GENETICS, NEUROGENETICS AND NEURODEGENERATION**
  Program Number 2713
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

Clinical non-motor manifestations in Parkinson disease patients carrying GBA mutations. *G. Lopez[1], J. Choi[1], B. McElroy[3], C. Crews[2], M. Brooks[3], N. Gupta[1], A. Velayati[1], A. Britton[2], M. Hallett[3], E. Sidransky[1]* 1) NHGRI/NIH, Bethesda, MD; 2) NIA/NIH, Bethesda, MD; 3) NINDS/NIH, Bethesda, MD.

The association between parkinsonism and mutations in the glucocerebrosidase gene (GBA) is now well established. Patients with parkinsonism have a five-fold increased likelihood of carrying a mutation in GBA . Non-motor symptoms described in Parkinson disease include depression, sleep disorders, fatigue, olfactory problems, and cognitive difficulties. Non-motor symptoms are considered non-dopaminergic and therefore, refractory to dopaminergic supplementation. Non-motor manifestations often precede motor symptoms, and tend to intensify with disease progression, dominating the clinical picture in late-stages of the disease. Systematic evaluation of non-motor manifestations in subjects with Gaucher disease and GBA mutation carriers has not been extensively studied. We performed a uniform cross-sectional clinical study of 155 consecutive patients evaluated at the Parkinson disease clinic at the National Institutes of Health by a single neurologist. All 11 exons of GBA were sequenced and subjects were screened for the three most common LRRK2 mutations (G2019S, R1441H/C, and Y1699C). Eight patients were found to be heterozygous for a GBA mutation. Clinical evaluations included a detailed history and physical examination, neurological and UPDRS evaluation, detailed review of systems interview, Geriatric Depression Scale, Fatigue Scale, Epworth Sleep Scale, and smell evaluation using the University of Pennsylvania Smell Identification Test. Our study showed that patients with Parkinson disease who carried a GBA mutation were more likely to complain of memory problems when compared to Parkinson disease patients without mutations, although objective measurements with clinical cognitive screening tools did not demonstrate a cognitive deficit. While the small sample size limits generalization of this finding, more formal neurocognitive assessments are warranted. Both patients with Gaucher disease and GBA carriers are being prospectively followed at the National Human Genome Research Institute in order to study motor and non-motor symptoms in patients at increased risk of parkinsonism.

- **POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY**
  Program Number 2963
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content

Quality Control Pipeline for Genome-Wide Association Studies in the eMERGE Network: Comparing Single Site QC to a Merged QC Approach. *M. Ritchie[1], L. Armstrong[2], Y. Bradford[1], C. Carlson[3,4], D. Crawford[1], A. Crenshaw[5], M. de Andrade[6], K. Doheny[7], J. Haines[1], G. Hayes[2], G. Jarvik[3,4], L. Jiang[1], H. Ling[7], I. Kullo[6], R. Li[8], T. Manolio[8], M. Matsumoto[6], C. McCarty[9], A. McDavid[3,4], D. Mirel[5], L. Olson[1], J. Paschall[10], E. Pugh[7], L. Rasmussen[9], R. Wilke[1], R. Zuvich[1], S. Turner[1]* 1) Molec Physiology & Biophysics, Vanderbilt Univ, Nashville, TN; 2) Northwestern University, Chicago, IL; 3) University of Washington, Seattle, WA; 4) Group Health Cooperative, Seattle, WA; 5) Broad Institute of MIT and Harvard, Cambridge, MA; 6) Mayo Clinic, Rochester, MN; 7) Center for Inherited Disease Research (CIDR), John Hopkins University, Baltimore, MD; 8) National Human Genome Research Institute (NHGRI), Bethesda, MD; 9) Marshfield Clinic, Marshfield, WI; 10) national Center for Biotechnology Information (NCBI).

Genome-wide association studies (GWAS) are being conducted at an unprecedented rate in disease-based cohorts and have increased our understanding of the pathophysiology of complex disease. Regardless of context, the practical utility of this information will ultimately depend upon the quality of the original data. Quality control (QC) procedures for GWAS are computationally intensive, operationally challenging, under constant evolution, and critically important. What has not yet been explored in detail are the challenges that emerge when multiple GWAS datasets, genotyped in different labs, are merged for downstream GWAS analysis; a scenario that is likely to increase in frequency with the advent of dbGaP. The genomics workgroup of the NHGRI funded electronic Medical Records and Genomics (eMERGE) network has spent a considerable amount of effort developing strategies for quality control of these data. eMERGE consists of five sites, each with DNA databanks linked to electronic health information. Approximately 17,000 samples have been genotyped (~15000 European Americans using Illumina 660W half performed at each (Broad and CIDR) and ~2000 African Americans using Illumina 1M performed at Broad), and phenotypes have been enumerated for ~20 diseases and traits. The lessons learned by this group of investigators will be valuable for the genomics community also dealing with the combining of large scale genomic datasets. We compare the characteristics of various quality control measures between each of the five eMERGE sites, and the merged dataset. Here we enumerate some of the challenges in QC of merged GWAS datasets, including population substructure, merging data from 2 genotyping centers, strand orientation, and other errors inherent from merging ~17000 samples GWAS data, and describe the approaches that the eMERGE network uses to guarantee quality assurance in GWAS data, thereby minimizing potential bias and error in GWAS

results. Finally, we describe the best practices that we have decided upon, such as having 2 sites duplicate efforts to eliminate errors early on in the process, and discuss areas of ongoing and future research.

- **POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY**
  Program Number 2971
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Replication of Genetic Variants associated with Serum Lipids in African Americans. *K. Meilleur, A. Adeyemo, A. Doumatey, D. Shriner, H. Huang, J. Zhou, E. Ramos, G. Chen, C. Rotimi* Center for Research on Genomics and Global Health, NHGRI, NIH, Bethesda, MD.

Statement of Purpose: Both candidate and genome-wide association studies (GWAS) have identified multiple genetic variants associated with serum lipid parameters including triglycerides, HDL and LDL-cholesterol. However, few of these studies have been conducted in African Americans (AA), who have been reported to display different lipid profiles compared to other US groups. We sought to identify novel susceptibility loci for dyslipidemia in AA and to assess whether observed differences in lipid parameters between AA and other ethnic groups could be explained by differences in genetic background. Methods: In the present study, we tested a set of genetic variants associated with lipid traits in GWAS in a sample of 927 African Americans from the Washington DC metropolitan region. Genotyping was conducted using Affymetric ® Genome-wide Human SNP Array 6.0 with genotyping calls determined by Birdseed, v2. Imputation was conducted using MACH and the HapMap reference panels for CEU and YRI. A total of 2,366,856 SNPs were assessed for association with lipid traits using PLINK under the additive model with adjustment for age, sex, BMI, and the first 2 principal components identified in the assessment of population stratification. Results: We identified a significant novel locus (rs1047163; p-value 6.4X10-8) associated with HDL on chromosome 2; this SNP is located in the 3 ' untranslated region (UTR) of HS1BP3 (hematopoietic-specific protein 1 binding protein 3 gene). An intergenic SNP (rs820042) also on chromosome 2 was identified to be associated with LDL. We replicated previous associations with SNPs in the following genes: LPL, APOB, [APOE, APOC1, APOC4, APOC2], PCSK9, [CELSR2, PSCRC1, SORT1], HMGCR, [NCAN, CILP2, PBX4], CETP, DOCK7, B3GALT4, TRIB1, and TOMM40. Conclusion: Overall, we identified novel susceptibility loci for HDL and LDL cholesterol and replicated several reported GWAS findings in this cohort of African Americans.

- **POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY**
  Program Number 2981
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 10:30AM-11:30AM

Abstract Content
Ordered Subset Analyses of Prostate Cancer Susceptibility in Finland Provides Evidence for Replication of *HPCX1* and *HPC10* Loci and Suggestive Evidence for Several Novel Loci. *C. D. Cropp[1], C. S. Simpson[1], T. Wahlfors[2], A. George[1, 3], H. Nati[2], T. Tammela[4], J. Schleutker[2], J. E. Bailey-Wilson[1]* 1) Statistical Gen Branch, IDRB/NHGRI/NIH, Baltimore, MD; 2) Institute of Medical Technology, University of Tampere and Tampere University Hospital, Tampere, Finland; 3) Fox Chase Cancer Center, Philadelphia, Pennsylvania; 4) Department of Urology, Tampere University Hospital, University of Tampere, Tampere, Finland.

Prostate cancer is the most common non-cutaneous cancer in men in America and European industrialized countries. It is a complex disorder, with a strong genetic component. Although better treatments and earlier detection have contributed to decreasing mortality rates for many countries, mortality rates are actually increasing in Asian countries such as Japan and Singapore. We recently reported a genome-wide linkage scan in 69 Finnish Hereditary Prostate Cancer (HPC) families, which replicated the *HPC9* locus on 17q21-q22 and identified a locus on 2q37. Using ordered subset analysis (OSA) and conditioning on non-parametric linkage to these loci, we sought to find other loci linked to HPC in subsets of families not detectable in the overall sample. Significance of the change in LOD scores (ΔLOD) due to OSA analysis was determined using permutation tests (empirical p-value). These analyses revealed a significant linkage peak with an OSA LOD score of 4.876 on Xq26.3-q27 (ΔLOD = 3.193, empirical p = 0.009)in a subset of 41 families weakly linked to 2q37. This region overlaps the *HPCX1* locus. Other linked loci were 12q21.1-q23.3 (OSA LOD = 3.67, ΔLOD = 2.526, p = 0.04) in a subset of 17 families unlinked to 2q37, and 8q24.22-q24.3 (OSA LOD = 3.195, ΔLOD = 2.963, p = 0.02) in a subset of 15 families weakly linked to 2q37, overlapping the *HPC10* locus. Another subset of 41 families most strongly linked to 17q21-q22 revealed a significant linkage to Xq25 with a peak OSA LOD score of 3.542 (ΔLOD = 1.484, p = 0.04). This subset contains many of the same families found in the subset linked to Xq26.3-q27 when conditioning on linkage to 2q37. Thus it is likely that these signals represent the same locus. Other strongly linked loci were found at 3q26.31-q27.1 (OSA LOD = 3.492, ΔLOD = 2.39, p = 0.02) in a subset of 47 families unlinked to 17, and 12q14.2-q21.31 (OSA LOD = 3.23, ΔLOD = 2.326, p = 0.02) in a subset of 34 families unlinked to 17. We also used the maximum of the family NPL scores for 2q37 and 17q21-q22 to condition on linkage to either of these loci. Two novel loci were found in this analysis; 18q12.1-q12.2 (OSA LOD = 2.541, ΔLOD = 1.651, p = 0.03) and 22q11.1-

q11.21 (OSA LOD = 2.395, ΔLOD = 2.36, p = 0.006), which is close to *HPC6*. Using ordered subset analysis allows us to find additional loci linked to HPC in subsets of families, which would not otherwise be detectable, thus contributing to the effort to untangle the complex genetic heterogeneity of HPC.

- **POSTER SESSION: CANCER GENETICS**
  Program Number 550
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Identification of loci associating with histiocytic-dendritic cell neoplasms using a canine genome wide study. *A. L. Shearin[1], H. G. Parker[1], E. Cadieu[1,2], B. Hedan[2], M. Breen[3], J. Cullen[4], A. Grone[5], G. Rutteman[5], C. Andre[2], E. A. Ostrander[1], E. V. Schmidt[1,6,7]* 1) Cancer Genetics Branch, NHGRI, NIH, Bethesda, MD; 2) Institut de Génétique et Développement de Rennes, Université de Rennes, Rennes, France; 3) Department of Molecular Biomedical Sciences, College of Veterinary Medicine, North Carolina State University, Raleigh, NC; 4) Department of Population Health and Pathobiology, College of Veterinary Medicine, North Carolina State University, Raleigh, NC; 5) Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands; 6) Cancer Research Center at Massachusetts General Hospital, Boston, MA; 7) Department of Pediatrics, Harvard Medical School, Boston, MA.

While comparatively rare, histiocytic-dendritic cell neoplasms remain devastating. Their status as human orphan diseases urges adaptation of novel genetic strategies to identify their pathogenesis. Breeding bottlenecks and strong selective pressures have caused strong genetic predipositions for many cancers in specific canine breeds. The 25% incidence of histiocytic sarcomas in Bernese Mountain dogs (BMD) offers a unique opportunity to understand the genetic basis of dendritic cell neoplasms. We performed sequential genome wide association studies (GWAS), initially using 240 BMD samples from the U.S. followed by a confirmatory study using 234 European BMDs. Comparing 19,000 informative SNPs, a single-marker chi-squared analysis corrected for population structure and kinship yielded significant associations with the disease phenotype at 2 loci ($p<1.1 \times 10^{-7}$ and $p<2.5 \times 10^{-6}$). Focused genotyping and sequencing of the most highly associated region identify a 300 kb candidate region. This region contains a disease-associated haplotype for which 75.0% of cases are homozygous versus 20.8% of controls. However, this haplotype is also tightly linked ($r2 \geq 0.8$) with additional individual segments spread over the whole region, making causative mutational analysis difficult. Indeed, ten linked SNPs over the whole region predict 75% of the cases with a false positive rate of 5%. Using GM-CSF/IL4-dependent dendritic cell cultures, we are performing functional analyses of candidate genes in the region. Notably, the segments spread across the region that are linked to its core haplotype contain predicted transcriptional regulatory elements. We are therefore testing the effects of these DNA segments using standard transcriptional analyses to evaluate the possibility that multiple associating regulatory SNPs are involved in the pathogenesis of the histiocytic sarcomas. Since the candidate region is syntenic with several genome-wide human disease associations, our results may offer interesting comparative insights into genetic association studies. A second locus identified in the BMD is largely confined to the European population, possibly because this locus has approached fixation in the U.S. BMD population. Future studies of genetic interactions between these two loci offer the potential to understand regulatory pathways that cause poorly understood histiocytic-dendritic cell neoplasms.

- **POSTER SESSION: CANCER GENETICS**
  Program Number 636
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

An Enhanced International Fanconi Anemia Registry (IFAR). *A. D. Auerbach[1], A. Smogorzewska[2], F. P. Lach[2], M. J. Wrobel[3], M. Sengupta[3], E. Barbour[3]* 1) Human Genetics & Hematology, Rockefeller Univ, New York, NY; 2) Laboratory of Genome Maintenance, Rockefeller Univ, New York, NY; 3) Hospital Informatics, Rockefeller Univ, New York, NY.

Fanconi anemia (FA) is a rare recessively inherited disorder characterized by genome instability, DNA crosslink hypersensitivity, congenital malformations, bone marrow failure, and predisposition to malignancy. The International Fanconi Anemia Registry (IFAR) was established at The Rockefeller Univ. in 1982 in order to collect data regarding patients with FA and their families (1). In addition, the Fanconi Anemia Mutation Database, http://www.rockefeller.edu/fanconi/mutate/, was established in 1998 to accelerate the availability of information on mutations in the 13 FA genes. The new IFAR project is developing a comprehensive, ontology-driven Phenotype Recording Instrument (PRI) and database for FA. The PRI integrates the existing IFAR data held in multiple disparate data sources for a unique look at the FA patient population, which includes the patient data available in the current IFAR database, the polymorphic (SNP) and mutation data available for patients representing the known FA genes. Furthermore, the NIH 's Genomics Center (NHGRI) is currently sequencing certain patient tissue samples using Next Generation technology, which will further enhance the current IFAR patient data. The new development effort will also include "deep phenotyping " from new patient history questionnaires. We are hypothesizing the addition of these two

data stores, full gene sequencing and deep phenotyping, will add significantly to the understanding of FA while providing a deeper basic scientific understanding of DNA repair mechanisms, and more prognostic ability for the current patient population. The enhanced data model resulting from the aforementioned data sources is being modeled using an OWL ontology defined with Protégé. We are building data mining and visualization tools that will be required to maximize the research view of this integrated patient data. As the name of the existing IFAR system implies, this registry is an International collaboration. The new system will enable worldwide international collaboration between FA researchers. To accomplish this, we are building a web-based system utilizing the Java JSP front-end coupled with an Oracle 10g database backend for performance and data integrity. This is truly a unique medical informatics opportunity made possible because of the 28 year collection of FA data combined with next generation sequencing and deep phenotyping data. (1) Kutler DI et al., Blood 101:1249-1256, 2003.

- **POSTER SESSION: CLINICAL GENETICS AND DYSMORPHOLOGY**
  Program Number 846
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

The Oculocutaneous Albinism Natural History Study at the National Institutes of Health. *D. R. Simeonov[1], B. P. Brooks[2], C. C. Brewer[3], W. M. Zein[2], C. K. Zalewski[3], T. D. Ngugyen[4], M. Huizing[1], W. A. Gahl[1], D. R. Adams[1]* 1) NHGRI, National Institutes of Health, Bethesda, MD; 2) NEI, National Institutes of Health, Bethesda, MD; 3) NIDCD, National Institutes of Health, Bethesda, MD; 4) Clinical Center, National Institutes of Health, Bethesda, MD.

Introduction: Autosomal recessive oculocutaneous albinism (OCA) causes low vision, hair, skin and eye hypopigmentation, and skin ultraviolet-light sensitivity in 1:18,000 newborns. Approximately 85% of OCA is caused by defects in one of four known genes: *TYR*, *OCA2*, *TYRP1* and *SLC45A2*, leaving a substantial minority of OCA cases without molecular confirmation. The pathogenic potential of many individual variants of the known genes has yet to be quantified. Key aspects of the natural history and cellular biology of OCA also remain to be elucidated.
Methods: The NIH OCA Natural History Study is actively recruiting individuals and families with OCA to participate in a 3-5 day evaluation at the NIH Clinical Center in Bethesda, Maryland. Participants are clinically evaluated at the NIH, focusing on comprehensive evaluation of the visual system with an attempt to uncover clinically relevant endpoints, and evaluation of hearing, auditory processing abilities, and low-vision adaptation. Functional assays are being developed to assess the pathogenic potential of individual mutations in known genes.
Results: Forty individuals have been recruited to date. Demographic and preliminary molecular data are presented along with examples of functional assessment of DNA variants.
Conclusions: The careful collection of clinical data, genetic data and cell biological data for persons with albinism has the potential to yield new albinism-related genes, new biological insights and novel therapeutic targets. Medical providers are encouraged to provide information about the study to their patients with albinism.

- **POSTER SESSION: CLINICAL GENETICS AND DYSMORPHOLOGY**
  Program Number 904
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

A SATB2 nonsense mutation causes Cleft Palate and Cognitive Defects through a dominant negative effect. *P. Leoyklang[1], K. Suphapeetiporn[2], M. Huizing[3], WA. Gahl[3], V. Shotelersuk[2]* 1) Biomedical Science Program, Faculty of Graduate School, Chulalongkorn University, Thailand; 2) Department of Pediatrics, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand; 3) Medical Genetics Branch, NHGRI (NIH), Bethesda, MD.

SATB2 (special AT-rich sequence-binding protein2) (*608148) encodes a DNA binding protein, forming a homodimer that specifically binds nuclear matrix attachment regions and is involveds in transcription regulation and chromatin remodeling. In 2007, we found a 36-year-old man with cleft palate, generalized osteoporosis, and profound mental retardation. He had a de novo germ-line nonsense mutation (c.7154C>T; p.R239X) of SATB2 (Human Mutation. 2007; 28: 732-738). Whether the mutation causes disease through haploinsufficiency, a dominant-negative or a gain-of-function effect was not determined. Here, we first show that the mutant protein is stable. Then, using immunofluorescence and subcellular fractionation, we demonstrate that both the wild-type and mutant SATB2 localize to the nucleus. In addition, protein-protein interaction studies show that the mutant truncated SATB2, which retains the dimerization domain, can form a dimer with the wild type SATB2. These findings strongly suggest that the severe clinical features of our patient are caused by a dominant negative effect of the SATB2 nonsense mutation, rather than to a haploinsufficiency, which associated with a milder phenotype.

- **POSTER SESSION: GENE STRUCTURE AND GENE PRODUCT FUNCTION**

Program Number 1586
Exhibit Hall A, Lower Concourse Level, Convention Center
Fri 7:00AM-12:30PM
Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

An evaluation of Limp-2 in patients with Gaucher disease and myoclonic epilepsy. *N. Gupta, A. Velayati, JH. Choi, W. Westbrook, J. Depaolo, BK. Stubblefield, E. Sidransky, N. Tayebi* Section on Molecular Neurogenetics, Medical Genetics Branch/NHGRI, NIH, Bethesda, MD.

Lysosomal integral membrane protein type 2 (LIMP-2) is responsible for the proper sorting of glucocerebrosidase, the enzyme deficient in Gaucher disease (GD), to the lysosome. Mutations in LIMP-2 were identified in patients with some forms of myoclonic epilepsy (ME). A subgroup of patients with neuronopathic Gaucher disease develop myoclonic epilepsy. We investigated whether development of ME in type 3 GD is related to alterations in Limp-2, studying two groups of patients with GD,13 patients with myoclonic epilepsy and 18 without myoclonic epilepsy, as well as 40 normal controls. The promoter region, all 12 exons and the flanking intronic regions of the Limp-2 gene (Scarb2) were sequenced in each of the patients and controls. One patient with GD/ME was found to have a heterozygous mutation in exon 12. Two other GD/ME patients had intronic mutations, although no effect on splicing was demonstrated using RT-PCR and cDNA sequencing. Furthermore, one polymorphism in exon 1 and 21 polymorphisms in introns were identified among patients and controls. Two polymorphisms are also present in the promoter region of Scarb2. The frequency of these polymorphic changes did not differ significantly between patients with GD with or without ME or controls. We also studied glucocerebrosidase and Limp-2 RNA expression in fibroblasts from patients with GD/ME, 20 with GD and 20 controls using real-time. The results demonstrated down-regulation of Limp-2 in the patient with the exonic mutation and up-regulation in individuals carrying an intronic mutation 1100bp upstream of exon 4 as compared to controls. Protein expression studies, cell-based assays and the confocal microscopy are being employed to evaluate the functional significance of the exonic and intronic mutations identified. These studies should clarify to what extent Limp-2 is associated with myoclonic epilepsy in patients with neuronopathic Gaucher disease.

- **POSTER SESSION: GENOMICS**
  Program Number 1946
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Admixture mapping of fasting glucose: Identification of a candidate locus associated with T2D. *G. Chen[1], D. Shriner[1], J. Zhou[1], A. Doumatey[1], H. Huang[1], N. Gerry[3], A. Herbert[4], M. Christman[3], G. Douston[2], M. Faruque[2], Y. Chen[2], A. Adeyemo[1], C. Rotimi[1]* 1) CRGGH/NHGRI/NIH, NIH, Bethersda, MD; 2) National Human Genome Center, Howard University, Washington DC 20060 USA; 3) Department of Genetics and Genomics, Coriell Coriell Institute for Medical Research, Camden, NJ 08103 USA; 4) Department of Genetics and Genomics, Boston University School of Medicine, Boston, Massachusetts 02118 USA.

Elevated fasting blood glucose is a feature of type 2 diabetes and insulin resistance. We have carried out a search for novel loci associated with fasting blood glucose (FBG) among African Americans using the mapping by admixture linkage disequilibrium (or “admixture mapping ”) approach. Using an ancestry informative marker (AIM) panel comprising 1800 SNPs, the genomes of 619 non diabetic and non hypertensive unrelated African Americans from the Howard University Family Study (HUFS) were screened for regions with elevated ancestry proportions from either of the ancestral populations (reference HapMap YRI and CEU). The mean proportion of African ancestry in the study sample was 0.81 ± 0.10. An increased fasting glucose was associated with increasing proportion of European ancestry with β = 0.087 and p value = 0.044. Using ADMIXMAP, we identified a ~12.36 Mb region (46.49 - 59.85 Mb) on Chromosome 14q22 - 23 associated with FBG with the best p value 6.4x10-5 (rs12895262). Ten AIMs in this region showed Bonferroni corrected p values < 0.05. Repeating the analysis with the STRUCTURE program yielded the similar results. In a follow up fine mapping study, 800 SNPs in this region (average density 9.8 kb) present on the Affymetrix 6.0 SNP array were tested for association with FBG. The best p values were 3.38x10-6 (rs10146136, position 51079426bp, ATL1) and 3.23x10-5 (rs11570807, 50830141bp, CDKL1).In summary, we found that increasing percent of European ancestry was associated with increasing FBG among African Americans. We identified a new locus on chromosome 14q22-23 that influences FBG levels and fine mapping confirmed this association.

- **POSTER SESSION: GENOMICS**
  Program Number 1980
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

***ABCA13*: the naming of a gene.** *R. Seal, S. Gordon, M. Lush, M. Wright, E. Bruford* HUGO Gene Nomenclature Committee (HGNC), European Bioinformatics Institute, Hinxton, Cambridgeshire, United Kingdom.

The HUGO Gene Nomenclature Committee (HGNC) aims to approve a gene symbol and name for every human gene. Standardisation of gene symbols is important as it allows researchers to refer to the same gene without ambiguity and facilitates data retrieval. The primary rule of the HGNC is that every approved gene symbol must be unique. Gene symbols should also be acceptable to researchers to ensure their widespread use, and should be based on structure, function or homology wherever possible. The HGNC encourages the development of a common root symbol for members of a gene family, with a hierarchical numbering system to distinguish the individual members, as this is an efficient way to name large numbers of related genes and makes each family member instantly recognisable. We provide individual web pages on our site for many established gene families and have over one hundred specialist advisors that help us to accurately maintain these families. Here we describe the naming of one particular gene family member, *ABCA13* (full name: ATP-binding cassette, sub-family A (ABC1), member 13). We explain how and why the gene was named and how the symbol has subsequently been used. The first step was the development of the ABC (ATP-binding cassette) gene superfamily and sub-family nomenclature scheme, following in depth discussions between the HGNC and the ABC research community. This was followed by the identification and naming of *ABCA13* as part of this family, its appearance in the biomedical literature and databases, its adoption for the mouse and rat orthologs as *Abca13*, and its subsequent breakthrough into the international media. For further information on human gene nomenclature, please email us at hgnc@genenames.org, or visit http://www.genenames.org/. The work of the HGNC is supported by the NHGRI and the Wellcome Trust.

- **POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2236
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Evidence of association of SNPs in *MSH6* with café-au-lait macule burden in neurofibromatosis type 1. *A. Pemov[1], H. Sung[2], J. L. Sloan[3], S. Ruppert[1], J. Mullikin[4], P. Cruz[4], A. F. Wilson[2], D. R. Stewart[1]* 1) Genetic Disease Research Branch, NHGRI, NIH, Bethesda, MD; 2) Inherited Disease Research Branch, NHGRI, NIH, Baltimore, MD; 3) Genetics and Molecular Biology Branch, NHGRI, NIH, Bethesda, MD; 4) Genome Technology Branch, NHGRI, NIH, Rockville, MD.

Background. The cause of the variation in phenotypic severity in neurofibromatosis type 1 (NF1) is unknown and may be due to genetic modifiers. We hypothesized that variation in *germline* gene expression of certain genes correlates with variation in the severity of quantifiable phenotypic features of NF1. **Methods.** We performed whole-genome transcriptional profiling (Illumina HumanRef-8 arrays) in lymphoblastoid cell lines from 79 individuals affected with NF1 and 23 controls. A single observer quantified severity in multiple NF1 sub-phenotypes, including café-au-lait macules (CALM) number. We examined the correlation of the 6 NF1 sub-phenotypes with the level of each of the 22,177 transcripts. To control for multiple testing, we calculated a False Discovery Rate (FDR), in addition to a nominal *P*-value of the significance of the regression. We filtered for FDR (< 0.3), expression range (~2X) and expression level (mean log2 > 6.0). We validated 22 QTTs by quantitative PCR on low-density microfluidic arrays (ABI). By qPCR, 9 QTTs remained statistically significant (nominal *P*-value < 0.05). Exons, 5 ’-UTR, 3 ’-UTR and limited intronic regions of three genes (*MSH6, MED21, DPH2*), whose expression significantly correlated with CALM burden were subject to Sanger sequencing in 99 patients (79 from original set and 20 new). Two additional genes (*MSH2, MLH1*) which products are known to form functional complexes with MSH6 were also subject to sequencing analysis. The genotypes of the sequenced SNP variants were regressed against CALM burden. **Results.** We found evidence of association for two non-coding *MSH6* SNPs (rs3136316, MAF ~0.20 and rs1800934, MAF ~0.20) with CALM burden (nominal *P* <0.05). We also identified four rare (MAF < 0.05) SNPs in the 3 ’-end of *MSH6*, which when collapsed in a single group, correlated significantly with CALM burden (*P* = 0.047). **Conclusions.** We identified six SNPs in *MSH6* that are associated with the number of café-au-lait macules in NF1. Functional tests and genotyping of rs3136316 and rs1800934 in a second independent cohort of phenotyped patients is underway. *MSH6* is involved in mismatch repair. Our findings are of interest since CALMs arise secondary to bi-allelic inactivation of *NF1*, the same pathogenic mechanism as NF1-associated tumors. If validated, *MSH6* would be the first identified modifier gene of NF1.

- **POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS**
  Program Number 2240
  Exhibit Hall A, Lower Concourse Level, Convention Center
  Fri 7:00AM-12:30PM
  Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Preliminary whole genome sequencing analysis of monozygotic twins with a concordant unique neurological phenotype. *K. V. Fuentes Fajardo[1], S. S. Ajay[3], T. C. Markello[1], D. A. Adams[1], C. Toro[1], M. Sincan[1], H. Carlson-Donohoe[1], P. F. Cherukuri[3], N. F. Hansen[3], H. Ozel Abaan[3], J. C. Mullikin[2], W. A. Gahl[1], E. H. Margulies[3], NISC Comparative Sequencing Program* 1) Undiagnosed Diseases Program, NHGRI/NIH, Bethesda, MD; 2) NIH Intramural Sequencing Center, NIH, Bethesda MD; 3) Genome Technology Branch, NHGRI, NIH Bethesda MD.

Advances in Next Generation Sequencing technology have made whole genome sequencing a practical reality for clinical research applications. Other investigators have reported on whole genome sequencing of monozygotic twins discordant for multiple sclerosis. No discordant variant was identified as the cause of MS in the affected twin. We have an opportunity to sequence concordant twins. Two of the NIH Undiagnosed Diseases Program 's initial choices for whole genome sequencing were a 24 year-old monozygotic twin pair concordant for the unique phenotype that includes early childhood neuro-cognitive deficits, rare seizures, and slowly progressive movement disorder characterized by myoclonus with ataxia. A detailed neurologic evaluation, including metabolic screening, candidate gene sequencing and mitochondrial DNA studies was not diagnostic. We will present the interim analysis of genomic variations we have found in one or both twins and contrast and compare them to the reference human genome sequence; an a priori unknown variant, that could be anywhere in the human genome, must be in both twins. The search will be one of the most rigorous tests of whether whole genome sequencing can be successfully used to identify a disease-causing variant, or whether the intrinsic background false positive rate will overwhelm our current analytic tools.

▪ POSTER SESSION: MOLECULAR BASIS OF MENDELIAN DISORDERS

Program Number 2276
Exhibit Hall A, Lower Concourse Level, Convention Center
Fri 7:00AM-12:30PM
Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Absence of Cyclophilin B Causes Autosomal Recessive Osteogenesis Imperfecta but does not Impair Type I Collagen Peptidyl-Prolyl Isomerization. *A. Barnes[1], E. Carter[2], W. Cabral[1], M. Weis[3], W. Chang[1], E. Makareeva[4], S. Leikin[4], C. Rotimi[5], D. Eyre[3], C. Raggio[2], J. Marini[1]* 1) BEMB, NICHD/NIH, Bethesda, MD; 2) Hospital for Special Surgery, New York, NY; 3) University of Washington, Seattle, WA; 4) SPB, NICHD/NIH, Bethesda, MD; 5) NHGRI/NIH, Bethesda, MD.

Osteogenesis imperfecta, or brittle bone disease, is a genetic disorder characterized by bone fragility and growth deficiency. Recently, recessive forms of osteogenesis imperfecta have been shown to be caused by mutations in the three genes coding for components of the collagen 3-hydroxylation complex. This complex post-translationally hydroxylates a single residue in type I collagen, Pro986, as well as residues on types II and V collagen. Null mutations in *CRTAP* or *LEPRE1* lead to severe/lethal forms of OI with white sclerae and rhizomelia (types VII and VIII OI). Collagen synthesized by patient fibroblasts has minimal Pro986 hydroxylation, but is overmodified, indicating delay in collagen folding. In a pair of Senegalese siblings born to consanguineous parents, we identified a homozygous mutation in the start codon of *PPIB*, which encodes cyclophilin B (CyPB) the third member of the 3-hydroxylation complex. These children are ambulatory with moderate OI and white sclerae, but without rhizomelia or severe growth deficiency. Proband fibroblast RNA had 55% of the normal level of *PPIB* transcripts by real-time RT-PCR. On Western blot of fibroblast lysates, no CyPB protein was detectable using 3 antibodies to full protein, C-terminal half and final 15 residues or after treating proband cells with a proteasome inhibitor. Both CRTAP and P3H1 protein levels were reduced to half of normal levels, suggesting that complex stability is decreased. Immunofluorescence microscopy confirmed these findings in vivo. CyPB is a well-known peptidyl-prolyl cis-trans isomerase which is thought to be involved in the rate-limiting step of type I collagen folding. Therefore, we examined the proband 's steady-state type I collagen protein as well as the levels of lysyl and 3- and 4-prolyl hydroxylation in the collagen helix. Pro986 hydroxylation was normal in proband collagen, indicating that the complex can complete its collagen modification function in the absence of CyPB. Surprisingly, steady-state collagen from our proband was not overmodified, with normal lysyl and 4-prolyl hydroxylation levels, in contrast to collagen synthesized by cells with recessive defects in *CRTAP* or *LEPRE1*, or with recessive *PPIB* mutations which allow synthesis of misfolded CyPB. Therefore, the complete absence of CyPB does not appear to delay collagen folding, strongly suggesting that there is redundancy for collagen isomerization, or CyPB may not be the major type I collagen folding isomerase.

▪ POSTER SESSION: STATISTICAL GENETICS AND GENETIC EPIDEMIOLOGY

Program Number 2982
Exhibit Hall A, Lower Concourse Level, Convention Center
Fri 7:00AM-12:30PM
Presentation Time: Fri, Nov 5, 2010, 11:30AM-12:30PM

Abstract Content

Genetic Heterogeneity of Myopia Susceptibility in an Ashkenazi Jewish Population. *C. Simpson[1], R. Wojciechowski[1], D. Stambolian[2], J. E. Bailey-Wilson[1]* 1) Inherited Disease Res Branch, NHGRI, NIH, Baltimore, MD; 2) Department of Ophthalmology, University of Pennsylvania.

Myopia affects at least one third of most populations, is a complex disorder, with both genetic and environmental etiological influences. It has a significant impact on the lives of affected individuals and carries high economic costs associated with treatment and with loss of productivity and co-morbidity from vision impairment. Despite many years of research, most of the factors contributing to myopia development remain unknown. Genetic studies have pointed to a strong inherited component, but although many loci have been found, few genes have been positively identified as causal agents. We have previously reported 2 genomewide linkage scans in a population of 49 highly aggregated Ashkenazi Jewish families which identified a locus on chromosome 22. Here we have used ordered subset analysis, conditioned on non-parametric linkage to chromosome 22 to detect other loci which also had evidence of linkage to myopia in subsets of the families, but not the overall sample. Similar analyses using parametric LOD scores in OSA are ongoing and may increase power. Suggestive linkage to a 25-cM linkage interval with a peak OSA nonparametric allele-sharing LOD score of 2.622 on 20p12.1-q11.23 (ΔLOD = 2.193, empirical P = 0.034) was identified in a subset of 15 families with strong evidence of linkage to chromosome 22, which represents 31% of the total dataset (15/49 families). Seven other loci also presented with suggestive LOD scores > 2.0 on chromosome 2q34-q35, 3p11.1-q24, 5q21.1-q23.3, 6q22.31-q24.2, 7p22.3-21.3, 9q31.1-q33.3 and 14q31.3-q32.3. However, none of the ΔLOD scores were significant by permutation testing. Results using model-based parametric LOD scores in OSA will also be presented. The chromosome 20 locus is entirely novel and appears only in a subset of families already known to be strongly linked to chromosome 22. Using ordered subset analysis allows us to find additional loci linked to myopia in subsets of families, and underlines the complex genetic heterogeneity of myopia even in highly aggregated families and genetically isolated populations such as the Ashkenazi Jews.

- **POSTER SESSION: 53. GENETIC ARCHITECTURE OF NEUROLOGICAL DISEASES**
  Program Number 287
  Room 202, Level 2, Convention Center
  Fri Nov 5, 2010 01:30PM-04:00PM
  Presentation Time: Fri, Nov 5, 2010, 01:30PM-01:45PM

Abstract Content

Identification of genetic variants associated with stuttering: Using inbred pedigrees to find rare genetic variants of large effect. *D. Drayna[1], S. Riazuddin[2], D. Krasnewich[3], P. Friedman[4], C. Kang[1]* 1) NIDCD, National Institutes of Health, Rockville, MD; 2) Alama Iqbal Medical College, Lahore, Pakistan; 3) NHGRI, National Institutes of Health, Bethesda, MD; 4) Clinical Center, National Institutes of Health, Bethesda, MD.

Stuttering is a common speech disorder present in all populations and language groups. Although the causes of this disorder have been unknown, twin and family studies have demonstrated that it is highly heritable. Segregation analyses and linkage studies, however, have failed to identify Mendelian inheritance of stuttering. We hypothesized that stuttering is caused by rare genetic variants of large effect, and we studied large, highly consanguineous families from Pakistan as a method to efficiently reveal such variants. We identified significant linkage on chromosome 12q in this population, and a search of the genes within this region identified a Glu1200Lys mutation in GNPTAB, which encodes the catalytic subunit of GlcNAc phosphotransferase, in affected members of several families in our collection. This mutation was identified in unrelated individuals who stutter from both Pakistan and India, and other mutations in this gene were identified in other cases but not in controls. Based on this, we evaluated the GNPTG gene, which encodes the recognition subunit of this enzyme. We identified a number of mutations in this gene in individuals who stutter that did not occur in matched controls. We then examined the NAGPA gene, which encodes the enzyme that acts immediately downstream of GlcNAc phosphotransferase in the same metabolic pathway, and identified a number of mutations in unrelated cases that did not occur in controls. These two enzymes act to generate the Mannose-6-phosphate lysosomal targeting signal that directs a diverse group of hydrolases to the lysosome. Deficits in GNPTAB and GNPTG have been associated with the mucolipidosis types II and III, which are rare recessive disorders with manifestations in the connective tissue, skeletal system, brain, liver, and spleen. No disorder has previously been associated with mutations in NAGPA. Detailed clinical examination of four stuttering subjects carrying mutations in these genes revealed no symptoms associated with mucolipidosis types II and III, and other than stuttering, these individuals were neurologically normal. We hypothesize that the mutations we 've identified affect a distinct group of neurons in the brain that are uniquely dedicated to speech production and uniquely sensitive to the metabolic deficit caused by these mutations, and that inbred families present a generally powerful strategy for identifying rare mutations of large effect that underlie common complex disorders.