Identifying Genetic Disorders and Implementing Genetic Testing for the Wisconsin Plain Community

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PURPOSE

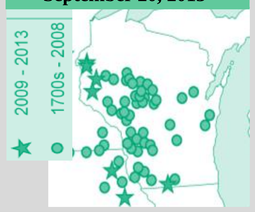
To develop an accessible, low cost genetic testing infrastructure to identify genetic disorders in Wisconsin Plain communities.

BACKGROUND

Wisconsin has the fourth largest Plain population in the United States with an estimated 17,000 Old Order Amish and 2,500 Old Order Mennonites and the population continues to have rapid growth. These isolated communities are subject to founder effects resulting in a higher frequency of autosomal recessive genetic disorders.

Members of the Plain communities have limited access to specialized health care due to lack of insurance, difficulties with travel, and the unavailability of local physicians with experience in identifying genetic disorders.

Existing Settlements as of September 20, 2013[1]



METHODS

Families were counseled and consented for genetic testing at:

- the La Farge Medical Clinic,
- the University of Wisconsin Genetics Clinic or within their homes.

Three approaches were used to identify pathogenic variants for a variety of disorders in the population:

1. Targeted Variant Sequence Analysis (TVAR)

for identification of characterized pathogenic variants associated with known genotype/phenotype correlations segregating within the Plain population

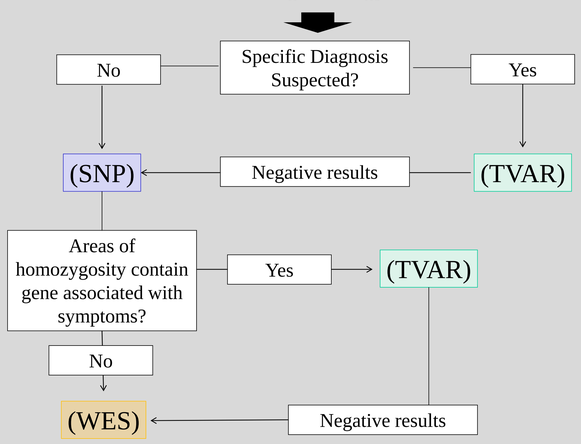
2. Single Nucleotide Polymorphism (SNP) microarrays

to identify recurrent genomic dosage aberrations and regions of homozygosity that may indicate disease associated genes or variants

3. Whole Exome Sequencing (WES)

to identify novel and characterized variants within the protein-coding regions of genes.

Testing Strategy:



RESULTS

Conditions Prevalent in Plain Population - with Classic Presentation

Condition Identified	Presenting Symptoms/ Family History	Test	Gene	Mutation/ Inheritance	Pathophysiology	Classic presentation	Treatment/ Prognosis
Galloway-Mowat syndrome	Sibs (2 year old and 4 month old) with global developmental delay (GDD), failure to thrive (FTT), microcephaly, spasticity, abnormal movements and nystagmus. Older child had proteinuria. 3 paternal aunts with GDD; 2 died from kidney failure in childhood. Parents are 2nd cousins.	TVAR	WDR73	Homozygous for: c.888delT p.Phe296Leufs*26 Autosomal Recessive (AR)	Neurodegenerative disorder caused by dysfunction of the WDR73 protein expressed in brain and kidney tissues.	Progressive microcephaly, visual impairment with nystagmus, stagnant psychomotor development, intellectual disability (ID), abnormal extrapyramidal movements, cerebellar atrophy, and steroid non-responsive nephrosis. Life limiting due to kidney failure.	Symptomatic care. Typically life limiting due to kidney failure.
Infantile Lethal Cardiomyopathy	2 week old with poor feeding, shortness of breath and tachypnea. Found to have restrictive cardiomyopathy and moderate muscular VSD. Family decline transplant. Patient died at 3 weeks of age. Parents and two grandparents found to be carriers.	TVAR	MYBPC3	Homozygous for: c.3330+2T>G p.Asp1064Glyfs*38 AR[a]	Dysfunction of myosin binding protein C results in severe hypertrophic cardiomyopathy.	Infantile onset cardiomyopathy resulting in heart failure and death within 3 to 4 months without transplant. Strongly suspected carriers are at risk for hypertrophic cardiomyopathy.	Heart transplantation or palliative care.
Cobalamin C Deficiency	2 week old with abnormal newborn screen - elevated C3. Had elevated methylmalonic acid (MMA) and homocysteine (HCY). Propionic acidemia(PA) testing was normal.	TVAR	MMACHC	Homozygous for: c.271dupA p.Arg91Lysfs*14[b] AR	Dysfunction of the MMACHC enzyme responsible for cobalamin trafficking resulting in decreased methionine and increased MMA and HCY levels.	Intrauterine growth retardation, microcephaly, congenital heart disease, poor feeding, lethargy, failure to thrive, hypotonia, seizures, nystagmus and GDD.	Betaine, carnitine and intramuscular B12 as well as fasting and illness precautions.
Sitosterolemia	9 year old with xanthomas on hands, knee and Achilles tendon, short stature, and failure to gain weight.	TVAR	ABCG8	Homozygous for: c.1720G>A p.Gly574Arg AR	Dysfunction of sterolin, a protein responsible for eliminating plant sterols that cannot be used by human cells.	Xanthomas, premature atherosclerosis, hemolytic anemia and thrombocytopenia.	Medication (Ezetimibe) and specialized diet to reduce sterol levels.
Propionic Acidemia	7 year old with complicated post-natal course including pulmonary hypertension. Continued to have seizures and developmental delays. Abnormal newborn screen during this evaluation.	TVAR	PCCB	Homozygous for: c.1606A>G p.Asn536Asp AR	Deficiency of the beta subunit of propionyl-CoA-carboxylase, which results in an accumulation of propionic acid and associated metabolites.	Lethargy, hyperammonemia, profound acidosis, coma and death in neonatal period. Subsequent symptoms include ataxia, GDD, and seizures. PCCB-related PA, previously thought to be milder, reported to result in cardiomyopathy, arrhythmia and sudden death.	Patient started on specialized diet and given illness precautions.

Conditions Prevalent in Plain Population - with Non-traditional Presentation

Condition Identified	Presenting Symptoms/ Family History	Test	Gene	Mutation/ Inheritance	Pathophysiology	Classic presentation	Treatment/ Prognosis
Cortical Dysplasia Focal Epilepsy syndrome	14 month old with focal epilepsy, mild developmental delays (DD) and a normal brain MRI.	aCGH, then TVAR	CNTNAP2	Homozygous for: c.3709delG p.Asp1237Ilefs*17 AR	Dysfunction of a protein important for nervous system development and function.	Seizures, DD, regression in language and social skills, ID, cortical dysplasia, and behavioral concerns (hyperactivity or impulsive aggression).	Symptomatic care.
Symptomatic Epilepsy and Skull Dysplasia	24 year old with profound ID, ptosis, robust stature, and well-controlled seizures. Walked with limited assistance.	aCGH, then WES	SNIP1	Homozygous for: c. 1097A>G p.Glu366Gly AR	Dysfunction of SNIP1 protein, likely involved in signal transduction cascade for brain, skull, craniofacial bones and distal limbs.	Severe DD with inability to walk or speak, hypotonia with poor feeding in infancy, brain abnormalities, intractable seizures, dysmorphic facial features and a "lumpy" skull surface.	Symptomatic care.
Mucolipidosis Type II (I-cell disease)	Amish 2 year old with coarse facial features, FTT, poor growth, small thoracic cavity, profound DD, respiratory distress and infections, dilated cardiomyopathy and dysplastic cardiac valves. Normal berry spot and alpha-L-Iduronidase enzyme testing. Child died at age 3.	aCGH, then specific gene	GNPTAB	Homozygous for: c.3503-3504delC[c] AR	Dysfunction of lysosomal enzyme (GlcNAc-1-phosphotransferase) resulting in accumulation of carbohydrates and lipids and cell damage.	Hypotonia, weak cry, poor growth, multiple bone abnormalities with dysostosis multiplex, heart valve abnormalities, coarse facial features, GDD and narrow airway which leads to respiratory concerns. Children usually die in early childhood. Previously seen in two Mennonite children, but no Amish patients.	Symptomatic care.

Conditions Not Thought to be More Prevalent in Plain Population

Condition Identified	Presenting Symptoms/ Family History	Test	Gene	Mutation/ Inheritance	Pathophysiology	Classic presentation	Treatment/ Prognosis
Rett syndrome	17 year old with central hand wringing, developmental regression at 18 mos., severe ID, febrile seizures, laughing fits, breath holding spells, and growth retardation. Teacher suspected Rett syndrome.	specific gene	MECP2	Heterozygous for: partial exon 4 deletion X-linked Dominant	Altered expression of MECP2 important for nervous system development and function.	Developmental regression, especially in the areas of communication and coordination, around age 6 to 18 months, loss of purposeful hand movements in early childhood, microcephaly, breathing abnormalities, seizures, scoliosis and sleep disturbances.	Symptomatic care.
16p11.2 duplication syndrome	19 year old with mild to moderate ID, childhood seizures, nonverbal until age 8, mildly dysmorphic facial features. AGA at birth. No concerns for growth, behavior or social concerns. Negative Fragile X testing.	aCGH		Heterozygous for duplication Autosomal Dominant (AD)		Variable presentation that include ID, DD - particularly speech, autism, ADHD, and mental health disorders. Recurrent seizures and poor growth have also been reported. Some people with this duplication have no health or developmental concerns.	Symptomatic care.
Ataxia Telangiectasia - Like Type 2	4 year old who was small for gestational age, poor growth (ht and wt < -3 SD), ataxia, GDD, and nonverbal until age 4. Clinical diagnosis of spastic paraplegia. Physical exam: hairline scarring, irregular pigmentation and freckles at temples bilaterally. No telangiectasias.	WES	PCNA	Homozygous for: c. 683G>T p.Ser228Ile AR	Neurodegenerative disorder due to defects in DNA excision repair.	Short stature (-3.8 to -5.2 SD), ataxia, DD, telangiectasias (conjunctival and cutaneous), sensorineural hearing loss and photophobia. One report of dermatologic malignancy. No reports of immunodeficiency[d]. PCNA mutation reported in 4 Amish children.	Symptomatic care.
Senior-Loken syndrome Type 5	18 year old with progressively poor vision since early childhood. Eye exam showed retinal dystrophy with rod and cone dysfunction - Retinitis Pigmentosa v. Leber Congenital Amaurosis. No other health concerns. Normal renal function tests. Many relatives with poor vision. Parents are 2nd cousins.	WES	IQCB1	Homozygous for: c.1213_1276del p.Lys405Leufs*12 AR	Interacts with calmodulin and may participate in common pathway of ciliary function.	Retinal dystrophy and nephronophthisis with ocular onset in infancy to early childhood. Renal symptoms have variable onset and, when present, progress to end stage renal disease.	Symptomatic care.

[a] - Carriers may be symptomatic, which would be associated with AD inheritance.
[b] - Associated with severe, early-onset disease. Accounts for 40% of all disease-causing alleles.
[c] - Common French Canadian mutation
[d] – Immunodeficiency can be seen in ataxia telangiectasia caused by ATM mutations (more common in Amish and Mennonite populations.

CONCLUSION

Collectively, we have provided diagnostic testing and disease-specific education and counseling in an accessible manner to the Plain communities of Wisconsin.

Through this process, we have increased our knowledge about

- the presence of specific disorders within the WI Plain population, and
- the phenotypic spectrum of these conditions.

We have also developed and validated several affordable clinically available TVAR tests for this population.

Altogether, this ultimately improves detection of and the medical management for these previously undiagnosed genetic disorders.

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