Utilization of Genetic Testing Among Children with Autism Spectrum Disorders

Master’s Thesis

Presented to

The Faculty of the Graduate School of Arts and Sciences
Brandeis University
Department of Biological Sciences, Program in Genetic Counseling

Beth Rosen-Sheidley, MS, CGC, Advisor

In Partial Fulfillment
of the Requirements for

Master’s of Science Degree

By
Whitney Hunter

May 2011
Copyright by
Whitney Hunter
© 2011
Acknowledgments

Many thanks and appreciation to the Autism Consortium for providing the dataset and the families who participated in the Autism Consortium research project

My committee members, Beth Rosen-Sheidley, Dr. Laurie Demmer and Dr. Julia O’Rourke, for their support and direction

The Brandeis Genetic Counseling Program for their support

Scott Motyka for statistical analysis support
ABSTRACT

Utilization of Genetic Testing Among Children with Autism Spectrum Disorders

A thesis presented to the Department of Biological Sciences, Program in Genetic Counseling

Graduate School of Arts and Sciences
Brandeis University
Waltham, Massachusetts

By Whitney Hunter

Autism spectrum disorders (ASDs) which include autistic disorder, Asperger disorder and pervasive developmental disorder-not otherwise specified (PDD-NOS) have gained significant attention because of an apparent increase in incidence. The etiology of ASDs has been an ongoing research endeavor, but compelling data thus far suggest a genetic basis. Several professional organizations, including the American Academy of Pediatrics and the American College of Medical Genetics, recommend genetic testing of children affected with ASDs. However, previous research suggests many children with ASDs are not receiving genetic services. The aim of the current study was to analyze data collected by the Autism Consortium, a collaborative research effort based in Boston, Massachusetts, regarding the frequency of genetic testing among children with ASDs. Specifically, we looked at medical, family and genetic testing histories that were collected between 2006 and 2010, to assess genetic test utilization and to identify factors
influencing receipt of genetic testing. In accordance with other studies, we found that approximately 50% of the 485 research participants did not receive genetic testing. Individuals with autistic disorder or PDD-NOS were more likely to have obtained genetic testing than individuals with Asperger disorder. Accompanying intellectual disability or seizures in the child and/or a family history of intellectual disability increased the likelihood that a child with an ASD would receive genetic testing. However, having a sibling with an ASD or a family history of ASDs did not increase the likelihood of obtaining genetic testing. It is unclear whether the reduced rates of testing overall were due to decision making on the part of the medical providers or the parents. Future research to elucidate the reasons for this lack of testing might lead to an improved ability to educate both care providers and families about the importance of genetic testing for this population.

Key Words: Autism Spectrum Disorders, Genetic Testing, Predictors of Genetic Testing
Table of Contents

Table of Contents ................................................................. Page vi

Introduction .................................................................................. Page 01

Materials & Methods .................................................................... Page 09

Results ....................................................................................... Page 12

Discussion .................................................................................. Page 27

Conclusion .................................................................................. Page 36

References .................................................................................. Page 38
List of Tables

Table 1: Participant Information Obtained From the Medical History Questionnaire of the Autism Consortium Dataset ........................................ Page 10

Table 2: Demographic Information .......................................................... Page 13

Table 3: Summary of Predictors that Increase Receipt of Genetic Testing........Page 22

Table 4: Summary of Predictors that Decrease Receipt of Genetic Testing........Page 25
List of Figures

Figure 1: Racial Distribution of Autism Consortium Enrollees......................... Page 14

Figure 2: Female Primary Caregiver’s Education Distribution of Autism Consortium Enrollees...........................................................................................................Page 15

Figure 3: Male Primary Caregiver’s Education Distribution of Autism Consortium Enrollees...........................................................................................................Page 16

Figure 4: Annual Household Income Distribution........................................Page 17

Figure 5: Autism Spectrum Disorder Distribution...........................................Page 18

Figure 6: Participant’s Receipt of Genetic Testing............................................Page 19

Figure 7: Percentage of Children Diagnosed with Autistic Disorder, Asperger Disorder and PDD-NOS Disorder Receiving Genetic Testing..............................Page 20
Introduction

Autism spectrum disorders (ASDs) are neurobehavioral pervasive developmental disorders with manifestations classified along a spectrum representing the range of symptom severity. Manifestations typically present before three years of age and include impairments of social skills, language and communication skills, repetitive behaviors and narrow range of interests (American Psychiatric Association [DSM-IV-TR], 2000). ASDs can be accompanied by regression of skills with reports between 19% (Siperstein & Volkmar, 2004) and 33% (Goldberg et al., 2003) of cases, intellectual disability in 50-70% of cases (Fombonne, 2005), and seizures with reported estimates between 5% (Bryson, Clark, & Smith, 1988) and 46% (Hughes & Melyn, 2005) of cases. ASD diagnoses include autistic disorder, pervasive developmental disorder- not otherwise specified (PDD-NOS) and Asperger disorder as specified by the Diagnostic and Statistical Manual of Mental Disorders, 4th Edition, Text Revision (American Psychiatric Association [DSM-IV-TR], 2000).

The DSM-IV-TR outlines the manifestations of ASDs along with the diagnostic criteria to classify symptomatic individuals to one of the three disorders. A diagnosis of an ASD is based on the degree of impairment in the three areas described above (i.e. qualitative impairment(s) in social interactions, communication and restricted, repetitive and stereotyped patterns of behavior, interests, and activities). For example, an autism diagnosis involves impairments in all three categories described above where as an individual with Asperger disorder does not have clinically significant delay in language
or cognitive development. PDD-NOS is considered atypical autism and serves to encompass individuals who do not fit the diagnostic criteria of the previous two disorders (DSM-IV-TR).

Prevalence

The estimated prevalence of ASDs has steadily been increasing according to epidemiological studies since 1966 (Fombonne, 2002). In 1966 the overall prevalence of ASDs was estimated at 4.1/10,000 (Lotter, 1966). Currently, it is estimated that every 6/1,000 individuals are diagnosed with an ASD with more males than females affected (Shen et al., 2010). There has been considerable speculation regarding an increasing prevalence of ASDs including broader diagnostic criteria, improved public and professional awareness of the disorder, and better knowledge of disease variability (Schaefer & Mendelsohn, 2008).

Researchers concerned with an ever increasing prevalence of ASDs are trying to identify the etiology or cause of ASDs. Genetic research has demonstrated many underlying genetic causes of ASDs. Uncovering a cause of ASDs will provide insight into the disorder as well as allow for the development of strategies to prevent and treat manifestations of ASDs.

Etiology of ASDs

Several discoveries have led many researchers to believe ASDs have a genetic etiology. Altered chromosomal loci (Martin & Ledbetter, 2007; Szatmari et al., 2007; Weiss et al., 2008), copy number variants (deletions or duplications of certain DNA sequences) and several specific gene mutations, for example PTEN (Varga, Pastore,
Prior, Herman, & McBride, 2009) and MET (Campbell et al., 2006), have been shown to cause ASDs. Twin studies have also supported a genetic etiology of ASDs because of a high concordance rate of autism spectrum disorders (Hallmayer et al., 2002). Specifically, one twin study demonstrated a 92% concordance rate of the broader autism spectrum phenotype among monozygotic twins and a 10% concordance rate among dizygotic twins (Bailey et al., 1995). Additionally, the heritability of ASDs is estimated to be about 90% (Freitag, 2007). These factors together, highlight the profound influence of genetic factors on causing ASDs.

Interestingly, several genetic disorders are associated with clinical symptoms of ASDs, further suggesting genetic abnormalities as the underlying cause of ASDs. These genetic disorders where autism is secondary to the diagnosis, account for about 10% of ASD cases (Freitag, 2007). Fragile X, tuberous sclerosis, Angelman syndrome, Rett syndrome and Smith-Lemli Opitz syndrome are some of the disorders associated with ASDs (Johnson & Myers, 2007). Some metabolic disorders, such as untreated phenylketonuria, can also account for some cases of ASD.

Identifying a genetic etiology for an individual with ASD allows for accurate recurrence risk estimations for other family members, medical monitoring of other manifestations if the ASD is secondary to a syndrome, and provides a more precise prognosis. An approach for uncovering a genetic etiology is through genetic evaluation and genetic testing.
Genetic Testing

There are several genetic tests used to identify the genetic etiology of an individual’s ASD. Karyotype (chromosome analysis), fragile X, and array comparative genomic hybridization (aCGH or CMA) are tests often performed on individuals with ASD. Choosing the appropriate test depends on the clinical presentation of the ASD, as well as the presence or absence of intellectual disability and abnormal physical features (dysmorphic features) (Mendelsohn & Schaefer, 2008). These clinical features can be ascertained through a genetic evaluation and can prompt certain testing strategies.

Shen et al. (2010) looked at the different types of tests available for diagnosing ASDs and compared their effectiveness in identifying an underlying genetic etiology. Researchers compared detection rates of genetic abnormalities between karyotyping, fragile X testing and chromosomal microarray (CMA) in patients diagnosed with an ASD. Of the individuals who received karyotyping, about 2.2% (19/852) of the individuals had an abnormal finding. Ten of these patients had balanced rearrangement not detectable by CMA. Four patients had abnormal fragile X results. About 18% of patients (154/848) had CNVs identified by CMA testing with 59 possibly being clinically significant. Of the individuals diagnosed with intellectual disability, 22% (12/54) had an abnormality detected by CMA. Karyotype only identified two genetic abnormalities in this cohort of people while fragile X testing identified three abnormalities. Among the individuals with dysmorphic features, 63% (10/16) had abnormalities detected by CMA while karyotype only detected two genetic abnormalities in this cohort. Thirty-six individuals were reported to have seizures, and 22% (8/36) of them had abnormalities detected by CMA while karyotype only detected 5.6% (2/36) of these genetic
abnormalities. Although CMA would not detect balanced rearrangements, it can detect more genetic abnormalities than karyotyping. The authors suggest that karyotyping should not be replaced because it may be helpful in looking for these balanced rearrangements. However, CMA should be a first tier test instead of fragile X testing and karyotyping (Shen, et al., 2010).

Recommendations for Genetic Evaluation and Testing

The ultimate goal is to identify an underlying genetic cause of the ASD using a combination of a clinical evaluation and genetic testing. A genetic evaluation serves to identify features that may be consistent with specific gene findings in order to tailor the sequence of the genetic tests (i.e. macrocephaly and \( PTEN \) gene mutations). Choosing the appropriate test should also take into account any family history of a specific condition and any significant personal medical history (Mendelsohn & Schaefer, 2008). Consequently, the sequence of testing would then be in order of usefulness, with the test that has the highest likelihood of obtaining an informative result first. Using a tier based approach limits the types and therefore the cost of testing because testing is tailored to an individual.

The American College of Medical Genetics and American Academy of Pediatrics have both published recommendations regarding medical evaluations of individuals diagnosed with ASDs. The American College of Medical Genetics recommends a tier based approach to evaluation and testing. In their approach, a genetics evaluation is first and should identify if the case is syndromic. If a syndrome is suspected, and ASD is secondary to the syndrome, targeted genetic testing should proceed (i.e. tuberous
sclerosis, Angelman syndrome, etc). If the ASD is not suspected to be syndromic, the first tier of testing should be karyotype, fragile X and metabolic screening. The second tier includes fibroblast karyotype (only if pigmentary changes are noted), CMA, MECP2 (females only), and PTEN (if head circumference is 2.5 SD above the mean). The third tier of genetic tests includes brain MRI and serum uric acid. They also emphasize the importance of tailoring the testing to the individual and the clinical situation (Schaefer & Mendelsohn, 2008). Similarly, the American Academy of Pediatrics endorsed the recommendations set by the American Academy of Neurology and the Child Neurology Society for genetic testing among individuals diagnosed with ASD and reaffirmed them in 2003. They too recommend karyotype and fragile X testing first. If metabolic symptoms are present, selective metabolic testing should ensue (Filipek et al., 2000).

Utilization of Genetic Testing

A study by Mercer et al. in 2006 evaluated parents of children with ASD about their perceptions of what caused their child’s disorder. Despite the fact that about 90% (37/41) of the parents believed genetics contributed to the diagnosis, none of the children were evaluated by genetics (Mercer, Creighton, Holden, & Lewis, 2006). In another study that looked at utilization of genetic services, only 36% (28/77) of the participants had met with a genetics professional (Tansey, Rosen-Sheidley, Picker, & Sastry, 2007). Similarly, McLennan et al. analyzed whether individuals with ASDs were receiving genetic tests in an attempt to evaluate whether this population was obtaining the recommended services and testing. Of the 64 families with a reported ASD diagnosis in a family member, only 31% (20 individuals) received genetic testing (McLennan, Huculak,
& Sheehan, 2008). There seems to be a large discrepancy between the recommendations and actual utilization of genetic testing among individuals diagnosed with ASD.

Aims of the study

It is unclear to what extent the guidelines for genetic testing in children with ASDs are being followed. There are recommendations for testing yet studies have shown that minimal genetic testing, if any, is being utilized. The aim of this study was to determine the genetic tests being performed on individuals with ASD. The goal was to classify the types of genetic tests actually being ordered for patients with ASD and if there are any patterns of which children with ASD are more or less likely to receive genetic testing.

The Autism Consortium

The Autism Consortium is a nonprofit organization that funded a multi-institutional collaborative study involving over 60 physicians and scientists from 14 different centers based in the Boston, MA area. These study centers collected medical history, family history and genetic testing information between 2006 and 2010 regarding children diagnosed with ASD and entered the information into a de-identified database. To accomplish our aim of analyzing genetic test utilization among children with ASD, we analyzed the medical, family and the genetic testing histories of the Autism Consortium participants. We wanted to see what percentage of individuals received genetic testing, which tests are being ordered, and if there are any predictors of who is more likely to receive genetic testing. We hypothesized that a significant portion of individuals
diagnosed with an ASD are not receiving genetic testing and those receiving genetic testing were more likely to have accompanying intellectual disability, seizures, birth defects, or a family history of ASD, intellectual disability, seizures or birth defects.

Hypotheses

1. A significant portion of patients with ASD are not receiving genetic testing.

2. Individuals with more severe ASD or that have accompanying intellectual disability and/or seizures are more likely to obtain genetic testing than those with milder ASD.

3. Individuals with birth defects or dysmorphic features are more likely to obtain genetic testing.

4. Individuals who have a sibling with an ASD are more likely to have had genetic testing.

5. Individuals who have a family history of an ASD are more likely to have had genetic testing.
Materials & Methods

For this project, de-identified research participant information was obtained from the Autism Consortium database and analyzed using a computer program called Statistical Package for the Social Sciences (SPSS). The Brandeis Institutional Review Board approved this study and the Autism Consortium Steering Committee granted us access to participant information.

The current study analyzed the data that was collected on probands and siblings through the Autism Consortium’s Phenotypic and Genetic Factors of Autism protocol. Questionnaires were either administered by a genetic counselor/study coordinator with a parent as the respondent, or were completed by the respondent and subsequently reviewed by a genetic counselor/study coordinator. The questionnaires were used to collect family, medical and genetic testing history regarding each of the participants. Specifically, the medical history information obtained from the database includes background information, demographics, neurological and metabolic problems, personal medical history, family history, and genetic testing information. The specific questionnaire items that we included in our dataset and responses obtained from the dataset are listed below in table 1.
Table 1: Participant Information Obtained From the Medical History Questionnaire of the Autism Consortium Dataset

A. Participant Demographic Information Obtained

- Is the participant the proband or the affected sibling of the proband?
- Participants gender: male/female
- A1. Age of child (Child’s date of birth)
- A2. Ethnicity of child
- A3. Race of child (Please check all that apply)
- A4. Please indicate your total annual household income
- A5. What is the child’s female primary caregiver’s highest level of education attained?
- A7. What is the child’s male primary caregiver’s highest level of education attained?

B. Participant Medical/Family History (Response Options: Yes, No, Unsure):

- J34. Has your child ever had seizures?

Please indicate whether your child has ever been diagnosed with or suspected of having any of the following metabolic symptoms (Response Options: Never Diagnosed, Suspected of Having, Diagnosed, Unsure):

- J35. Unusual body odor
- J36. Urine with unusual smell or odor
- J37. Compared to other children his/her age, does your child have an unusually low energy level or low physical endurance most of the time?

Please indicate if there is a family history of the disorder and if YES, indicate whether or not the child’s blood relatives listed have been diagnosed (If any of the blood relatives listed do not exist, check “no”) (Response Options: Yes, No, Unsure; If yes, indicate affected individual (i.e. Child (being evaluated), biological mother, biological father, sibling(s), maternal ½ siblings, paternal ½ siblings, maternal 1st cousins, paternal 1st cousins, maternal aunt(s)/uncle(s), paternal aunt(s)/uncle(s), maternal grandparent(s), paternal grandparent(s), other blood relative):

- N1. Autistic Disorder (Not including Asperger’s Disorder or Pervasive Developmental Disorder- Not Otherwise Specified)
- N2. Asperger’s Disorder
- N3. Pervasive Developmental Disorder-Not Otherwise Specified (PDD-NOS)
- N16. Mental Retardation
- N33. Congenital Heart Defect
- N34. Kidney Defect (From Birth)
- N35. Other Birth Defects (e.g. cleft lip or cleft palate, open spine)
C. Genetic Testing (Response Options: Yes, No, Unsure):

- B2. Was Fragile X DNA testing performed?
- B5. Was karyotyping (chromosome analysis) performed?
- B8. Was Comparative Genomic Hybridization (Chromosomal Microarray Analysis) testing performed?
- B11. Was other clinical genetic testing performed?

The statistical analysis included binary logistic regression analyzing what factors predict whether or not genetic testing was performed. Intellectual disability in the child or a family member and seizures in the child or a family member were independent regression models with genetic testing as the outcome. In comparison, demographic questions, the types of ASD diagnoses in the child or a family member, birth defects in the child or family member and metabolic symptoms were each used in their own regression model with genetic testing as the outcome. For the latter model, when there were greater than one predictor in the model, the odds ratios were adjusted for the number of predictors in the model. There were several types of genetic tests that were analyzed including, fragile X, karyotype, chromosomal microarray (CMA) and “other” genetic testing. Another variable was created that looked at whether at least one of the above genetic tests was performed or none of these genetic tests were performed. Therefore a total of five “types” of tests were used as dependent variables. These predictors are outlined in table 1 part C above. We were attempting to identify if certain characteristics of an individual or their family history predicted whether or not that individual received genetic testing. The predictors we used are outlined in table 1 parts B and C above.
Results

A total of 485 participants fully completed the questionnaires and were included in the statistical analysis. The mean age for the cohort was 96.35 months (8.03 years) of age. There was roughly a 4:1 ratio of males (80.2% of cohort) to females (19.8% of cohort). This coincides with previous research indicating ASDs are up to four times more common in males than females (Schaefer & Mendelsohn, 2008). The majority of the participants were probands (87.8% of cohort), non Hispanic (92.6% of cohort) and white (91.3% of cohort). The demographics of the entire cohort are detailed in table 2 below.
Table 2: Demographic Information

<table>
<thead>
<tr>
<th>Demographic Information</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (Mean Months)</td>
<td>96.35 (8.03 years)</td>
</tr>
<tr>
<td>Probands (%)</td>
<td>87.8</td>
</tr>
<tr>
<td>Affected Sibling of a Proband (%)</td>
<td>12.2</td>
</tr>
<tr>
<td>Male (%)</td>
<td>80.2</td>
</tr>
<tr>
<td>Female (%)</td>
<td>19.8</td>
</tr>
<tr>
<td>Hispanic (%)</td>
<td>7.4</td>
</tr>
<tr>
<td>Non-Hispanic (%)</td>
<td>92.6</td>
</tr>
<tr>
<td>White (%)</td>
<td>91.3</td>
</tr>
<tr>
<td>Black (%)</td>
<td>5.4</td>
</tr>
<tr>
<td>Asian (%)</td>
<td>4.9</td>
</tr>
<tr>
<td>Native Hawaiian or Other Pacific Islander (%)</td>
<td>0.2</td>
</tr>
<tr>
<td>American Indian/Alaska Native (%)</td>
<td>1.6</td>
</tr>
<tr>
<td>Other Race (%)</td>
<td>3.3</td>
</tr>
<tr>
<td>Median Household Income</td>
<td>$81,000-$100,999</td>
</tr>
<tr>
<td>Female Primary Caregiver's Education Level (Mean)</td>
<td>Some College/Associates - Bachelors</td>
</tr>
<tr>
<td>Male Primary Caregiver's Education Level (Mean)</td>
<td>Some College/Associates - Bachelors</td>
</tr>
</tbody>
</table>

Figures 1-6 featured below demonstrate the race, caregiver’s education, annual household income and autism diagnosis distributions of the entire cohort to show the characteristics of the participant population enrolled in the Autism Consortium. Figure 1 below is the racial distribution of the cohort analyzed.
The most common race was white and made up 91.3% of the cohort while black was 5.4% of the cohort and Asian was 4.9% of the cohort.

Figure 2 below is the distribution of the female primary caregiver’s education level of our cohort. The majority of respondents had a bachelor’s degree.
Figure 2: Female Primary Caregiver’s Education Distribution of Autism Consortium Enrollees

Figure 3 below is the distribution of the male primary caregiver’s education level of our cohort. The majority of respondents had a bachelor’s degree. More male caregiver’s had post graduate degrees than female primary caregivers.
Figure 3: Male Primary Caregiver’s Education Distribution of Autism Consortium Enrollees

![Male Primary Caregiver’s Education Distribution](chart.png)

Figure 4 below is the distribution of annual household incomes in our cohort. The majority of the respondents indicated annual household incomes between $81,000-$130,000 but income ranged from less than $20,000 to over $160,000.
Figure 4: Annual Household Income Distribution

Figure 5 below is the distribution of the autism spectrum diagnoses in the participants. Individuals diagnosed with PDD-NOS made up 47.1% of the cohort, individuals diagnosed with autistic disorder made up 36.3% of the cohort and individuals diagnosed with Asperger’s disorder made up 16.7% of the cohort.
Receipt of Genetic Testing Performed

We analyzed the genetic testing performed within the entire cohort. Of the entire cohort, approximately 53.6% of the participants received at least one genetic test while 46.4% did not receive any testing. Figure 6 below shows the overall percentage of participants that received the different types of genetic testing.
We also looked at the receipt of genetic testing among the specific ASD diagnoses. In general, children with an Asperger’s diagnosis were less likely to obtain genetic testing than those with an autistic disorder or PDD-NOS diagnosis. Only 22.5% of the children with an Asperger’s diagnosis in the cohort, received at least one genetic test. Therefore, in this cohort, 77.5% of children with an Asperger’s diagnosis did not receive any genetic testing. Of the children with an autistic disorder diagnosis, 62.1% of them received at least one genetic test. Similar to the children with an autistic diagnosis, 55.3% of the children with PDD-NOS received at least one genetic test. This information is summarized in figure 7 below.
Significant Predictors That Increase Receipt of Genetic Testing

Children with an accompanying diagnosis of intellectual disability were significantly more likely to have obtained at least one genetic test (p=0.009), fragile X testing (p=0.014), karyotype (p=0.031), CMA (p=0.014) and “other” genetic testing (p=0.001). In fact, the odds ratio that a child with intellectual disability obtained at least one genetic test was 3.3, 2.7 for fragile X testing, 2.2 for karyotype, 2.6 for microarray and 3.6 for “other” genetic testing. Similarly, children that have had at least one seizure were significantly more likely to have obtained at least one genetic test (p=0.024), fragile X (p=0.025), karyotype (p=0.042) and “other” genetic testing (p=0.011). The odds ratio that a child who has had at least one seizure would obtain at least one genetic test was 1.57, 1.54 for fragile X testing, 1.48 for karyotype and 1.68 for “other” genetic testing.
Accompanying seizures in a child was not a significant predictor for obtaining CMA testing. These predictors, along with the odds ratio are summarized below in table 3 parts A and B below.

Another characteristic that increased the likelihood of having obtained testing across all types of testing was having a family history of intellectual disability. Having a family history of intellectual disability significantly increased the likelihood of having obtained at least one genetic test (p=0.004), fragile X testing (p=0.007), karyotype (p=0.013), CMA (p=0.038) and “other” genetic testing (p=0.014) as compared to a participant without a family history of intellectual disability. Having a family history of intellectual disability increased the chance of obtaining any genetic testing by 0.85 times, fragile testing by 0.75 times, karyotype by 0.67 times, CMA by 0.55 times and “other” genetic testing by 0.75 times. These predictors along with the odds ratios are summarized in table 3 part C below.

Having any metabolic symptoms in the participant including, abnormal smell of the urine, abnormal body odor or having abnormally low energy, was a significant predictor of having obtained “other” genetic testing. A child with any of the above symptoms had an increased likelihood of having obtained “other” genetic testing (p=0.022) and were 80% more likely to have obtained “other” genetic testing than a child without any of these symptoms. These results are outlined in table 3 part D.

Lastly, females were significantly more likely to have obtained “other” genetic testing (p=0.000) than males. This was a strong predictor of receiving “other” genetic testing and is also summarized in table 3 part E below.
Table 3: Summary of Predictors that Increase Receipt of Genetic Testing

<table>
<thead>
<tr>
<th>Predictor</th>
<th>Tests</th>
<th>P-Value</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. Intellectual Disability in Child*</td>
<td>At Least One Genetic Test</td>
<td>0.009</td>
<td>3.3</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.014</td>
<td>2.7</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.031</td>
<td>2.2</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.014</td>
<td>2.6</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.001</td>
<td>3.6</td>
</tr>
</tbody>
</table>

| B. Seizures in Child* | At Least One Genetic Test     | 0.024   | 1.57       |
|                       | Fragile X                     | 0.025   | 1.54       |
|                       | Karyotype                     | 0.042   | 1.48       |
|                       | CMA                           | 0.379   | -          |
|                       | "Other" Genetic Test          | 0.011   | 1.68       |

| C. Intellectual Disability in Blood Relative* | At Least One Genetic Test     | 0.004   | 1.85       |
|                                              | Fragile X                     | 0.007   | 1.75       |
|                                              | Karyotype                     | 0.013   | 1.67       |
|                                              | CMA                           | 0.038   | 1.55       |
|                                              | "Other" Genetic Test          | 0.014   | 1.75       |

| D. Any Metabolic Disorder Symptoms in Child* | "Other" Genetic Test          | 0.022   | 1.8        |

| E. Females | "Other" Genetic Test          | 0.000   | 2.8        |

* Indicates independent regression model meaning this was the only predictor used when calculating the odds ratio of whether or not a child would receive genetic testing.

Significant Predictors That Decrease Receipt of Genetic Testing

Demographic predictors that significantly decrease the likelihood of obtaining any of the five types of testing were age of the participant and whether the participant was a proband or an affected sibling of a proband. As age increased, participants were less likely to have obtained at least one genetic test (p=0.001), fragile X (p=0.001), karyotype (p=0.002), CMA (p=0.000) and “other” genetic testing (p=0.034). Being an affected
sibling of a proband, significantly reduced the likelihood of having obtained at least one genetic test (p=0.001), fragile X (p=0.001), karyotype (p=0.004), CMA (p=0.035) and “other” genetic testing (p=0.046). Neither of these were strong predictors. These predictors along with their odds ratios are summarized in table 4 parts A and B below.

Another factor that significantly reduced the likelihood that a child on the spectrum received genetic testing was having a family history of Asperger disorder. This was only significant for receipt of at least one genetic test and CMA testing. These also were weak predictors of decreasing the likelihood of having obtained genetic testing. These predictors are summarized in table 4 part C below.

The diagnosis of the child also influenced receipt of genetic testing. An Asperger diagnosis in the child significantly reduced the likelihood that a child would have received genetic testing. Although a weak predictor, this diagnosis significantly reduced the likelihood for having obtained at least 1 genetic test, (p=0.000), fragile X (p=0.000), karyotype (p=0.000), CMA (p=0.000) and “other” genetic testing (p=0.001). An autistic disorder diagnosis reduced the likelihood of having obtained CMA testing (p=0.022) as compared to a child without this ASD diagnosis. A PDD-NOS diagnosis reduced the likelihood of having obtained at least one genetic test (p=0.009) and CMA testing (p=0.01). These were also weak predictors and are summarized in table 4 parts D, E and F below.

A child that had an affected sibling with any ASD diagnosis also reduced the likelihood of having obtained genetic testing as compared to a child without any ASD affected siblings. This was significant for at least one genetic test (p=0.000), fragile X
(p=0.000), karyotype (p=0.001) and CMA (p=0.02). These are summarized in table 4 part G below.

Lastly, a family history of “other” birth defects significantly reduced the likelihood of having obtained fragile X testing (p=0.036). This was not a strong predictor and is also summarized in table 4 part H below.
Table 4: Summary of Predictors that Decrease Receipt of Genetic Testing

<table>
<thead>
<tr>
<th>Predictor</th>
<th>Tests</th>
<th>P-Value</th>
<th>Odds Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. Age of Child</td>
<td>At Least One Genetic Test</td>
<td>0.001</td>
<td>0.995</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.001</td>
<td>0.994</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.002</td>
<td>0.994</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.000</td>
<td>0.992</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.034</td>
<td>0.995</td>
</tr>
<tr>
<td>B. Proband vs. Sibling</td>
<td>At Least One Genetic Test</td>
<td>0.001</td>
<td>0.283</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.001</td>
<td>0.266</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.004</td>
<td>0.387</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.035</td>
<td>0.472</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.046</td>
<td>0.416</td>
</tr>
<tr>
<td>C. Family History of Aspergers</td>
<td>At Least One Genetic Test</td>
<td>0.05</td>
<td>0.41</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.583</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.61</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.017</td>
<td>0.336</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.1</td>
<td>-</td>
</tr>
<tr>
<td>D. Asperger's Diagnosis in Child</td>
<td>At Least One Genetic Test</td>
<td>0.000</td>
<td>0.085</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.000</td>
<td>0.15</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.000</td>
<td>0.169</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.000</td>
<td>0.033</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.001</td>
<td>0.098</td>
</tr>
<tr>
<td>E. Autistic Disorder Diagnosed in Child</td>
<td>At Least One Genetic Test</td>
<td>0.059</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.768</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.665</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.022</td>
<td>0.373</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.932</td>
<td>-</td>
</tr>
<tr>
<td>F. PDD-NOS Diagnosed in Child</td>
<td>At Least One Genetic Test</td>
<td>0.009</td>
<td>0.364</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.271</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.332</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.01</td>
<td>0.328</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.133</td>
<td>-</td>
</tr>
<tr>
<td>G. Child with Any ASD Affected Sibling*</td>
<td>At Least One Genetic Test</td>
<td>0.000</td>
<td>0.391</td>
</tr>
<tr>
<td></td>
<td>Fragile X</td>
<td>0.000</td>
<td>0.402</td>
</tr>
<tr>
<td></td>
<td>Karyotype</td>
<td>0.001</td>
<td>0.448</td>
</tr>
<tr>
<td></td>
<td>CMA</td>
<td>0.02</td>
<td>0.02</td>
</tr>
<tr>
<td></td>
<td>&quot;Other&quot; Genetic Test</td>
<td>0.359</td>
<td>-</td>
</tr>
<tr>
<td>H. Family History of &quot;Other&quot; Birth Defects</td>
<td>Fragile X</td>
<td>0.036</td>
<td>0.566</td>
</tr>
</tbody>
</table>

* Indicates independent regression model meaning this was the only predictor used when calculating the odds ratio of whether or not a child would receive genetic testing
Non-significant Predictors of Genetic Test Utilization

Across all types of testing, the demographic characteristics not significant for predicting genetic test utilization included child ethnicity, race, household income level and primary caretaker’s education. Gender of the child was only significant for females in increasing “other testing”, and was not a significant predictor for the remainder of the tests analyzed.

Several of the family history and child characteristics that were analyzed were not significant predictors of genetic test utilization. A family history of seizures was not a significant predictor for any of the test types. Likewise, birth defects in the child were not significant predictors for any test types. A family history of “other” birth defects was only a significant predictor of fragile X testing. The remaining birth defect history in a family was not significant at predicting utilization of any of the other genetic tests.
Discussion

Overall, about half of the research participants had not obtained genetic testing. There were many predictors that significantly reduced the likelihood that a child would obtain a genetic test and fewer that would significantly increase the likelihood of obtaining genetic testing. Some of these predictors were surprising and unexpected from what we originally hypothesized. Overall, it represents the variability of services that a child on the spectrum receives. This study begins to identify factors that influence receipt of genetic testing among children with an ASD.

Hypothesis 1: A Significant Portion of Patients with ASD Are Not Receiving Any Genetic Testing

There is a large discrepancy between the recommendations of genetically testing children with autism and the utilization of genetic tests for this population of children. Of the children enrolled in the study, 46.4% had not received any genetic testing. Although receipt of genetic testing is slightly higher in this study, our findings were similar to those of McLennan et al. in which 1/3 of the children with autism received genetic testing (McLennan, Huculak, & Sheehan, 2008). Both the American College of Medical Genetics and American Academy of Pediatrics published guidelines for a genetic evaluation of children on the spectrum in 2008 and 2000, respectively, so it is surprising that despite the guidelines, many children with an ASD had not received genetic testing.
when the Autism consortium data was collected between 2006 and 2010. There are several possible reasons why we observed a low uptake of genetic testing. One of the reasons we may have seen low uptake of genetic testing is because there are still a large portion of families who do not believe autism is caused by genetic factor. In fact, 41.8% of families believe autism is caused by other factors (i.e. environmental factors, genetic susceptibility to environmental trigger, immune system differences/weaknesses) (Selkirk, McCarthy Veach, Lian, Schimmenti, & LeRoy, 2009). However, the families who participated in the Autism Consortium research were aware that this study was trying to uncover genetic factors causing ASDs. Therefore, we would expect that families who participated in this study believed that the etiology of ASDs were genetic. Another reason may be that physicians caring for these individuals may not know or understand the utility of a genetic evaluation and therefore are not referring their patients to a genetics team. There are many possible explanations for this discrepancy and future research is implicated to decipher why we are seeing this.

Hypothesis 2: Individuals with More Severe ASD or With Accompanying Intellectual Disability and/or Seizures Are More Likely to Obtain Genetic Testing Than Those with Milder ASD

We hypothesized some factors would increase the receipt of genetic testing among children with an ASD. We expected that individuals with a more severe diagnosis of ASD or that have accompanying intellectual disability and/or seizures would be more likely to obtain genetic testing as compared with children without these characteristics. This hypothesis was found to be true for the presence of intellectual disability across all
of the different types of tests. The presence of seizures also increased the likelihood of having obtained genetic testing significantly except for CMA. A child who was showing any metabolic symptoms significantly increased the likelihood that they would have obtained “other” genetic testing, also supporting the hypothesis that receipt of genetic testing increases as severity of the case increases.

When looking at the different diagnoses in the participant, Asperger syndrome is considered to be at the milder end of the spectrum. We did find that participant’s with Asperger syndrome were significantly less likely to obtain genetic testing across all of the different tests, thus confirming the hypothesis. Surprisingly, a participant with autistic disorder or a PDD-NOS diagnosis in comparison to participants without these diagnoses decreased the likelihood of obtaining CMA even though these diagnoses are considered to be the more severe forms of ASD. A PDD-NOS diagnosis in the participant also decreased the likelihood of obtaining at least one genetic test as compared to children without this diagnosis. It is unclear why a participant with a diagnosis considered to be more severe would be less likely to obtain genetic testing. Therefore, in the case of CMA testing among participants diagnosed with ASD, our hypothesis was disproved.

**Hypothesis 3: Individuals with Birth Defects or Dysmorphic Features Are More Likely to Obtain Genetic Testing**

We also hypothesized that individuals with birth defects or dysmorphic features would be more likely to obtain genetic testing. Neither having at least one birth defect (i.e. heart, kidney, “other”) or any of the specific birth defects asked about in the questionnaire significantly increased or decreased receipt of any of the genetic tests
analyzed. This was a surprising finding especially because in clinical practice, birth defects usually warrant either a CMA or karyotype or both to rule out large chromosome abnormalities or small duplications or deletions (Pletcher et al., 2007). Interestingly, a child with a family history of “other” birth defects was significantly less likely to have obtained fragile X testing. Perhaps the data demonstrates that physicians are ordering CMA and karyotype and not fragile X testing, although we would have expected significance in this case.

**Hypothesis 4: Individuals Who Have a Sibling with an ASD Will Be More Likely To Have Had Genetic Testing**

This hypothesis was disproved. In comparison to a child without affected siblings, a child with affected siblings was significantly less likely to obtain all types of genetic testing except for “other” genetic testing. We would expect having multiple children in a family affected with an ASD increased the chance of a genetic link. The reason for this finding is unclear; however, this may also be explained by testing only the most severely affected child in the family and only testing subsequent affected individuals if a genetic abnormality is identified.

**Hypothesis 5: Individuals Who Have a Family History of an ASD Will Be More Likely to Have Had Genetic Testing**

Surprisingly, a family history of ASDs was not significant at predicting receipt of genetic testing except for a family history of Asperger disorder. If a child had a family history of Asperger disorder, the likelihood of this child to obtain genetic testing was
significantly reduced as compared to participants without a family history of Asperger disorder. We would expect a family history of ASDs to contribute to case severity, such as in the case of a family history of intellectual disability, because multiple individuals affected in the same family would signify a genetic link between the cases. Physicians may have more of a chance of identifying a genetic abnormality in these families because they may have more family members to evaluate and could perhaps shed light on to which tests should be ordered. Since a family history of Asperger disorder decreased the likelihood of obtaining testing, it could be that these tend to be the more mild forms of ASD and this alone could decrease receipt of genetic testing as we saw in the first hypothesis. As for the other ASD diagnoses, it is unclear why a family history of ASDs did not significantly increase the likelihood that an individual would have received genetic testing.

Additional Findings

One surprising finding was that affected siblings of probands were significantly less likely to obtain genetic testing across all types of testing analyzed than if they were not siblings of probands. One possible explanation of this finding is that if a proband in a family was tested and was negative for the tests, geneticists may be less inclined to test an affected sibling of that proband. The proband may be the most severely affected individual, hence why he/she presented first, and therefore if he/she is negative for any of the genetic tests ordered, the likelihood of finding a positive result in a more mildly affected sibling would be low. During practice, if a physician is following multiple children from the same family they may be more inclined to continue testing the most
severely affected child because this would increase the chance of finding a genetic abnormality. However, evaluating an affected sibling may be helpful for a clinical geneticist to try and identify a genetic etiology (Schaefer & Mendelsohn, 2008).

We found that the older participants in the study were significantly less likely to have obtained genetic testing. We would expect that this would be relatively constant for children on the spectrum because as they age, different genetic tests become available and we would expect that as these new tests are developed, children on the spectrum would be tested. However, this was not the case in this cohort of people. Perhaps when children start showing symptoms of ASD, typically before three years old, they are more likely to receive genetic testing versus waiting several years before testing these children. It is also possible that these children are not being re-evaluated after these new tests become available when they are older, which could also explain this finding.

Another significant predictor increasing the likelihood of obtaining genetic testing was the presence of a family history of intellectual disability. This was significant across all types of genetic testing. It is understandable why a family history of intellectual disability would increase the likelihood of obtaining genetic testing in a child. The American Academy of Pediatrics also published guidelines for evaluating a child with intellectual disability and the among the recommendations were taking a detailed family history and obtaining a karyotype, fragile X testing and metabolic testing (Moeschler & Shevell, 2006). Therefore, a family history of intellectual disability should warrant genetic testing. In general, as the severity of the case increases, the likelihood of utilizing genetic tests increases. We would expect that a family history of intellectual disability would be considered when deciding which type of testing is warranted. However, a
family history of seizures was not a significant predictor of having obtained genetic testing. We would expect if a family history of intellectual disability would increase the likelihood of obtaining genetic testing, a family history of seizures would too because it may be taken into account by a physician ordering genetic testing. However, having a family history of seizures was not a significant factor for increasing genetic test utilization and the reason for this is unclear.

Limitations

For this study we had a large set of participants. The diversity of the sample was limited because the majority of the participants were white, non-Hispanic, and had educated parents. The sample size of each of the different ASD diagnoses was small and may have influenced the statistical significance identified in the study.

There are other attributes of the dataset that may have influenced the resulting statistics. For example, patients were recruited in different ways to participate in the Autism Consortium including clinical referrals and community referrals. Each of these referral patterns may have influenced the findings and/or the medical, family and genetic testing history responses of the participants. Participants who were clinically referred came from a variety of institutions that may have had varying protocols regarding genetic evaluation of children on the spectrum. In addition, each physician performs their evaluation of children with an ASD differently which may have also influenced which genetic tests were performed. In comparison, the community referred families had children whose medical providers were not affiliated with any of the participating medical centers, and we therefore do not have any information about their usual practices.
regarding genetic testing and/or referral to genetic services. Therefore, looking at the entire dataset it would be important to separate participants by referral type to the Autism Consortium database. At this point, we have not determined if the children who did not receive genetic testing came primarily from community referrals or from clinical referrals. This information would help us further characterize which children are not receiving any genetic testing.

Additionally, since this study collected information over several years. The institutional guidelines may have changed during this time and may have been influenced by the introduction of the American College of Medical Genetics guidelines in regard to genetic evaluations of children on the spectrum that were also published during this time frame.

Another limitation of this study is the fact that participant data was obtained through self-report and not through medical record analysis. There may have been inconsistencies between events that actually happened and what was reported. There may have also been recall bias among the respondents filling out the questionnaire.

**Future Research**

This study examined the rate of genetic testing in a group of children with ASDs and identified different factors that may influence which types of genetic testing children on the spectrum are receiving. It would be interesting to see the reasons behind some of these decisions both from a physician and parent’s point of view. From this data, we cannot determine if the decision to forgo genetic testing was made by the child’s physician or by their parents. Perhaps, a physician’s decisions in regard to ordering
genetic testing may be influenced by institutional policy as well as published practice
guidelines. Some physicians, not specifically trained in genetics, may not see the clinical
utility of testing children on the spectrum. Similarly, parents may not feel that genetic
testing would change clinical care for their child. Physicians may also be influenced by
parental perspectives of the cause of their child’s ASD and/or the feasibility for a family
to pay for genetic testing or if testing is covered by insurance. It would interesting to see
what influences physicians decisions with regard to genetic testing among children with
ASDs. In addition to seeing how physician’s are influenced, it would be just as important
to see what influences the decisions of parents of children on the spectrum. If a parent
does not believe there is an underlying genetic abnormality that is causing their child’s
ASD, they may be less apt to follow through with a genetics appointment referral. Parents
who believe that ASD is caused by environmental factors may be more apt to decline
genetic testing. By identifying the influences on parents and physicians, it may help us
further classify why a large percentage of these children are not receiving testing. Future
studies to parse out the discrepancy between the recommendations and the actual
utilization of genetic testing would help further classify this disconnect and would be a
way to find out where efforts should be focused.
Conclusion

This study not only confirmed previous studies findings that a large portion of children with ASD are not receiving genetic testing, but also suggests that there may be specific factors that influence receipt of genetic testing among these children. In this study, almost 50% of the participants did not receive any genetic testing. We were able to identify several factors that influenced whether or not a child would receive genetic testing. In general, a more severe case of ASD, such as the presence of intellectual disability or seizures, increased the likelihood of obtaining genetic testing. However, the presence of birth defects in the child or a family history of birth defects did not seem to influence the receipt of genetic testing. Therefore these factors did not seem to attribute to case severity. We found that fewer children with Asperger disorder were receiving testing as compared to autistic disorder and PDD-NOS disorder. Other factors that reduced the likelihood of obtaining testing included, having a sibling affected with an ASD and having a family history of ASDs. This was unexpected, as a family history usually alerts clinicians that a genetic etiology is more likely. Although we had a large sample size, when we divided up by type of ASD, each cohort was small. If the cohorts of each type of ASD were larger, the findings that were specific to ASD diagnosis may have been different. Although about half of the participants in the cohort did not receive any genetic testing, it is unclear if this lack of uptake of genetic testing among children with ASD is because of physician’s not ordering the tests or referring these children for a
genetics evaluation or if it is the parents of children not following through with recommendations, declining testing because of their beliefs or because of financial restrictions for the family. Future research efforts should focus on determining why it appears that children with ASD are not receiving genetic testing since the guidelines published by the American College of Medical Genetics and American Academy of Pediatrics with regard to genetic evaluation of children on the spectrum, have been in the literature since 2009 and 2000, respectively.
References:


