Chromosomal Microarray for the Genetic Evaluation of Patients With Global Developmental Delay, Intellectual Disability, and Autism Spectrum Disorder

EXECUTIVE SUMMARY

Background

Global developmental delay (GDD) is diagnosed in children 5 years or younger who show significant delay in 2 or more developmental domains: gross or fine motor, speech/language, cognitive, social/personal, and activities of daily living. GDD can precede the development of intellectual disability (ID) as the child ages. ID is accompanied by significant limitations in intellectual functioning and adaptive behavior, diagnosed at or after age 5, but prior to age 18 and is life-long. Prevalence estimates of GDD and ID range from 1% to 3%. Both are influenced by environmental, infectious, perinatal factors, and genetics (~450 implicated genes). Congenital anomalies are often present in children with GDD and ID. A suspected etiology can often be established from history and physical examination (in skilled hands as much as 20%-40% of cases) without genetic testing. If no diagnosis is suspected, fragile X syndrome testing, metabolic testing for inborn errors of metabolism, and chromosomal microarray (CMA) testing (without karyotyping) are part of the initial recommendations—regardless of presence or absence of dysmorphic features or congenital anomalies.

In autism spectrum disorder (ASD) communication and social interaction deficits are apparent along with restricted repetitive behavior, interests, or activities. In 2010, the estimated prevalence ASD among 8-year-olds was 14.7 per 1000 or 1 in 68. An accurate diagnosis can generally be made by age 2 with evaluation including developmental screening and diagnostic testing (i.e., hearing, vision, neurologic, laboratory evaluation for metabolic disorders, and genetic testing). A substantial body of evidence supports a genetic etiology in ASD.

The goal of a genetic evaluation is to identify chromosomal imbalances that cause a disorder. The most common genetic imbalances are copy number variants (CNVs) or deletions and duplications of large segments of genomic material. CNVs are common in GDD/ID and ASD but can often reflect normal genetic variation. However, de novo CNVs are observed about 4 times more frequently in children with ASD than in normal individuals.

Until recently, the first step in cytogenetic analysis was karyotyping, which evaluates all chromosomes. Karyotyping offers limited diagnostic ability—e.g., in ID establishing a genetic etiology in about 3% of cases. Molecular cytogenetic techniques including array comparative genomic hybridization and single-nucleotide polymorphisms arrays (both referred to as a chromosomal microarrays) can detect small CNVs not evident with karyotyping. Fluorescent in situ hybridization, a targeted approach, is used to identify specific chromosomal abnormalities associated with suspected diagnoses such as DiGeorge syndrome.

Objective

The objective of this Special Report is to summarize the evidence on chromosomal microarray testing to identify CNVs in children with GDD, ID, and ASD.
Search Strategy
MEDLINE® was searched (via PubMed) through February 24, 2015, and updated on June 24, 2015, to identify studies of diagnostic yield and clinical utility in children with GDD/DD or ASD undergoing CMA testing.

Selection Criteria
We included case series or cohort studies that enrolled 20 or more patients with clinical diagnoses of GDD/ID or ASD with known or suspicion of genetic abnormalities, with or without negative results by conventional cytogenetic evaluation, and that conducted CMA testing on enrolled patients. Studies were also included if they examined management decisions and/or patient outcomes based on genetic evaluation results. We also sought to identify studies examining CMA technical performance.

Main Results

Analytic Validity
Acceptable analytic validity is generally assumed for CMA testing based on laboratories meeting quality standards under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), including participation in proficiency testing. Reports of analytic validity specific to CMA are not readily identified. Data supporting the analytic validity of the CytoScan Dx® Assay for use in children with ID and GDD were included as part of the U.S. Food and Drug Administration 510(k) clearance.

Clinical Validity
For common phenotypes and syndromes, the pathogenicity of CNVs is supported by considerable evidence; for uncommon phenotypes and uncommon CNVs, determining pathogenicity requires a systematic evaluation that includes parental studies, examining databases for reported associations, and considering the molecular consequences of the identified variant. The recommend categories of clinical significance for reporting are: pathogenic, uncertain clinical significance (likely pathogenic, likely benign, or no subclassification), or benign.

Included studies of diagnostic yield were heterogeneous with respect to inclusion criteria, dates, prior normal studies (most series), and array platform. There was variability of diagnostic yields: studies including patients with GDD/ID ± ASD (median, 13.6%; 55 studies; interquartile range [IQR], 9.5%-17.2%; median from 21 studies published 2012 or later, 19.0%), and from those categorized as examining primarily ASD (median, 8.4%; 12 studies; IQR, 7.2%-17.3%; median from 4 studies published 2012 or later, 12.3%). When examined according to year of publication, there was on average just over 1% annual improvement in diagnostic yield.

Clinical Utility
We identified 5 retrospective studies and 1 database analysis that examined a potential impact of CMA results on clinical decisions. Collectively, the studies found identified pathogenic variants can prompt clinical actions potentially impacting morbidity. Less clear is how often outcomes will be improved and in which cases interventions might have occurred in the absence of testing. The proportion that may benefit will depend on the variants identified as well as diagnostic yield, which in turn depends on phenotype severity. Saam et al noted that CMA testing may impact the search for a diagnosis (further genetic testing avoided in 12/48 patients). Two studies concluded that having a child with ASD increases the risk of reproductive stoppage or avoiding future children. Another study suggested that genetic evaluation of families at increased risk of having a child with ID affects reproductive decisions and fertility rate.
Author Conclusions and Comment

The ability to detect pathogenic CNVs underlying GDD/ID and ASD is improving. This improvement is likely due to higher CMA resolution along with increasingly extensive data concerning CNV pathogenicity and associated phenotypes and availability of those data. Professional societies have recommended CMA testing as first-line evaluation when genetic evaluation is desired as opposed to first obtaining a karyotype.\textsuperscript{3,10,20} Data supporting analytic validity are readily available only for the Affymetrix CytoScan® Dx assay, but laboratories meeting CLIA standards but using other platforms would be expected to achieve adequate technical performance. There is consistent evidence that the diagnostic yield obtained from CMA testing is higher than with karyotyping in children with GDD/ID or ASD, with or without congenital anomalies.

Establishing the pathogenicity of detected CNVs relies on evidence, informatics, and genetics expertise. A particular challenge when considering the evidence and methods used to determine variant pathogenicity is that, outside the more common syndromes, diagnoses include a large number of rare disorders a clinician, even a specialist, might not encounter during a lifetime. Identifying a pathogenic variant can: (1) impact the search for a diagnosis, (2) inform follow-up that can benefit a child by reducing morbidity, and (3) affect reproductive planning for parents and potentially the affected patient. Finally, we were unable to identify case reports of incorrect diagnoses; how often they might occur is unclear.

For families desiring a genetic diagnosis and what might follow from one, CMA testing can establish a diagnosis more often than other approaches such as karyotyping. Still, other assays identify genetic alterations not detected by an array. The complexities of CMA testing, interpretation, understanding its limitations, and the potential implications requires that testing is obtained by clinicians with genetic expertise, that families receive genetic counseling, and that testing be performed in laboratories meeting recommended molecular pathology standards.

References


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Chromosomal Microarray for the Genetic Evaluation of Patients With Global Developmental Delay, Intellectual Disability, and Autism Spectrum Disorder

OBJECTIVE

The objective of this Special Report is to summarize the evidence on chromosomal microarray (CMA) testing to identify copy number variants (CNVs) in children with global developmental delay (GDD), intellectual disability (ID), and autism spectrum disorder (ASD).

BACKGROUND

Global Developmental Delay and Intellectual Disability

GDD is diagnosed in children 5 years or younger who show significant delay in 2 or more developmental domains: gross or fine motor, speech/language, cognitive, social/personal, and activities of daily living. GDD can precede the development of ID as the child ages.

ID is manifest by significant limitations in intellectual functioning and adaptive behavior. It is diagnosed at or after age 5 (when intelligence testing is considered valid and reliable) but prior to age 18 and is lifelong. The Diagnostic and Statistical Manual of Mental Disorders: Fifth Edition (DSM-5), of the American Psychiatric Association, defines ID as occurring during the developmental period and involving impairments of general mental abilities (eg, IQ <70 or 75) that impact adaptive functioning in the conceptual, social, and practical domains.

Prevalence estimates of GDD and ID range from 1% to 3%. Both are influenced by genetic, environmental, infectious, and perinatal factors. Approximately 450 genes have been causally related to ID—most genes (>90%) associated with syndromes. Inheritance of ID can be autosomal dominant, recessive, or X-linked; and most nonsyndromic genes are located on the X chromosome. Prior to the advent of whole exome and genome sequencing, Willemsen and Kleefstra concluded that 20% to 40% of ID cases could be attributed to a genetic variant. With use of whole genome sequencing, they estimated almost 60% of cases have an identifiable genetic etiology.

Congenital anomalies are frequently present in children with GDD and ID. In addition, a suspected etiology can often be established from history and physical examination (in skilled hands as much as 20% to 40% of cases) without genetic testing. The recommended approach to evaluation in GDD/ID begins with history including a 3-generation family history and physical exam (including neurologic). Subsequent testing is used to confirm a suspected diagnosis (eg, targeted fluorescent in situ hybridization [FISH] testing for DiGeorge or cri-du-chat syndromes). If no diagnosis is suspected, fragile X syndrome testing, metabolic testing for inborn errors of metabolism, and CMA testing (without karyotyping) are

<table>
<thead>
<tr>
<th>Abbreviations and Acronyms</th>
<th>Descriptions</th>
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<tbody>
<tr>
<td>aCGH</td>
<td>array comparative genomic hybridization</td>
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<td>ACMG</td>
<td>American College of Medical Genetics and Genomics</td>
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<tr>
<td>ASD</td>
<td>autism spectrum disorder</td>
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<tr>
<td>CLIA</td>
<td>Clinical Laboratory Improvement Amendments of 1988</td>
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<tr>
<td>CMA</td>
<td>chromosomal microarray</td>
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<tr>
<td>CNV</td>
<td>copy number variant</td>
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<tr>
<td>DSM-5</td>
<td>Diagnostic and Statistical Manual of Mental Disorders: Fifth Edition</td>
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<tr>
<td>FDA</td>
<td>Food and Drug Administration</td>
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<tr>
<td>FISH</td>
<td>fluorescent in situ hybridization</td>
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<tr>
<td>GDD</td>
<td>global developmental delay</td>
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<tr>
<td>ID</td>
<td>intellectual disability</td>
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<tr>
<td>IQR</td>
<td>interquartile range</td>
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<tr>
<td>ISCA</td>
<td>International Standard Cytogenomic Array Consortium</td>
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<tr>
<td>LDT</td>
<td>laboratory-developed test</td>
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<tr>
<td>MCA</td>
<td>multiple congenital anomaly</td>
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<tr>
<td>SNP</td>
<td>single-nucleotide polymorphism</td>
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<tr>
<td>UP</td>
<td>uniparental disomy (UP)</td>
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recommended—regardless of the presence or absence of dysmorphologic features or congenital anomalies.¹

### Autism Spectrum Disorders

*DSM-5*³ defines ASD⁸ by the presence of:

- Persistent deficits in social communication and social interaction across multiple contexts,
- Restricted, repetitive patterns of behavior, interests, or activities,
- Symptoms must be present in the early developmental period (typically recognized in the first 2 years of life), and
- Symptoms cause clinically significant impairment in social, occupational, or other important areas of current functioning.

In 2010, the estimated prevalence of ASD among 8-year-olds was 14.7 per 1000 or 1 in 68.⁶ ASD is 4 to 5 times more common in boys than girls, and white children are more often identified with ASD than black or Hispanic children. An accurate diagnosis can generally be made by age 2. Evaluation includes developmental screening and diagnostic evaluation (ie, hearing, vision, neurologic, laboratory testing for metabolic disorders, and genetic testing).

A large body of evidence supports a genetic etiology in ASD. Twin studies estimate heritability between 60% and 90%.⁹ A family with an affected child has a 13% to 19% risk for recurrence in subsequent children.¹⁰ Based on Swedish genetic studies, Gaugler et al recently concluded that “the bulk of autism arises from genetic variation” (as opposed to environmental causes).¹¹ Still, although genetic determinants can be heritable, most appear to arise de novo.⁹

For these reasons, a child with ASD is often evaluated with genetic testing. Testing may be targeted when a child has a recognizable syndrome such as those shown in Table 1. Alternatively, high-resolution cytogenetic analysis evaluating multiple genes—the focus of this Special Report—is used.

### Table 1. Examples of Specific Genes Associated With Disorders That Include Autistic Behaviors

<table>
<thead>
<tr>
<th>Gene (Syndrome)</th>
<th>Patient Selection</th>
<th>Yield</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>FMR1</em> (fragile X)</td>
<td>Unselected autism</td>
<td>3%-10%</td>
<td>Schaefer and Mendelsohn (2008)¹²</td>
</tr>
<tr>
<td><em>MECP2</em> (Rett)</td>
<td>Females with nonsyndromic autism, intellectual disability, and cerebral palsy</td>
<td>3%-13%</td>
<td></td>
</tr>
<tr>
<td><em>PTEN</em></td>
<td>Autism with macrocephaly</td>
<td>≤17%</td>
<td>Butler et al (2005)¹³</td>
</tr>
</tbody>
</table>

### Cytogenetic Analysis

#### Karyotyping and FISH

The goal of a cytogenetic evaluation is to identify chromosomal imbalances that cause a disorder. The most common imbalances are CNVs or deletions and duplications of large segments of genomic material. CNVs are common in GDD/ID and ASD but more often reflect normal genetic variation.¹⁴ However, de novo CNVs are observed about 4 times more frequently in children with ASD than in normal individuals.⁹ Less frequently, other abnormalities such as balanced translocations (ie, exchanges of equally sized DNA loci between chromosomes) may be pathogenic. For many well-described syndromes, the type and location of the associated chromosomal abnormality have been established by studying large patient samples. For others, few patients with similar abnormalities may have been evaluated to establish genotype-phenotype correlation. Finally, in some patients, cytogenetic analysis will discover chromosomal abnormalities that require study to determine their significance.

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Prior to the advent of CMAs, the initial step in cytogenetic analysis was G-banded karyotyping, which evaluates all chromosomes. High-resolution G-banding can detect changes as small as 3 to 5 megabases (Mb) in size, although standard G-banding evaluates more than 10-Mb changes. In children with GDD/ID, 1 review found G-banded karyotyping diagnostic in approximately 3% to 5%. In ASD, high-resolution karyotyping appears to identify abnormalities in up to 5% of cases.

In contrast, molecular cytogenetic techniques can detect small submicroscopic chromosomal alterations. FISH, a targeted approach, is used to identify specific chromosomal abnormalities associated with suspected diagnoses such as DiGeorge syndrome. Prior to CMAs, FISH was also used to screen the rearrangement-prone subtelomeric regions. Subtelomeric FISH was found to identify abnormalities in children with GDD and ID, diagnostic in approximately 5% to 6% of those with negative karyotypes, but uncommonly in ASD.

**Chromosomal Microarrays**

Two types of CMAs are considered here: array comparative genomic hybridization (aCGH) and single-nucleotide polymorphism (SNP) arrays. The aCGH approach uses DNA samples from a patient and a normal control. Each is labeled with distinct fluorescent dyes (red or green). The labeled samples are then mixed and hybridized to thousands of cloned or synthesized reference (normal) DNA fragments of known genomic locus immobilized on a glass slide (microarray) to conduct thousands of comparative reactions simultaneously. CNVs are determined by computer analysis of the array patterns and intensities of the hybridization signals. If the patient sequence is missing part of the normal sequence (deletion) or has the normal sequence plus additional genomic material within that genomic location (eg, a duplication of the same sequence), the sequence imbalance is detected as a difference in fluorescence intensity (Korf and Rehm offer an illustrative graphic). For this reason, aCGH cannot detect balanced chromosomal translations (equal exchange of material between chromosomes) or sequence inversions (same sequence is present in reverse base pair order) because the fluorescence intensity would not change.

Like aCGH, SNP arrays detect CNVs. In a SNP array, the 2 alleles for genes of interest are tagged with different fluorescent dyes. Comparative florescence intensity will be increased when there are duplications and diminished with deletions. The resolution provided by aCGH is higher than that with SNP arrays. In addition, aCGH has better signal-to-background characteristics than SNP arrays. In contrast to aCGH, SNP arrays will also identify long stretches of DNA homozygosity, which may suggest uniparental disomy (UPD) or consanguinity. UPD occurs when a child inherits 2 copies of a chromosome from 1 parent and no copies from the other parent. UPD can lead to syndromes such as Angelman and Prader-Willi.

Table 2 summarizes the cytogenetic tests used to evaluate children with GDD/ID and autism. The table emphasizes the large difference in resolution between karyotyping and CMA.

**Table 2. Resolution and Analysis Comparison of FISH, Karyotyping, and CMA**

<table>
<thead>
<tr>
<th>Test</th>
<th>Resolution in Kilobases</th>
<th>Analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Karyotyping</td>
<td>3000-5000 kb</td>
<td>Genome-wide</td>
</tr>
<tr>
<td>CMA</td>
<td>≈50 kb</td>
<td>Genome-wide</td>
</tr>
<tr>
<td>FISH</td>
<td>≈500 to 1000 kb (depending on probe)</td>
<td>Targeted</td>
</tr>
</tbody>
</table>

CMA: chromosomal microarray; FISH: fluorescent in situ hybridization; kb: kilobases.

a One kb = 1000 bases, 1000 kb = 1 Mb.

Finally, although current diagnostic strategies using CMA focus primarily on identification of CNVs underlying GDD/ID and ASD, sequence variants detectable by exome sequencing or other methods, are also implicated.
Guidelines and Consensus Statements

American Academy of Neurology and Child Neurology Society

The American Academy of Neurology and the Child Neurology Society updated their guidelines on the evaluation of unexplained GDD/ID with information on genetic and metabolic (biochemical) testing in order to accommodate advances in the field. The guidelines conclude that CMA testing has the highest diagnostic yield in children with GDD/ID and that the often complex results require confirmation and careful interpretation, often with the assistance of a medical geneticist, and that CMA testing should be considered first line. The guidelines note that “Research is sorely lacking on the medical, social, and financial benefits of having an accurate etiologic diagnosis.”

American College of Medical Genetics and Genomics

The American College of Medical Genetics and Genomics (ACMG) published guidelines on array-based technologies and their clinical use for detecting chromosomal abnormalities. The guidelines note: “The increased resolution of microarray technology over conventional cytogenetic analysis allows for identification of chromosomal imbalances with greater precision, accuracy, and technical sensitivity.” CMA testing for CNV is recommended as a first-line test in the initial postnatal evaluation of individuals with the following:

a. Multiple anomalies not specific to a well-delineated genetic syndrome
b. Apparently nonsyndromic GDD/ID
c. ASD

Additional ACMG guidelines have been published for the design and analytical performance expectations of microarrays, software, interpretation, and reporting of CNVs applicable to the postnatal setting. A 2013 update includes recommendations for validation of microarray methodologies for both prenatal and postnatal specimens.

A 2013 ACMG guideline revision states that a stepwise or tiered approach to the clinical genetic diagnostic evaluation of ASD is recommended, with the recommendation being that first tier include fragile X syndrome and CMA, and that second tier include MECP2 and PTEN testing. The guideline notes that

“This approach will evolve with continued advancements in diagnostic testing and improved understanding of the ASD phenotype. Multiple additional conditions have been reported in association with an ASD phenotype, but none of these has been evaluated in a large prospective cohort. Therefore, a future third tier of evaluation is a distinct possibility. Further studies would be needed to elevate the evidence to the point of recommended testing. Alternatively, advances in technology may permit bundling of individual tests into an extended, more readily accessible, and less expensive platform…. the accumulating evidence … [using] ‘next-generation’ sequencing [third-tier testing] … will increase the diagnostic yield even more over the next few years.”

International Standard Cytogenomic Array Consortium

The International Standard Cytogenomic Array Consortium (ISCA) Consensus Statement recommends offering CMA as the first-tier genetic test, in place of G-banded karyotype, for patients with unexplained GDD/ID, ASD, or multiple congenital anomalies (MCA). “Except in special cases, such as those involving family history of multiple miscarriages, a karyotype is not cost effective in a child with DD/ID, ASD, or MCA and a negative array study. CMA testing is not inexpensive, but the cost is less than the cost of a G-banded karyotype plus a customized FISH test such as subtelomeric FISH, and the yield is greater.”
METHODS

Search Strategy

MEDLINE® was searched (via PubMed) through February 24, 2015, and updated on June 24, 2015, to identify studies of diagnostic yield and clinical utility in children with GDD/DD or ASD undergoing CMA testing using the search strategy in Appendix Table 1.

Study Selection

We included case series or cohort studies that enrolled 20 or more patients with clinical diagnoses GDD/ID or ASD with known or suspicion of genetic abnormalities, with or without negative results by conventional cytogenetic evaluation, and performed CMA testing on enrolled patients. Studies were also included if they examined management decisions and/or patient outcomes based on genetic evaluation results.

Medical Advisory Panel Review

This Special Report was reviewed by the Blue Cross Blue Shield Association Medical Advisory Panel (MAP) on June 10, 2015. To maintain the timeliness of the scientific information in this Special Report, literature search updates were performed subsequent to the Panel's review (see Search Strategy section above). No additional studies were identified that would change the conclusions of this Special Report.

A prior Special Report, Array Comparative Genomic Hybridization (aCGH) for the Genetic Evaluation of Patients with Developmental Delay/Mental Retardation and Autism Spectrum Disorder, was reviewed by the MAP on December 16, 2008.

REVIEW OF EVIDENCE

Overview of Available Evidence

Analytic Validity

CMA testing is available from laboratories licensed under the Clinical Laboratory Improvement Amendments of 1988 (CLIA). Testing is performed as a laboratory-developed test (LDT) under CLIA regulations but not cleared by the U.S. Food and Drug Administration (FDA). CLIA regulations require in-house validation (eg, comparing G-banded karyotype with CMA); public reporting of technical performance data is not required by CLIA.

Affymetrix CytoScan® Dx has been FDA-cleared through the de novo 510(k) process. FDA's review of the CytoScan Dx® Assay included an analytic evaluation of the test's ability to accurately detect numerous chromosomal imbalances of different types, sizes, and genome locations when compared with several analytically validated test methods. FDA found that the CytoScan Dx® Assay could analyze a patient's entire genome and adequately detect chromosome imbalances in regions of the genome associated with ID and GDD. Still, reproducibility decreased with the CNV size (gain or loss), particularly when less than approximately 400 kb (generally recommended as the lower reporting limit). FDA documents offer insight into the extent of technical performance data that can be required to support technical performance.

ACMG offers detailed standards for constitutional cytogenomic microarray analysis. Recommendations for reporting how standards are met are not included. Confirmation of an identified CNV (postnatal) is considered unnecessary but may often be performed.

The College of American Pathologists

25 ACMG offers detailed standards for constitutional cytogenomic microarray analysis. Recommendations for reporting how standards are met are not included. Confirmation of an identified CNV (postnatal) is considered unnecessary but may often be performed.

27 The College of American Pathologists

* "With proper technical performance and analytical validation, it should not be necessary for the performing laboratory to further confirm a CNV called with the laboratory-validated parameters, after the validation stage."
proficiency testing program includes challenge testing for CMA. Participation in a proficiency testing program is recommended by ACMG and required under CLIA; these accuracy data are not, however, publically available.

In summary, acceptable analytic validity is generally assumed for CMA testing based on laboratories meeting quality standards under CLIA, including participation in proficiency testing. Reports of analytic validity specific to CMA are not readily identified. Data supporting the analytic validity of the CytoScan Dx® Assay for use in children with ID and GDD were included as part of the FDA clearance.

Clinical Validity and Diagnostic Yield

For common phenotypes and syndromes, the pathogenicity of CNVs may be supported by considerable evidence; for uncommon phenotypes and uncommon CNVs determining pathogenicity requires a systematic evaluation that includes parental studies, examining databases for reported associations, and considering the molecular consequences of the identified variant. Parental studies (eg, “trio” testing of affected child, father, and mother) can identify an inherited CNV from an unaffected parent and therefore considered benign. A variety of databases index the clinical implications of CNVs their associations with a particular phenotype. CNVs are continuously cataloged and, with growth in CMA testing and improved resolution, databases have become increasingly extensive (eg, DECIPHER [https://decipher.sanger.ac.uk], ClinVar [http://www.ncbi.nlm.nih.gov/clinvar/]). For uncommon CNVs, in addition to reports of CNV-phenotype associations, the location and size of the CNV can offer clues to pathogenicity; larger CNVs are more often pathogenic and the role of affected genes in brain circuitry and effect of CNV on gene expression can implicate pathogenicity. Although uncommon, an observed phenotype can result from unmasking a mutated recessive allele on the unaffected (non-CNV) chromosome. Other considerations when determining pathogenicity include CNV dosage, X linkage, number of reports in the literature of association between CNV and phenotype, and findings in “normal” individuals.

ACMG has published guidelines for evaluating, interpreting, and reporting pathogenicity reflecting these principles. The recommended categories of clinical significance for reporting are: pathogenic, uncertain clinical significance (likely pathogenic, likely benign, or no subclassification), or benign. ISCA recently proposed “an evidence-based approach to guide the development of content on chromosomal microarrays and to support interpretation of clinically significant copy number variation.” The proposal defines levels of evidence (from the literature and/or the ISCA databases and other public databases) that describe how well or how poorly detected mutations or CNVs are correlated with phenotype.

Diagnostic Yield

Diagnostic yield is defined as the proportion of tested patients with clinically relevant genomic abnormalities. Figure 1 summarizes the diagnostic yields from 67 case series (≥20 patients) according to diagnostic categories of included children (Appendix Table 2 provides detailed study characteristics). Variability is apparent in the diagnostic yield from the GDD/ID ± ASD studies of 13.6% (55 studies [N=26,301]; interquartile range [IQR], 9.5%-17.2%) and for ASD a median of 8.4% (12 studies [N=4077]; IQR, 7.2%-17.3%). Although not examined here, diagnostic yield generally increases with phenotype severity and is highest in children with congenital anomalies. Most studies included patients with prior normal studies (eg, karyotype and FMR1 testing). However, it is difficult to assess phenotype severity across studies owing to reporting and how samples were often assembled. Arrays used also differed across studies. For a recent comparison, investigators reported diagnostic yield from 1133 children enrolled in the UK Deciphering Developmental Disorders study for whom a diagnosis was not established prior to CMA testing. Using both CMA and exome sequencing, a diagnostic yield of 27% was achieved.
CMA technologies and diagnostic yield have improved over time. For example, in 2013, Palmer et al.\textsuperscript{32} reanalyzed samples tested in 2010 from 67 children with ID. In 2010, 13 (19\%) “potentially pathogenic” variants were detected; in 2013, results from an additional 8 children were added. We explored secular changes in diagnostic yield among the studies identified for this Special Report. Because studies inconsistently reported dates when specimens were collected, the analysis was limited to using the publication date acknowledging some attendant imprecision. Figure 2 displays results suggesting just over a 1\% average annual improvement in yield when all studies were analyzed in a weighted regression model (results being nearly identical including a covariate for diagnostic category). For studies published in 2012 or later, the median diagnostic yield in GDD/ID ± ASD studies was 19.0\% (n=21), and from those categorized as examining primarily ASD 12.3\% (n=4).
Figure 2. Diagnostic Yield Reported in Included Studies According to Publication Dates (Regression Line and 95% Confidence Interval)a

Reasons Cited for CMA Testing

In 2006, the Cambridge Genetics Knowledge Park released a report culminating from the Learning Disability & the Interface with Genetics Project, an interdisciplinary effort that sought to develop expertise and services for the genetic investigation of learning disability. The project employed a collaborative process that involved families affected by learning disability; the report summarized the parent/caregiver perspective. Parents’ most commonly cited reason for pursuing diagnostic testing was “to know”—to establish a medical cause for their child’s condition, and to end the uncertainty of not knowing. Although few studies have examined the impact of “knowing,” potential improvements in parental quality of life, decreased maternal anxiety, and qualitative psychological benefits have been suggested. Additional reasons and expected outcomes of establishing a diagnosis in the Cambridge report were:

- To understand prognosis and future needs;
- To improve response (be taken seriously) by medical and educational service providers;
- To gain improved and early access to educational and social services and support;
- To test other offspring with questionable symptoms; and
- To make future reproductive decisions based on estimated recurrence rate.

a Symbols scaled to sample sizes.
The U.K. Genetic Testing Network, which advises the National Health Service on genetic testing, requested a working group to evaluate use of aCGH for investigating etiological factors in learning disability. The working group was composed of clinical and public health experts who provided insight on the clinical utility of aCGH. The final report\(^3\) identified the clinical benefits of achieving a specific genetic diagnosis from the clinical viewpoint, as follows:

- End the diagnostic odyssey and allay parents’ fears about other causes;
- Refer patients to an appropriate specialist;
- Determine possible prognosis;
- Guide optimal management and surveillance (eg, of associated comorbidities);
- Advise on risk of recurrence in future offspring or in extended family;
- Increase knowledge about precise genotype-phenotype correlation; and
- Provide potential insight into disease mechanisms and eventual development of therapeutic interventions.

**Clinical Utility**

Genetic testing of children with GDD/ID or ASD has potential to benefit a child or family as displayed in the analytic framework (see Figure 3). Benefits can accrue by identifying comorbidities requiring treatment and addressing risks (eg, screening for cancers), ending a diagnostic odyssey, assisting reproductive planning, and potentially improving treatment of the conditions (eg, through better access to services). Long periods between presentation and diagnosis have been described and can extend a decade or more.\(^38\)\(^-\)\(^40\) Lacking systematic study, how often a diagnostic odyssey is ended is uncertain. Uncovering nonpaternity and incidental findings are possible harms. An incorrect diagnosis is possible, but, given guidance interpreting variants and the ability to confirm of abnormal results, errors are likely uncommon. Following the structure of the analytic framework, we review evidence identified supporting clinical utility.

**Figure 3. Analytic Framework for Outcomes of CMA Testing in Children With GDD/ID or ASD**

Dotted lines indicate likely limited, if any, potential benefit.

ASD: autism spectrum disorder; CMA: chromosomal microarray; GDD: global developmental delay; ID: intellectual disability.

\(^a\) For example, from earlier treatment and screening.

\(^b\) Incidental findings may result in benefit.\(^41\)
**Improved Health Outcomes in Child Tested**

We identified 5 retrospective studies and 1 database analysis that examined a potential impact of CMA results on clinical decisions (see Tables 3 and 4). Saam et al invited 22 physicians to participate in a survey of management changes in patients with positive aCGH results. A total of 14 (12 geneticists, 2 neurologists) agreed to be interviewed. Management changes were recommended for 34 (70.8%) of the patients. Among the actions reported taken in 48 individuals were: referral (14.6%), screening for associated morbidity (16.7%), avoiding further genetic testing (25.0%) or other diagnostic testing (16.7%), and enhanced access to educational and insurance services (25.0%). From the 48 patients included, 17 (35.4%), families were provided information on recurrence risk.

Coulter et al retrospectively reviewed CMA test results ordered for 1792 patients with GDD/DD and ASD or congenital anomalies. Abnormal variants were identified in 131 (7.3%) (two-thirds in those with GDD/DD and ASD) and variants of possible significance in 104 (5.8%); follow-up was available in 121 and 73 patients with corresponding findings. Some clinical action (referral, imaging, laboratory) was taken in 53.7% of children with abnormal variants and in 34.2% of those with variants of possible significance. When abnormal variants were identified, action was recommended in 7 (26.9%) of 26 with ASD and in 34 (61.8%) of 55 with GDD/DD. Based on CMA results in the 194 patients with abnormal or possibly abnormal variants, the authors suggested 356 prior studies could have been avoided. Two patients with GDD/DD and ASD were described where testing potentially impacted outcomes: a 15-year-old girl with GDD and newly identified protein C deficiency and a 9-year-old boy with suspected Asperger's and toe walking found to have Becker muscular dystrophy.

Ellison et al examined a database of CMA results obtained from 46,298 patients. The most common indications for testing were GDD/DD, dysmorphic features, and neurobehavioral issues, but the proportions of each were not reported. Diagnostic yield was 5.4%, and in 2088 (4.5%) the identified disorder was judged to require follow-up. In 122 of these cases, physicians were contacted about actions taken. From the 81 responses received, actions taken were judged appropriate in 76, ranging from referrals to laboratory evaluation. Examples included evaluation for potential arrhythmias (long QT or Brugada syndromes), hematologic (thrombocytopenia, anemia), meningocele (Currarino syndrome), and hearing loss.

Henderson et al conducted a retrospective review of electronic medical records from 1780 patients undergoing CMA testing. GDD/DD or ASD were present in 66.1% of those with abnormal results, and just over half of the total sample had congenital anomalies. Abnormal variants were identified in 12.7% (227 patients), and 13.5% were of uncertain significance. Of the 227 patients with abnormal findings, clinic follow-up was available in 187 (82.4%). Clinical management was reportedly impacted in 54.5% of the 187 with abnormal variants—eg, medication, screening or surveillance (or avoided), referral, imaging, or laboratory testing.

Tao et al retrospectively reviewed CMA results from 327 patients tested for unexplained GDD/DD and ASD (65.7%) or congenital anomalies, dysmorphia, and other reasons (34.3%). Overall, 11.3% of variants were judged pathogenic or likely pathogenic. In those with unexplained GDD/DD and ASD, pathogenic or likely pathogenic variants were found in 4.2%—clinical actions were taken in 66.7% (referral, surveillance, diagnostic tests, procedures, medication, lifestyle). Variants of unknown clinical significance were identified in 13.0%.

Finally, Riggs et al (not included in Table 3) first distinguished 186 phenotypes associated with genetic abnormalities and identifiable by CMA that were judged "clinically actionable"—defined by surgical intervention indicated or contraindicated, surveillance, medication potentially indicated, lifestyle changes to reduce risk, and “other.” Counseling on recurrence risk was not considered an intervention. Next variants corresponding to the phenotypes were determined. The authors then identified when those variants were present in 28,526 CNVs submitted to the ISCA database through March 2012—7% were...
deemed “actionable” or 46.3% (1908/4125) of the pathogenic variants detected. Those phenotypes were, however, not limited to patients with GDD/DD and ASD.

These studies collectively indicate that identified pathogenic variants can prompt clinical actions potentially impacting morbidity. Less clear is how often outcomes will be improved and in which cases interventions might have occurred in the absence of testing. The proportion that may benefit will depend on the variants identified as well as diagnostic yield, which in turn depends on phenotype severity. Studies did not report on any follow-up or management changes in patients without identified pathogenic variants. In addition to reducing morbidity, bringing closure to a diagnostic odyssey is a reason for genetic testing cited by parents and noted as an outcome in case series and reports. For example, Turner et al found a median of 16.5 years to diagnosis from the initial medical contact to identify a causal mutation in 38 extended families with fragile X syndrome. Saam et al also noted that CMA testing may influence that odyssey. Parents cite obtaining services and support as a reason for testing, but how the frequency can impact outcomes is difficult to quantify. The studies reviewed convey a set of intermediate outcomes likely to favorably affect the health of some children. Lacking are end-to-end studies following children at presentation to final outcomes.

**Reproductive Planning**

Having a child with ASD appears to impact reproductive decision making, or so-called reproductive stoppage. For example, Hoffmann et al examined reproductive stoppage in families with ASD using the California Department of Developmental Services database linked to birth certificates. Between 1990 and 2003, 19,710 families with 39,361 siblings and half-siblings were identified. Birth histories in these families were then compared with a control group (matched 2:1 by sex, birth year, maternal age, ethnicity/race, and county). Investigators found fertility rates in case and control families similar in the 2 years following birth of a child with ASD, but, in the subsequent years, the rate was 33% (95% confidence interval, 30 to 37) lower in families having a child affected by ASD.

Wood et al analyzed reproductive stoppage and ASD recurrence rates within 2 U.K. family databases—299 families including 660 children (327 diagnosed with ASD). In 10% of families, there was more than 1 ASD-affected child with an estimated 24.7% recurrence risk. Reproductive stoppage was examined by statistically comparing whether children with ASD were born later in families than their unaffected siblings. In 132 of the 180 complete families analyzed, the last-born child was more often affected (p<0.05); 40 families had a single child (affected) and 62 families 2 children with only the second affected. Any potential confounding by maternal or paternal age was not reported.

These results are consistent with an impact of ASD-affected child on reproductive planning. Whether it can be attributed to concerns over having another affected child or the caregiving burden of the first affected child is unclear. Regardless, quantifying recurrence risk may assist reproductive decision making, particularly given that recurrence risk may be high—eg, in ASD, as high as 18%. However, establishing a genetic cause may revise the estimated risk considerably, as shown in Table 5.

Consistent with this notion, Turner et al reported the impact of genetic counseling on reproductive decisions in 38 extended families having a male with X-linked mental retardation. Prior to variant testing, the fertility rate in families with an affected child was 1 in 27 reproductive years (48 women, 673 reproductive years). After variant testing became available, the fertility rate in families undergoing testing followed by counseling increased to 1 in 7 reproductive years (42 women, 304 reproductive years)—74% of carriers choosing prenatal genetic evaluation. In the entire district (United Kingdom-New South Wales) over the same period the fertility rate was 1 in 11 reproductive years. Although the results of this study are consistent with an impact of testing on fertility, they show only an indirect effect of recurrence risk on reproductive decisions.

Having a child affected by ASD, and likely GDD/DD, can affect reproductive decision making. When recurrence risk can be estimated for an identified variant (eg, by including parent testing), future reproductive decisions can be affected. In addition, individuals with mild phenotypes and pathogenic variants may desire families, and recurrence risk could potentially inform later reproductive decisions.
Table 3. Studies Reporting Management Changes Following CMA Testing

<table>
<thead>
<tr>
<th>Study</th>
<th>Dates Testing</th>
<th>Design</th>
<th>Patients (Tests)</th>
<th>Sample</th>
<th>Diagnostic Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saam (2008)</td>
<td>01/2005-03/2007</td>
<td>Retrospective chart review (and 14 physician surveys)</td>
<td>490</td>
<td>GDD/ID</td>
<td>17.6%</td>
</tr>
<tr>
<td>Coulter (2011)</td>
<td>07/2009-07/2010</td>
<td>Retrospective chart review</td>
<td>1792</td>
<td>GDD/ID/ASD or congenital anomalies</td>
<td>7.3% (abnormal)</td>
</tr>
<tr>
<td>Ellison (2012)</td>
<td>04/2004-10/2011</td>
<td>Retrospective laboratory results database (and physician survey)</td>
<td>46,298</td>
<td>ID/GDD, dysmorphic, neurobehavioral, others</td>
<td>5.4%</td>
</tr>
<tr>
<td>Henderson (2014)</td>
<td>07/2009-07/2012</td>
<td>Retrospective electronic medical record</td>
<td>1780</td>
<td>GDD/ID/ASD (81.5% of 227 abnormal)</td>
<td>12.7%</td>
</tr>
<tr>
<td>Tao (2014)</td>
<td>01/2011-05/2013</td>
<td>Retrospective</td>
<td>327</td>
<td>GDD/ID/ASD (66% or 215)</td>
<td>11.3%</td>
</tr>
</tbody>
</table>

ASD: autism spectrum disorder; CMA: chromosomal microarray; GDD: global developmental delay; ID: intellectual disability.

Table 4. Clinical Management Changes When Variants Detected

<table>
<thead>
<tr>
<th>Study</th>
<th>Pathogenic n</th>
<th>Actionable, n (%)</th>
<th>Referral, n (%)</th>
<th>Screening, n (%)</th>
<th>Avoid Genetic Testing, n (%)</th>
<th>Improved Access to Services, n (%)</th>
<th>Reproductive Recurrence Risk, n (%)</th>
<th>Imaging, n (%)</th>
<th>Laboratory Testing, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saam (2008)</td>
<td>48</td>
<td>34 (70.8%)</td>
<td>7 (14.6%)</td>
<td>8 (16.7%)</td>
<td>12 (25.0%)</td>
<td>12 (25.0%)</td>
<td>17 (35.4%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coulter (2011)</td>
<td>121 Abnormal</td>
<td>65 (53.7%)</td>
<td>67 (60%)</td>
<td>7 (13.3%)</td>
<td>12 (25.0%)</td>
<td>25 (22%)</td>
<td>20 (18%)</td>
<td>9 (24%)</td>
<td>18 (47%)</td>
</tr>
<tr>
<td></td>
<td>73 Possible significance</td>
<td>25 (34.2%)</td>
<td>11 (29%)</td>
<td>2 (3.5%)</td>
<td>15 (30.0%)</td>
<td>18 (29%)</td>
<td>12 (24%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ellison (2012)</td>
<td>1259</td>
<td>441 (35.5%)</td>
<td></td>
<td></td>
<td>20 (25%)</td>
<td>12 (30%)</td>
<td>38 (45%)</td>
<td>15 (20%)</td>
<td></td>
</tr>
<tr>
<td>Henderson (2014)</td>
<td>187</td>
<td>102 (54.5%)</td>
<td>84 (44.9%)</td>
<td>11 (5.9%)</td>
<td>38 (20.3%)</td>
<td>29 (15.5%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tao (2014)</td>
<td>9</td>
<td>6 (66.7%)</td>
<td></td>
<td></td>
<td>9 (100%)</td>
<td>3 (100%)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a Percentages as reported in the publication—denominators vary from 121 and 73.
b Assumed to be from pathogenic results oligonucleotide arrays.
c Of the 215 patients with global developmental delay/intellectual disability or autism spectrum disorder.
### Table 5. Revised Sibling Recurrence Risk After Identification of Different Types of Genomic Abnormalities Associated With ASD

<table>
<thead>
<tr>
<th>Type of Genetic Abnormality</th>
<th>Clinical Example</th>
<th>Sibling Recurrence Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dominant single-gene disorder with full penetrance</td>
<td>Tuberous sclerosis: involves abnormalities of the skin, brain, and heart; associated with ID and autism</td>
<td>50% if parent carries the disease-causing variant (ie, not a de novo mutation)</td>
</tr>
<tr>
<td>Recessive single-gene disorder</td>
<td>Smith-Lemli-Opitz syndrome: congenital multiple anomaly syndrome; associated with ASD</td>
<td>25%</td>
</tr>
</tbody>
</table>
| X-linked single-gene disorder | Fragile X syndrome: most common cause of mental retardation; associated with ASD | **Brother:** 50%  
Sister: up to 50% will be a carrier or might be mildly affected | Same as population prevalence if de novo (ie, not found in parents)                |
| Copy number variation | Prader-Willi syndrome/Angelman syndrome (15q11-q13 duplication) |                                                                                       |

ASD: autism spectrum disorder; ID: intellectual disability.
DISCUSSION

The ability to detect pathogenic CNVs underlying GDD/ID and ASD using CMA assays has improved over time. The improvement can be attributed to higher CMA resolution along with sharing and availability of data documenting CNV pathogenicity. Professional societies and organizations have recommended CMA as the initial test when genetic evaluation is desired.42,43 At least 2 cohort studies have found a CMA-first strategy may be the least costly,51,49 consistent with cost-effectiveness analysis.

A recent Agency for Healthcare Research and Quality Technical Brief summarized information on genetic tests (not limited to CMA) available in the United States for GDD/ID and ASD. The report also sought to identify studies supporting clinical utility, but did “not systematically review, existing evidence addressing the tests’ clinical utility” or suggest an analytic framework for an indirect chain of evidence. The Technical Brief therefore lacked a synthesis of the body of evidence on clinical utility. No direct evidence supporting clinical utility was identified—and emphasized as an important research gap. The authors also noted the challenges in obtaining direct evidence including rapidly changing technologies, cost, and issues surrounding rare disorders.51

Data supporting analytic validity are readily available only for the Affymetrix CytoScan® Dx assay, but laboratories meeting CLIA standards and using other platforms would be expected to achieve adequate, and likely similar, technical performance. Still, analytical performance characteristics decline with small CNV size, but small variants (eg, <400 kb) are generally unreported. Detected variants can also be confirmed using other methods to enhance analytic validity. Still, making quality metrics available, including proficiency testing results across platforms and laboratories would be desirable.

There is consistent evidence that the diagnostic yield obtained from CMA testing is higher than that with karyotyping in children with GDD/ID or ASD, with or without congenital anomalies. Diagnostic yield also appears to be improving. Establishing the pathogenicity of detected CNVs relies on evidence, informatics, and genetics expertise. The process, databases queried, and functional genetic information have complexities, yet the underlying logic used to determine pathogenicity mirrors in many ways a clinician’s diagnostic approach. A challenge when considering the process and supporting evidence is that, outside the more common syndromes, diagnoses include a large number of rare disorders a clinician, even a specialist, might not encounter during a lifetime. In addition, defining standard diagnostic performance characteristics is not possible from the available data. From an evidence perspective, the existence of multiple public and proprietary databases—in the absence of an open science imperative for data sharing—is a potential gap in the ability to fully delineate the clinical significance of identified CNVs. Transparency across databases could accelerate the ability to confirm pathogenic CNVs and resolve those of uncertain significance. Access across all databases would also reduce the potential for false-positive diagnoses. Lastly, we were unable to identify case reports of incorrect diagnoses; how often they might occur is unclear. Although involved, following recommended approaches when determining pathogenicity allows conveying uncertainty.

Assessing clinical utility is challenging. End-to-end cohort studies of children from presentation to outcomes have not been reported. There are considerable challenges conducting studies of sufficient size given the underlying genetic heterogeneity, and including follow-up adequate to observe final health outcomes. Some potential benefits—eg, from cancer screening or disease monitoring—can take years to be apparent. Studies examining clinical utility have reported intermediate outcomes and indirect evidence. In addition, outside readily recognizable syndromes, pathogenic variants identified represent a collection of rare disorders. Ascertaining improved net health outcome for rare diseases32 is not easy. Both conditions and outcomes can be heterogeneous. The evidence reviewed here reflects the accompanying uncertainty, but supports a perspective that identifying a pathogenic variant can: (1) impact the search for a diagnosis, (2) inform follow-up that can benefit a child by reducing morbidity and rarely potential mortality, and (3) affect reproductive planning for parents and later potentially the affected child. There are also likely circumstances where other family members may be impacted owing to the nature of the variant and subsequent cascade (family member) testing. The downsides to testing can include detecting
nonpaternity, an incorrect diagnosis, and findings of uncertain significance—how often they occur is uncertain. It is difficult to quantify lower or upper bounds for any potential improvement in the net health outcome owing in part to heterogeneity of disorders, rarity, and outcome importance that may differ according to identified pathogenic variants. The strong expert opinion in recommending initial CMA testing over other approaches (prior beliefs in the decision problem), together with the indirect evidence for benefit following testing, supports concluding that the net health outcome can be improved.

For families that desire a genetic diagnosis and what might follow from one, CMA testing can establish a diagnosis more often than other approaches such as karyotyping. Still, other assays will identify some genetic abnormalities not detected by an array. The complexities of CMA testing, interpretation, understanding limitations, and the potential implications require that testing is obtained by clinicians with genetic expertise, alongside genetic counseling, and performed in laboratories meeting molecular pathology standards.
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40. Mroch AR, Flanagan JD, Stein OP. Solving the puzzle: case examples of array comparative genomic hybridization as a tool to end the diagnostic odyssey. Curr Probl Pediat Adolesc Health Care. Mar 2012;42(3):74-78. PMID 22325475


APPENDIX

Appendix Table 1. Search Strategy

<table>
<thead>
<tr>
<th>Search Term</th>
</tr>
</thead>
<tbody>
<tr>
<td>A literature search was conducted in February 2015 of PubMed/MEDLINE</td>
</tr>
<tr>
<td>Search terms:</td>
</tr>
</tbody>
</table>
### Appendix Table 2. Diagnostic Yields of 67 Case Series

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Diagnoses</th>
<th>Patient Description</th>
<th>Previous Normal Studies</th>
<th>Array Type</th>
<th>Minimum Array Resolution</th>
<th>Confirmation</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aradhya (2007)93</td>
<td>20</td>
<td>GDD/ID</td>
<td>GDD/ID and either dysmorphic features, CAs, or growth retardation</td>
<td>G-banded karyotype, FISH</td>
<td>BAC (Spectral Chip 2600 array)</td>
<td>1000 kb</td>
<td>FISH</td>
<td>30.0%</td>
</tr>
<tr>
<td>Aradhya (2007)93</td>
<td>20</td>
<td>GDD/ID</td>
<td>As above</td>
<td>As above</td>
<td>Oligo (Agilent)</td>
<td>&lt;35 kb</td>
<td>FISH</td>
<td>50.0%</td>
</tr>
<tr>
<td>Baldwin (2008)</td>
<td>211</td>
<td>GDD/ID/ASD</td>
<td>Various, including idiopathic GDD/ID, dysmorphic features, CAs, ASD, or syndromal phenotype</td>
<td>G-banded karyotype (&quot;many&quot;)</td>
<td>Oligo (Agilent 44K)</td>
<td>300 kb</td>
<td>FISH</td>
<td>15.6%</td>
</tr>
<tr>
<td>Ballif (2007)105</td>
<td>6946</td>
<td>GDD/ID</td>
<td>Variety of clinical presentations, most commonly GDD, dysmorphic features, and/or MCAs</td>
<td>Karyotype, subtelomere FISH</td>
<td>BAC (Signature Chip v.1-3)</td>
<td>No DNA backbone coverage</td>
<td>FISH</td>
<td>2.4%</td>
</tr>
<tr>
<td>Ballif (2006)96</td>
<td>3600</td>
<td>GDD/ID</td>
<td>Consecutive cases with diverse range of GDD or MR features</td>
<td>Not specified</td>
<td>BAC (Signature Chip v.3)</td>
<td>No DNA backbone coverage</td>
<td>FISH</td>
<td>5.1%</td>
</tr>
<tr>
<td>Bartnik (2014)117</td>
<td>256</td>
<td>GDD/ID</td>
<td>GDD/ID present with or without dysmorphic features, additional neurodevelopmental abnormalities, and/or CAs</td>
<td>G-banded karyotype, fragile X testing</td>
<td>Oligo (Agilent 105K or 180K)</td>
<td>10-30 kb</td>
<td>FISH/PCR</td>
<td>27.0%</td>
</tr>
<tr>
<td>Bartnik (2014)117</td>
<td>112</td>
<td>GDD/ID</td>
<td>ID present and accompanied by dysmorphic features and/or CAs</td>
<td>G-banded karyotype, fragile X testing, MLPA</td>
<td>Oligo (Agilent 60K or CytoSure ISCA v.2)</td>
<td>40 kb</td>
<td>MLPA, FISH, or karyotyping</td>
<td>21.4%</td>
</tr>
<tr>
<td>Battaglia (2013)119</td>
<td>349</td>
<td>GDD/ID/ASD</td>
<td>Idiopathic GDD/ID/ASD or dysmophia</td>
<td>FISH or karyotype</td>
<td>Oligo (Agilent 44K or 180K) or SNP (Affymetrix v.6.0)</td>
<td>40 kb</td>
<td>FISH or qPCR</td>
<td>22.1%</td>
</tr>
<tr>
<td>Bremer (2011)120</td>
<td>223</td>
<td>ASD</td>
<td>151 diagnosed ASD with normal karyotype, 1 nonpathogenic inherited balanced translocation, 72 patients who had not received karyotyping</td>
<td>Karyotype</td>
<td>Oligo (Agilent 244K and 180K)</td>
<td>30-50 kb</td>
<td>FISH, MLPA</td>
<td>8.1%</td>
</tr>
<tr>
<td>Bruno (2009)121</td>
<td>117</td>
<td>GDD/ID</td>
<td>Idiopathic MR and/or CAs</td>
<td>Karyotype (400 to 650–band level)</td>
<td>SNP (Affymetrix 250K array)</td>
<td>Not specified</td>
<td>FISH, MLPA</td>
<td>15.4%</td>
</tr>
<tr>
<td>Chong (2014)122</td>
<td>115</td>
<td>GDD/ID/ASD/CA</td>
<td>105 patients with GDD/ID/ASD/CA were recruited by clinical genetics services</td>
<td>Karyotype</td>
<td>Oligo (Agilent 44K and 180K)</td>
<td>25-100 kb</td>
<td>NimbleGen CGX-135K array</td>
<td>19.0%</td>
</tr>
<tr>
<td>Christian (2008)123</td>
<td>397</td>
<td>ASD</td>
<td>Nonsyndromic autism, subset of AGRE subjects (Roswell Park Cancer Institute)</td>
<td>Karyotype</td>
<td>BAC (19K)</td>
<td>Tiling</td>
<td>FISH, PCR</td>
<td>11.6%</td>
</tr>
<tr>
<td>Coulter (2011)124</td>
<td>1792</td>
<td>GDD/ID/ASD</td>
<td>ASD, GDD/ID, CAs, dysmorphic features, seizures, hypotonia</td>
<td>&quot;Majority&quot; karyotype</td>
<td>Not specified</td>
<td>Not specified</td>
<td>Not specified</td>
<td>7.3%</td>
</tr>
<tr>
<td>D'Amours (2014)125</td>
<td>21</td>
<td>CA</td>
<td>21 children with GDD/ID with or without CA</td>
<td>Karyotype</td>
<td>SNP (Affymetrix Genome-Wide Human SNP Array 6.0, Affymetrix Cytogenetics Whole-Genome 2.7)</td>
<td>Various high-resolution</td>
<td>NimbleGen CGX-12</td>
<td>14.3%</td>
</tr>
<tr>
<td>Study</td>
<td>N</td>
<td>Diagnoses</td>
<td>Patient Description</td>
<td>Previous Normal Studies</td>
<td>Array Type</td>
<td>Minimum Array Resolution</td>
<td>Confirmation</td>
<td>Yield</td>
</tr>
<tr>
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<td>-------------------------------------------------</td>
<td>----------------------------------------------------------------------------</td>
<td>----------------------------</td>
<td>--------------</td>
<td>-------</td>
</tr>
<tr>
<td>de Vries (2005)</td>
<td>100</td>
<td>GDD/ID</td>
<td>Idiopathic MR</td>
<td>G-banded karyotype, subtelomere MPLA</td>
<td>M array; Illumina HumanOmni1-Quad BeadChip, Illumina HumanCytoSNP-12 DNA Analysis BeadChip</td>
<td>BAC (32K)</td>
<td>Tiling</td>
<td>10.0%</td>
</tr>
<tr>
<td>Eriksson (2015)</td>
<td>162</td>
<td>ASD</td>
<td>Diagnosed ASD</td>
<td>Karyotype (unclear precise proportion but &lt; half)</td>
<td>Oligo (Agilent 244K and 180K; Oxford Gene Technology 180K)</td>
<td>Not specified</td>
<td>NR</td>
<td>8.6%</td>
</tr>
<tr>
<td>Fliges (2012)</td>
<td>131</td>
<td>GDD/ID/ASD</td>
<td>Consecutive patients with normal karyotype but presenting with chromosomal phenotypes: malformation syndromes, syndromic and nonsyndromic ID, and ASD</td>
<td>Karyotype</td>
<td>SNP (NimbleGen 385K and 720K; Oxford Gene Technology 180K)</td>
<td>50-200 kb</td>
<td>FISH</td>
<td>25.2%</td>
</tr>
<tr>
<td>Friedman (2006)</td>
<td>100</td>
<td>GDD/ID</td>
<td>Idiopathic ID</td>
<td>Karyotype</td>
<td>SNP (Affymetrix 100K GeneChip)</td>
<td>400-1000 kb</td>
<td>FISH</td>
<td>11.0%</td>
</tr>
<tr>
<td>Friedman (2009)</td>
<td>100</td>
<td>ID</td>
<td>Moderate-severe Idiopathic MR/GDD with CAs</td>
<td>Uncertain</td>
<td>SNP (Affymetrix 500K GeneChip)</td>
<td>Not specified</td>
<td>FISH, MPLA</td>
<td>16.0%</td>
</tr>
<tr>
<td>Froyen (2007)</td>
<td>108</td>
<td>GDD/ID</td>
<td>Suspicious for X-linked MR</td>
<td>G-banded karyotype, FMR1</td>
<td>BAC (X-chromosome)</td>
<td>80 kb</td>
<td>PCR</td>
<td>13.0%</td>
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<tr>
<td>Harada (2004)</td>
<td>69</td>
<td>GDD/ID</td>
<td>Idiopathic MR, with or without MCAs</td>
<td>Karyotype (400-band level)</td>
<td>Illumina Human610-Quad BeadChip or HumanOmni 1M BeadChip</td>
<td>1 Mb</td>
<td>Not specified</td>
<td>5.8%</td>
</tr>
<tr>
<td>Henderson (2014)</td>
<td>1780</td>
<td>GDD/ID/ASD</td>
<td>Patients referred to laboratory for CMA</td>
<td>Not specified</td>
<td>Affymetrix 100K GeneChip</td>
<td>400-1000 kb</td>
<td>FISH</td>
<td>9.1%</td>
</tr>
<tr>
<td>Hoyer (2007)</td>
<td>104</td>
<td>GDD/ID</td>
<td>Unselected patients with idiopathic MR</td>
<td>G-banded karyotype</td>
<td>SNP (Affymetrix 100K GeneChip)</td>
<td>400-1000 kb</td>
<td>FISH</td>
<td>9.1%</td>
</tr>
<tr>
<td>Iourov (2012)</td>
<td>54</td>
<td>ID/ASD/CA</td>
<td>Patients were highly selected from group of 2426 patients according to clinical and cytogenic data</td>
<td>G-banded karyotype</td>
<td>BAC (Human BAC Array-System 12K)</td>
<td>300-1000 kb</td>
<td>FISH</td>
<td>28.0%</td>
</tr>
<tr>
<td>Jacquemont (2006)</td>
<td>29</td>
<td>ASD</td>
<td>Syndromic ASD</td>
<td>Karyotype, biochemical tests</td>
<td>BAC/PAC</td>
<td>1000 kb</td>
<td>FISH</td>
<td>28.0%</td>
</tr>
<tr>
<td>Krepschi-Santos (2006)</td>
<td>95</td>
<td>GDD/ID</td>
<td>Syndromic MR or other</td>
<td>G-banded karyotype, FMR1 (in some)</td>
<td>BAC</td>
<td>1000 or 3000 kb</td>
<td>FISH or MLPA</td>
<td>15.8%</td>
</tr>
<tr>
<td>Lay-Son (2015)</td>
<td>40</td>
<td>GDD/ID/Other</td>
<td>Patients had at least 2 of the following: MCAs, facial dysmorphism, GDD, or ID</td>
<td>Karyotype, 4 patients (10%) had abnormality on karyotype but it did not convey a definite cause of patients’</td>
<td>Oligo and SNP (Affymetrix 2.7M)</td>
<td>100 kb</td>
<td>FISH</td>
<td>25.0%</td>
</tr>
</tbody>
</table>

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## TEC Special Report

**Chromosomal Microarray for the Genetic Evaluation of Patients With Global Developmental Delay, Intellectual Disability, and Autism Spectrum Disorder**

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Diagnoses</th>
<th>Patient Description</th>
<th>Previous Normal Studies</th>
<th>Array Type</th>
<th>Minimum Array Resolution</th>
<th>Confirmation</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lee (2013)</td>
<td>190</td>
<td>GDD/ID</td>
<td>Retrospective chart review of patients at single center with idiopathic ID/GDD</td>
<td>G-banded karyotype</td>
<td>BAC (MacArray Karyo 1440 BAC-chip)</td>
<td>Not specified</td>
<td>FISH</td>
<td>13.7%</td>
</tr>
<tr>
<td>Lu (2007)</td>
<td>1726</td>
<td>GDD/ID</td>
<td>Diverse range of GDD/ID, dysmorphic, or MCA features</td>
<td>G-banded karyotype and/or FISH</td>
<td>BAC/PAC</td>
<td>Not specified</td>
<td>FISH</td>
<td>5.2%</td>
</tr>
<tr>
<td>Lugtenberg (2006)</td>
<td>40</td>
<td>GDD/ID</td>
<td>Idiopathic MR, suspicious for X-linked abnormality</td>
<td>Karyotype</td>
<td>BAC (X-chromosome, 1460)</td>
<td>100 kb tiling</td>
<td>MLPA</td>
<td>7.5%</td>
</tr>
<tr>
<td>Madrigal (2007)</td>
<td>54</td>
<td>GDD/ID</td>
<td>Idiopathic MR; 52 from families c/w X-linked inherited MR; 2 with suspicion of X chromosome deletion</td>
<td>Karyotype, FMR1</td>
<td>BAC (X-chromosome, 1460)</td>
<td>100 kb</td>
<td>MLPA, PCR</td>
<td>11.6%</td>
</tr>
<tr>
<td>Manolakos (2010)</td>
<td>82</td>
<td>ID</td>
<td>Idiopathic MR</td>
<td>G-banded karyotype</td>
<td>Oligo (Agilent 180K)</td>
<td>22 kb</td>
<td>FISH, MLPA</td>
<td>3.6%</td>
</tr>
<tr>
<td>Marshall (2008)</td>
<td>427</td>
<td>ASD</td>
<td>Diagnosed ASD</td>
<td>Karyotype (32 with known abnormality)</td>
<td>SNP (Affymetrix 500K)</td>
<td>Not specified</td>
<td>PCR</td>
<td>6.3%</td>
</tr>
<tr>
<td>McGrew (2012)</td>
<td>97</td>
<td>ASD</td>
<td>Retrospective review of EMR for patients. Diagnosed ASD or pervasive DD NOS</td>
<td>Karyotype?</td>
<td>Athena Diagnostics v.5-v.8.5</td>
<td>Not specified</td>
<td>Not specified</td>
<td>6.2%</td>
</tr>
<tr>
<td>Menten (2006)</td>
<td>140</td>
<td>GDD/ID</td>
<td>Idiopathic MR and MCAs</td>
<td>Karyotype, subtelomere MPLA (n=31)</td>
<td>BAC/PAC (3431)</td>
<td>1000 kb</td>
<td>FISH and/or PCR</td>
<td>13.6%</td>
</tr>
<tr>
<td>Miyake (2006)</td>
<td>30</td>
<td>GDD/ID</td>
<td>Idiopathic MR with some dysmorphic features</td>
<td>G-banded karyotype</td>
<td>BAC (2505)</td>
<td>1400 kb</td>
<td>FISH</td>
<td>16.7%</td>
</tr>
<tr>
<td>Nava (2014)</td>
<td>194</td>
<td>ASD</td>
<td>Diagnosed ASD</td>
<td>Karyotype, fragile X testing, FISH</td>
<td>SNP (Illumina 370/CNV-370 Quad, 660W-Quad, or CytoSNP-12)</td>
<td>35 kb</td>
<td>FISH</td>
<td>1.5%</td>
</tr>
<tr>
<td>Nicholl (2014)</td>
<td>1700</td>
<td>GDD/ID/ASD</td>
<td>1453 unrelated patients prospectively referred to study for investigation of ID/GDD/ASD and 247 epilepsy cases</td>
<td>Uncertain</td>
<td>Oligo (BlueGnome CytoChip ISCA 60k)</td>
<td>35 kb</td>
<td>FISH or MLPA</td>
<td>11.5%</td>
</tr>
<tr>
<td>Palmer (2014)</td>
<td>67</td>
<td>ID</td>
<td>Idiopathic ID</td>
<td>Karyotype, fragile X, subtelomeric MPLA</td>
<td>Oligo and SNP (Affymetrix 2.7M)</td>
<td>50 kb</td>
<td>MPLA</td>
<td>19.0%</td>
</tr>
<tr>
<td>Pickering (2008)</td>
<td>1176</td>
<td>GDD/ID/CA</td>
<td>Consecutive cases referred for idiopathic GDD/MR/MCA or other dysmorphia</td>
<td>Oligo and MPLA (Affymetrix 500K)</td>
<td>Oligo (BlueGnome CytoChip ISCA 60k)</td>
<td>1 Mb</td>
<td>FISH</td>
<td>9.8%</td>
</tr>
<tr>
<td>Preiksaštienė (2014)</td>
<td>211</td>
<td>GDD/ID</td>
<td>Syndromic and nonsyndromic cases of unknown etiology of GDD/ID</td>
<td>FISH, MLPA, or karyotype</td>
<td>Oligo (Agilent 44K, 105K, and 400K) or SNP (Illumina 300K and 700K)</td>
<td>&lt;500 kb</td>
<td>FISH or PCR</td>
<td>13.7%</td>
</tr>
<tr>
<td>Redin (2014)</td>
<td>106</td>
<td>GDD/ID</td>
<td>Idiopathic ID</td>
<td>Karyotype</td>
<td>SNP (Affymetrix Array 6.0)</td>
<td>Not specified</td>
<td>Sanger sequencing</td>
<td>24.5%</td>
</tr>
<tr>
<td>Roberts (2014)</td>
<td>215</td>
<td>GDD/ID/ASD</td>
<td>Diagnosed ASD or ID</td>
<td>Uncertain</td>
<td>Oligo (CombiMatrix 105K and 180K)</td>
<td>&lt;21 kb</td>
<td>BAC, aCGH, or FISH</td>
<td>14.9%</td>
</tr>
</tbody>
</table>
## Chromosomal Microarray for the Genetic Evaluation of Patients With Global Developmental Delay, Intellectual Disability, and Autism Spectrum Disorder

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Diagnoses</th>
<th>Patient Description</th>
<th>Previous Normal Studies</th>
<th>Array Type</th>
<th>Minimum Array Resolution</th>
<th>Confirmation</th>
<th>Yield</th>
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<tbody>
<tr>
<td>Rosenberg (2006)</td>
<td>81</td>
<td>GDD/ID</td>
<td>Idiopathic MR and CAs</td>
<td>Karyotype</td>
<td>BAC (3.5K)</td>
<td>1000 kb</td>
<td>FISH</td>
<td>16.0%</td>
</tr>
<tr>
<td>Rosenfeld (2010)</td>
<td>1461</td>
<td>ASD</td>
<td>Retrospective review of putative ASD submitted for clinical testing</td>
<td>Not specified</td>
<td>Oligo (Agilent 105K)</td>
<td>Not specified</td>
<td>FISH</td>
<td>7.7%</td>
</tr>
<tr>
<td>Saam (2008)</td>
<td>490</td>
<td>GDD/ID</td>
<td>Diagnosed GDD/ID</td>
<td>Karyotype</td>
<td>BAC (Spectral Genomics Constitutional Array or Spectral Chip 2600 array)</td>
<td>1 Mb</td>
<td>FISH</td>
<td>17.6%</td>
</tr>
<tr>
<td>Schaefer (2010)</td>
<td>68</td>
<td>ASD</td>
<td>Retrospective review of patients who had received aCGH for autism</td>
<td>Uncertain</td>
<td>BAC (Spectral Genomics Constitutional Array or Spectral Chip 2600 array)</td>
<td>1 Mb</td>
<td>FISH</td>
<td>22.0%</td>
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<tr>
<td>Schoumans (2005)</td>
<td>41</td>
<td>GDD/ID</td>
<td>Mild-to-severe idiopathic MR and dysmorphism and/or family history; patients scored &gt;3 on de Vries Checklist (2001)</td>
<td>Spectral karyotype (n=11), subtelomere (n=30)</td>
<td>BAC (Spectral Chip 2600 array)</td>
<td>1000 kb</td>
<td>FISH</td>
<td>9.8%</td>
</tr>
<tr>
<td>Sebat (2007)</td>
<td>195</td>
<td>ASD</td>
<td>Nonsyndromic autism; majority from AGRE or NIMH Center for Collaborative Genetic Studies on Mental Disorders</td>
<td>Karyotype</td>
<td>Oligo</td>
<td>35 kb</td>
<td>aCGH dye-swap replicate, other CGH arrays</td>
<td>7.2%</td>
</tr>
<tr>
<td>Schoenfeld (2006)</td>
<td>1500</td>
<td>GDD/ID</td>
<td>Consecutive patients with diverse range of DD or MR diagnoses</td>
<td>Karyotype (94%), FISH (20%) where prior testing available</td>
<td>BAC (SignatureChip v7, 831)</td>
<td>1000 kb</td>
<td>FISH</td>
<td>5.6%</td>
</tr>
<tr>
<td>Sharp (2006)</td>
<td>290</td>
<td>GDD/ID</td>
<td>Idiopathic MR with or without dysmorphism or MCAs</td>
<td>Karyotype, subtelomere FISH (n=255)</td>
<td>BAC/PAC (2007)</td>
<td>Not specified</td>
<td>FISH and/or oligo array targeted to same hotspots</td>
<td>5.5%</td>
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<tr>
<td>Shaw-Smith (2004)</td>
<td>50</td>
<td>GDD/ID</td>
<td>Idiopathic MR and dysmorphism or other features</td>
<td>Karyotype, subtelomere (n=41)</td>
<td>BAC</td>
<td>1000 kb</td>
<td>FISH</td>
<td>14.0%</td>
</tr>
<tr>
<td>Shen (2007)</td>
<td>211</td>
<td>GDD/ID</td>
<td>Diagnosed ASD</td>
<td>Not selected by prior results</td>
<td>Oligo (Agilent &gt;10K)</td>
<td>&lt;35 in targeted region not specified</td>
<td>FISH, MLPA, BAC-aCGH, aCGH dye-swap replicate</td>
<td>8.1%</td>
</tr>
<tr>
<td>Shen (2010)</td>
<td>848</td>
<td>ASD</td>
<td>Idiopathic MR and/or dysmorphism or MCAs</td>
<td>G-banded karyotype, fragile X</td>
<td>SNP (Agilent 244K or Affymetrix 500k v.5)</td>
<td>Not specified</td>
<td>Not specified</td>
<td>7.0%</td>
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<tr>
<td>Shevell (2008)</td>
<td>94</td>
<td>GDD/ID</td>
<td>Final diagnosis of GDD, &lt;5 years old at diagnosis</td>
<td>G-banded karyotype, fragile X, FMR1, neuroimaging</td>
<td>BAC (Signature Chip v4.4, 1887)</td>
<td>Not specified</td>
<td>FISH</td>
<td>6.4%</td>
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<tr>
<td>Shoukier (2013)</td>
<td>342</td>
<td>GDD/ID</td>
<td>Retrospective review of idiopathic GDD/ID</td>
<td>Karyotype</td>
<td>Oligo (Agilent 180K or 244K)</td>
<td>Not specified</td>
<td>MPLA or qPCR</td>
<td>13.2%</td>
</tr>
<tr>
<td>Sorte (2013)</td>
<td>50</td>
<td>ASD</td>
<td>Diagnosed ASD</td>
<td>G-banded karyotype</td>
<td>Oligo (Agilent 105K)</td>
<td>Not specified</td>
<td>MPLA or qPCR</td>
<td>16.0%</td>
</tr>
<tr>
<td>Stobbe (2014)</td>
<td>23</td>
<td>ASD</td>
<td>Retrospective review of patients referred for autism</td>
<td>Karyotype (some patient &lt;44%, 1)</td>
<td>Oligo (NimbleScan 2.5)</td>
<td>Not specified</td>
<td>FISH</td>
<td>21.7%</td>
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</table>
## Study

<table>
<thead>
<tr>
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<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tao (2014)</td>
<td>327</td>
<td>GDD/ID/ASD</td>
<td>Patients seen by clinical genetician</td>
<td>Not specified</td>
<td>Oligo (NimbleGen 135K array)</td>
<td>140 kb</td>
<td>FISH/qPCR/ karyotype</td>
<td>11.3%</td>
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<tr>
<td>Thuresson (2007)</td>
<td>48</td>
<td>GDD/ID</td>
<td>Idiopathic MR and CAs</td>
<td>G-banded karyotype, subtelomere FISH</td>
<td>BAC</td>
<td>1000 kb</td>
<td>FISH, MPLA</td>
<td>6.0%</td>
</tr>
<tr>
<td>Tyson (2005)</td>
<td>22</td>
<td>GDD/ID</td>
<td>Mild-to-moderate MR and nonsyndromic dysmorphic features; patients scored &gt;3 on de Vries Checklist (2001)</td>
<td>G-banded karyotype, subtelomere FISH (n=13)</td>
<td>BAC (Spectral Genomics)</td>
<td>1000 kb</td>
<td>FISH</td>
<td>13.6%</td>
</tr>
<tr>
<td>Tzetis (2012)</td>
<td>334</td>
<td>DD/ID/ASD</td>
<td>Classified as GDD/ID/ASD or with major CA or dysmorphic features</td>
<td>Karyotype, FISH, fragile X and Rett syndromes</td>
<td>Oligo (Agilent 180K or 244K)</td>
<td>8.9-25 kb</td>
<td>Not specified</td>
<td>25.1%</td>
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<tr>
<td>Utine (2014)</td>
<td>100</td>
<td>ID</td>
<td>Idiopathic ID</td>
<td>Karyotype, FISH</td>
<td>SNP (Affymetrix Array 6.0)</td>
<td>Not specified</td>
<td>PCR</td>
<td>12.0%</td>
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<tr>
<td>Uwineza (2014)</td>
<td>50</td>
<td>GDD/ID/MCA</td>
<td>GDD/ID/MCA</td>
<td>Karyotype</td>
<td>Oligo (Agilent 180K)</td>
<td>13 kb</td>
<td>FISH, MPLA</td>
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<tr>
<td>Vissers (2003)</td>
<td>20</td>
<td>GDD/ID</td>
<td>Idiopathic MR and dysmorphism; patients scored &gt;3 on de Vries Checklist (2001)</td>
<td>Karyotype</td>
<td>BAC (3569)</td>
<td>&lt;1000 kb</td>
<td>FISH, aCGH dye-swap replicate</td>
<td>10.0%</td>
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<tr>
<td>Wagenstaller (2007)</td>
<td>67</td>
<td>GDD/ID</td>
<td>Idiopathic MR</td>
<td>G-banded karyotype, FISH (n=42)</td>
<td>SNP (Affymetrix GeneChip 100K array)</td>
<td>23.6 kb</td>
<td>PCR</td>
<td>16.4%</td>
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<tr>
<td>Wincent (2011)</td>
<td>160</td>
<td>GDD/CA</td>
<td>Idiopathic GDD/CA</td>
<td>Karyotype, fragile X, FISH/MPLA</td>
<td>BAC (32K/38K) or oligo (Agilent 244K)</td>
<td>50 -300 kb</td>
<td>FISH or MLPA</td>
<td>13.1%</td>
</tr>
</tbody>
</table>

aCGH: array comparative genomic hybridization; AGRE: Autism Genetic Resource Exchange; ASD: autism spectrum disorder; BAC: bacterial artificial chromosome; CA: congenital anomaly; CGH: comparative genomic hybridization; CMA: chromosomal microarray; c/w: consistent with; DD: developmental delay; EMR: electromagnetic radiation; FISH: fluorescent in situ hybridization; GDD: global developmental delay; ID: intellectual disability; kb: kilobases; Mb: megabases; MCA: multiple congenital anomaly; MPLA: multiplex ligation-dependent probe amplication; MR: mental retardation; NIMH: National Institute of Mental Health; NOS: not otherwise specified; NR: not reported; PAC: plasmid artificial chromosome; PCR: polymerase chain reaction; qPCR: quantitative polymerase chain reaction; SNP: single-nucleotide polymorphism.