

## GENETIC TESTING IN PATIENTS WITH GLOBAL DEVELOPMENTAL DELAY / INTELLECTUAL DISABILITIES. A REVIEW

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### Abstract

*Genetic factors are responsible for up to 40% developmental disability cases, such as global developmental delay/intellectual disability (GDD/DI). The American and more recently the European guidelines on this group of diseases state that genetic testing is essential and should become a standardized diagnostic practice. The main arguments for the necessity of implementing such a practice are: (1) the high prevalence of developmental disabilities (3% of the population); (2) the high genetic contribution to this type of pathology; (3) insufficient referral for genetic consultation. In an attempt to address these issues, the purpose of this paper is to present the genetic etiology of global developmental delay / intellectual disability with emphasis on the need to implement a genetic testing protocol for the patients with GDD/DI, as indicated by the current guidelines. Chromosomal abnormalities and fragile X syndrome are the most frequent causes of developmental disabilities and the techniques employed to detect such genetic disorders should be used as first line investigations of GDD/DI.*

**Keywords:** global developmental delay, intellectual disabilities, genetic testing, CGH array, X fragile syndrome

### Introduction

Genetic testing should become a standardized diagnostic practice in patients affected by global developmental delay (GDD)/intellectual disability (ID) [1].

According to the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) and the American Association of Psychiatry (APA), GDD/ID is characterized by significant functional mental impairment and adaptive functioning deficits in conceptual, social and practical domains [2].

This category of disabilities has a high prevalence, affecting about 3% of the general population [3-5].

### Clinical diagnostic tests

The first step for the evaluation of the genetic etiology is a correct clinical diagnosis of GDD/ID. The proportion of the patients diagnosed with developmental disabilities depends on the utilization and validity of the diagnostic tests (measured by specificity and sensitivity). The application of well-standardized, reliable and valid instruments for the assessment of GDD/ID is essential and must precede genetic testing.

The most commonly tests for identifying patients with GDD/ID are: *Wechsler Preschool and Primary Scale of Intelligence-Revised (WPPSI-R)* for children 3-7 years old, *Wechsler Intelligence Scale for Children-IV (WISC-IV)* for the 6-16 years old interval, and *Wechsler Adult Intelligence Scale (WAIS)*, for children above 16 years old

[6,7]. Before the age of five years, we rather talk about a developmental coefficient, rather than an intelligence coefficient, therefore, the diagnosis of ID is correct only after the age of five years. Before the age of five years the diagnosis of global developmental delay is more adequate. The developmental coefficient is frequently measured with the *Brunet-Lézine revised scale*, *Bayley Scales of Infant Development (BSID)* or *Denver Developmental Screening Test (DDST)* and specifies if the developmental age of the child is equal or different compared with the chronological age [8].

A correct evaluation of GDD/ID includes a complete anamnesis and a clinical examination, and is able to indicate the etiology in up to 70% of cases [9]. Special attention should be paid to: personal (data on prenatal, perinatal and postnatal period, developmental history) and familial history (three-generation pedigree). The clinical exam should include: head circumference evaluation (microcephaly or macrocephaly may provide useful clues to etiology) and height measurement (short stature could indicate fetal alcohol syndrome or tall stature may suggest fragile X syndrome or Sotos syndrome), neurological consultation (evaluation of muscle tone, coordination, tendon reflexes, abnormal movements, or other modifications), ophthalmologic and otolaryngology evaluation, skin changes (pigmentation abnormalities), facial and extremity dysmorphism or internal malformations assessment [10].

### Etiology

Once the clinical diagnosis of a developmental disability is established, the next step is represented by the identification of the etiology. The genetic factors are responsible for up to 40% of the cases of intellectual disability [4,9-12]. The etiologic role of the following environmental factors is well known: infectious agents (*cytomegalovirus*, *rubella virus*, *Trichomonas*) or toxic substances (alcohol or lead exposure), perinatal events (periventricular hemorrhage in extreme prematurity, hypoxia-ischemia at preterm gestation, congenital hypothyroidism) or postnatal events (meningitis, neurodegenerative disorders, traumatic brain injury). These latter events account for less than 15% of GDD/ID cases, most often being revealed by complete anamnesis and clinical examination.

### Genetic etiology

Some patients present phenotypic signs associated to developmental delay (dysmorphism, internal organ malformations) that may suggest the presence of a syndromic genetic disorder (15% of cases), but the majority of the patients do not present signs or present only few minor signs associated with GDD/ID [9]. In this last case, the intellectual disability is considered non-syndromic. However, even in the case of the syndromic forms, the clinical signs are not always obvious enough to permit a

straightforward diagnosis [4]. Usually the presence of syndromic form indicates a larger genetic defect than in isolated developmental disabilities. The genetic etiology of GDD/ID is very heterogeneous [4].

Among the genetic causes, chromosomal abnormalities are responsible for 25% of cases [4,9]. Trisomy 21 is the most frequent chromosomal abnormality associated with GDD/ID. Other chromosomal abnormalities frequently associated with this pathology are the following: other types of trisomies (trisomy 13 or trisomy 18), structural chromosomal abnormalities which often include microdeletions of the following regions: 1p36; 2q37, 4p16, 5p15, 7q11.2, 8p23.1, 8q23q24, 9q34.3, 11p13, 11p11.2, 15q11.2, 15q11.2, 16p13.3, 17p13.3, 17p11.2, 17q12, 17q21.3, 18q23, 22q11.2, 22q13.3, some of them being associated with pathology also in duplicated state, such as: 7q11.2; 8p23.1; 15q11.2; 17p11.2; 22q11.2 [4,13].

Monogenic causes are responsible for up to 10% of the GDD/ID cases. Fragile X syndrome is the most common monogenic defect associated with GDD/ID, being responsible for about 5% of the cases of intellectual disability [9,14,15]. Adult males present a characteristic phenotype with long face and macroorchidism, but in children the phenotype is rather nonspecific, therefore testing for fragile X syndrome should be included in addition to chromosomal testing in the initial genetic evaluation of GDD/ID [9,14,15].

In the following, we present a very useful classification of the genes implicated in GDD/ID, according to the main pathogenic pathways to which they belong:

1. genes implicated in metabolic pathways (organic acid metabolism - *ALDH5A1*, *L2HGDH* genes; polysaccharide metabolism - *NAGLU*, *SGSH* genes; purine metabolism - *ADSL* gene; protein glycosylation- *PMM1* gene; monocarboxylate transporter - *SLC16A2* gene; creatine transporter - *SLC6A8* gene) [4,16];

2. genes implicated in neurogenesis (mitotic spindle regulation in neuroblast - *ASPM*, *CDK5RAP2*, *CENPJ* genes; DNA repair and mitotic arrest in neuroblast - *MCPHI* gene) [4,17];

3. genes implicated in neuronal migration (protein glycosylation - *POMGNT1*, *POMT1*, *POMT2*, *FKTN*, *FKRP*, *LARGE* genes; microtubule subunits - *TUBA1A*, *TUBB2B* genes; microtubule regulation - *DCX* gene; microtubule associated proteins - *PAFAH1B1*; transcription factors implicated in neuronal migration - *ARX*) [4,18];

4. genes implicated in the synaptic function:

- a. genes implicated in presynaptic function (adhesion between pre and postsynaptic membranes - *NRXN1*, *CDH15* genes; vesicle traffic - *RAB3GAP1*, *STXBP1*, *GDII*, *RAB39B* genes; exocytosis inhibition - *IL1RAPL1*, *CASK* genes) [4,19];

- b. genes implicated in postsynaptic density organization (adhesion between pre and postsynaptic membranes - *CNTNAP2*, *NLGN3*, *NLGN4* genes;

neurotransmitter receptor interaction with membrane proteins - *SHANK2*, *SHANK3* genes; subunits of NMDA receptor - *GRIN2A*, *GRIN2B* genes) [4,19];

c. genes implicated in the regulation of postsynaptic proteins (ubiquitin ligase of UPS proteolysis - *UBE3A*, *UBE2A*, *UBR1*; *HUWE1*, *CUL4B* genes; transport of mRNA from the nucleus to the cytoplasm - *FMR1* gene) [4,19];

d. genes implicated in cytoskeletal dynamics of dendritic cells (activation in Rho-GTPase pathway - *MEGAP*, *OCRL1*, *OPHN1*, *FGD1*, *ARHGEF6*, *ARHGEF9* genes; regulation of actin polymerization and vesicles endocytosis- *LIMK1*, *APIS2*, *IQSEC2* genes; Rho-GTPase and cytoskeleton interaction - *PAK3* gene) [4,19];

5. genes implicated in intracellular signalization (Ras-MAPK-ERK pathway - *SOS1*, *RAF1*, *BRAF*, *SHOC2*, *HRAS*, *KRAS*, *PTPN11*, *SPRED1*, *MAP2K1*, *MAP2K2*, *NF1*, *DYRK1A*, *RPS6KA3* genes; PI3K-AKTmTOR pathway - *TSC1*, *TSC2*, *PTEN* genes) [4,19];

6. genes implicated in epigenetic regulation of transcription (histone deacetylase - *HDAC4* gene; histone acetyl-transferase - *CREBBP*, *EP300* genes; histone methyltransferase - *NSD1*, *EHMT1*, *MLL2* genes; histone demethylase - *KDM5C* gene; subunits of Mediator complex (transcription pre-initiation) *MED12*, *MED17* and *MED23* genes; transcription factors - *TCF4*, *RAI1*, *ZNF711*, *ZNF41*, *ZNF674*, *ZNF81*, *PHF6*, *PHF8* genes; DNA replication - *SETBP1* gene; DNA methyltransferase - *DNMT3B* gene; repression of transcription factors - *BCOR*, *MECP2* genes; chromatin modification - *ATRX*, *BRWD3* genes) [4,19].

### Genetic testing

The chromosomal abnormalities may be observed in about 25-30 % of the patients with GDD/ID [4,9]. The monogenic disorders are responsible for 10% of the GDD/ID cases. The fragile X syndrome is the most frequent cause, being observed in 5% of the patients with GDD/ID [4,9]. When the clinical picture suggests a syndromic form of GDD/ID, a diagnostic genetic test should be applied in order to confirm or rule out a certain condition: classical cytogenetic analysis or FISH (for chromosomal abnormalities) or molecular biology testing (for monogenic disorders). If the clinical picture does not suggest a syndromic genetic form of GDD/ID, the most recent diagnostic guidelines recommend the global assessment of the genomic alterations by classical and/or molecular karyotyping (CGH array), as well as the assessments of mutations in the fragile X mental retardation *FMR1* gene [1].

The global assessment of the genomic alterations can be done by classical karyotyping (for anomalies larger than 5Mb), and by a high resolution new technology of molecular karyotyping or array analysis (*CGH array or SNP-array* technology). Microarray technology allows for whole genome analysis of patient samples and has the

advantage of detecting sub-microscopic anomalies as low as 1 Kb, known as CNV (*copy number variants*) [4,20]. This technology is currently used for the elucidation of the etiology of nearly 30% of the GDD/ID cases [4,20].

The actual evaluation of GDD/ID in clinical practice should follow the assessment protocol shown in Figure 1.

### Genetic consultation and genetic counseling

Since up to 15% of the GDD/ID cases are syndromic forms, associated with other clinical signs, a genetic consultation performed by a clinical geneticist trained in identifying the etiology based on clinical findings is highly recommended [9,10]. Also, because of the high percentage of GDD/ID cases having a genetic etiology (40%), genetic consultation is mandatory for all the cases with this specific clinical picture [9].

In the case of a patient with GDD/ID, the clarification of the etiological diagnosis is necessary in order to answer the questions regarding the possibilities for therapeutic intervention and the risk of recurrence. The elucidation of the etiological diagnosis of the condition provides a coherent explanation for the affected child's parents and may provide certain psychological relief, being an essential part of the adaptation process. Through the process of genetic counseling, families are offered information regarding the mode of inheritance and are supported to take informed family planning decisions, based on the implied risk of recurrence and the available reproductive options [10].

We would like to emphasize the importance of multidisciplinary teams consisting of specialists from different domains such as pediatrics, psychiatry, psychology, neurology, and the establishment of a well-defined referral route for the patients with GDD/ID, as well as implementing an adequate protocol for clinical diagnosis and genetic testing [1,9,10]. The primary purpose of the multidisciplinary team is to offer integrated services on clinical and genetic testing, as well as genetic counseling, reducing therefore the patient's waiting time between the required consultations, as well as "breaking the chain" of unnecessary medical investigations.

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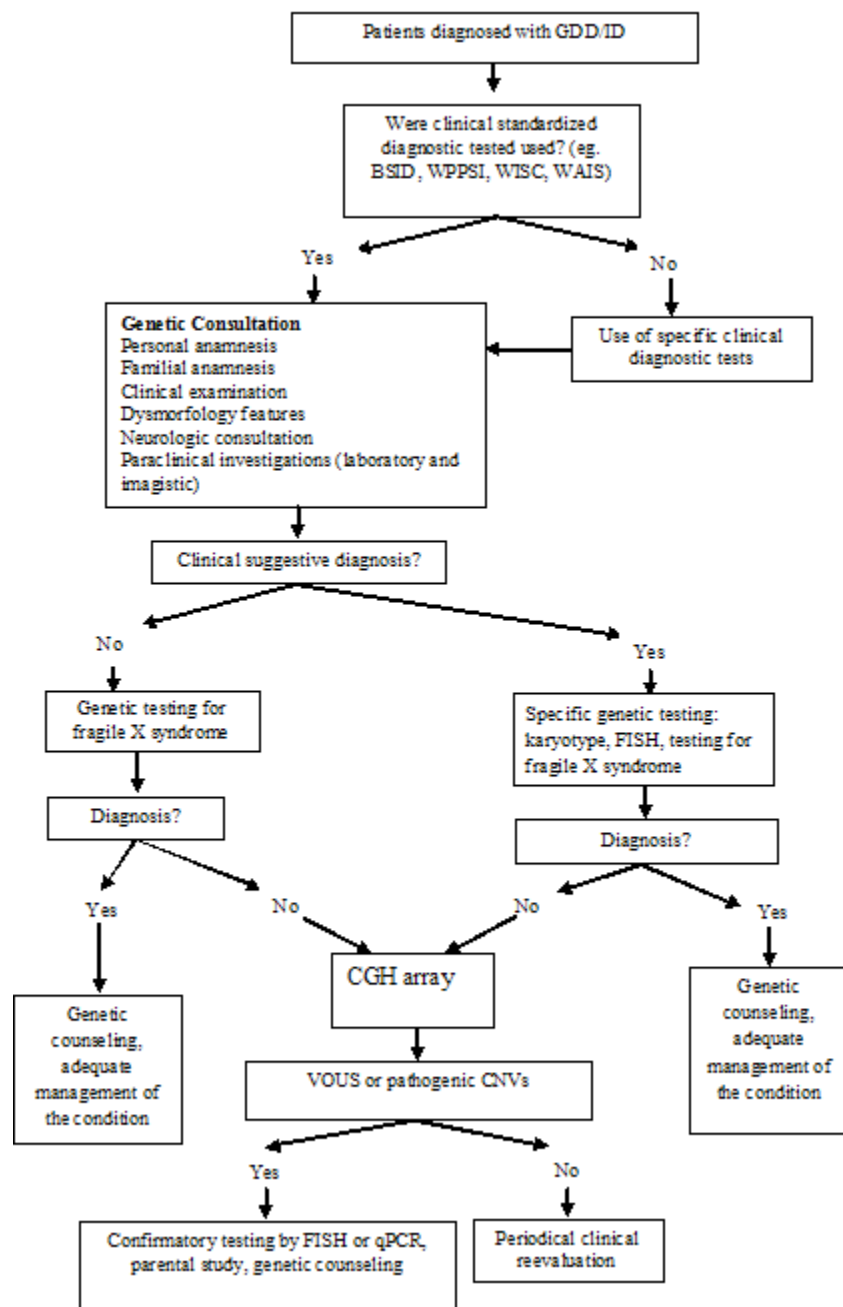


Figure 1. Assessment protocol for genetic testing in patients with GDD/ID diagnosis [10].

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