

# Down Syndrome, Molecular Genetics Of Clinical Findings

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## Abstract

In spite of the disabilities that Down Syndrome children have, it is important to emphasize that with appropriate medical care, training, teaching methods, personal skills development, and the establishment of the special needs; these children will be very effective, self-dependent and productive in society. They can perform difficult tasks and become very successful in their careers. Many of Down syndrome children have accomplished a great future by becoming painters, musicians and artists. In this review we are focusing on the link between genetic changes and the clinical presentation of Down Syndrome (DS), the risk factors behind the chromosomal imbalance, the future of diagnostic methods, the prevention and the appropriate management.

## INTRODUCTION

Down Syndrome (DS) is the most common genetic cause for mental retardation. In 1866, Doctor John Langdon Down was the first who diagnosed a case of DS. In the 1930s, Waardenburg and Bleyer hypothesized that DS might be due to chromosomal abnormalities. But it was not until 1959 when Jerome Lejeune and Patricia Jacobs discovered trisomy (triplication) of the 21st chromosome-which is the smallest human chromosome-that the cause was determined. Because of the extra full or partial copy of chromosome 21, the most structural, phenotypic, functional and metabolic changes can be attributed to the chromosomal imbalance, gene dysregulation and gene interactions. Trisomy 21 due to meiotic non-disjunction constitutes 95.4% of cases, Robertsonian translocation 2.7%, mosaicism 0.7%, and in 2% of cases, it is caused by a rearrangement of chromosomal material between chromosome 21 and another acrocentric chromosome such as chromosome 14. The translocation was parentally inherited for 33.3% of cases and maternal transmission was twice as common as paternal. In mosaicism type, there are trisomic and normal cell lines that presumably result from a mitotic error. Other change in chromosome 21 includes circular structure called ring chromosome. There is an excess of males in all groups except the mosaic group where the male: female ratio was 0.67 (1-3).

Studies revealed that nondisjunction chromosome 21 requires "two hits"; first, the establishment in prophase I of a "vulnerable" bivalent and second, abnormal processing of the bivalent at metaphase I or II.

Freeman SB et al and through the national Down syndrome project found that nondisjunction errors during oogenesis accounted for 93.2% of the cases, errors in spermatogenesis 4.1% and 2.7% in post-zygotic errors (4-5).

Chromosome 21 is the second human chromosome to be fully sequenced and it contains between 200 and 400 genes.

Most studies of Down syndrome etiology and pathology are focusing on the extra copy of the proximal part of 21q22.3, the interaction with other genes on different chromosomes and the consequences thereafter (1-5).

## GEOGRAPHICAL DISTRIBUTION OF DOWN SYNDROME

The association between geographical regions and Down Syndrome distribution was not fully established, and if any difference, it would be related to the mother's age on time of pregnancy in certain areas compared to others. However, recent increase in the incidence of Down's syndrome in young mothers was thought to be related to environment and yet to be determined.

Down's syndrome revealed no aggregation of cases, except for a slight seasonal peak in the summer. However, maternal age-adjusted rates vary little across human populations, but no differences of other factors were found such as paternal age, birth order, ancestral origin, country of birth, maternal educational level, maternal ABO and Rhesus blood groups, interval to and outcome of mother's previous pregnancy, and parental consanguinity (6-11).

Since 1980, the proportion of births to mothers of 35 years of age and over has risen quite dramatically from 8 to 14% for the European Union as a whole, with steeper rises in some regions. By 1995-1999, the proportion of “older” mothers varied between regions from 10% to 25%, and the total prevalence (including terminations of pregnancy) of Down syndrome varied from 1 to 3 per 1000 births. Some European regions have shown a more than twofold increase in total prevalence of Down syndrome since 1980 (12).

On the other hand, geographical differences influence uptake of prenatal diagnosis which probably related to access to services. Women from Africa were 61% less likely to have testing than women born in Australia. While overall uptake of testing was lower than average in rural regions. A socioeconomic index of place of residence showed no association with uptake (13).

### **PHYLOGENESIS AND EVOLUTION OF THE DOWN SYNDROME.**

It was theorized that the pathogenesis of Down syndrome is best viewed in terms of the mechanisms of speciation. Doctor Down named a syndrome “Mongoloid idiocy” because he thought it represented a “throwback” to the “Mongolian stage” in human evolution (14).

In fact, for living organisms, the starting point is cell division -regardless if this is a fertilized egg or a single cell- followed by differentiation and growth, and upon progression, it follows the direction to a specific species. The course in this journey depends on the genetic program carried on related chromosomes. In every certain stage there is a switch on and switch off genes that produce chemicals with different interactions which eventually result in the physical structures that belong to a certain organism. The development of the embryo is supposed to retrace the evolution of its group. It was even once believed that the fertilized egg, for example, would represent our one-celled ancestors, sort of the “amoeba stage”. However, the embryonic development itself is not analogous to evolution (15). In some opinions, Down syndrome neotenes the brain and the body to the fetal state “i.e.”developmental variability with many anomalies of incomplete morphogenesis (vestigia), atavisms, increased morphometric variability and increased fluctuating asymmetry. Some evidences for the physical neoteny of people with Down syndrome are: round in shape, bowed legs which tend to be short, slanty eyes, long tongue and short fingers. And evidences for their mental neoteny: unsexual, playful, affectionate, mischievous

and imitative (15-18).

Comparative genomic studies between species found many genetic changes such as gene duplications, deletions and mytelations that are related to evolutionary concept, and the relationship between down syndrome phenotype and the DNA program is not that simple and we cannot approach this explanation on the light of Neo-Darwinism which is under strong attack; most genetic changes accumulated over time may very well be of neutral effect, and detailed studies in several related groups of vertebrate species has shown that molecular and organismal evolution are largely independent of one another (15-18).

### **SYMPTOMS AND SIGNS OF DOWN SYNDROME:**

It was estimated that there are over 100 characteristics of Down syndrome and these are due to either structural or metabolic disturbances that result in the unique phenotype of DS, and the most common characteristics include: Flat facial profile, flattened back of head, an upward slant to the eye, almond shaped eyes, white spots on the iris of the eye (called Brushfield spots), small ears set low on head with a fold at the top, flattened bridge across nose, small mouth, protruding tongue, short neck, single, deep transverse crease on the palm of the hand, shortened fingers, gap between 1st and 2nd toe, joint looseness, broad feet with short toes, poor muscle tone (hypotonia), and learning disabilities. However, the final confirmation for the diagnosis of DS is by testing for trisomy 21 or by new molecular methods.

### **DOWN SYNDROME CRITICAL REGION**

The distal 10 Mb region of the long arm of chromosome 21 has been referred to be the Down syndrome critical region (DSCR). The duplication of genes located within DSCR may lead to the major phenotypic features of Down syndrome (19, 20). Details about these genes will be discussed in separate paragraphs and summary of these are;

DSCR1: It regulates the angiogenic genes that are activated by the calcineurin-NFAT signaling pathway in endothelial cells and the imbalance of the DSCR1 gene dosage contributes to an important pathogenesis of Down's syndrome (21, 22).

DSCR2 (located in chromosome 21q22.3): It is related to cell proliferation; the WDR9 gene (WD Repeat 9) is located in the (DCR-2) and has several tissue-specific transcripts. DSCR2 protein is targeted to the cytoplasmic compartment as a soluble form which has negative interaction with

peroxisome proliferator-activated receptor beta (PPARbeta) i.e. PPARbeta decreases the solubility of DSCR2 and increases the levels of insoluble DSCR2 (23,26).

DSCR3: It is located between carbonyl reductase (CBR) and Ets Related Gene (ERG) (CBR-ERG region), may contain 8 exons, is expressed in most tissues, spans 2.5 Mb on 21q22.2, and contributes significantly to the pathogenesis of many characteristics of Down syndrome, including morphological features, hypotonia, and mental retardation (27-28).

DSCR4: It is located between CBR and ERG genes (CBR-ERG region) in the band q22.2 of human chromosome 21, which spans 2.5 Mb and encodes a protein of 118 amino acids. It contributes significantly to the pathogenesis of many characteristics of Down syndrome, including morphological features, hypotonia, and mental retardation. Expression of DSCR4 mainly occurs in the human placenta and placental choriocarcinoma cell lines (BeWo and JEG3) and multiple transcripts may exist. Transcription of DSCR4 gene is regulated positively by binding to OLF-1-like transcription factor and negatively by binding to E47-like transcription factor (29-31).

DSCR5 (PIG-P): It is a component of glycosylphosphatidylinositol- N-acetylglucosaminyltransferase (GPI-GnT) gene which is expressed in various human tissues and considered a candidate for pathogenesis of Down syndrome (32-34).

DSCR6: It is expressed only in limited tissues at low levels and it is also a candidate for the pathogenesis of Down syndrome (34).

DSCR9: DSCR9 was found only in the human genome.

### RISK FOR HAVING A CHILD WITH DOWN SYNDROME

The maternal age is the main high risk factor for regular trisomy 21 that results in meiotic non-disjunction (the mean age 38.2 years) but not in translocation (the mean age is 25.3 years). In addition, the geographic variation in gene polymorphism, gene-nutritional, gene-gene or gene-nutritional-environmental factor interactions are considered other risk factors. The lack of the correlation between allopolyploid DNA variation and non-disjunction of chromosome 21 was already established (35).

On the other hand, Abnormal folate and methyl metabolism can lead to DNA hypomethylation and abnormal

chromosomal segregation, and mothers with mutation in MTHFR (C677T) and MTRR (A66G) gene have elevated levels of plasma homocysteine. These mothers have 2.6 to 2.9 fold increased risk of having a child with DS compared to mothers without that mutation (36).

Socio-genetic analysis revealed the association of intensive drug therapy of infectious diseases during the periconceptual period and maternal meiotic non-disjunction of chromosome 21. The correlation between non-disjunction of chromosome 21 and increased parental age as well as exposure to radiation, alcohol, tobacco and mutagenic substances were not found (35-37).

### DOWN SYNDROME IN PROXIMITY WITH OTHER CHROMOSOMAL ABNORMAL DISEASES

Double aneuploidy involving both autosomal and sex chromosomes is rare. Double aneuploidy has been reported in Down-Turner (female, mosaic (46,X,+21/47,XX,+21)) and Down-Klinefelter (48,XXY,+21) Syndromes and in male mosaic with 2 cell lines (45,X/47,XY,+21).

Robertsonian translocation is seen in 13;14 of maternal origin combined with regular trisomy 21(46, XX, der (13; 14) (q10; q10) mat, +21) of Down syndrome phenotype. Cryptic subtelomeric rearrangements was found in patients with mental retardation and inherited submicroscopic translocation (18;21)(q22.1q21.3) were found in multiple congenital anomalies/mental retardation (MCA/MR) syndrome (38-42).

### GENE EXPRESSION

Having a third copy of chromosome 21 in Down syndrome apparently triplicates the number of genes, but expression of the protein is different. There are up and down regulation of sequences that consist of chromosomal transcripts, enzymes of intermediary metabolism, hormones, transporters, channels and transcription factors (TFs). Nevertheless, trisomy 21 may leads to deterioration or absence of gene expression of different chromosomes other than chromosome 21, and this may explain the metabolic imbalances in DS patients.

Down syndrome phenotype can be attributed to the gene dosage imbalance that could be either due to “gene dosage effect” hypothesis which claims that DS critical region contains a subset of dosage-sensitive genes that determines DS phenotypes, or the “amplified developmental instability” hypothesis stating that 21 trisomy determines general alteration in developmental homeostasis.

In spite of imbalances were found between ubiquitinated and deubiquitinated substrates which most likely result from increase in USP25 gene dosage. However, the “gene dosage effect hypothesis” is not sufficient to fully explain DS phenotype (43-48).

In fetal cerebral cortex, there is increase expression of DYRK1A, alpha A-crystallin, FTCD, GARS-AIRS-GART, and CBS, while there is no overexpression for Tiam1 (T-cell lymphoma invasion and metastasis inducing) protein, holocarboxylase synthetase, human interferon-regulated resistance GTP-binding protein MxA, Pbx regulating protein 1, autoimmune regulator and pericentrin.

Neither the expression of sAPP alpha nor sAPP beta showed any detectable changes. The ETS-2{(V-ets erythroblastosis virus E26 oncogene homolog 2 (avian))}, Collagen alpha 1 (VI) chain precursor and HACS1 are significantly decreased in DS (49-50).

Other studies showed that ETS-2 proto-oncogene is transcribed at constant levels in neural tissue between the 13th and 24th weeks and its expression appeared to be slightly increased in the DS brain compared with that of normal controls of the same gestational age (51).

In parallel to the gene dose effect; increased gene expression and elevated protein levels in the blood of a child with Down syndrome are not necessarily due to functioning of chromosome 21 genes. The following proteins are found to be elevated and belong to genes found on other chromosomes.

These are; Uric acid, Xanthine and Hypoxanthine, Catabolites of purine metabolism, increased S100 beta (which is important for synaptogenesis and dendritic development) and GluR5 (which has been mapped to chromosome 11 cytogenetic position 11q14) (52, 53).

Finally, 29% of the expressed chromosome 21 transcripts are overexpressed in Down syndrome and correspond to either genes or open reading frames. And among these, 22% are increased proportional to the gene-dosage effect, 7% are amplified, the other 71% of expressed sequences are either compensated (56%, with a large proportion of predicted genes and antisense transcripts) or highly variable among individuals (15%).

Thus, most of the chromosome 21 transcripts are compensated for the gene-dosage effect and overexpressed genes are likely to be involved in the DS phenotype in

contrast to the compensated genes.

Highly variable genes could account for phenotypic variations observed in patients of DS and the alternative transcripts belonging to the same gene are similarly regulated but not the sense and antisense transcripts (54).

In summary, increased expression of genes on chromosome 21 altered transcriptional regulation of a subset of genes throughout the entire genome.

### **STRUCTURAL ABNORMALITIES IN DOWN SYNDROMES**

Phenotype of Down syndrome represents a wide range of structural abnormalities and defects. This phenotype is underlined by a variety of dysregulated genes and pathways at the molecular levels (55). These defects involve the soft and hard tissues. Most common are seen in the cardiovascular system that represents 40% to 60% of infants with Down syndrome. The most common anomalies are atrioventricular septal defects followed by tetralogy of Fallot (56). The Down syndrome cell adhesion molecule (Dscam, located on chromosome 21q22.2-22.3) is expressed in the heart during cardiac development and considered the candidate gene for these anomalies. Other genes, the DS-CHD candidate region has been suggested to span between PFKL (defined by tetralogy of Fallot) and D21S3 (defined by ventricular septal defect) with possible involvement of DSCR1L2 in cardiac contraction (57-59).

Gender and ethnic differences exist for atrioventricular septal defects showing twice as many affected females and twice as many affected Caucasian as African Americans and half as many Hispanics (60).

The Neural tube defect (NTD) in Down syndrome is influenced by one-carbon metabolism. The distinct data produced in different geographical areas may be explained by differences in the nutritional environment and genetic characteristics of the populations. The one-carbon metabolism is under the influence of folate, vitamin B12 and genetic polymorphisms of methylenetetrahydrofolate reductase (MTHFR 677 C --> T and 1298 A --> C) of methionine synthase (MTR 2756 C --> G), methionine synthase reductase (MTRR 66 A --> G) and transcobalamin (TCN 776 C --> G) (49). An example of geographical changes is represented by the heterozygous genotype MTR 2756AG that is associated with the increase in plasma homocysteine (Hcy) concentrations in Brazilian patients with DS while it is decreased in other Down syndrome

patients (61). Reduction in brain weight and size of specific brain regions are evident from birth and these are due to the altered number of neurons, dendrites, and dendritic branching that result from a reduction in the level of developmental signals or regulators, such as serotonin, gamma-aminobutyric acid, taurine, and dopamine (62). On the other hand, Dscam that plays a role in CHD as mentioned above is involved in neural differentiation and contributes to the central and peripheral nervous system defects in DS (63-66). Deregulation of DSCR5 gene may be the first candidate to elucidate the pathophysiology of tongue malformation observed in DS (67).

Congenital gastrointestinal defects were present in 6.7% of DS, and no significant differences were found between Down syndrome patients and control population. Defects included esophageal atresia/tracheoesophageal fistula (0.4%), pyloric stenosis (0.3%), duodenal stenosis/atresia (3.9%), Hirschsprung disease (0.8%), and anal stenosis/atresia (1.0%). Although not significant, esophageal atresia was observed more often in infants of younger mothers and Hispanics. Hirschsprung disease was more frequent in males and in infants of younger mothers and African Americans. Anal stenosis/atresia was found more often among females and Asians (68-70).

### DOWN SYNDROME AND HEMATOLOGICAL DISORDERS

Trisomy 21 is a predisposing factor for childhood leukemia due to altering the function of responsible genes on chromosome 21 and related genes on other chromosomes. Studies revealed that congenital malformations in DS do not increase the risk of leukemia beyond the attributable risk of trisomy 21 in this population (71).

Acute megakaryoblastic leukemia (AMKL) is very common in Down syndrome, and death from leukaemia, in part, account for the excess mortality associated with Down syndrome and transient myeloproliferative disorder (TMD). However, the TMD is self-limited leukemia occurs almost exclusively in neonates with Down syndrome (72, 73).

The mutation in GATA1 (megakaryocyte transcription factor) that occurs in utero along with trisomy 21 and other yet undefined genetic alteration need to occur to produce leukemia. The GATA 1 gene mutation is acquired in exon 2.5. Hence, the trisomy 21 and GATA1 mutation are found to be restricted to leukemic cells (74-78). GATA1 has several binding sites and it regulates the expression of cytidine deaminase (CDA) and the CDAsf promoter and it

could be a stable marker for minimal residual disease (MRD) (79-80).

The gain-and loss-of function mutations of Janus kinase 3 (jak3) can be acquired in DS-TMD/AMKL, and Activation of the JAK-STAT pathway contribute to lymphoid and hematologic disorders are observed in children with DS (81-82).

The RUNX1/AML1 (runt-related transcription factor 1/acute myeloid leukemia 1) gene is located on chromosome 21 and it is implicated in human leukemias. Chromosome translocations and point mutations are well-documented genetic alterations in RUNX leukemia, and RUNX1 over dosage in trisomy 21 can result in Down syndrome related leukemia (83).

### DAWN SYNDROME AND IMMUNE SYSTEM

Frequent infections and increased autoimmune phenomena in persons with DS are presumably due to impaired immune responses. Several transcripts have been down-regulated in DS; some of these are proteins involved in T-cell and B-cell receptor signaling e.g., PI3Kdelta, RGS2, LY6E, FOS, TAGAP and CD46. Overexpression of the following proteins; CuZn-superoxide dismutase (SOD-1), interferon receptor, APP-amyloid precursor protein, protein S-100 beta may contribute to thymic derangements. In addition, increased expression and abnormal distribution of adhesion molecules in DS thymuses alters the interaction between developing thymocytes and the thymic stroma which results in the abnormal thymocyte maturation and eventually the perturbation of cellular immunity (84-87).

### GENES RELATED TO THE METABOLIC DISTURBANCES IN DOWN SYNDROME

Metabolic changes in Down syndrome range from intracellular enzymatic defects through hormone level changes and functions. These changes occur due to genetic unbalances, mutations, rearrangements, inhibition or activation.

One important observation belongs to the HLCS “holocarboxylase synthetase (biotin-(propionyl-CoA-carboxylase (ATP-hydrolysing)) ligase)” gene that activates biotin-dependent carboxylases. The HLCS gene is located on the long arm of chromosome 21 at position 22.1; it is involved in many critical cellular functions, including the production and breakdown of proteins, fats, and carbohydrates. In the nucleus, the carboxylase enzyme attaches biotin molecules to histones, which are structural

proteins that bind to DNA and give chromosomes their shape, and the changes in the shape of histones determine whether certain genes are turned on or off; Hence, mutations in the HLCS gene causes holocarboxylase synthetase deficiency, and without biotin, carboxylases remain inactive and are unable to process proteins, fats, and carbohydrates effectively. A lack of holocarboxylase synthetase activity may also alter the regulation of certain genes that are important for normal development (88).

Another metabolic alteration was observed in DS fetal cerebral cortex, which is related to the decrease in sialic acid synthase (SAS) and result in abnormal brain development and sialylation defect in essential brain proteins such as neuronal cell adhesion molecule and myelin associated glycoprotein (89).

Disequilibrium of the antioxidant enzyme activities was found in DS patients and that may explain the susceptibility of cells for oxidative stress and premature atherosclerosis, cell aging, and neurologic disorders. However, the decrease in all glutathione forms, including glutathionyl-hemoglobin reduces the detoxification of harmful molecules (90). Hypothyroidism and rare Hashimoto thyroiditis are increased in Down syndrome. Thyroid hormone abnormalities are strongly associated with DS and the most consistent finding is the elevation of TSH level with over-expression of mRNA of TSH-receptor (TSH-R) in brain of a fetus with DS. Because the thyroid hormone system is well-documented in controlling the programmed cell death; the up-regulation in TSH-R is not specific for DS but may reflect the apoptosis which is the hallmark of neurodegenerative disorders as in Alzheimer's disease (91, 92). On the other hand, children with leukemic Down syndrome have decreased buffering of metabolic processes that result in a predisposition to hyperuricemia and increased insulin resistance and perhaps develop diabetes mellitus.

### **STERILIZATION AND SPERMATOGENESIS**

Significantly impaired fertility and sterility of both sexes is common in Down syndrome population. In male it is most likely due the consequence of the behavior of the extra chromosome in the meiotic prophase and spermatogenesis which could be due to disturbance of small transcript (expressed only in tissue of adult testis) of PKNOX1 gene that is located on chromosome 21q22.3 (93, 94).

### **GENE REGULATION MEDIATE NEUROLOGICAL DYSFUNCTION IN DS**

Dysfunctional brain development in Down syndrome is

observed from birth as reduction in brain weight, decrease volume of specific brain regions, altered number of neurons, dendrites, and dendritic branching.

These abnormalities are contributed to the phenotypic changes observed in Down syndrome patients such as dementia, cognitive development, delay fine motor skills, speech delay and learning deficits. These can be linked to changes in gene expression noticed in different areas of Down syndrome brain which is in one aspect resulted in reduction levels of neurotransmitters and eventually neuronal development in the Down syndrome fetal brain (95).

Damage and disruption of neural differentiation in Down syndrome fetal brain can be attributed to variety of genetic disturbances such as over-expression of TTC3 (inhibits neuronal differentiation via RhoA and Citron kinase), increase expression of metabotropic glutamate receptor 5 (GluR5), over-expression of Ets2 (transcription factor), dysregulation of Single-minded 2 (SIM2) gene which is regulated by protein transcription factor c-myb, BACE1 and BACE2 (related to production of amyloid beta), upregulation of GIRK2 gene (G-protein-activated inwardly rectifying potassium channel 2) at the hippocampus region, down-regulation of complex I and NADH3 (mitochondrial enzyme) at the DS cerebellum (96-99).

DYRK1A is expressed in the cerebellum and functionally related structures as in brainstem motor nuclei and spinal cord, and this supports the role of DYRK1A in controlling motor function and impairment of motor learning as well as alteration of the organization of locomotor behavior. DYRK1A seems to play a role in proliferation of neural progenitor cells, neurogenesis, cell cycle regulation or apoptosis, neuronal differentiation and may be involved in learning and memory. Nevertheless, most of the molecular mechanisms of MNB/DYRK1A are involved in several neuropathologies and cognitive deficits of Down syndrome. Also, DYRK1 may disrupt the function of fully differentiated neurons and this disruption is significantly reversible (100-105).

### **DOWN SYNDROME AND ALZHEIMER DISEASES**

After middle age, most Down Syndrome patients develop Alzheimer's disease (AD) with associated neuropathology of neuritic plaques and neurofibrillary tangles (tau). The dementia onset varies from ages 40 to 70 years and that is highly due to the abnormal phosphorylation of tau which

develops prematurely in Down syndrome patients. Several pathways and gene dysfunctions are involved in this pathogenesis.

S100B (a calcium-binding protein, localized to astroglial cells) is overexpressed in brain of both DS and Alzheimer's disease (AD), the S100B activates the stress response kinase and results in formation of reactive oxygen species (ROS) which may consider an important factor for mental retardation. The ROS may increase the expression of water channel aquaporin 4 (AQP4) and inhibition of AQP4 by siRNA increase the ROS. Hence, elevated levels of S100B-induced ROS and loss of AQP4 expression led to increased programmed cell death (106-110). Amyloid beta (A $\beta$  or Abeta) is a peptide of 39–43 amino acids that is the central component of neuritic plaques and is generated from  $\beta$ -amyloid precursor protein (APP) after cleaved by beta (BACE1) - and gamma-secretases.

BACE1 is ( $\beta$ -Secretase) and also called ( $\beta$ -site of APP cleaving enzyme) or memapsin-2 which is an aspartic-acid protease that is important in the formation of myelin sheaths in peripheral nerve cells. BACE1 is overexpressed (abnormal trafficking) in Golgi apparatus (which is immobile in Golgi in DS cells) than normal controls, and overproduction of Abeta in DS is caused by abnormal BACE1 protein trafficking and maturation. Interestingly, inhibition of BACE1 has therapeutic potential in AD and DS.

BACE2 gene is located on chromosome 21 and encodes for Beta-site APP-cleaving enzyme 2 (glycosylated protein), it is localizes mainly intracellularly but to some extent to the plasma membrane.

BACE1 and BACE2 are overexpressed in Down syndrome and by their cleaving activity of APP, they increase the formation of amyloid plaques that can be fortified by Tau phosphorylation due to increase expression of MAPKs (mitogen-activated protein kinases) and apolipoprotein E (APOE) that are interact with the brain deposition of Abeta in DS brains (1110-1116).

Furthermore, the cystathionine beta-synthase (CBS) is encoded on chromosome 21 and the levels of CBS in DS brains are approximately three times greater than those in the normal individuals. The overexpression of CBS may cause the developmental abnormality in cognition in DS children and that may lead to early AD in DS adults (117).

Overexpression of RCAN1 (Regulator of calcineurin 1) has

been postulated to contribute to mental retardation in Down syndrome and expedite neuronal degeneration observed in Alzheimer's due to reducing both the number of synaptic vesicles and the amount of neurotransmitter.

A protein known as nascent polypeptide-associated complex (NAC protein) -heterodimeric complex of alpha-and beta-subunits- prevents mistargeting of nascent polypeptide chains to the endoplasmic reticulum membranes, and in fetal DS brain, the decrease in alpha-NAC results in neurodegenerative changes including mistargeting, mistranslation, and proteolysis of proteins by affecting overall NAC function.

The significant decrease of alpha-NAC protein, which was even more pronounced when related to either actin or neuron-specific enolase levels, was also observed in both DS and AD (118-122).

### PSYCHOLOGICAL FEATURES IN DOWN SYNDROME

Bipolar affective disorder is observed in Down syndrome and it may relate to two cytoplasmic proteins (UMODL1L and UMODL1S) and ZNF298 gene that are located on chromosome 21q22.3. Increased prevalence of seizure disorders and impairments of short-term and long-term memory also observed in DS (123).

### DOWN SYNDROME AND MALIGNANCY

Interestingly, impairment of the immune system in DS Patients favors high rate of malignancies, but on the other hand, overexpression of some protein encoded by genes found on chromosome 21 blocks the formation of new blood vessels and reduces tumor growth in general (124).

### GENES INVOLVED IN DIAGNOSIS OF DOWN SYNDROME

Increase our knowledge about human genome will optimize the diagnosis of Down syndrome. It was found that the number of fetal cells in maternal blood in trisomic 21 pregnancies is higher than that in normal pregnancies and this can facilitate the using of these cells as alternative resource for traditional samples.

The current samples for clinical testing are amniotic fluid, amniocyte culture, chorionic villous or amniotic fluid supernatant. The molecular base noninvasive prenatal diagnostic tests for Down syndrome can provide fast, accurate, and economical alternation for the traditional cytogenetic diagnostic method (124).

One test depends on the presence of six specific tetranucleotide repeat loci on chromosome 21 that can be amplified using PCR. All Down's syndrome patients were identified as having at least two loci with three alleles (125-127).

One of the promising methods is the quantitative polymerase chain reaction (Q-PCR) to determine the gene dose effects. The method is based on quantitative assessment of PCR products after using primers amplifying DNA fragments located in the pericentromeric, heterochromatic, euchromatic and telomeric regions of chromosome 21 (127-128).

Gene signature for Down syndrome can be based on DNA micro array for down regulation of 10 genes and up-regulation of 1 gene which are located on chromosome 21 in amniotic fluid cells of DS. Down regulated genes are 1COL6A1, CASP5, AKT2, JUN, PYGM, BNIP1, OSF-2, PRSS7, COL3A1, and MBLL. The GSTT1 is the only up-regulated gene in this profile (128-129).

In addition, using SNP analysis technique and matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry is reproducible method for the detection of trisomy 21 (129-130).

Practically, molecular cytogenetic aneuploidy testing by using one-tube Quantitative Fluorescence QF-PCR test for trisomies 13, 18, and 21 is considered a reliable and fast method as well (130-131). Another easy and reproducible technique is using the multiplex ligation-dependent probe amplification (MLPA) in chorionic villous samples that can replace the traditional karyotyping for the detection of euploidies of chromosomes 21, 18, 13, X, and Y (13-132).

Recently a novel method was prescribed to detect trisomy 21 that combined the fetal-specific epigenetic and genetic markers by detecting the hypermethylation of putative promoter of the HLCS (holocarboxylase synthetase) gene in the placenta which is hypomethylated in maternal blood cells. A chromosome-dosage comparison of the hypermethylated HLCS and ZFY loci could distinguish samples of T21 and euploid placental DNA (132-135).

Finally, when gonadal mosaicism is suspected, cytogenetic testing cannot be entirely replaced by molecular testing and a combination of both methods should be applied (135-136).

### MANAGEMENT

Preventive measures consider the main step to decrease the frequency of Down syndrome and the current management is

based on behavioral and rehabilitation training in addition to symptomatic treatments. Never the less, novel therapeutic interventions for Down syndrome will be designed based on disease pathogenesis resulting from the genomic analysis. Recently, it was found that inhibiting of BACE1 in AD and DS may carry therapeutic promise for both diseases (137,138).

### CONCLUSION

Down Syndrome is still the most common genetic cause for mental retardation. However, the advance in molecular sciences added significant progress in understanding the etiology of this syndrome and related diseases.

The scientists at this field are focusing on decreasing the incidence of Down Syndrome worldwide by approaching the following issues; Prevention the occurrence of Down syndrome by avoiding the high risk factors, emphasizing on the education of both couples, performing chromosomal karyotyping or molecular testing in the first 8 weeks of pregnancy, and discussing the pregnancy outcome with both parents with respect to cultural and religious backgrounds.

Nevertheless, more research and investigation on molecular pathology of DS will facilitate applying new diagnostic tools that are more cost effective, less invasive, and safe with high accuracy.

And the promising is to use the genetic maneuvers on prenatal or postnatal stage to treat or improve some or all clinical findings that accompany DS by changing the genetic program.

### References

1. Len Leshin, Trisomy 21: The Story of Down Syndrome. Published on line, <http://www.ds-health.com/trisomy.htm>
2. Mokhtar MM, Abd el-Aziz AM, Nazmy NA, Mahrous HS. Cytogenetic profile of Down syndrome in Alexandria, Egypt. *East Mediterr Health J.* 2003 Jan-Mar; 9(1-2):37-44.
3. Dutta S, Nandagopal K, Gangopadhyay PK, Mukhopadhyay K. Molecular aspects of Down syndrome. *Indian Pediatric.* 2005 Apr;42(4):339-44.
4. Hassold T, Sherman S. Down syndrome: genetic recombination and the origin of the extra chromosome 21. *Clin Genet.* 2000 Feb; 57(2):95-100.
5. Freeman SB, Allen EG, Oxford-Wright CL, Tinker SW, Druschel C, Hobbs CA, O'Leary LA, Romitti PA, Royle MH, Torfs CP, Sherman SL. The National Down Syndrome Project: design and implementation. *Public Health Rep.* 2007 Jan-Feb; 122(1):62-72.
6. Read SG. The distribution of Down's syndrome. *J Ment Defic Res.* 1982 Dec;26 (Pt 4):215-2
7. Jarup L, Morris S, Richardson S, Briggs D, Cobley N, de Hoogh C, Gorog K, Elliott P. Down syndrome in births near landfill sites. *Prenat Diagn.* 2007 Dec;27(13):1191-6.
8. O'Nuallain S, Flanagan O, Raffat I, Avalos G, Dineen B.



- The prevalence of Down syndrome in County Galway. *Ir Med J.* 2007 Jan;100(1):329-31.
9. Rothman KJ, Fabia JJ. Place and time aspects of the occurrence of Down's syndrome. *Am J Epidemiol.* 1976 Jun;103(6):560-4.
10. Muggli EE, Collins VR, Halliday JL. Mapping uptake of prenatal diagnosis for Down syndrome and other chromosome abnormalities across Victoria, Australia. *Aust N Z J Obstet Gynaecol.* 2006 Dec;46(6):492-500.
11. Carothers AD, Castilla EE, Dutra MG, Hook EB. Search for ethnic, geographic, and other factors in the epidemiology of Down syndrome in South America: analysis of data from the ECLAMC project, 1967-1997. *Am J Med Genet.* 2001 Oct 1;103(2):149-56.
12. Dolk H, Loane M, Garne E, De Walle H, Queisser-Luft A, De Vigan C, Addor MC, Gener B, Haeusler M, Jordan H, Tucker D, Stoll C, Feijoo M, Lillis D, Bianchi F. Trends and geographic inequalities in the prevalence of Down syndrome in Europe, 1980-1999. *Rev Epidemiol Sante Publique.* 2005 Nov;53 Spec No 2:2S87-95.
13. Carothers AD, Castilla EE, Dutra MG, Hook EB. Search for ethnic, geographic, and other factors in the epidemiology of Down syndrome in South America: analysis of data from the ECLAMC project, 1967-1997. *Am J Med Genet.* 2001 Oct 1;103(2):149-56.
14. John Davy, "Once Upon A Time," *Observer-Review* (London: August 16, 1981).
15. Stephen Jay Gould, "Dr. Down's Syndrome," *Natural History*, Vol. 89 (April 1980), p. 144.
16. Opitz JM, Gilbert-Barnes EF. Reflections on the Pathogenesis of Down Syndrome. *American Journal of Medical Genetics.* 1990; 7:38-51. Review
17. Rachidi M, Lopes C, Costantine M, Delabar JM. C21orf5, a new member of Dopey family involved in morphogenesis, could participate in neurological alterations and mental retardation in Down syndrome. *DNA Res.* 2005;12(3):203-10.
18. Richards, M.C. *Toward wholeness: Rudolf Steiner education in America.* 1980. University Press of New England, N.H.
19. C J Epstein, J R Korenberg, G Annerén, S E Antonarakis, S Aymé, E Courchesne, L B Epstein, A Fowler, Y Groner, J L Huret. Protocols to establish genotype-phenotype correlations in Down syndrome. *Am J Hum Genet.* 1991 July; 49(1): 207-235.
20. Berto G, Camera P, Fusco C, Imarisio S, Ambrogio C, Chiarle R, Silengo L, Di Cunto F. The Down syndrome critical region protein TTC3 inhibits neuronal differentiation via RhoA and Citron kinase. *J Cell Sci.* 2007 Jun 1; 120(Pt 11):1859-67.
21. Yao YG, Duh EJ. VEGF selectively induces Down syndrome critical region 1 gene expression in endothelial cells: a mechanism for feedback regulation of angiogenesis? *Biochem Biophys Res Commun.* 2004 Aug 27; 321(3):648-56.
22. Eki T, Abe M, Naitou M, Sasanuma SI, Nohata J, Kawashima K, Ahmad I, Hanaoka F, Murakami Y. Cloning and characterization of novel gene, DCRR1, expressed from Down's syndrome critical region of human chromosome 21q22.2. *DNA Seq.* 1997; 7(3-4):153-64.
23. Ramos VC, Vidal-Taboada J, Bergeón S, Egea A, Fisher EM, Scartezzini P, Oliva R. Characterization and expression analysis of the WDR9 gene, located in the Down critical region-2 of the human chromosome 21. *Biochim Biophys Acta.* 2002 Sep 27; 1577(3):377-83.
24. Vidal-Taboada JM, Lu A, Pique M, Pons G, Gil J, Oliva R. Down syndrome critical region gene 2: expression during mouse development and in human cell lines indicates a function related to cell proliferation. *Biochem Biophys Res Commun.* 2000 May 27; 272(1):156-63.
25. Vesa J, Brown Y, Greenfield D, Korenberg JR. Molecular and cellular characterization of the Down syndrome critical region protein 2. *Biochem Biophys Res Commun.* 2005 Mar 4; 328(1):235-42.
26. Song HJ, Park J, Seo SR, Kim J, Paik SR, Chung KC. Down syndrome critical region 2 protein inhibits the transcriptional activity of peroxisome proliferator-activated receptor beta in HEK293 cells. *Biochem Biophys Res Commun.* 2008 Nov 21; 376(3):478-82.
27. Nakamura, A., Hattori, M., Sakaki, Y. Isolation of a novel human gene from the Down syndrome critical region of chromosome 21q22.2. *J. Biochem.* 122: 872-877, 1997.
28. Down syndrome critical region gene 3; dscr3. On line publication, <http://omim.org/entry/605298>
29. Asai S, Yamaki A, Kudoh J, Shimizu N, Shimizu Y. Analysis of the promoter region of human placenta-specific DSCR4 gene. *Biochim Biophys Acta.* 2008 Jan; 1779(1):40-50.
30. Asai S, Yamaki A, Kudoh J, Shimizu N, Shimizu Y. WITHDRAWN: Analysis of the promoter region of human placenta-specific DSCR4 gene. *Biochim Biophys Acta.* 2007 Oct 16.
31. The GeneCards Human Gene Database, on line, <http://www.genecards.org/cgi-bin/carddisp.pl?gene=DSCR4>.
32. Shibuya K, Kudoh J, Minoshima S, Kawasaki K, Asakawa S, Shimizu N. Isolation of two novel genes, DSCR5 and DSCR6, from Down syndrome critical region on human chromosome 21q22.2. *Biochem Biophys Res Commun.* 2000 May 19; 271(3):693-8.
33. Ferrando-Miguel R, Cheon MS, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down Syndrome brain (Part V): overexpression of phosphatidylinositol-glycan class P protein (DSCR5). *Amino Acids.* 2004 Jun; 26(3):255-61. Epub 2004 Mar 16.
34. Shibuya K, Kudoh J, Minoshima S, Kawasaki K, Asakawa S, Shimizu N. Isolation of two novel genes, DSCR5 and DSCR6, from Down syndrome critical region on human chromosome 21q22.2. *Biochem Biophys Res Commun.* 2000 May 19; 271(3):693-8.
35. Vorsanova SG, Iourov IY, Beresheva AK, Demidova IA, Monakhov VV, Kravets VS, Bartseva OB, Goyko EA, Soloviev IV, Yurov YB. Non-disjunction of chromosome 21, alphoid DNA variation, and sociogenetic features of Down syndrome. *Tsitol Genet.* 2005 Nov-Dec; 39(6):30-6.
36. Sheth JJ, Sheth FJ. Gene polymorphism and folate metabolism: a maternal risk factor for Down syndrome. *Indian Pediatr.* 2003 Feb; 40(2):115-23.
37. Kovaleva NV. Germ-line transmission of trisomy 21: Data from 80 families suggest an implication of grandmaternal age and a high frequency of female-specific trisomy rescue. *Mol Cytogenet.* 2010 Mar 18;3:7.
38. Zaki MS, Kamel AA, El-RubyM. Double aneuploidy in three Egyptian patients: Down-Turner and Down-Klinefelter syndromes. *Genet Couns.* 2005; 16(4):393-402.
39. Iurov Iu, Vorsanova SG, Monakhov VV, Beresheva AK, Solov'ev IV, Iurov IuB. Molecular cytogenetic study of Robertsonian translocation 13;14 and Down syndrome in a 3-year-old infant. *Tsitol Genet.* 2004 Nov-Dec;38(6):54-9.
40. Horn D, Neitzel H, Tönnies H, Kalscheuer V, Kunze J, Hinkel GK, Bartsch O. Familial MCA/MR syndrome due to inherited submicroscopic translocation t(18;21)(q22.1q21.3) with breakpoint at the Down syndrome critical region. *Am J Med Genet A.* 2003 Mar 15; 117A (3):236-44.
41. Cyrus Cyril, Chandra N, Jegatheesan T, Chandralekha K, Ramesh A, Gopinath P. M., Marimuthu K. M. Down syndrome child with 48,XXY,+21 karyotype. *Indian Journal*

- of Human Genetics. January-April 2005 Volume 11 Issue 1.
42. Iliopoulos D, Poultsides G, Peristeri V, Kouri G, Andreou A, Voyiatzis N. Double trisomy (48,XXY,+21) in monozygotic twins: case report and review of the literature. *Ann Genet.* 2004 Jan-Mar;47(1):95-8.
43. Labudova O, Kitzmueller E, Rink H, Cairns N, Lubec G. Gene expression in fetal Down syndrome brain as revealed by subtractive hybridization. *Neural Transm Suppl.* 1999;57:125-36.
44. Valero R, Marfany G, González-Angulo O, González-González G, Puelles L, González-Duarte R. USP25, A novel gene encoding a deubiquitinating enzyme, is located in the gene-poor region 21q11.2. *Genomics.* 1999 Dec 15; 62(3):395-405.
45. Rachidi M, Lopes C. Mental retardation in Down syndrome: from gene dosage imbalance to molecular and cellular mechanisms. *Neurosci Res.* 2007 Dec; 59(4):349-69.
46. Lana-Elola E, Watson-Scales SD, Fisher EM, Tybulewicz VL. Down syndrome: searching for the genetic culprits. *Dis Model Mech.* 2011 Sep-Oct;4(5):586-95.
47. Cheon MS, Kim SH, Yaspo ML, Blasi F, Aoki Y, Melen K, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down syndrome brain: challenging the gene dosage effect hypothesis (Part I). *Amino Acids.* 2003; 24(1-2):111-7.
48. Cheon MS, Shim KS, Kim SH, Hara A, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down syndrome brain: Challenging the gene dosage effect hypothesis (Part IV). *Amino Acids.* 2003 Jul; 25(1):41-7.
49. Cheon MS, Bajo M, Kim SH, Claudio JO, Stewart AK, Patterson D, Kruger WD, Kondoh H, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down syndrome brain: challenging the gene dosage effect hypothesis (Part II). *Amino Acids.* 2003; 24(1-2):119-25.
50. Cheon MS, Kim SH, Yaspo ML, Blasi F, Aoki Y, Melen K, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down syndrome brain: challenging the gene dosage effect hypothesis (Part I). *Amino Acids.* 2003; 24(1-2):111-7.
51. Engidawork E, Baiic N, Fountoulakis M, Dierssen M, Greber-Platzer S, Lubec G.J. Beta-amyloid precursor protein, ETS-2 and collagen alpha 1 (VI) chain precursor, encoded on chromosome 21, are not overexpressed in fetal Down syndrome: further evidence against gene dosage effect. *Neural Transm Suppl.* 2001 ;( 61):335-46.
52. Baffico M, Perroni L, Rasore-Quartino A, Scartezzini P. Expression of the human ETS-2 oncogene in normal fetal tissues and in the brain of a fetus with trisomy 21. *Hum Genet.* 1989 Oct; 83(3):295-6.
53. Corti C, Clarkson RW, Crepaldi L, Sala CF, Xuereb JH, Ferraguti F. Gene structure of the human metabotropic glutamate receptor 5 and functional analysis of its multiple promoters in neuroblastoma and astrogloma cells. *J Biol Chem.* 2003 Aug 29;278(35):33105-19.
54. Rampoldi L, Zimbello R, Bortoluzzi S, Tiso N, Valle G, Lanfranchi G, Danieli GA. Chromosomal localization of four MAPK signaling cascade genes: MEK1, MEK3, MEK4 and MEK5. *Cytogenet Cell Genet.* 1997; 78(3-4):301-3.
55. Ait Yahya-Graison E, Aubert J, Dauphinot L, Rivals I, Prieur M, Golfier G, Rossier J, Personnaz L, Creau N, Bléhaut H, Robin S, Delabar JM, Potier MC. Classification of human chromosome 21 gene-expression variations in Down syndrome: impact on disease phenotypes. *Am J Hum Genet.* 2007 Sep; 81(3):475-91.
56. Mao R, Wang X, Spitznagel EL Jr, Frelin LP, Ting JC, Ding H, Kim JW, Ruczinski I, Downey TJ, Pevsner J. Primary and secondary transcriptional effects in the developing human Down syndrome brain and heart. *Genome Biol.* 2005; 6(13):R107.
57. L. Robinson SW, Morris CD, Goldmuntz E, Reller MD, Jones MA, Steiner RD, Maslen C. Missense mutations in CRELD1 are associated with cardiac atrioventricular septal defects. *Am J Hum Genet.* 2003 Apr; 72(4):1047-52.
58. Barlow GM, Chen XN, Shi ZY, Lyons GE, Kurnit DM, Celle L, Spinner NB, Zackai E, Pettenati MJ, Van Riper AJ, Vekemans MJ, Mjaatvedt CH, Korenberg JR. Down syndrome congenital heart disease: a narrowed region and a candidate gene. *Genet Med.* 2001 Mar-Apr;3(2):91-101.
59. Kazuki Y, Kimura M, Nishigaki R, Kai Y, Abe S, Okita C, Shirayoshi Y, Schulz TC, Tomizuka K, Hanaoka K, Inoue T, Oshimura M. Human chromosome 21q22.2-qter carries a gene(s) responsible for downregulation of mlc2a and PEBP in Down syndrome model mice. *Biochem Biophys Res Commun.* 2004 Apr 30;317(2):491-9.
60. Canaider S, Facchin F, Griffoni C, Casadei R, Vitale L, Lenzi L, Frabetti F, D'Addabbo P, Carinci P, Zannotti M, Strippoli P. Proteins encoded by human Down syndrome critical region gene 1-like 2 (DSCR1L2) mRNA and by a novel DSCR1L2 mRNA isoform interact with cardiac troponin I (TNNI3). *Gene.* 2006 May 10; 372:128-36.
61. Freeman SB, Bean LH, Allen EG, Tinker SW, Locke AE, Druschel C, Hobbs CA, Romitti PA, Royle MH, Torfs CP, Dooley KJ, Sherman SL. Ethnicity, sex, and the incidence of congenital heart defects: a report from the National Down Syndrome Project. *Genet Med.* 2008 Mar; 10(3):173-80.
62. Guéant JL, Guéant-Rodriguez RM, Anello G, Bosco P, Brunaud L, Romano C, Ferri R, Romano A, Candito M, Namour B. Genetic determinants of folate and vitamin B12 metabolism: a common pathway in neural tube defect and Down syndrome. *Clin Chem Lab Med.* 2003 Nov; 41(11):1473-7.
63. Biselli JM, Goloni-Bertollo EM, Haddad R, Eberlin MN, Pavarino-Bertelli EC. The MTR A2756G polymorphism is associated with an increase of plasma homocysteine concentration in Brazilian individuals with Down syndrome. *Braz J Med Biol Res.* 2008 Jan; 41(1):34-40.
64. Whittle N, Sartori SB, Dierssen M, Lubec G, Singewald N. Fetal Down syndrome brains exhibit aberrant levels of neurotransmitters critical for normal brain development. *Pediatrics.* 2007 Dec; 120(6):e1465-71.
65. Schmucker D. Molecular diversity of Dscam: recognition of molecular identity in neuronal wiring. *Nat Rev Neurosci.* 2007 Dec;8(12):915-20.
66. Chen BE, Kondo M, Garnier A, Watson FL, Püettmann-Holgado R, Lamar DR, Schmucker D. The molecular diversity of Dscam is functionally required for neuronal wiring specificity in *Drosophila*. *Cell.* 2006 May 5;125(3):607-20.
67. Amano K, Yamada K, Iwayama Y, Detera-Wadleigh SD, Hattori E, Toyota T, Tokunaga K, Yoshikawa T, Yamakawa K. Association study between the Down syndrome cell adhesion molecule (DSCAM) gene and bipolar disorder. *Psychiatr Genet.* 2008 Feb; 18(1):1-10.
68. Hattori D, Millard SS, Wojtowicz WM, Zipursky SL. Dscam-mediated cell recognition regulates neural circuit formation. *Annu Rev Cell Dev Biol.* 2008; 24:597-620.
69. Freeman SB, Torfs CP, Romitti PA, Royle MH, Druschel C, Hobbs CA, Sherman SL. Congenital gastrointestinal defects in Down syndrome: a report from the Atlanta and National Down Syndrome Projects. *Clin Genet.* 2009 Feb; 75(2):180-4.
70. Choi DK, Suzuki Y, Yoshimura S, Togashi T, Hida M, Taylor TD, Wang Y, Sugano S, Hattori M, Sakaki Y. Molecular cloning and characterization of a gene expressed in mouse developing tongue, mDscr5 gene, a homolog of

- human DSCR5 (Down syndrome Critical Region gene 5). *Mamm Genome*. 2001 May; 12(5):347-51.
71. Linabery AM, Blair CK, Gamis AS, Olshan AF, Heerema NA, Ross JA. Congenital abnormalities and acute leukemia among children with Down syndrome: a Children's Oncology Group study. *Cancer Epidemiol Biomarkers Prev*. 2008 Oct;17(10):2572-7.
72. Webb D, Roberts I, Vyas P. Haematology of Down syndrome. *Arch Dis Child Fetal Neonatal Ed*. 2007 Nov; 92(6):F503-7.
73. Magalhães IQ, Splendore A, Emerenciano M, Córdoba MS, Córdoba JC, Allemand PA, Ferrari I, Pombo-de-Oliveira MS. Transient neonatal myeloproliferative disorder without Down syndrome and detection of GATA1 mutation. *J Pediatr Hematol Oncol*. 2005 Jan;27(1):50-2.
74. Ahmed M, Sternberg A, Hall G, Thomas A, Smith O, O'Marcaigh A, Wynn R, Stevens R, Addison M, King D, Stewart B, Gibson B, Roberts I, Vyas P. Natural history of GATA1 mutations in Down syndrome. *Blood*. 2004 Apr 1;103(7):2480-9.
75. Vyas P, Crispino JD. Molecular insights into Down syndrome-associated leukemia. *Curr Opin Pediatr*. 2007 Feb; 19(1):9-14.
76. Magalhães IQ, Splendore A, Emerenciano M, Córdoba MS, Córdoba JC, Allemand PA, Ferrari I, Pombo-de-Oliveira MS. Transient neonatal myeloproliferative disorder without Down syndrome and detection of GATA1 mutation. *J Pediatr Hematol Oncol*. 2005 Jan; 27(1):50-2.
77. Pine SR, Guo Q, Yin C, Jayabose S, Levendoglu-Tugal O, Ozkaynak MF, Sandoval C. GATA1 as a new target to detect minimal residual disease in both transient leukemia and megakaryoblastic leukemia of Down syndrome. *Leuk Res*. 2005 Nov; 29(11):1353-6.
78. Shimada A, Xu G, Toki T, Kimura H, Hayashi Y, Ito E. Fetal origin of the GATA1 mutation in identical twins with transient myeloproliferative disorder and acute megakaryoblastic leukemia accompanying Down syndrome. *Blood*. 2004 Jan 1; 103(1):366.
79. Ge Y, Jensen TL, Stout ML, Flatley RM, Grohar PJ, Ravindranath Y, Matherly LH, Taub JW. The role of cytidine deaminase and GATA1 mutations in the increased cytosine arabinoside sensitivity of Down syndrome myeloblasts and leukemia cell lines. *Cancer Res*. 2004 Jan 15; 64(2):728-35.
80. Khan I, Malinge S, Crispino J. Myeloid leukemia in down syndrome. *Crit Rev* 0. 2011;16(1-2):25-36.
81. De Vita S, Mulligan C, McElwaine S, Dagna-Bricarelli F, Spinelli M, Basso G, Nizetic D, Groet J. *Br J Haematol*. Loss-of-function JAK3 mutations in TMD and AMKL of Down syndrome. 2007 May; 137(4):337-41.
82. Malinge S, Ben-Abdelali R, Settegrana C, Radford-Weiss I, Debre M, Beldjord K, Macintyre EA, Villeval JL, Vainchenker W, Berger R, Bernard OA, Delabesse E, Penard-Lacronique V. Novel activating JAK2 mutation in a patient with Down syndrome and B-cell precursor acute lymphoblastic leukemia. *Blood*. 2007 Mar 1; 109(5):2202-4.
83. Osato M, Ito Y. Increased dosage of the RUNX1/AML1 gene: a third mode of RUNX leukemia. *Crit Rev Eukaryot Gene Expr*. 2005; 15(3):217-28.
84. Sommer CA, Pavarino-Bertelli EC, Goloni-Bertollo EM, Henrique-Silva F. Identification of dysregulated genes in lymphocytes from children with Down syndrome. *Genome*. 2008 Jan;51(1):19-29.
85. Murphy M, Insoft RM, Pike-Nobile L, Derbin KS, Epstein LB. Overexpression of LFA-1 and ICAM-1 in Down syndrome thymus. Implications for abnormal thymocyte maturation. *J Immunol*. 1993 Jun 15;150(12):5696-703.
86. Sustrová M, Saríková V. *Bratisl Lek Listy*. Down's syndrome--effect of increased gene expression in chromosome 21 on the function of the immune and nervous system. 1997 Apr;98(4):221-8.
87. Barrena MJ, Echaniz P, Garcia-Serrano C, Zubillaga P, Cuadrad E. *Clin Exp Immunol*. Differential expression of lymphocyte function-associated antigen (LFA-1) on peripheral blood leucocytes from individuals with Down's syndrome. 1992 Apr;88(1):41-4.
88. Blouin JL, Duriaux Sail G, Antonarakis SE. This HCS gene may contribute in a gene dosage-dependent manner to the phenotype of Down syndrome. Mapping of the human holocarboxylase synthetase gene (HCS) to the Down syndrome critical region of chromosome 21q22. *Ann Gene*. 1996; 39(3):185-8.
89. Gulesserian T, Engidawork E, Fountoulakis M, Lubec G. Manifold decrease of sialic acid synthase in fetal Down syndrome brain. *Amino Acids*. 2007 Jan;32(1):141-4.
90. Pastore A, Tozzi G, Gaeta LM, Giannotti A, Bertini E, Federici G, Digilio MC, Piemonte F. Glutathione metabolism and antioxidant enzymes in children with Down syndrome. *J Pediatr*. 2003 May; 142(5):583-5.
91. Labudova O, Cairns N, Koeck T, Kitzmueller E, Rink H, Lubec G. Thyroid stimulating hormone-receptor overexpression in brain of patients with Down syndrome and Alzheimer's disease. *Life Sci*. 1999; 64(12):1037-44.
92. Yoo BC, Fountoulakis M, Cairns N, Lubec G. Changes of voltage-dependent anion-selective channel proteins VDAC1 and VDAC2 brain levels in patients with Alzheimer's disease and Down syndrome. *Electrophoresis*. 2001 Jan; 22(1):172-9.
93. Johannisson R, Gropp A, Winking H, Coerdts W, Rehder H, Schwinger E. Down's syndrome in the male. Reproductive pathology and meiotic studies. *Hum Genet*. 1983; 63(2):132-8.
94. Ni B, Li LY, Yin ZC, Zou YH, Li H. Molecular cloning for an alternatively splicing cDNA of human PKNOX1 gene and its expression analysis. *Yi Chuan Xue Bao*. 2004 Jan;31(1):19-25.
95. Whittle N, Sartori SB, Dierssen M, Lubec G, Singewald N. Fetal Down syndrome brains exhibit aberrant levels of neurotransmitters critical for normal brain development. *Pediatrics*. 2007 Dec; 120(6):e1465-71.
96. Berto G, Camera P, Fusco C, Imarisio S, Ambrogio C, Chiarle R, Silengo L, Di Cunto F. The Down syndrome critical region protein TTC3 inhibits neuronal differentiation via RhoA and Citron kinase. *J Cell Sci*. 2007 Jun 1; 120(Pt 11):1859-67.
97. Yamaki A, Tochigi J, Kudoh J, Minoshima S, Shimizu N, Shimizu Y. Molecular mechanisms of human single-minded 2 (SIM2) gene expression: identification of a promoter site in the SIM2 genomic sequence. *Gene*. 2001 May 30; 270(1-2):265-75.
98. Krapfenbauer K, Yoo BC, Cairns N, Lubec G. Differential display reveals deteriorated mRNA levels of NADH3 (complex I) in cerebellum of patients with Down syndrome. *J Neural Transm Suppl*. 1999; 57:211-20.
99. Labudova O, Kitzmueller E, Rink H, Cairns N, Lubec G. Gene expression in fetal Down syndrome brain as revealed by subtractive hybridization. *J Neural Transm Suppl*. 1999; 57:125-36.
100. Murakami N, Xie W, Lu RC, Chen-Hwang MC, Wieraszko A, Hwang YW. Phosphorylation of amphiphysin I by minibrain kinase/dual-specificity tyrosine phosphorylation-regulated kinase, a kinase implicated in Down syndrome. *J Biol Chem*. 2006 Aug 18; 281(33):23712-24.
101. Martínez de Lagrán M, Altafaj X, Gallego X, Martí E,

- Estivill X, Sahún I, Fillat C, Dierssen M. Motor phenotypic alterations in TgDyrk1a transgenic mice implicate DYRK1A in Down syndrome motor dysfunction. *Neurobiol Dis.* 2004 Feb; 15(1):132-42.
102. Hämmerle B, Elizalde C, Tejedor FJ. The spatio-temporal and subcellular expression of the candidate Down syndrome gene Mnb/Dyrk1A in the developing mouse brain suggests distinct sequential roles in neuronal development. *Eur J Neurosci.* 2008 Mar; 27(5):1061-74.
103. Hämmerle B, Elizalde C, Galceran J, Becker W, Tejedor FJ. The MNB/DYRK1A protein kinase: neurobiological functions and Down syndrome implications. *J Neural Transm Suppl.* 2003 ;( 67):129-37.
104. Maenz B, Hekerman P, Vela EM, Galceran J, Becker W. Characterization of the human DYRK1A promoter and its regulation by the transcription factor E2F1. *BMC Mol Biol.* 2008 Mar 26;9:30.
105. Raich WB, Moorman C, Laceyfield CO, Lehrer J, Bartsch D, Plasterk RH, Kandel ER, Hobert O. Characterization of *Caenorhabditis elegans* homologs of the Down syndrome candidate gene DYRK1A. *Genetics.* 2003 Feb; 163(2):571-80.
106. Cheon MS, Kim SH, Yaspo ML, Blasi F, Aoki Y, Melen K, Lubec G. Protein levels of genes encoded on chromosome 21 in fetal Down syndrome brain: challenging the gene dosage effect hypothesis (Part I). *Amino Acids.* 2003; 24(1-2):111-7.
107. Esposito G, Imitola J, Lu J, De Filippis D, Scuderi C, Ganesh VS, Folkert R, Hecht J, Shin S, Iuvone T, Chesnut J, Steardo L, Sheen V. Genomic and functional profiling of human Down syndrome neural progenitors implicates S100B and aquaporin 4 in cell injury. *Hum Mol Genet.* 2008 Feb 1; 17(3):440-57.
108. Shapiro LA, Marks A, Whitaker-Azmitia PM. Increased clusterin expression in old but not young adult S100B transgenic mice: evidence of neuropathological aging in a model of Down Syndrome. *Brain Res.* 2004 Jun 4; 1010(1-2):17-21.
109. Lai F, Kammann E, Rebeck GW, Anderson A, Chen Y, Nixon RA. Neurology. APOE genotype and gender effects on Alzheimer disease in 100 adults with Down syndrome. *1999 Jul 22; 53(2):331-6.*
110. Maslen CL, Babcock D, Robinson SW, Bean LJ, Dooley KJ, Willour VL, Sherman SL. CRELD1 mutations contribute to the occurrence of cardiac atrioventricular septal defects in Down syndrome. *Am J Med Genet A.* 2006 Nov15; 140(22):2501-5.
111. Wikipedia, Amyloid beta (A $\beta$  or A $\beta$ eta), on line, <http://en.wikipedia.org/wiki/Special:Search?search=Amyloid+beta+%28A%CE%B2+or+Abeta%29+&go=Go>
112. Grimm MO, Rothhaar TL, Hartmann T. The role of APP proteolytic processing in lipid metabolism. *Exp Brain Res.* 2011 Dec 17.
113. Tomita S, Kirino Y, Suzuki T. Cleavage of Alzheimer's amyloid precursor protein (APP) by secretases occurs after O-glycosylation of APP in the protein secretory pathway. Identification of intracellular compartments in which APP cleavage occurs without using toxic agents that interfere with protein metabolism. *J Biol Chem.* 1998 Mar 13;273(11):6277-84.
114. Wolvetang EW, Bradfield OM, Tymms M, Zavarsek S, Hatzistavrou T, Kola I, Hertzog PJ. The chromosome 21 transcription factor ETS2 transactivates the beta-APP promoter: implications for Down syndrome. *Biochim Biophys Acta.* 2003 Jul 28;1628(2):105-10.
115. Sun X, Tong Y, Qing H, Chen CH, Song W. Increased BACE1 maturation contributes to the pathogenesis of Alzheimer's disease in Down syndrome. *FASEB J.* 2006 Jul; 20(9):1361-8.
116. Swatton JE, Sellers LA, Faull RL, Holland A, Iritani S, Bahn S. *Eur J Neurosci.* Increased MAP kinase activity in Alzheimer's and Down syndrome but not in schizophrenia human brain. 2004 May; 19(10):2711-9.
117. Lucarelli P, Piciullo A, Verdecchia M, Palmarino M, Arpino C, Curatolo P. The role of -850 tumor necrosis factor-alpha and apolipoprotein E genetic polymorphism in patients with Down's syndrome-related dementia. *Neurosci Lett.* 2003 Nov 27; 352(1):29-32.
118. Ichinohe A, Kanaumi T, Takashima S, Enokido Y, Nagai Y, Kimura H. Cystathionine beta-synthase is enriched in the brains of Down's patients. *Biochem Biophys Res Commun.* 2005 Dec 23; 338(3):1547-50.
119. Hoeffler CA, Dey A, Sachan N, Wong H, Patterson RJ, Shelton JM, Richardson JA, Klann E, Rothermel BA. The Down syndrome critical region protein RCAN1 regulates long-term potentiation and memory via inhibition of phosphatase signaling. *J Neurosci.* 2007 Nov 28; 27(48):13161-72.
120. Keating DJ, Dubach D, Zanin MP, Yu Y, Martin K, Zhao YF, Chen C, Porta S, Arbonés ML, Mittaz L, Pritchard MA. *Hum Mol Genet.* DSCR1/RCAN1 regulates vesicle exocytosis and fusion pore kinetics: implications for Down syndrome and Alzheimer's disease. 2008 Apr 1; 17(7):1020-30.
121. Yeghiazaryan K, Turhani-Schatzmann D, Labudova O, Schuller E, Olson EN, Cairns N, Lubec G. Downregulation of the transcription factor scleraxis in brain of patients with Down syndrome. *J Neural Transm Suppl.* 1999; 57:305-14.
122. Kim SH, Shim KS, Lubec G. Human brain nascent polypeptide-associated complex alpha subunit is decreased in patients with Alzheimer's disease and Down syndrome. *J Investig Med.* 2002 Jul; 50(4):293-301.
123. Shibuya K, Nagamine K, Okui M, Ohsawa Y, Asakawa S, Minoshima S, Hase T, Kudoh J, Shimizu N. Initial characterization of an uromodulin-like 1 gene on human chromosome 21q22.3. *Biochem Biophys Res Commun.* 2004 Jul 9; 319(4):1181-9.
124. Tracy Hampton. Down Syndrome Protein Deters Cancer. *JAMA.* 2005; 293:284-285.
125. Krabchi K, Gadji M, Samassekou O, Grégoire MC, Forest JC, Drouin R. Quantification of fetal nucleated cells in maternal blood of pregnant women with a male trisomy 21 fetus using molecular cytogenetic techniques. *Prenat Diagn.* 2006 Jan; 26(1):28-34.
126. Kazunori Shibuya, Jun Kudoh, Michiyo Okui and Nobuyoshi Shimizu. Identification of a novel zinc finger protein gene (ZNF298) in the GAP2 of human chromosome 21q. *Biochemical and Biophysical Research Communications*, Volume 332, Issue 2, 1 July 2005, Pages 557-568.
127. Cirigliano V, Voglino G, Adinolfi M. Non-invasive screening and rapid QF-PCR assay can greatly reduce the need for conventional cytogenetic analyses in prenatal diagnosis. *Reprod Biomed Online.* 2005 Dec; 11(6):671-3.
128. Wang S, Jia C, Ren G, Ma Y, Lü W, Ding F, Han J. Molecular diagnosis of Down's syndrome. *Chin Med J.* 2003 Nov; 116(11):1773-5.
129. Nowacka J, Helszer Z, Walter Z, Płucienniczak A, Kazewski B. Adaptation of PCR technique for quantitative estimation of genetic material from different regions of chromosome 21 in cases of trisomy 21. *Acta Biochim Pol.* 2004; 51(4):995-1001.
130. Chung IH, Lee SH, Lee KW, Park SH, Cha KY, Kim NS, Yoo HS, Kim YS, Lee S. Gene expression analysis of cultured amniotic fluid cell with Down syndrome by DNA microarray. *J Korean Med Sci.* 2005 Feb; 20(1):82-7.

131. Huang DJ, Nelson MR, Zimmermann B, Dudarewicz L, Wenzel F, Spiegel R, Nagy B, Holzgreve W, Hahn S. Reliable detection of trisomy 21 using MALDI-TOF mass spectrometry. *Genet Med*. 2006 Nov;8(11):728-34.
132. Ogilvie CM, Donaghue C, Fox SP, Docherty Z, Mann K. Rapid prenatal diagnosis of aneuploidy using quantitative fluorescence-PCR (QF-PCR). *0*. 2005 Mar;53(3):285-8.
133. Kooper AJ, Faas BH, Feuth T, Creemers JW, Zondervan HH, Boekkooi PF, Quartero RW, Rijnders RJ, van der Burgt I, van Kessel AG, Smits AP. Detection of chromosome aneuploidies in chorionic villus samples by multiplex ligation-dependent probe amplification. *J Mol Diagn*. 2009 Jan; 11(1):17-24.
134. Fauret AL, Bilan F, Patri S, Couet D, Marechaud M, Pierre F, Gilbert-Dussardier B, Kitzis A. Molecular biology usefulness for rapid diagnosis of Down's syndrome and common aneuploidies. *Gynecol Obstet Fertil*. 2009 Jul-Aug;37(7-8):611-9.
135. Yu K. Tong, Shengnan Jin, Rossa W.K. Chiu, Chunming Ding, K.C. Allen Chan, Tak Y. Leung, Ling Yu, Tze K. Lau, and Y.M. Dennis Lo. Noninvasive Prenatal Detection of Trisomy 21 by an Epigenetic-Genetic Chromosome-Dosage Approach. *Clin Chem* 2010 56: 90-98.
136. Kovaleva NV, Tahmasebi-Hesari M. Detection of gonadal mosaicism in parents of children with Down syndrome. *Tsitol Genet*. 2007 Sep-Oct; 41(5):36-42.
137. Antonarakis SE. 10 years of Genomics, chromosome 21, and Down syndrome. *Genomics*. 1998 Jul 1; 51(1):1-16.
138. Sun X, Tong Y, Qing H, Chen CH, Song W. Increased BACE1 maturation contributes to the pathogenesis of Alzheimer's disease in Down syndrome. *FASEB J*. 2006 Jul;20(9):1361-8.

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