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DOWN SYNDROME AND LEUKEMIA – AN OVERVIEW OF CYTOGENETIC AND MOLECULAR EVENTS*

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Down syndrome (DS) is the most common trisomic disorder seen in humans with an incidence of 1/700-1/1000 live births. In addition to its characteristic phenotypic features, heart defects, gastrointestinal system abnormalities, premature aging, and hematological disorders, including an increased risk of leukemia, are associated with DS. The relation between DS and leukemia has been known for many years and has continued to be a focus of interest not only for the probable role of trisomy 21 in the development of leukemia at the molecular level but also for the unique features o fthe leukemia. These include the predominance of acute myeloid leukemia (AML) in younger children with the megakaryoblastic subtype and spontaneous regression of myeloproliferation in newborns.

Acute Leukemia

Down syndrome patients have a 10-to 20-fold increased risk of developing both acute lymphoblastic leukemia (ALL) and AML compared to non-DS children; one percent of DS individuals will develop leukemia during their lifetime[1]. Although most cases of leukemia in DS individuals occur early in childhood, the risk spans into adulthood[2]. AML predominates in DS patients under three years of age, while the relative proportions of ALL and AML are the same in older children with or without DS[3-4]. A large proportion of AML cases in DS children have been classified as "megakaryoblastic" (AML-M7) by positive electron microscopic detection of platelet peroxidases and/or staining with monoclonal antibodies against platelet antigens[5]. In DS patients, the incidence of AML-M7 is approximately 600 times higher than that in non-DS children with AML[6]. AML-M7 is usually associated with a myelodysplastic phase either in the neonatal period or immediately preceding the development of AML, and myelofibrosis is common in patients with DS[3]. Although extramedullary involvement, especially bone infiltration, is relatively common in non-DS patients with AML-M7, the development of granulocytic sarcoma is very rare in DS patients with AML-M7[7].

A significant number of AML patients with DS have additional cytogenetic changes similar to the cytogenetic abnormalities of de novo cases of AML in non-DS patients[4, 8-9]. Trisomies of chromosomes 1q, 8, and monosomy 7 are the most

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common numerical abnormalities, while tetrasomy 21 is the most frequent quantitative microscopic abnormality in AML-M7 in DS[8-10]. Non-DS AML-M7 is associated with t (1;22) (p13;q13), while this is not seen in DS cases[11-12]. However, numerical abnormalities of chromosome 21 have been demonstrated in AML-M7 patients without Down syndrome as either primary or nonrandom secondary changes. Favorable treatment outcomes have been reported in DS patients with ALL using intensive regimens which were associated with excessive methotrexate toxicity[13]. DS patients with AML have a superior response rate to intensive chemotherapy with high dose cytosine arabinoside (AraC) containing regimens compared to non-DS patients[9]. Low dose AraC therapy has also been reported to be effective in some DS patients with myelodysplastic syndrome or AML-M7[6, 14].

Transient Myeloproliferative Disorder

Transient myeloproliferative disorder (TMD), also known as transient leukemia, transient leukemoid reaction, or transient abnormal myelopoiesis, occurs in neonates with DS. Cases of TMD may not be recognized due to its mild symptoms, uncomplicated course and self-limiting nature; some cases may even resolve in utero. For these reasons, it is very hard to estimate the true incidence of this disorder in the DS population. It is usually recognized during the first month of life; prenatal diagnosis has also been reported[3, 15]. The most fascinating characteristic of this disorder is that spontaneous regression occurs within several months with supportive care alone, which may include red cell and platelet transfusions. TMD is frequently suspected when a high white blood cell (WBC) count with blasts are observed on the peripheral blood smear. The bone marrow findings of TMD show increased blasts without evidence of myelofibrosis, while the ratio of the blasts in the bone marrow is lower compared to that in the peripheral blood. Skin involvement which occurs frequently in neonatal AML, does not occur in TMD.

Although TMD is a self-limiting disorder, infants can have life-threatening complications, including hyperviscosity secondary to hyperleukocytosis, disseminated intravascular coagulation and pericardial tamponade, which all require rapid intervention such as exchange transfusions[4, 16-17]. Hepatofibrosis, a rare complication, has been the leading cause of death in TMD[18-19]. Hydrops observed in some DS fetuses is hypothesized to be due to hepatofibrosis associated with TMD[3]. The majority of DS neonates with TMD do not have clonal chromosomal abnormalities which is the opposite of DS patients with AML[8-20]. The most frequent cytogenetic abnormality observed in infant leukemias, 11q23 rearrangement, has not been reported in TMD. Cases of TMD have also been reported in phenotypically normal infants with trisomy 21 mosaicism and in neonates without any constitutional cytogenetic abnormalities[17, 21-23]. Interestingly, in the latter

cases, trisomy 21 has been identified in the blast cells alone while following resolution of TMD, trisomy 21 is no longer identified in either peripheral or bone marrow cells. These findings strongly suggest a relationship between TMD and trisomy 21. Although spontaneous resolution of the hematological abnormalities occurs in DS patients with TMD, about one-fourth of these patients later develop AML. There is no clear-cut evidence of clonal evolution of TMD to AML, though it may be likely in view of the increased risk of developing particularly AML-M7.

The blast cells of TMD morphologically resemble AML-M7 cells with cytoplasmic projections and bluish staining of the cytoplasm with the Giemsa stain. These cells have been shown to express some platelet antigens and contain platelet associated enzymes. Erythroid antigens are expressed in some cases and, recently, the same expression pattern of the transcription factors GATA-1 and GATA-2 in normal erythroid precursors has been reported in blasts of TMD and AML-M7 patients with DS[24]. These findings suggest that TMD and AML-M7 arise from a common megakaryocytic-erythroid progenitor or from a myeloid progenitor with the capacity to differentiate into erythroid and megakaryocytic lineages. Studies with fluorescence in situ hybridization in MDS patients with DS further support the erythroid origin of malignant cells by showing the trisomy 8 in blasts and normoblasts but not in myeloid and lymphoid cells[25]. Leukemic blasts from DS patients with AML-M7 produce both megakaryocyte and basophil/mast cell precursors in culture[26]. The observation of scattered hemopoietic cells in fibrous tissues, including pleomorphic megakaryocytes in the livers of patients with liver fibrosis, provided evidence of a mechanisms to explain hepatic fibrosis seen in some DS patients with TMD. Japanese investigators proposed that liver fibrosis in TMD results from the secretion of the cytokines platelet-derived growth factor (PDGF), transforming growth factor-β (TGF-β), and platelet factor 4, which stimulate collagen synthesis, by abnormal hemopoietic cells in the liver and may be analogous to myelofibrosis in AML-M7[27]. Recently, slight fibrotic changes in the bone marrow of two DS patients with TMD and hepatic fibrosis have been reported[28].

Is TMD a true neoplastic process? Despite the monoclonal nature of TMD in DS which has been shown by analysis of the methylation patterns of the X chromosomes using restriction fragment length polymorphisms in female patients, Miyauchi et al.[27] proposed that TMD was not a true malignancy based on the following observations: i) TMD undergoes spontaneous regression in most cases without chemotherapy, ii) trisomy 21 is the only chromosomal abnormality in most TMD cases, while DS patients with either MDS or AML often have additional chromosomal abnormalities, iii) extramedullary hematopoiesis consists of both blast cells and mature trilineage hematopoietic cells as opposed to a homogenous blast infiltration in AML cases, and iv) mature hematopoietic colonies with normal features which are usually absent in acute leukemia grow

in conjunction with the abnormal blast colonies in vitro[29-30]. As a result, TMD can be defined as a self-limited, monoclonal proliferation of hematopoietic progenitors with trisomy 21 which undergoes spontaneous maturation in tissues.

Hematological Abnormalities in Down Syndrome

Newborn infants with DS frequently have hematological abnormalities besides TMD including polycythemia, macrocytosis, and thrombocytosis. Polcythemia is common soon after birth, while macrocytosis and thrombocytosis may develop later in infancy[31]. Thrombocytosis usually subsides by the end of the first year of life, while DS individuals continue to have higher red cell mean corpuscular volumes (MCV) throughout their lives[31-32]. Increased red blood cell (RBC) turnover secondary to accelerated cell membrane aging or other unexplained intrinsic defects giving rise to a younger red cell population has been proposed as a mechanism to explain high MCV in DS patients. A recent study suggests that DS patients with high MCV have a normal RBC life span as RBC hexokinase activity and creatine levels were normal[33]. It has also been speculated that alterations in the folate remethylation pathway secondary to increased cystathionine β-synthase (CBS) activity (localized to 21q22.3) may play a part. These hematological changes suggest that trisomy 21 has an effect on hematopoiesis, particularly the erythroid and megakaryocytic lineages, especially during the perinatal period.

Leukemogenesis and Trisomy 21

How does trisomy 21 contribute to the development of neoplasia? It is known that patients with constitutional trisomies of chromosomes 8, 9, 13, 18, and 21 are at an increased risk of developing various hematological malignancies and solid tumors compared to the general population[34]. This suggests a relationship between the underlying mechanism giving rise to trisomy and the development of the neoplastic process. This may not be true based on the following arguments. A high percentage of patients with constitutional trisomies do not develop cancer; this may be because additional mutational events (hits) must occur for the neoplasia to develop. The types of tumors observed in constitutional trisomies more frequently affect certain organs (leukemia and cutaneous syringoma in trisomy 21, kidney and liver tumors in trisomy 18, kidney tumors and neuroblastoma in trisomy 13). This suggests that an increased gene dosage of genes located on a particular trisomic chromosome may play a central role in the development of neoplasia rather than the underlying mechanisms leading to trisomy itself. This is further supported by the fact that acquired trisomies of the same chromosomes are occasionally detected in patients with the same types of tumors without constitutional abnormalities. But it must be kept in mind that there may be significant differences in the contribution to the development of neoplasia by the trisomic chromosome in a constitutionally trisomic cell and in a cell with an acquired trisomy.

Oncogenes and Chromosome 21

The oncogenes AML1, ERG and ETS-2 have been localized to chromosome 21 and play an important role in the neoplastic processes. The AML1 gene is associated with AML-M2 with the t(8;21) (q22;q22) cytogenetic abnormality and with the cryptic TEL-AML1 chimeric transcript, which has been identified in 25 percent of cases of pediatric B-precursor ALL[35-37]. These observations suggest that the AML1 gene may be involved in both TMD and leukemia in DS patients. AML1 is a transcription factor which binds to the enhancer core motif in the transcriptional regulatory region of the granulocyte-macrophage colony-stimulating factor (GM-CSF), interleukin-3 (IL-3), and the colony stimulating factor 1 (CSF-1) receptor. The 8;21 translocation of AML-M2 results in the fusion of the AML1 gene to the transcription factor ETO gene, located on chromosome 8, and production of the AML-ETO chimeric protein. This protein interacts with the enhancer core DNA sequence and can alter the transcriptional regulation of AML1 target genes via interference with AML1-dependent transactivation, leading to leukemogenesis.

AML1-deficient mice (generated through gene knock-out targeting in embryonal stem cells), have complete absence of fetal liver derived hematopoiesis[38]. This finding strongly suggests that the AML1 gene plays an essential role in definitive hematopoiesis along with the other transcription factors[39]. Linkage analysis studies have identified that the gene of a familial platelet disorder (FPD), characterized by both quantitative and qualitative platelet abnormalities with a propensity to develop myeloid malignancies, is localized to 21q22[40]. This gene lies in close proximity to the AML1 gene on chromosome 21 and the hematological abnormalities observed in FPD resemble the clinical picture of DS patients with TMD in several aspects.

Genomic Imprinting, Chromosome 21 and Cancer

Since a large proportion of patients with DS or other constitutional trisomies do not develop cancer, it can be postulated that additional mutational steps are required for the multistep process of carcinogenesis. This hypothesis raises the following question. what are those additional hits? Studies have focused on the characteristics of the extra copy of chromosome 21 in cases of DS with leukemia. The increased incidence of TMD or leukemia in DS is not associated with increasing maternal age. "Atypical" constitutional karyotypes, including isochromosome 21, double-ring 21, and mosaic trisomy, each of which generates identical copies of the entire long arm of chromosome, have been reported with increased frequency in DS patients with TMD and AML-M7[41]. This condition, known as "disomic homozygosity", in which two identical copies of a single allele are inherited, may have some important implications in the development of the neoplastic process.

Some of the characteristics of DS may be due to overexpression of a critical susceptibility allele of a gene by disomic homozygosity (the presence of two identical copies of a gene from the parent of origin of the trisomic chromosome) as proposed by Feingold et al[42]. Trisomy 21 arising from mitotic nondisjunction would result in disomic homozygosity of the entire chromosome 21, while nondisjunction occurring during either meiosis I or II would result in disomic homozygosity of a region on the chromosome depending upon the extent of crossovers between the nondisjoined chromosomes (Fig. 1). In accordance with

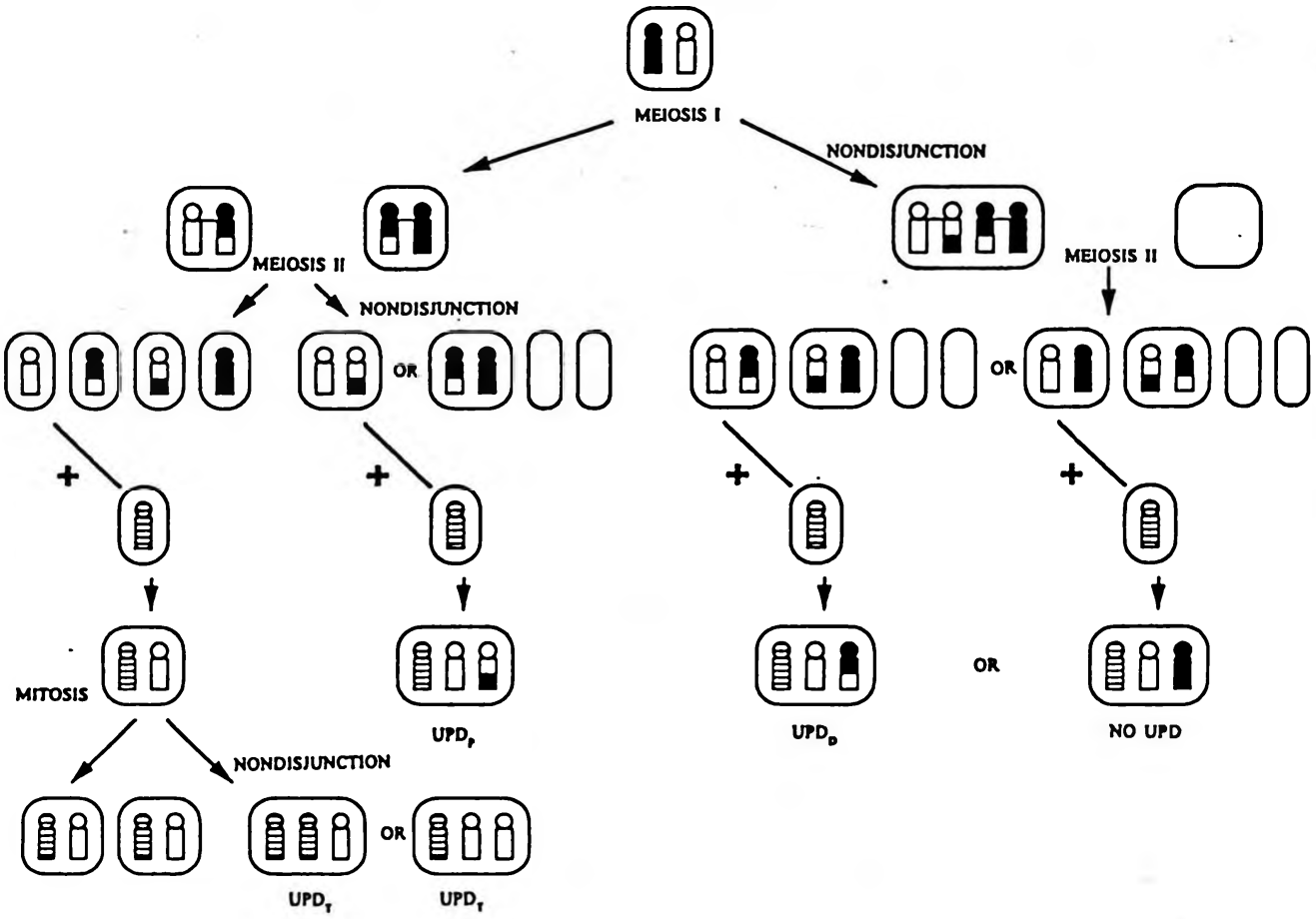

Fig. 1: Generation of uniparental disomy (UPD) in different types of nondisjunction. Abbreviations: UPD_T: uniparental disomy-total; UPD_P: uniparental disomy-proximal; UPD_D: uniparental disomy-distal.

the above hypothesis, earlier studies showed that the copies of chromosome 21 from the parent of origin were identical at the centromere (interpreted as being consistent with the meiosis II or mitotic nondisjunction) in TMD patients with DS[43-44]. A more recent study provided additional evidence of an increased frequency of disomic homozygosity in DS individuals with TMD and AML-M7, especially in the proximal 21q region[41]. These findings suggest a possible contributory role of disomic homozygosity in some TD or AML patients with DS.

Genomic imprinting is the differential expression of a gene on a chromosome which is dependent on whether the gene is inherited from the father or mother.

The parental origin of the extra chromosome in trisomy 21 is similar in patients with or without leukemia[45-46]. Maternal and paternal uniparental disomies for chromosome 21 result in normal phenotype, providing evidence that genomic imprinting on chromosome 21 does not occur[47-48]. These two lines of evidence argue against genomic imprinting as a possible mechanism contributing to the development of neoplasia in DS.

Alternatively, the significance of the disomic homozygosity can be based on the genomic imprinting mechanism giving rise to inactivation of several critical genes with identical base-pair sequences located on chromosome 21. This would lead to decreased expression of these genes while leaving a possibly defective allele on the third copy of chromosome 21 dominant (Fig. 2). Evidence against this assumption is that meiosis II and mitotic nondisjunctions ending in identical or near-identical copies of chromosome 21 are known to occur in both parents of infants with TMD and also AML patients with DS[43-44].

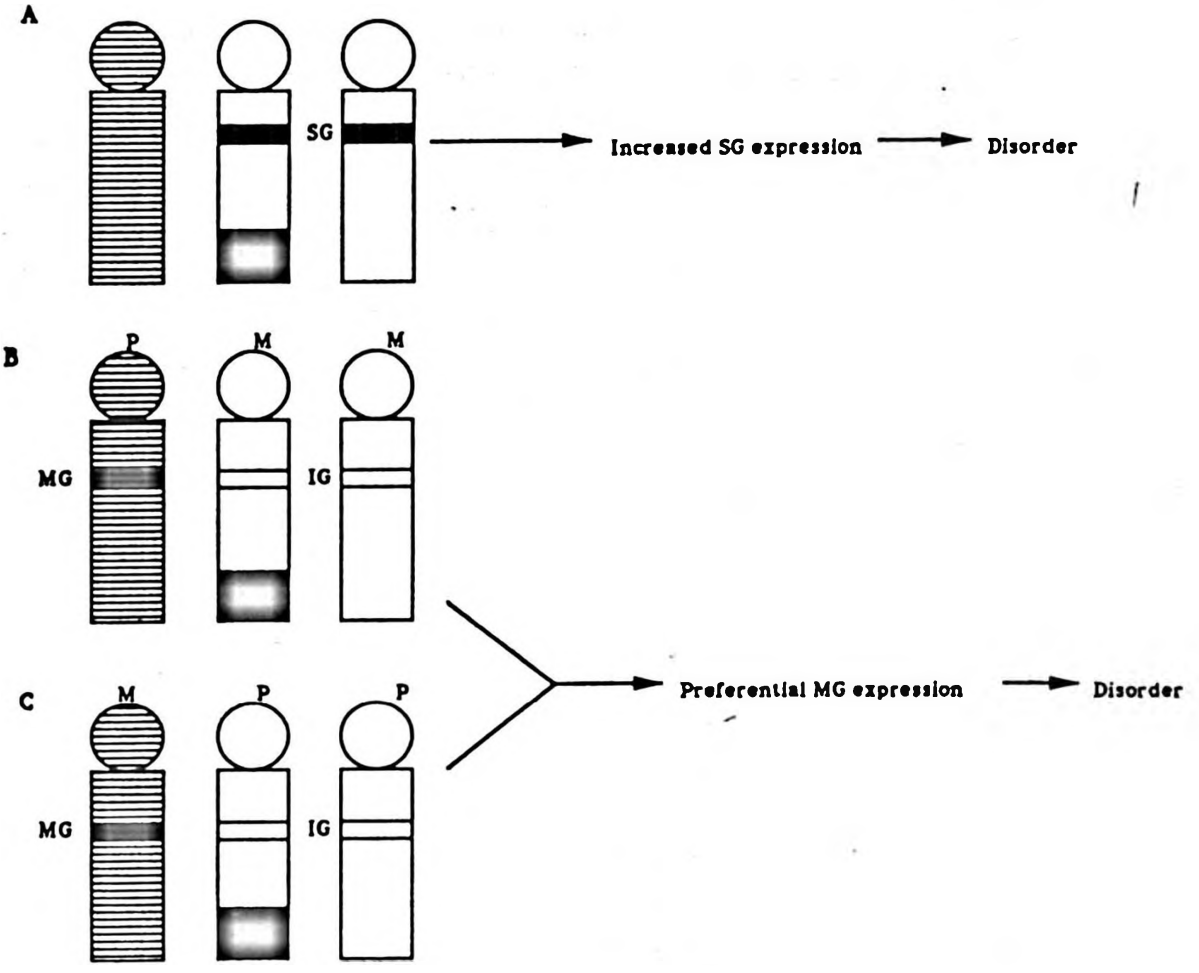

Fig. 2: Different pathophysiologic pathways for the contribution of uniparental disomy to the development of myeloproliferation. Abbreviations: SG: susceptibility gene; MG: mutated gene; IG: imprinted gene; P: paternal; M: maternal.

TMD, AML and Neonatal Tumors

Miyauchi et al.[27] proposed that abnormal blasts seen in TMD may originate from the fetal liver due to the frequent finding of a higher percentage of blast cells in the peripheral blood compared to bone marrow. AML1 appears to be a crucial transcription factor in fetal liver hematopoiesis[38]. Carolyn et al.[40] hypothesized that patients with a certain subtype of familial platelet disorder may have a defective AML1 gene which results in qualitative and quantitative platelet abnormalities and may lead to the development of AML after additional genetic defects are acquired. This suggests that an increased gene dosage of genes such as AML1 plays an important role in the development of TMD. In contrast to FPD, increased expression of the AML1 gene in TMD may lead to overstimulated proliferation of critical progenitor cells in the presence of additional submicroscopic abnormalities involving genes regulating erythro-megakaryocytopoiesis, without affecting their capacity to differentiate into mature blood cells. The expression of these genes with hematopoiesis-controlling properties may be regulated in an age-dependent manner. This can explain the "transient" nature of TMD, the responsible gene(s) being expressed late in fetal life or through the early months of the postnatal period.

Spontaneous regression of neonatal tumors is not confined to trisomy 21-carrying neoplasias and has been reported in neonatal fibrosarcoma, congenital myofibromatosis, and stage IV-S neuroblastoma patients without constitutional genetic abnormalities[49-51]. A striking, common pattern has been observed in patients with certain types of neonatal tumors, particularly in patients with constitutional trisomies. As pointed out by Satge and Van den Berghe[34], spontaneous resolving tumors are characteristic of the perinatal period in which the tumors are either in situ or incompletely developed; spontaneous resolution of well-developed tumors does not occur in children and adults as seen in the case of TMD and AML-M7 in DS in which overproduction of immature fetal tissues is due to a dysregulation of normal embryological development. A similar scenario was proposed to explain intratubular germ cell neoplasia seen in trisomy 21 fetuses and the excess of testicular tumors seen in DS children or adults[52-53]. The influence of the intrauterine milieu and qualitative and quantitative changes of the developing immune system following birth must be taken into consideration in the development and the regression of these tumors. This theory may be very relevant in the case of DS. Down syndrome patients have immunological abnormalities and the interferon α, β, and γ receptor genes are located on chromosome 21. Several hypotheses have been proposed to illuminate the relation between immune dysfunction and trisomy 21[54-55]. The observation of lymphoid depletion in two fatal cases of DS with TMD and visceral fibrosis further supports the role of an antileukemic mechanism of the immune system in TMD[18].

In other words, in some DS patients with TMD, a severely disturbed immune system may not be able to control the proliferation of clonal cells causing fibrosis of visceral organs. A hypothetical schema of development of clonal proliferative disorders in DS is sumarized in Figure 3.

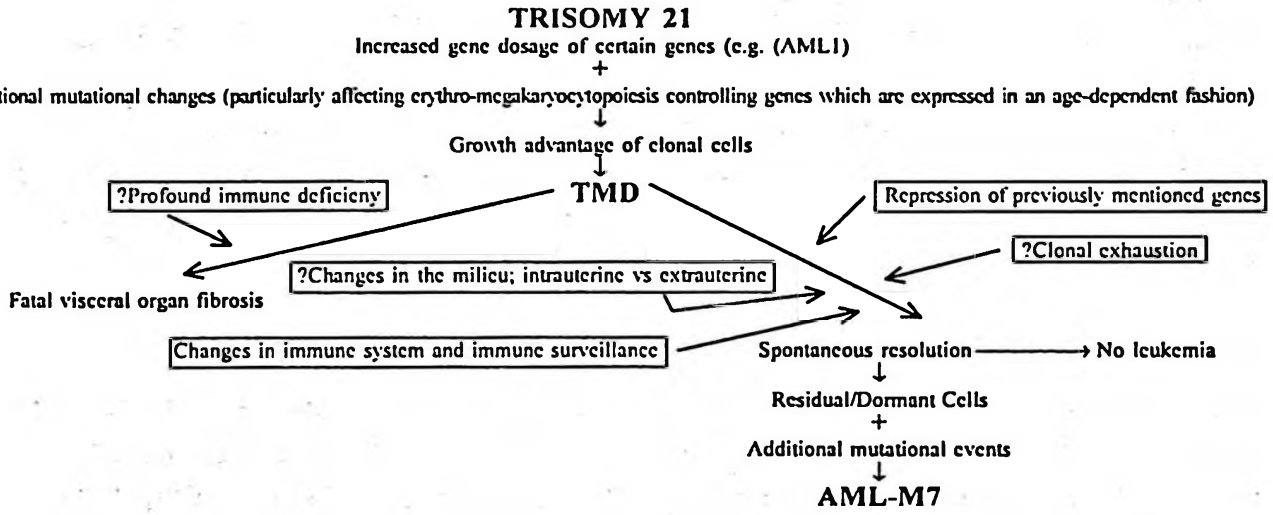

Fig. 3: Hypothetical steps in the development of TMD and AML-M7 in Down syndrome.

Recent Developments

The development of AML-M7 in 20 to 25 percent of TMD patients with DS may support the hypothesis that some residual cells may progress into a true malignant neoplasm by gaining new genetic abnormalities. Development of AML-M7 following TMD in a patient without Down syndrome can be explained by the same mechanism[56]. It is impossible to refute the central role played by different cytogenetic abnormalities in the development and progression of particular cancer, though the relative significance and contribution of each mutation can be quentioned, especially in the temporal relationship to the cancer. The persistence of multipotent progenitors expressing AML1/ETO transcripts in long-term remission patients with t(8;21) AML; spontaneous regression of congenital acute monoblastic leukemia with t(8;16) (p11;p13), which is associated with a poor prognosis; detection of t(14;18) (characteristic cytogenetic abnormality in follicular lymphomas); and expression of BCR-ABL (hallmark of chronic myeloid leukemia) in some healthy adults support the above concept[57-60].

Recent progress in the field of molecular genetics and molecular biology has prompted investigators to take advantage of these leukemogenic mutations as targets for therapy. All-trans retinoic acid has been used successfully for induction therapy of patients with acute promyelocytic leukemia characterized by the 15;17 translocation, which involves the retinoic acid receptor-α gene on chromosome 15[61]. Favorable results have been achieved with anti-sense oligonucleotide therapy directed to specific gene sequences involved in that particular cancer[62]. This

approach is based on the genetic abnormalities associated with the oncogenic mutation. It has been shown that patients with AML-M4 with inversion of chromosome 16 have a better prognosis if the multi drug resistance associated protein gene (MRP), localized to chromosome 16p13, was deleted during the process of chromosomal inversion[63]. The favorable response to intensive regimens and the moderate-to-severe methotrexate toxicity in ALL patients with DS have prompted investigators to study drug metabolism in AML patients with DS. Recently Taub et al.[64] showed that enhanced metabolism of AraC in DS cells is a contributing factor in the superior outcome of AML patients with DS. The gene dosage effect of genes localized to chromosome 21, such as cystathionine β-synthase, was hypothesized to be a mechanism which leads to altered drug metabolism.

Conclusion

The field of oncogenesis has expanded tremendously in the last decade, yet there are still several unanswered questions. Recent molecular oncologic inventions not only shed light on the pathogenesis of the neoplastic disorders but also contribute to the treatment of these disorders. The development of TMD and AML in patients with DS (characterized by trisomy 21) is an interesting model for studying the relationship between molecular changes and leukemogenesis. Despite the significant progress in the field of oncology, the treatment of non-DS AML patients remains a challenge[65]. Solutions may arise from observations which initially appear straightforward.

REFERENCES

1. Robinson L. Down syndrome and leukemia 1992; 6 (Suppl 1): 5-7.
2. Fong CT, Brodeur GM. Down's syndrome and leukemia: epidemiology, genetics, cytogenetics and mechanisms of leukemogenesis. Cancer Genet Cytogenet 1987; 28: 55-76.
3. Zipursky A, Poon A, Doyle J. Leukemia in Down syndrome: a review. Pediatr Hematol Oncol 1992; 9: 139-149.
4. Avet-Loiseau H, Mechinaud F, Harousseau JL. Clonal hematologic disorders in Down syndrome. J Pediatr Hematol Oncol 1995; 17: 19-24.
5. Kojima S,, Matsuyama T, Sato T, et al. Down's syndrome and acute leukemia in children: an analysis of phenotype by use of monoclonal antibodies and electron microscopic platelet peroxidase reaction. Blood 1990; 76: 2348-2353.
6. Zipursky A, Thorner P, DeHarven E, et al. Myelodysplasia and acute megakaryoblastic leukemia in Down's syndrome. Leuk Res 1994; 18: 163-171.
7. Slavc I, Urban C, Haas OA, et al. Acute megakaryocytic leukemia in children. Clinical immunologic, and cytogenetic findings in two patients. Cancer 1991; 68: 2266-2272.
8. Litz CE, Davies S, Brunning RD, et al. Acute leukemia and the transient myeloproliferative disorder associated with Down syndrome: morphologic, immunophenotypic and cytogenetic manifestations. Leukemia 1995; 9: 1432-1439.
9. Ravindranath Y, Abella E, Krischer JP, et al. Acute myeloid leukemia (AML) in Down's syndrome is highly responsive to chemotherapy: experience on Pediatric Oncology Group AML Study 8498. Blood 1992; 80: 2210-2214.

10. Lu G, Altman AJ, Benn PA. Review of cytogenetic changes in acute megakaryoblastic leukemia: one disease or several? Cancer Genet Cytogenet 1993; 67: 81-89.

11. Carroll A, Civinn C, Schneider N, et al. The t(1;22) (p13;q13) is nonrandom and restricted to infants with acute megakaryoblastic leukemia: a Pediatric Oncology Group Study. Blood 1991; 78: 748-752.

12. Lion T, Haas OA. Acute megakaryocytic leukemia with the t(1;22) (p13;q13). Leuk Lymphoma 1993; 11: 15-20.

13. Abdel-mageed AH, Ragab AJ, Shuster J, et al. Children with Down syndrome and acute lymphoblastic leukemia (DS-ALL). Presenting features and outcome of therapy: a Pediatric Oncology Group Study. Cancer 1991; 67: 1057-1063.

14. Tcherina G, Lejeune F, Boccara J-F, et al. Erythroblastic and/or megakaryoblastic leukemia in Down syndrome: treatment with low-dose arabinosyl cytosine. Pediatr Hematol Oncol 1996; 18: 59-62.

15. Zerres K, Schwanitz G, Niesen M, et al. Prenatal diagnosis of acute non-lymphoblastic leukaemia in Down syndrome. Lancet 1990; 335: 117.

16. Nakagawa T, Nishida H, Arai T, et al. Hyperviscosity syndrome with transient abnormal myelopoiesis in Down syndrome. J Pediatr 1988; 112: 58-61.

17. Jones GR, Weaver M, Laug WE. Transient blastemia in phenotypically normal newborns. Am J Pediatr Hematol Oncol 1987; 9: 153-157.

18. Becroft DM, Zwi LJ. Perinatal visceral fibrosis accompanying the megakaryoblastic leukemoid reaction of Down syndrome. Pediatr Pathol 1990; 10: 397-406.

19. Becroft DM. Fetal megakaryocytic dyshemopoiesis in Down syndrome: association with hepatic and pancreatic fibrosis. Pediatr Pathol 1993; 13: 811-820.

20. Hayashi Y, Eguchi M, Sugita K, et al. Cytogenetic findings and clinical features in acute leukemia and transient myeloproliferative disorder in Down's syndrome. Blood 1988; 72: 15-23.

21. Ridgway D, Benda GI, Magenis E, et al. Transient myeloproliferative disorder of the Down type in the normal newborn. Am J Dis Child 1990; 144: 1117-1119.

22. Hanna MD, Melvin SL, Dow LW, et al. Transient myeloproliferative syndrome in a phenotypically normal infant. Am J Pediatr Hematol Oncol 1985; 7: 79-81.

23. Faed MJ, Robertson J, Todd AS, et al. Trisomy 21 in transient myeloproliferative disorder. Cancer Genet Cytogenet 1990; 48: 259-264.

24. Ito E, Kasai M, Hayashi Y, et al. Expression of erythroid-specific genes in acute megakaryoblastic leukemia and transient myeloproliferative disorder in Down's syndrome. Br J Haematol 1995; 90: 607-614.

25. Zipursky A, Wang H, Brown EJ, Squire J. Interphase cytogenetic analysis of in vivo differentiation in the myelodysplasia of Down syndrome. Blood 1994; 84: 2278-2282.

26. Suda T, Suda J, Miura Y, et al. Clonal analysis of basophil differentiation in bone marrow cultures from a Down's syndrome patient with megakaryoblastic leukemia. Blood 1985; 66: 1278-1283.

27. Miyauchi J, Ito Y, Kawano T, et al. Unusual diffuse liver fibrosis accompanying transient myeloproliferative disorder in Down's syndrome: a report of four autopsy cases and proposal of a hypothesis. Blood 1992; 80: 1521-1527.

28. Yagihashi N, Watanabe K, Yagihashi S. Transient abnormal myelopoiesis accompanied by hepatic fibrosis in two infants with Down syndrome. J Clin Pathol 1995; 48: 973-975.

29. Kurahashi H, Hara J, Yumura-Yagi K, et al. Monoclonal nature of transient abnormal myelopoiesis in Down's syndrome. Blood 1991; 77: 1161-1163.

30. Alarcon PA, Patil S, Golberg J, et al. Infants with Down's syndrome. Use of cytogenetic studies and in vitro colony assay for granulocyte progenitor to distinguish acute nonlymphocytic leukemia from a transient myeloproliferative disorder. Cancer 1987; 60: 987-993.

31. Kivivuori SM, Rajantie J, Siimes MA. Peripheral blood cell counts in infants with Down's syndrome. Clin Genet 1996; 49: 15-19.

32. Roizen NJ, Amarose AP. Hematologic abnormalities in children with Down syndrome. Am J Med Genet 1993; 46: 510-512.

33. David O, Fiorucci GC, Tosi MT, et al. Hematologic studies in children with Down syndrome. Pediatr Hematol Oncol 1996; 13: 271-275.

34. Satge D, Van den Berghe H. Aspects of the neoplasms observed in patients with constitutional autosomal trisomy. Cancer Genet Cytogenet 1996; 87: 63-70.

35. Nucifora G, Rowley JD. AML1 and the 8;21 and 3;21 translocations in acute and chronic myeloid leukemia. Blood 1995; 86: 1-14.

36. Romana SP, Poirel H, Le Coniat M, et al. High frequency of t(12;21) in childhood B lineage acute lymphoblastic leukemia. Blood 1995; 86: 4263-4269.

37. Shurtleff SA, Buijs A, Behm FG, et al. TEL/AML1 fusion resulting from a cryptic t(12;21) is the most common genetic lesion in pediatric ALL and defines a subgroup of patients with an excellent prognosis. Leukemia 1995; 9: 1985-1989.

38. Okuda T, van Deursen J, Hiebert SW, et al. AML1, the target of multiple chromosomal translocations in human leukemia, is essential for normal fetal liver hematopoiesis. Cell 1996; 84: 321-330.

39. Ramesh AS, Orkin SH. The transcriptional control of hematopoiesis. Blood 1996; 87: 4025-4039.

40. Carolyn YH, Otterud B, Legare RD, et al. Linkage of a familial platelet disorder with a propensity to develop myeloid malignancies to human chromosome 21q22.1-22.2. Blood 1996; 87: 5218-5224.

41. Shen JJ, Williams BJ, Zipursky A, et al. Cytogenetic and molecular studies of Down syndrome individuals with leukemia. Am J Hum Genet 1995; 56: 915-925.

42. Feingold E, Lamb NE, Sherman SL. Methods for genetic linkage analysis using trisomies. Am J Hum Genet 1995; 56: 475-483.

43. Abe K, Kajii T, Niikawa N. Disomic homozygosity in 21-trisomic cells: a mechanism responsible for transient myeloproliferative syndrome. Hum Genet 1989; 82: 313-316.

44. Iselius L, Jacobs P, Morton N. Leukaemia and transient leukaemia in Down syndrome. Hum Genet 1990; 85: 477-485.

45. Lorber BJ, Freeman SB, Hassold T, et al. Characterization and molecular analysis of nondisjunction in 18 cases of trisomy 21 and leukemia. Genes Chromosom Cancer 1992; 4: 222-227.

46. Antonarakis SE. Parental origin of the extra chromosome in trisomy 21 as indicated by analysis of DNA polymorphisms. Down Syndrome Collaborative Group. N Engl J Med 1991; 324: 872-876.

47. Créau-Goldberg N, Gegonne A, Delabar J, et al. Maternal origin of a de novo balanced t(21q21q) identified by ets-2 polymorphism. Hum Genet 1987; 76: 396-398.

48. Blouin JL, Avramopoulos D, Pangalos C, Antonarahis SE. Normal phenotype with paternal uniparental isodisomy for chromosome 21. Am J Hum Genet 1993; 53: 1074-1078.

49. Madden NP, Spicer RD, Allibone EB, Lewis IJ, Spontaneous regression of neonatal fibrosarcoma. Br J Cancer 1992; 18 (Suppl): S72-75.

50. Jenkins EA, Cawley MI. Infantile myofibromatosis: a cause of severe bone pain in a neonate. Br J Rheumatol 1993; 32: 849-852.

51. Guglielmi M, De Bernardi B, Rizzo A, et al. Resection of primary tumor at diagnosis in stage IV-S neuroblastoma: does it affect the clinical course? J Clin Oncol 1996; 14: 1537-1544.

52. Jacobsen GK, Henriques UV. A fetal testis with intratubular germ cell neoplasia (ITGCN). Mod Pathol 1992; 5: 547-549.

53. Satge D, Jacobsen GK, Cessot F, et al. A fetus with Down syndrome and intratubular germ cell neoplasia. Pediatr Pathol 1996; 16: 107-112.

54. Cuadrado E, Barrena MJ. Immune dysfunction in Down's syndrome: primary immune deficiency or early senescence of the immune system? Clin Immunol Immunopathol 1996; 78: 209-214.

55. Zihni L. Down's syndrome, interferon sensitivity and the development of leukaemia. Leuk Res 1994; 18: 1-6.

56. Brissette MD, Duval-Arnould BJ, Gordon BG, Cotelingan JD. Acute megakaryoblastic leukemia following transient myeloproliferative disorder in a patient without Down syndrome. Am J Hematol 1994; 47: 316-319.

57. Miyamoto T, Nagafuji K, Akashi K, et al. Persistence of multipotent progenitors expressing AML1/ETO transcripts in long-term remission patients with t(8;21) acute myelogenous leukemia. Blood 1996; 87: 4789-4796.

58. Sainati L, Bolcato S, Cocito MG, et al. Transient acute monoblastic leukemia with reciprocal (8;16) (p11;p13) translocation. Pediatr Hematol Oncol 1996; 13: 151-157.

59. Liu Y, Hernandez AM, Shibata D, Cortopassi GA. BCL2 translocation frequency rises with age in humans. Proc Natl Acad Sci USA 1994; 91: 8910-8914.

60. Biernaux C, Sels A, Huez G, et al. Very low level of major BCR-ABL expression in blood of some healthy individuals. Bone Marrow Transplant 1996; 17 (Suppl 3): S45-47.

61. Warrell RPJr, de The H, Wang ZY, Degos L. Acute promyelocytic leukemia. N Engl J Med 1993; 329: 177-189.

62. Askari FK, McDonnel WM. Antisense-oligonucleotide therapy. N Engl J Med 1996; 334: 316-318.

63. Kuss BJ, Deeley RG, Cole SP, et al. Deletion of gene for multidrug resistance in acute myeloid leukaemia with inversion in chromosome 16: prognostic implications. Lancet 1994; 343: 1531-1534.

64. Taub JW, Matherly LH, Stout ML, et al. Enhanced metabolism of 1-β-D-Arabinofuranosylcytosine in Down syndrome cells: a contributing factor to the superior event free survival of Down syndrome children with acute myeloid leukemia. Blood 1996; 87: 3395-3403.

65. Ravindranath Y, Yeager AM, Chang MN, et al. Autologous bone marrow transplantation versus intensive consolidation chemotherapy for acute myeloid leukemia in childhood. New Engl J Med 1996; 334: 1428-1434.