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Molecular genetics of Down syndrome: transgenic models for Down syndrome related gene dosage effects

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Research Objectives

Down syndrome (DS), the phenotypic manifestation of trisomy 21, is one of the most common genetic abnormalities. DS patients suffer from various defects including high incidence of acute myeloid leukemia and early onset of Alzheimer disease. The syndrome results from a gene-dosage effect created by genes residing on the extra chromosome 21 and may thus represent a prototype for the study of human aneuploidy. The long-term objective of our research is to elucidate, at the molecular level, how an extra copy of otherwise normal gene produces the phenotypic features found in DS. For this purpose, we cloned, characterized and examined the expression of candidate genes residing at - 21q22, the chromosome region involved in DS. Transgenic models and transfected cells overexpressing a single candidate gene were developed. Three genes are currently investigated: the copper zinc superoxide dismutase (CuZnSOD), the amyloid-ß precursor protein (APP) and the acute myelogenous leukemia gene (AML1/RUNX1).The transgenic approach enabled us to correlate gene-dosage with specific phenotypic features found in the syndrome thus providing insights into the genesis of the abnormalities.

Our findings demonstrated, for the first time, a direct link between a clinical symptom of DS and overexpression of individual gene from the DS locus and led to a better understanding of the way imbalanced expression of these genes contributes to phenotypic features.

Results and Significance

A) Oxidative Stress, Neurodegeneration and early onset of Alzheimer Disease in Down syndrome (in collaboration with Menahem Segal). A growing body of evidence implicates oxygen radicals in a broad range of neuropathologies including Parkinson's disease Alzheimer's disease (AD) and amyotrophic lateral sclerosis (ALS). The genes for CuZnSOD, a key enzyme in the metabolism of oxygen free radicals, and for the amyloid-ß precursor protein (APP) reside on chromosome 21 and are overexpressed in DS patients who usually develop AD pathology early in life.
Transgenic CuZnSOD (Tg-SOD) mice with elevated activity of CuZnSOD and double transgenic (Tg-SOD/APP) mice with elevated SOD and APP are used to investigate the hypothesis that constitutive overexpression of CuZnSOD creates an indigenous oxidative stress that predisposes the Tg-SOD neurons to added insults such as increased level of amyloid ß-peptide (Aß). We postulate that the combined elevation of CuZnSOD and Aß predisposes DS patients to neurodegeneration and thereby causes the high incidence and early onset of AD in DS.

Our in vitro and in vivo data demonstrate a combined deleterious effect of Cu/ZnSOD and APP on neuronal survival (in hippocampal cultures) and plasticity (spatial learning/memory and tLTP deficits), a potential reflection of the in vivo situation observed in DS patients.

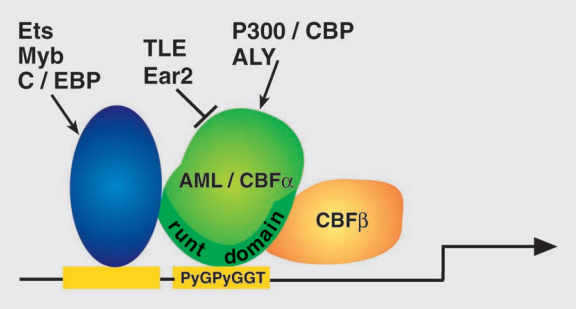

Fig.1. *Schematic drawing of AML/CBFß transcription complex*

B) The AML/RUNX gene family of transcription factors: Structure/function and role in Down Syndrome leukemia. The AML 1 & 2 genes (now termed RUNX1 and 3) are members of a gene family of heterodimeric transcription factors. AML1/RUNX1 plays a crucial role in hematopoiesis while the biological function of AML2/RUNX3 is not known yet. AML1 is one of the most frequently targeted genes in leukemia-associated translocations. Chromosomal translocations represent one way by which perturbation in AML1 function cause leukemia, but other alteration in its activity may prove to be leukemogenic as well. One such example is trisomy 21 (DS) where AML1 is overexpressed due to gene dosage. Moreover, AML1 and AML2 bind to the same consensus sequence, thus gene dosage of AML1 may

lead to alteration in expression of AML2. As part of our program to study DS related gene dosage effects, we set out to resolve the molecular structure and expression of the AML1 and its family member AML2 and to investigate their role in leukemia, in particular DS leukemia. Utilizing cell transfection experiments and creation of genetically modified (transgenic and knock-out) mice we aimed at gaining a better understanding of the molecular mechanisms underlying AML1 and AML2 mediated leukemogenesis. It is our hope that such knowledge will facilitate the development of better diagnostic and treatment procedures.

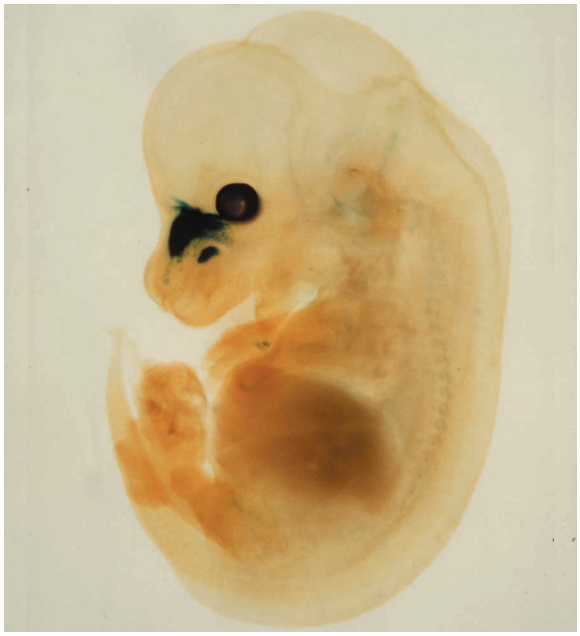

Fig. 2. *Tg-AML1-Lacz mouse embryo.*

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Acknowledgments

Yoram Groner holds the Dr. Barnet Berris Chair of Cancer Research