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Biological Functions of RUNX Transcription Factors in Neurogenesis and Hematopoiesis

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Opening Remark

We use cellular, biochemical and gene targeting approaches to investigate disease conditions in which genetic predisposition play role. The broad, long-term objective of our research is to elucidate, at the molecular level, how genetic mutations or gene dosage of otherwise normal genes produces pathophysiological conditions in humans particularly in patients with Down syndrome (DS).

For this purpose, we over the years cloned and characterized candidate genes residing at DS chromosomal regions and developed transgenic models for *in vivo* study of gene dosage effects. Gene-Knockouts of individual genes are used to investigate consequences of functional inactivation of candidate genes.

RUNX Biology

Currently we study the biology of two transcription factors Runx1 and Runx3 that belong to RUNX gene family (Fig 1). RUNX1 reside on chromosome 21 and could be involved in DS leukemia and RUNX3 reside on chromosome 1 at a region known to be involved in several human diseases.

The RUNX transcription factors are master regulator of linage specific gene expression in several developmental pathways. One of the exciting questions in molecular biology is how differential gene expression patterns are established and maintained during development. We address this question through investigating the biology of Runx1 and Runx3 at the molecular level and *in vivo* using genetically modified mouse models (Fig 2). Below is a spotlight account summarizing our past two years research on Runx3.

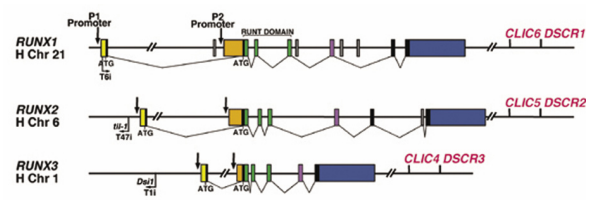

Fig. 1 *Genomic organization of the human RUNX genes (common exons are shown in similar color). Chromosomal localizations are depicted. The two promoters P1 and P2 and initiator ATGs are indicated. 5'UTRs, yellow and orange. 3'UTRs, sblue.*

We generated Runx3 knockout (KO) mice by inserting a LacZ-*neo* cassette so that the targeted allele also provided means for examining Runx3 expression by LacZ staining (Fig. 2). While heterozygous Runx3-mutant mice were phenotypically normal, the KO mice unexpectedly developed severe limb ataxia. What is the molecular basis of this limb defect? We found that in embryos Runx3 is highly expressed in a subset of sensory neurons, localized in dorsal root ganglia (DRG) (Fig.2). These neurons form connections with both muscle spindles and motor neurons to generate the monosynaptic stretch reflex circuit. When Runx3 is mutated these neurons do not gain their full identity and die by apoptosis due to frustration. These findings provide new genetic insights into neurogenesis of sensory DRG neurons and may help elucidate the molecular mechanisms underlying somatosensory-related ataxia in humans.

During thymopoiesis Runx3 regulates T-cell development. In KO mice T cells display abnormal expression of CD4 and impairment of CD8 T cells maturation. In a compound mutant mice, (Runx3-/-;Runx1+/-), null for Runx3 and heterozygous for Runx1 all peripheral single positive (SP) CD8 T cells also expressed CD4, resulting in a complete lack of SP CD8 T cells in the spleen. The results indicate that Runx3 acts as a negative regulator of CD4 expression during T cell lineage decision.

Runx3 is also highly expressed in dendritic cells

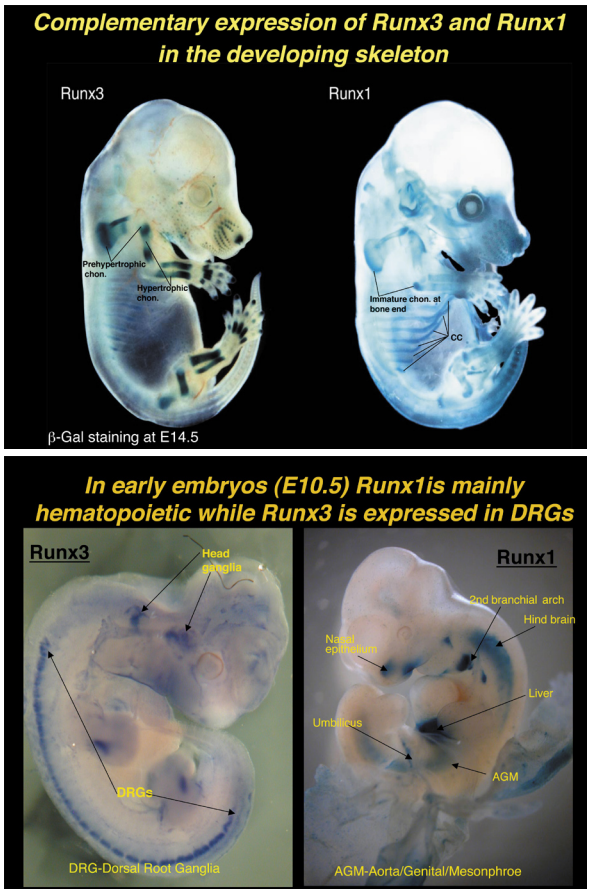

Fig. 2 *Expression patterns of Runx1 and Runx3 in developing mouse embryos. A-expression of β-galactosidase from Runx1 and Runx3 knock-in alleles in skeletal elements. B-expression of Runx1 in early hematopoiesis and Runx3 in the DRG.*

(DC), where it mediates their response to TGF-β. When Runx3 is mutated DC become insensitive to TGF-β and acquire highly active phenotype with increased potency to stimulate T cells. These hyperactive KO DC over-respond to otherwise innocuous airborne antigens and consequently induce an eosinophilic lung inflammation in the KO. Intriguingly, RUNX3 resides in 1p36.1 chromosome region that contains susceptibility genes for asthma and its deficiency may constitute an asthma risk factor in humans.

Selected Publications (2002-2004)

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Acknowledgements:

We thank the excellent assistance of Dorit Nathan, Tamara Berkuzki, Judith Chermesh, Rafi Saka, and Shoshana Grossfeled from the department of veterinarian recourses and Ayala Sharp and Eitan Ariel from the FACS unit. The work was done in collaboration with Steffen Jung and Ori Brenner. Supported by grants from the EU QLRT and QLK6-CT, the Israel Science Foundation and Minerva Foundation Germany.