http://www.ncbi.nlm.nih.gov/pubmed/12560239

Blood. 2003 Jun 1;101(11):4322-32. Epub 2003 Jan 30.

The in vivo profile of transcription factors during neutrophil differentiation in human bone marrow.

Bjerregaard MD, **Jurlander J**, **Klausen P**, **Borregaard N**, **Cowland JB**.

Department of Hematology, Rigshospitalet, University of Copenhagen, Denmark.
malene.bjerregaard@rh.dk

In vivo distribution of myeloid transcription factors during granulopoiesis was investigated by Northern and Western blotting in 3 neutrophil precursor populations from human bone marrow: immature (myeloblasts [MBs] and promyelocytes [PMs]); intermediate mature (myelocytes [MCs] and metamyelocytes [MMs]); and mature neutrophil cells (band cells [BCs] and segmented neutrophil cells [SCs]). Nonneutrophil cells were removed with magnetic-bead-coupled antibodies against CD2, CD3, CD14, CD19, CD56, CD61, glycophorin-A, and CD49d (BCs/SCs) before RNA and protein extraction. Polymorphonuclear neutrophils (PMNs) from peripheral blood depleted with anti-CD49d antibodies were also included. Expression of acute myeloid leukemia 1b (AML-1b), c-myb, GATA-1, and CCAAT/enhancer binding protein gamma (C/EBP-gamma) was seen primarily in MBs/PMs, and little expression was found in more mature cells. The level of C/EBP-alpha was constant in the bone marrow-derived cells and decreased in PMNs. C/EBP-epsilon was found primarily in MCs/MMs and was almost absent in more mature cells. Expression of C/EBP-beta, C/EBP-delta, and C/EBP-zeta was observed from the MC/MM stage onward, with peak levels in the most mature cells. The amount of PU.1 increased throughout maturation whereas the level of Elf-1 reached a nadir in MCs/MMs The PU.1 coactivator c-jun and c-jun's dimerization partner c-fos were both detectable in MCs/MMs and increased in amount with maturity. CCAAT displacement protein (CDP) was found at comparable levels at all stages of differentiation. This demonstrates a highly individualized expression of the transcription factors, which can form the basis for the heterogeneous expression of granule proteins during granulopoiesis and cell cycle arrest in metamyelocytes.

Full free study available

http://www.ncbi.nlm.nih.gov/pubmed/12620289

Leuk Res. 2003 May;27(5):387-92.

The level of MEF but not ELF-1 correlates with FAB subtype of acute myeloid leukemia and is low in good prognosis cases.

Fukushima T, **Miyazaki Y**, **Tsushima H**, **Tsutsumi C**, **Taguchi J**, **Yoshida S**, **Kuriyama K**, **Scadden D**, **Nimer S**, **Tomonaga M**.

Department of Hematology and Molecular Medicine Unit, Atomic Bomb Disease Institute, Nagasaki University School of Medicine, Nagasaki, Japan.

ETS proteins (such as PU.1, Fli-1 and ETS-1) have been shown to play important roles in normal and abnormal hematopoiesis. We examined the expression of the ELF subfamily of ETS genes (ELF-1, MEF and NERF) in acute myeloid leukemia (AML) cells using Northern blot analysis. ELF-1 and MEF were expressed in all samples, whereas NERF was not. The relative expression (RE) of MEF, but not ELF-1, was significantly lower ($P<0.0001$) in AML with t(8;21) and t(15;17) compared with AML with normal karyotype. The pattern of MEF expression was not uniform among cells with CD34(+)/CD33(+). It is suggested that the low RE of MEF might be part of a gene expression profile characterizing AML with a good prognosis.

http://www.ncbi.nlm.nih.gov/pubmed/8952536

Am J Pathol. 1996 Dec;149(6):2023-35.

Heterogeneous nuclear expression of the promyelocytic leukemia (PML) protein in normal and neoplastic human tissues.

Gambacorta M, **Flenghi L**, **Fagioli M**, **Pileri S**, **Leoncini L**, **Bigerna B**, **Pacini R**, **Tanci LN**, **Pasqualucci L**, **Ascani S**, **Mencarelli A**, **Liso A**, **Pelicci PG**, **Falini B**.

Institute of Pathology, Ospedale Niguarda, Milan, Italy.

The RING-finger promyelocytic leukemia (PML) protein is the product of the PML gene that fuses with the retinoic acid receptor-alpha gene in the t(15; 17) translocation of acute promyelocytic leukemia. Wild-type PML localizes in the nucleus with a typical speckled pattern that is a consequence of the concentration of the protein within discrete subnuclear domains known as nuclear bodies. Delocalization of PML from nuclear bodies has been documented in acute promyelocytic leukemia cells and suggested to contribute to leukemogenesis. In an attempt to get new insights into the function of the wild-type PML protein and to investigate whether it displays an altered expression pattern in neoplasms other than acute promyelocytic leukemia, we stained a large number of normal and neoplastic human tissues with a new murine monoclonal antibody (PG-M3) directed against the amino-terminal region of PML. As the PG-M3 epitope is partially resistant to fixatives, only cells that overexpress PML are detected by the antibody in microwave-heated paraffin sections. Among normal tissues, PML was characteristically up-regulated in activated epithelioid histiocytes and fibroblasts in a variety of pathological conditions, columnar epithelium in small active thyroid follicles, well differentiated foamy cells in the center of sebaceous glands, and hypersecretory endometria (Arias-Stella). Interferons, the PML of which is a primary target gene, and estrogens are likely to represent some of the cytokines and/or hormones that may be involved in the up-regulation of PML under these circumstances. In keeping with this concept, we found that PML is frequently overexpressed in Hodgkin and Reed-Sternberg cells of Hodgkin's disease, a tumor of cytokine-producing cells. Among solid tumors, overexpression of PML was frequently found in carcinomas of larynx and thyroid (papillary), epithelial thymomas, and Kaposi's sarcoma, whereas carcinomas of the lung, thyroid (follicular), breast, and colon were frequently negative or weakly PML+. We did not observe any changes in the levels of PML expression as the lesion progressed from benign dysplasia to carcinoma. Our immunohistological data are consistent with the hypothesized growth suppressor function of PML and strongly suggest that PML expression levels are likely to be modulated by a variety of stimuli, including cytokines and hormones.

Full free study available

http://www.ncbi.nlm.nih.gov/pubmed/16002702

J Immunol. 2005 Jul 15;175(2):1022-9.

Transcriptional regulation of CD1D1 by Ets family transcription factors.

Geng Y, **Laslo P**, **Barton K**, **Wang CR**.

Department of Pathology, and Howard Hughes Medical Institute, University of Chicago, 924 East 57th Street, Chicago, IL 60637, USA.

CD1 molecules are MHC class I-like glycoproteins specialized in presenting lipid/glycolipid Ags to T cells. The distinct cell-type specific expression of CD1D1 plays an important role in the development and function of NKT cells, a unique subset of immunoregulatory T cells. However, the mechanisms regulating CD1D1 expression are largely unknown. In this study, we have characterized the upstream region of the CD1D1 gene and identified a minimal promoter region within 200 bp from the translational start site of CD1D1 that exhibits cell-type specific promoter activity. Analysis of this region revealed an Ets binding site critical for CD1D1 promoter activity. Gel shift assays and chromatin immunoprecipitation experiments showed that Elf-1 and PU.1 bind to the CD1D1 promoter. Furthermore, we found that gene disruption of Elf-1 resulted in decreased CD1D1 expression on B cells but not other cell types, whereas conditional activation of PU.1 negatively regulated CD1D1 expression in PU.1-**deficient myeloid cells**. These findings are the first to demonstrate that Ets proteins are involved in the transcriptional regulation of CD1D1 and that they may function uniquely in different cell types.

Full free study available

http://www.ncbi.nlm.nih.gov/pubmed/10763819

Oncogene. 2000 Mar 23;19(13):1623-34.

The promyelocytic leukemia (PML) protein suppresses cyclin D1 protein production by altering the nuclear cytoplasmic distribution of cyclin D1 mRNA.

Lai HK, **Borden KL**.

Department of Physiology & Biophysics, Mount Sinai School of Medicine, New York, NY 10029, USA.

The majority of the promyelocytic leukemia (PML) protein is present in nuclear bodies which are altered in several pathogenic conditions including acute promyelocytic leukemia. PML nuclear bodies are found in nearly all cells yet their function remains unknown. Here, we demonstrate that PML and the eukaryotic initiation factor 4E (elF-4E) co-localize and co-immunopurify. eIF-4E is involved in nucleocytoplasmic transport of specific mRNAs including cyclin D1. eIF-4E overexpression leads to increased cyclin D1 protein levels; whereas, overexpression of PML leads to decreased cyclin D1 levels. Neither PML nor eIF-4E cause significant changes in cyclin D1 mRNA levels. The association with eIF-4E led us to investigate if PML could alter mRNA distribution as a possible post-transcriptional mechanism for suppressing cyclin D1 production. We show that overexpression of PML results in nuclear retention of cyclin D1 mRNA and that intact PML nuclear bodies are required. Addition of eIF-4E overcomes PML induced retention and alters the morphology of PML bodies suggesting a mechanism by which eIF-4E can modulate PML function. These results raise the possibility that PML nuclear bodies may participate in the regulation of nucleocytoplasmic transport of specific mRNAs.

Full free study available

http://www.ncbi.nlm.nih.gov/pubmed/19204326

Blood. 2009 Apr 9;113(15):3558-67. Epub 2009 Feb 9.

Structure of the AML1-ETO eTAFH domain-HEB peptide complex and its contribution to AML1-ETO activity.

Park S, **Chen W**, **Cierpicki T**, **Tonelli M**, **Cai X**, **Speck NA**, **Bushweller JH**.

Department of Molecular Physiology and Biological Physics, University of Virginia, Charlottesville, VA 22908, USA.

AML1-ETO is the chimeric protein product of the t(8;21) in acute myeloid leukemia. The ETO portion of the fusion protein includes the eTAFH domain, which is homologous to several TATA binding protein-associated factors (TAFs) and interacts with E proteins (E2A and HEB). It has been proposed that AML1-ETO-mediated silencing of E protein function might be important for t(8;21) leukemogenesis. Here, we determined the solution structure of a complex between the AML1-ETO eTAFH domain and an interacting peptide from HEB. On the basis of the structure, key residues in AML1-ETO for HEB association were mutated. These mutations do not impair the ability of AML1-ETO to enhance the clonogenic capacity of primary mouse bone marrow cells and do not eliminate its ability to repress proliferation or granulocyte differentiation. Therefore, the eTAFH-E protein interaction appears to contribute relatively little to the activity of AML1-ETO.

http://www.ncbi.nlm.nih.gov/pubmed/12819347

Proc Natl Acad Sci U S A. 2003 Jul 8;100(14):8448-53. Epub 2003 Jun 20.

Specific protein redirection as a transcriptional therapy approach for t(8;21) leukemia.

Steffen B, **Serve H**, **Berdel WE**, **Agrawal S**, **Linggi B**, **Büchner T**, **Hiebert SW**, **Müller-Tidow C**.

Department of Medicine, Hematology/Oncology, University of Münster, 48129 Münster, Germany.

Important progress has been achieved in the knowledge about the pathogenesis of cancer. However, despite these advances, the therapeutic strategies are still limited. Leukemias are often characterized by specific balanced translocations, with the t(8;21) balanced translocation being the most frequent chromosomal aberration in acute myeloid leukemia (AML). This translocation produces the AML1-ETO fusion protein, which binds to AML1 target promoter sequences. Transcriptional repression of AML1-dependent genes by AML1-ETO and associated corepressors represents the pathogenetic mechanisms of t(8;21). Here, we show that targeting of AML1-ETO to essential, MYB-dependent gene promoters induces t(8;21)-restricted cell death. We constructed a chimeric protein that contained the MYB DNA-binding domain and the AML1-binding domain of myeloid Elf-1-like factor (MEF). This protein associated with AML1-ETO and directed the complex to MYB-responsive promoters in vitro and in vivo. In the presence of AML1-ETO, the chimeric protein repressed the activity of MYB-responsive promoters, rapidly induced apoptosis, and specifically inhibited colony growth. All these effects occurred only in AML1-ETO-positive cells, whereas no adverse effects were observed in cells not expressing AML1-ETO. Taken together, this study demonstrates that redirection of oncogenic proteins can be used as a strategy to dramatically influence their cellular effects, with the ultimate goal to design highly specific therapies for cancer.

Full free study available