

Abstract:

Hematopoiesis is coordinated by a complex regulatory network of transcription factors among them PU.1 (Spi1, Sfp1) and GATA-1 represent key molecules. GATA-1 and PU.1 bind each other on DNA to block each others transcriptional programs to prevent development of undesired lineage during hematopoietic commitment. Murine erythroleukemia (MEL) cells, transformed erythroid precursors that are blocked from completing the late stages of erythroid differentiation, co-express GATA-1 and PU.1 and as my and others data document, are able to respond to molecular removal (down-regulation) of PU.1 or addition (up-regulation) of GATA-1 by inducing terminal erythroid differentiation. We provide novel evidence that downregulation of GATA-1 or upregulation of PU.1 induces incompletely differentiation into cell cycle arrested monocytic-like cells. Furthermore, PU.1- dependent transcriptome is negatively regulated by GATA-1 in MEL cells, including CCAAT/enhancer binding protein alpha (Cebpa) and Core-binding factor, beta subunit (Cbfb) that encode additional key hematopoietic transcription factors. Chromatin immunoprecipitation and reporter assays identified PU.1 motif sequences near Cebpa and Cbfb that are co-occupied by PU.1 and GATA-1 in the leukemic blasts. Furthermore, transcriptional regulation of these loci by manipulating the levels of PU.1 and GATA-1 involves quantitative increases in a transcriptionally active chromatin mark: acetylation of histone H3K9. Our data are further supported by experiments documenting that significant derepression of Cebpa and Cbfb is achieved in MEL cells by either activation of PU.1 encoded transgene or knockdown of GATA-1 expression.