Abstract

T cell development in the thymus occurs in a series of tightly regulated stages and is controlled by a complex network of transcription factors. E proteins, part of the class I basic-helix-loop-helix (bHLH) transcription factors, have been shown to be critical for T cell specification and commitment, as E protein deficiencies lead to major breaches in the T cell developmental program. There are three members of the E protein family: HEB, E2A and E2-2. HEBAlt, the shorter alternative isoform of HEB, lacks the first activation domain and instead has a unique and highly conserved alternative domain, the function of which still remains elusive. HEBCan, the longer canonical form, is expressed throughout T cell development whereas the expression of HEBAlt is limited to the DN2 and DN3 stages. This suggests that HEBAlt might have a specific role in programming early thymocytes. Not much is known about the target genes of HEB factors, but one group has shown that HEBCan and E2A are both able to upregulate the transcription of RORγt, an orphan nuclear receptor, in the context of EL4 cells. I was able to show that HEBAlt is also able to positively regulate the RORγt promoter in the context of 293T cells, and that the alternative domain is only partially responsible for this function. Also, there does not seem to be any synergistic or combinatorial effect with other E proteins for the regulation of RORγt. In addition, HEBAlt is not able to activate the putative Sox13 promoter in 293T cells. Furthermore, E proteins function as obligate dimers and it has been previously shown through co-immunoprecipitation experiments with tagged HEB vectors that HEBAlt is able to form homodimers with itself. I showed that HEBCan is able to form homodimers with itself and heterodimers with HEBAlt, E2A and Id2. HEBAlt is also able to form heterodimers with HEBCan, E2A and Id2. Further research is needed to elucidate the function of the alternative domain, the role of HEBAlt in T lymphopoiesis and its interactions with other co-activators and co-repressors.