Work Package 2: Genetic and epigenetic control of transcription
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Malignant hematopoiesis arises after the expansion of primitive cells that undergo defined genetic alterations, the probability of which is associated to the uninterrupted production of blood cells. These mutations involve a large variety of genes encoding from signalling molecules to transcription factors and epigenetic regulators. In general, the loss of function or the aberrant new functions associated to the products of these genetic alterations, interfere with the physiological transition between cells stages towards the production of fully differentiated blood cell types.

Evidence has accumulated about the involvement of chromatin regulators (often known under the term epigenetic) on normal hematopoiesis but also on its maligned variations. Among these, the Polycomb system, a heterogenous set of protein complexes organised around two catalytic modules that can modify nucleosomal histones, is known to play important functions.

We are interested in the subset of Polycomb complexes containing E3 ligases that monoubiquitylate histone H2A, also known as PRC1 complexes. Genetic manipulation of the subunits that make these ligases, as well as that of other Polycomb subunits underlines decisive contributions to the establishment and maintenance of transformed hematopoietic cells. Classically associated with cell memory functions keeping repressed genes with a relevance in developmental processes, recent work identifies among their targets a larger number of transcriptionally active genes. Overall, current understanding of hematopoietic Polycomb functions is still very limited.

Interested in PRC1 functions, we intend to investigate malignant and non-malignant immortalisation within myeloid lineages, which can be routinely modelled in vitro and in vivo through the expression of known fusion proteins identified in patients or homeotic products. Specifically we will use our mouse models of (inducible) loss of function of Ring1B - a key component of the Polycomb E3 ligase common to all PRC1 assemblies - and of RYBP, a subunit present in the functionally distinctive subset of non-canonical PRC1 complexes. Chromatin landscapes of immortal cells in the presence/absence of these will be analysed, by using histone marks and accessibility approaches. Preliminary observations suggest that appropriate genetic background may have to be devised that allow for viability of mutant cells. Also, complementation assays would be set up to assist not only in delineating structure-function relationships but also in discerning phenotypes obscured by uncontrollable effects secondary to simple gene inactivation. It is anticipated that most work will be confined to ex-vivo expanded cells leaving in vivo assays essentially as a validation tool.