

P015 Modelling early haemopoietic development using murine embryonic stem (ES) cells: the involvement of PI3Ks
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Regulation of early embryonic development and specifically development of the lympho-haemopoietic system has been shown to require PI3K-dependent signals. However, the inaccessibility of early embryonic material and the embryonic lethality of several PI3K gene knock-outs have made it difficult to further characterise the requirement for PI3K-mediated signals during early developmental processes. Here we have used an *in vitro* ES cell differentiation system, based on embryoid body (EB) formation, as a model to study early differentiation events and progression towards the haemopoietic lineage. We have used the pharmacological inhibitor of PI3Ks, LY294002, together with PDK1^{-/-} ES cells to demonstrate that PI3K-mediated signals, via PDK1, are required for proliferation of cells within developing EBs. Interestingly, the haemopoietic potential of EB-derived cells was not blocked upon PI3K inhibition, but rather enhanced, correlating with modest increases in expression of haemopoietic marker genes. In contrast, PDK1-deficient EB-derived progeny failed to generate terminally differentiated haemopoietic lineages, although small erythroid-like colonies did form. PI3K-dependent signalling was found to be critical for proliferation of the earliest haemopoietic progenitor, the blast-colony forming cell (BL-CFC). In addition, PI3K-dependent signalling was also required for the generation of terminally differentiated haemopoietic cells from haemopoietic progenitors present within EBs and mouse bone marrow.