Abstract

Blood is a necessary aspect of the human body, responsible for oxygen transport, immunity, and wound healing. Understanding the molecular mechanism of its regulation allows a better understanding of the development of blood disorders and malignancies. The process of regenerating new blood cells derived from Hematopoietic Stem Cells (HSCs) is known as hematopoiesis. This process involves multiple key signaling molecules and pathways, allowing the HSCs to differentiate into specialized effector cells. One of these signaling pathways includes the Akt pathway, associated with the regulation of the quiescence and activation states of HSCs. Although the Akt pathway is well studied, the role of its downstream effector Akt-Associated Protein (AAP), a negative regulator of mTORC1, in vertebrate hematopoiesis remains poorly understood. This study aims to characterize the function of AAP in zebrafish hematopoiesis by generating AAP knockout mutants using CRISPR/Cas9 and observing the changes in hematopoietic development by performing *in situ* hybridization of hematopoietic-related genes, followed by gene expression analysis using qPCR. Zebrafish was used due to its genetic tractability, conserved hematopoietic program, and transparent embryos, which will facilitate optimal *in vivo* visualizations. Results suggested impairments of the AAP gene resulted in disruption in early hematopoietic specification of Scl and Gata2 genes, which may result in alterations in lineage specification, as shown by increased Mpo myeloid marker and reduced Rag1 lymphoid marker. However, due to the lack of biological replicates, these results can only be interpreted as preliminary descriptive data. Hence, it is important to increase the sample size and biological replicates for more accurate analysis.

Keywords: Akt-Associated Protein (AAP), Hematopoiesis, Hematopoietic Stem Cells (HSCs), Zebrafish