Regulation of erythropoiesis in zebrafish model by the kinase of ribosomal protein S19

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In Diamond-Blackfan anemia (DBA), about half of the patients have mutations in one of several ribosomal protein (RP) genes. The most frequently mutated gene (～25%) is the ribosomal protein S19 (*RPS19*), in which a hot spot for mutations between residues 52 and 62 has been reported. However, it is not clear why mutations in the ubiquitously expressed *RPS19* gene specifically affect erythropoiesis. We previously showed in vitro that the 59th serine residue (Ser59) of RPS19 is phosphorylated by PIM1 kinase. Here we study the involvement of RPS19 and PIM1 in erythropoiesis using zebrafish to determine whether phosphorylation could affect red blood cells production. We generated the *rps19* knockdown zebrafish by injection of morpholino antisense-oligo (MO) at the one-cell stage. The rps19-deficient embryos (morphants) showed abnormal morphologies and a decreased number of red blood cells. Although *in vitro* synthesized *rps19* mRNA rescued the aberrant phenotypes in morphants, the recuperation was not shown by substitution of Ser59 residue with alanine or aspartic acid. These observations suggest that reversible phosphorylation of Ser59 is important for the function of rps19. Therefore, we injected the MO against *pim1*, which phosphorylates Ser59 of rps19. The pim1 morphants showed abnormal head and tail, and a decrease in the number of red blood cells. Co-injection with synthetic *pim1* mRNA restored morphology and red blood cell count. These findings suggested that pim1 was related to erythropoiesis. Further consideration will be needed to yield any findings about the relationship between phosphorylation and erythropoiesis by using pim1 deficient fish.