We aim at investigating vertebrate neural progenitor cells behavior in vivo by focusing on the peripheral midbrain layer (PML) which is a midbrain sub-territory wrapping the optic tectum (OT) posteriorly, laterally and medially. We performed a multilevel morphogenetic description of PML and OT progenitor cells between 24 and 48 hpf, e.g. 1/ lineage and fate, 2/ proliferation modes, and 3/ tissue morphogenesis. This was achieved by long term two-photon in vivo imaging of fluorescent proteins in transgenic fish lines and subsequent image analysis with homemade software.

We showed that PML cells are neuroepithelial and have big and elongated nuclei suggesting relatively decondensed chromatin, in contrast with OT cells that have a small rounded shape. In addition, 90% of the analyzed PML proliferate by means of symmetric divisions. The PML cells proliferate three times slower (1 mitosis every 2h) than the OT (1 mitosis every 6h40). Furthermore, individual PML cells tracking and global displacement field indicated their contribution to the OT. This suggests that during the cell population displacement of PML cells toward the OT, cells are undergoing a major change in their gene expression profile. Indeed, the analysis of gene expression patterns showed that PML cells express a specific subset of genes. The latter includes genes involved in nucleotides synthesis and genes encoding nucleolar or ribosomal proteins as reported for Drosophila neuroblasts. This idea is also supported by the analysis of a mutant line where PML and OT organization is disrupted and which exhibits extensive cell death.

This characterization of PML cells as slow cycling neural progenitors displaced toward their post mitotic location through global cell population movements by 2 dpf serves as a reference for further investigation of PML contribution to regeneration of OT after photoablation.