# Role of the scaffold protein Tks4 in cancer cell movement

Using the CRISPR/CAS9 technology, Tks4 protein was knockdown in HCT116 colon carcinoma cells. We found that the morphology and gene expression levels of the knockout cells were changed, likely due to the epithelioid-mesenchymal transition (EMT). The Reduced cell attachment of cell lacking Tks4 was also noticed. We explored the 2D motility of tks4 mutant cells in several parallel experiments. By image analysis techniques we demonstrated that the tks4 mutant cells move faster and perform a more persistent random walk. Cluster-size analysis of the segmented images reveals that Tks4 mutant cells move independently in small clusters or as individual cells. In contrast, wild type HCT116 cells form large, multicellular epithelioid patches. We also explored the 3D motility of tks4 mutant cells in collagen gels. HCT116 cells could invade the collagen by forming multicellular sprouts. We demonstrated that the multicellular sprouts formed by tks4 mutants are less stable with individual cells often leaving the sprout. The motility of tks4 mutants, however, is reduced in collagen gels, likely due to a defect in podosome formation.