A major role of active Kras in gastric chief cells during gastric carcinogenesis

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Gastric cancer, the second highest cause of cancer-related death in the world, develops in the setting of predisposing mucosal changes including parietal cell atrophy and two metaplastic processes: Spasmolytic polypeptide-expressing metaplasia (SPEM) and intestinal metaplasia (IM). Recent investigations indicate that SPEM arises from transdifferentiation of chief cells and that SPEM can then give rise to IM. Previous studies have noted activating mutations in Ras proteins in up to 15% of gastric cancers, but activation of Ras activity is observed in up to 40% of gastric cancers. While several studies in humans have indicated that SPEM progresses into IM and gastric cancer, the signaling pathways involved in metaplastic transitions remain obscure. We hypothesized that Ras activation in gastric chief cells influences the evolution and progression of metaplasia and dysplasia. We have utilized a mouse model, which develops metaplasia and dysplasia following induction of active Kras(G12D) specifically in gastric chief cells driven by the tamoxifen-inducible Mist1-CerERT2. The active Kras-induced chief cells transdifferentiated into SPEM within a month after activation and progressed to IM and invasive metaplasia within 3 to 4 months. Lineage tracing of active Kras-induced chief cells confirmed that the metaplasia and dysplasia developed directly from Kras-induced mature chief cells. We also treated this mouse model with selumetinib, a MEK inhibitor, to investigate whether the progression of IM to dysplasia can be controlled by inhibition of the Kras signaling pathway. The treatment with a MEK inhibitor for 2 weeks led to the regression of IM and reestablishment of normal mucosal lineage cells. Additionally, we have isolated and cultured metaplastic organoids from gastric mucosa in Mist1-Kras mice at 3 and 4 months after tamoxifen induction. The cultured metaplastic organoids have undergone continuous passaging for months and formed distinguishable glandular structures in 3-dimensional cultures and maintained a stable phenotype observed in metaplastic glands in the Mist1-Kras mouse stomachs. Therefore, our results indicate that Ras activation in gastric chief cells plays a major role in driving the evolution of gastric carcinogenesis. Also, our study suggests that MEK inhibitors may provide a novel treatment modality for patients with metaplasia.

Regulation and Function of Exosomes to Directional Cancer Cell Motility

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Directional cell movement through tissues is critical for multiple biological processes and requires maintenance of polarity in the face of complex environmental cues. Migration of cells toward chemical cues, or chemotaxis, is important for many biologic processes such as immune defense, wound healing and cancer metastasis. Although chemotaxis is thought to occur in cancer cells, it is less well characterized than chemotaxis of professional immune cells such as neutrophils. Here, we use intravital imaging to demonstrate that secretion of exosomes from late endosomes is required for directionally persistent and efficient *in vivo* movement of cancer cells. Inhibiting exosome secretion or biogenesis leads to defective tumour cell migration associated with increased formation of unstable protrusions and excessive directional switching. *In vitro* rescue experiments with purified exosomes and matrix coating identify adhesion assembly as a critical exosome function that promotes efficient cell motility. Live-cell imaging reveals that exosome secretion directly precedes and promotes adhesion assembly. Fibronectin is found to be a critical motility-promoting cargo whose sorting into exosomes depends on binding to integrins. *In vitro* chemotaxis experiments show that cancer cell chemotaxis relies on secretion of exosomes. We propose that autocrine secretion of exosomes powerfully promotes directionally persistent and effective cell motility by delivering multiple motility-promoting cargoes that contribute to different aspects of cancer cell motility.