**Supplementary Fig. S1.** (A): Immunohistochemical analysis of other human normal colon and colon cancer specimens. DCLK1-positive epithelial cells were not found at the crypt bottoms. Normal tissues are partially positive but cancer tissues were widely positive for DCLK1. Scale bar: 100 μm. (B): Representative metastatic lymph node. DCLK1-positive cells were dominant in the metastatic lymph node. Scale bar: 100 μm.

**Supplementary Fig. S2.** (A): SW480 cells are EpCAM-positive and low CD66a expression. (B): Representative flow cytometry plot. GFP+/mCherry+/CD44+ SW480 cells were collected by flow cytometry. (C): DCLK1 expression was suppressed in the SW480 cells transduced with the pGIPZ lentiviral shRNA against *DCLK1*. (D): (Top) Representative images of the organoids. The left and middle panels are the fluorescent microscopic images for the detection of GFP and mCherry, respectively. The right panels are the phase-contrast images of the organoids merged with the images of the green fluorescent images (left) and the red fluorescent images (middle). Scale bar: 100 μm. (Bottom) Co-expression of the exogenous DCLK1 substantially rescued the defect in organoid formation of shDCLK1-SW480 cells (\*p<0.05).

**Supplementary Fig. S3.** (A): Almost all hPCCs express EpCAM. (B): MiR-137 knockdown enhanced expression of DCLK1 of GFP+/EpCAM+/CD44+/CD66a- hPCCs. (C): Aline the two sequences between human *DCLK1* and murine *Dclk1*. Target 1 of human *DCLK1* is preserved.

**Supplementary Fig. S4.** (A): GFP+/CD44+ SW480 cells were sorted by flow cytometry and 2.0 × 106 cells were injected subcutaneously into immunodeficient mice. The tumor volume and incidence at eight weeks after injection into nude mice (top) or NOD/SCID mice (bottom) (\*p<0.05). (B): MiR-137 does not affect the proliferation of SW480 cells.