**Figure W1.** MicroRNA-200 expression is reduced at the invasive front of colorectal cancers with degraded basement membranes. ISH of miR-200b and miR-200c was performed on FFPE human colorectal adenocarcinomas. Adjacent sections were immunohistochemically stained for laminin and ZEB1. (A) Consecutive serial sections of an adenocarcinoma with a degraded basement membrane stained with H&E (top), miR-200c (middle), and laminin counterstained with methyl green (bottom). (B) H&E, miR-200b, ZEB1, and laminin at the invasive front of an adenocarcinoma with a degraded basement membrane. Scale bars represent 100 $\mu$m.
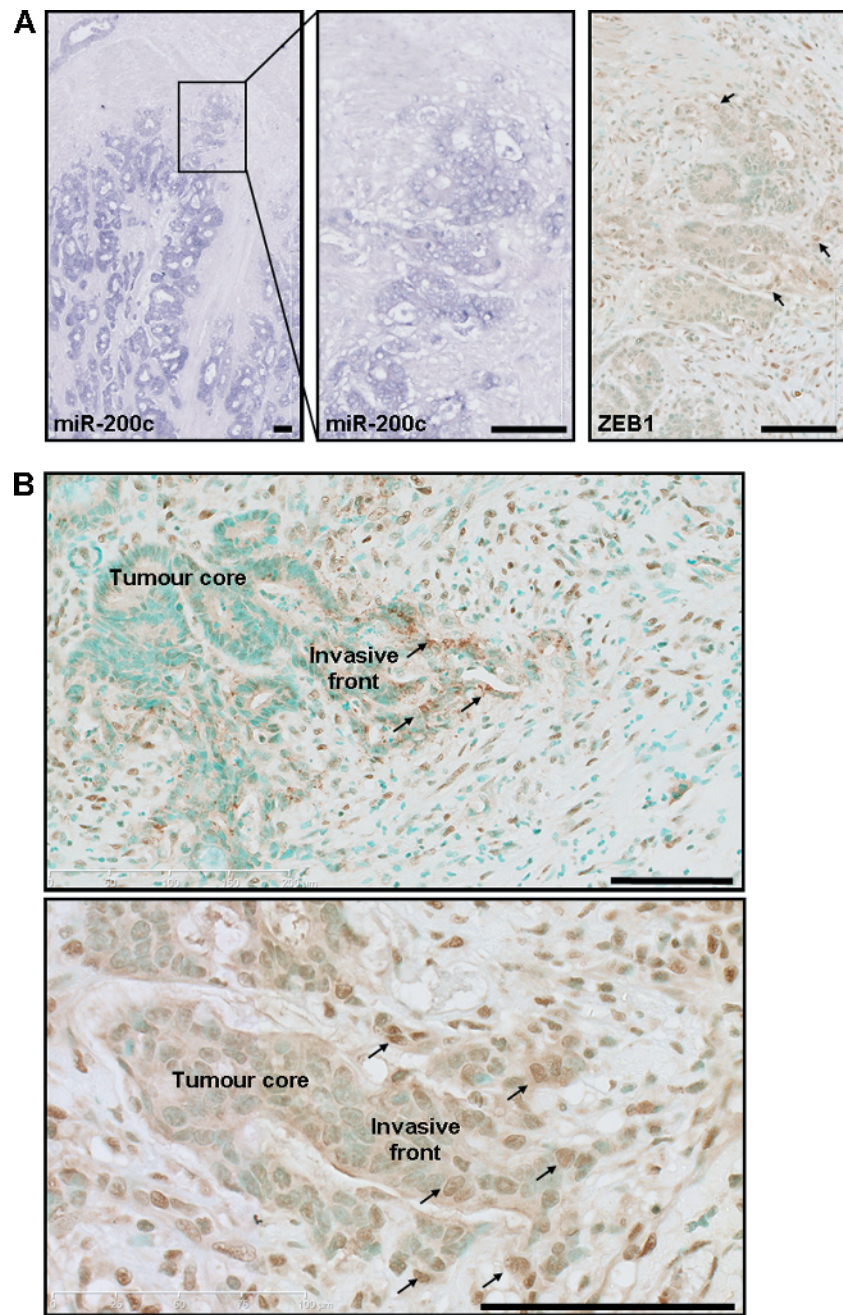
**Figure W2.** MicroRNA-200 expression is reduced at the invasive front of colorectal cancer where ZEB1 is upregulated. (A) Adjacent sections of an adenocarcinoma with a degraded basement membrane stained for miR-200c and ZEB1 (counterstained with methyl green). (B) Examples of ZEB1 expression in adenocarcinomas with degraded basement membranes (counterstained with methyl green). Arrows indicate ZEB1 expression. Scale bars represent 100 $\mu$m.
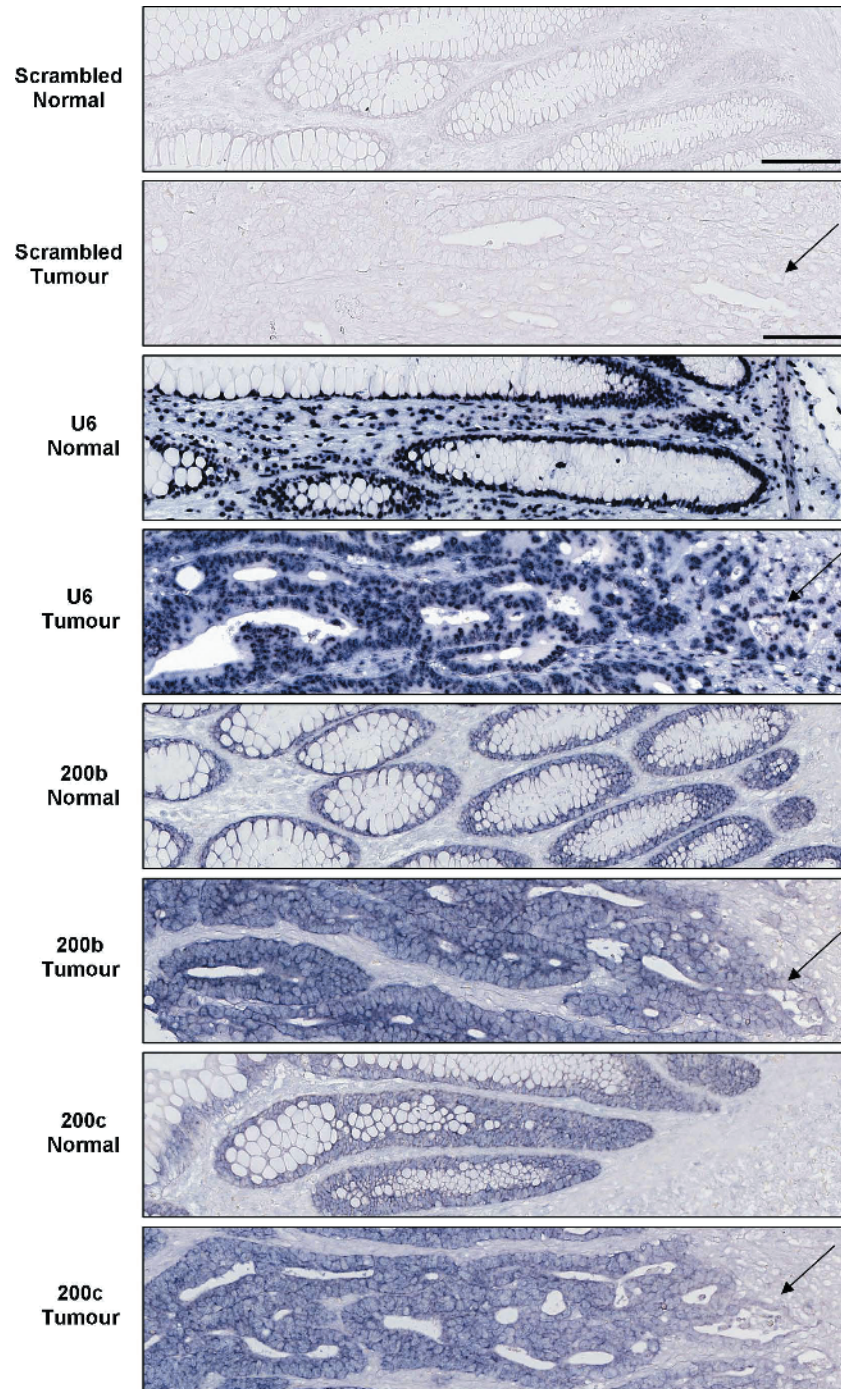
**Figure W3.** Positive and negative controls for ISH. Normal and tumor colon tissues stained through ISH with 3′DIG-labeled LNA probes for a negative scrambled control, the ubiquitous small nuclear RNA U6 (positive control), miR-200b, and miR-200c. Arrows point to the invasive front. Scale bars represent 100 $\mu$m.
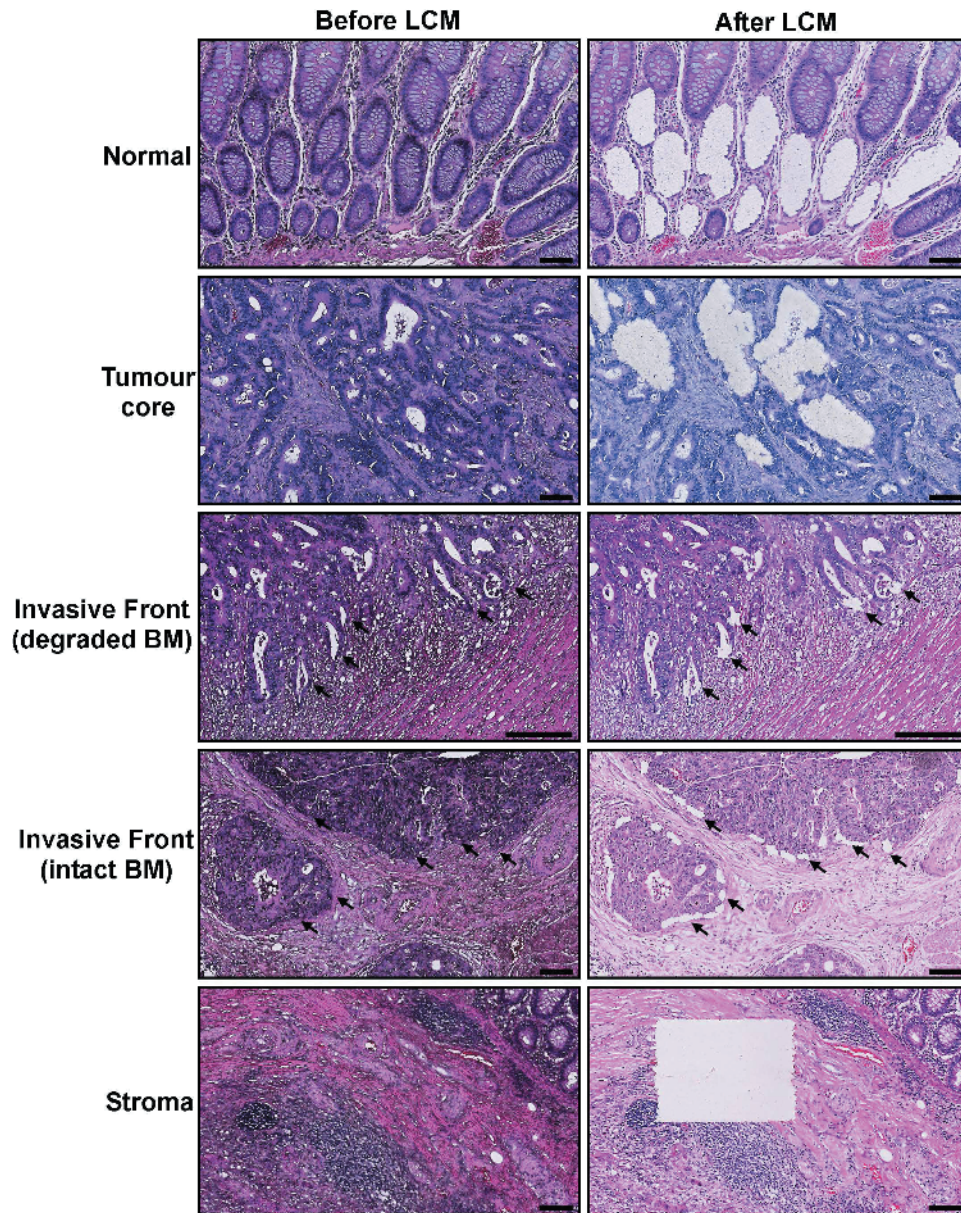
**Figure W4.** Examples of laser microdissected regions. Images of normal, tumor, invasive front, and stromal regions taken before LCM (left column) and after LCM (right column). Two 0.1-mm$^2$ samples were captured from each region and separately column-purified for miRNA. Arrows point to microdissected leading edge regions in tumors with intact and degraded basement membranes. The stroma image demonstrates the amount of tissue required per region. Scale bars represent 100 $\mu$m.

**Table W1.** Summary of miR-200 ISH and qPCR Data from Colorectal Cancers.

| TNM Status | Defining Morphologic Features | ISH of miR-200 at Invasive Front | Vascular Invasion | qPCR of miR-200a/b/c at the Invasive Front Compared to the Tumor Core | | | qPCR of miR-200a/b/c in the Tumor Compared to Normal Crypts | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | a | b | c | a | b | c |
| pT3N0M0 | Budding cells, loss of basement membrane | Loss | Absent | ↓ | ↓ | ↓ | – | – | – |
| pT3N0M0 | at leading edge, tubular invasion poles | | Present | – | – | ↓ | ↓ | ↓ | – |
| pT4aN2M0 | | | Present | – | ↓ | ↓ | ↓ | – | – |
| pT4aN0M0 | | | Present | ↓ | ↓ | ↓ | – | – | – |
| pT4aN0M0 | | | Present | – | ↓ | ↓ | – | – | – |
| pT4bN1M1 | | | Present | – | ↓ | ↓ | ↓ | – | – |
| pT4aN2M0 | | | Present | | | | | | |
| pT3N2aM1 | | | Present | | | | | | |
| pT1N0MX | | | Not identified | | | | | | |
| pT3N0MX | | | Present | | | | | | |
| pT3N2aM0 | | | Present | | | | | | |
| pT4aN1aM0 | | | Present | | | | | | |
| pT3N0M0 | | | Present | | | | | | |
| pT4aN2bM0 | | | Present | | | | | | |
| pT3N0M0 | | | Not identified | | | | | | |
| pT3N0M0 | | | Not identified | | | | | | |
| pT4aN2MX | | | Not identified | | | | | | |
| pT4aN1bM0 | | | Not identified | | | | | | |
| pT1N1M0 | No budding cells, intact basement membrane, | No loss | Present | – | – | – | – | – | – |
| pT3N0M0 | smooth tumor-host interface | | Present | – | – | – | – | – | – |
| pT2N1bM0 | | | Absent | | | | | | |
| pT0N0M0 | | | Not identified | | | | | | |
| pT3N0M0 | | | Suspicious | | | | | | |
| pT0N0M0 | | | Present | | | | | | |
| pT4bN1M0 | | Too weak to detect | Present | – | – | ↓ | – | – | – |
| pT4bNXMX | | | Present | – | – | – | – | ↓ | ↓ |
| pT3N2bMX | | | Present | | | | | | |
| pT1N1bM0 | | | Present | | | | | | |

Comparison of morphologic features common to adenocarcinomas with either loss or maintenance of miR-200 at the tumor-host interface. Pathologic assessment of Tumor-Node-Metastasis (TNM) status shown in column 3. Column 4 summarizes the ISH results. Quantitative real-time RT-PCR comparison between the tumor and invasive front and between tumor and normal epithelia is shown in columns 5 and 6, respectively (arrow denotes loss of miR-200 expression, and dash denotes no loss).