

Supplementary Methods:

Primary antibodies used for immunostaining were: E-cadherin (BD Transduction Laboratories, 610182), Lysozyme (used for staining, Biogenex, AR024-5R), LysozymeC (used for Western blot, Santa Cruz, sc27958), pHH3 (Millipore, 06-570), β -catenin (Cell Signaling, 9587), Laminin (Abcam 11575), Ki67 (Vector Lab, VP-K451), Asef1 (Aviva Systems Biology Corp, ARP59254_P050), PAK1 (Cell Signaling, 2602), GFP (Abcam, 6673), Gpr177 (Fu et al., 2011; Fu et al., 2009), c-Myc (Santa Cruz, sc-764), and BrdU (Accurate Chemical OBT0030G). For double staining of BrdU and Gpr177, rat anti-BrdU (1:1000) and rabbit anti-Gpr177 (1:1500) were applied to tissue sections, followed by incubation with secondary antibodies provided in VECTASTAIN ABC-AP (Vector Laboratory, SK-5000) and VECTASTAIN Elite ABC (Vector Laboratory, PK-6100) kits. Colors were sequentially developed for Gpr177 using Vector Red Alkaline Phosphatase Substrate Kit I (Vector Laboratory, SK-5100), followed by BrdU using DAB Peroxidase Substrate Kit (SK-4100). Western blot antibodies include Cdc42 (Cell Signaling, 2462), β -catenin (Cell Signaling, 9587; BD Transduction Lab, 610153), phosphor- β -catenin (Cell Signaling, 9561), Tcf4 (Cell Signaling, 2765), Tcf1 (Cell Signaling, 2203), CD44 (V6) (Bender Medsystems), Sox9 (Santa Cruz, sc-17341), β -actin (Santa Cruz, sc-47778), Cleaved Caspase-3 (Asp 175) (Cell Signaling, 9664).

RT-PCR primers:

Lgr5, forward-TAAAGACGACGGCAACAGTG; reverse- GATTCGGATCAGCCAGCTAC
CD44, forward- TGCATCGCGGTCAATAGTAG; reverse- TGATGGTCCCTGTTCACCA
Sox9, forward-AGTCGGTGAAAGAACGGACAA; reverse-CCCTCTCGCTTCAGATCAAC

Supplementary References:

- Fu, J., Ivy Yu, H.M., Maruyama, T., Mirando, A.J., and Hsu, W. (2011). Gpr177/mouse Wntless is essential for Wnt-mediated craniofacial and brain development. *Developmental dynamics : an official publication of the American Association of Anatomists* *240*, 365-371.
- Fu, J., Jiang, M., Mirando, A.J., Yu, H.M., and Hsu, W. (2009). Reciprocal regulation of Wnt and Gpr177/mouse Wntless is required for embryonic axis formation. *Proceedings of the National Academy of Sciences of the United States of America* *106*, 18598-18603.