
Supplementary information

**Single-nucleus and spatial transcriptome
profiling of pancreatic cancer identifies
multicellular dynamics associated with
neoadjuvant treatment**

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authors and unedited

L-WRN conditioned media (CM) was generated by collecting eight days of supernatant from L-WRN cells, grown in Advanced DMEM/F12 (Gibco) supplemented with 20% fetal bovine serum (Hyclone), 2 mM GlutaMAX, 100 U/mL of penicillin, 100 mg/mL of streptomycin, and 0.25 mg/mL amphotericin. L-WRN CM was diluted 1:1 in Advanced DMEM/F12 (Gibco) and supplemented with additional RSPO-1 conditioned media (10% v/v), generated using Cultrex HA-R-Spondin1-Fc 293T cells. Complete media were supplemented with the following additives: B27 (Gibco), 1 mM N-acetylcysteine (Sigma-Aldrich), 10 mM nicotinamide (Sigma-Aldrich), 50 ng/mL EGF (Novus Biologicals), 500 nM A83-01 (Cayman Chemical), 10 mM SB202190 (Cayman Chemical), and 500 nM PGE2 (Cayman Chemical). Wnt activity of the conditioned media was assessed and normalized between batches via luciferase reporter activity of TCF/LEF activation (Enzo Leading Light Wnt reporter cell line).