

Figure S1

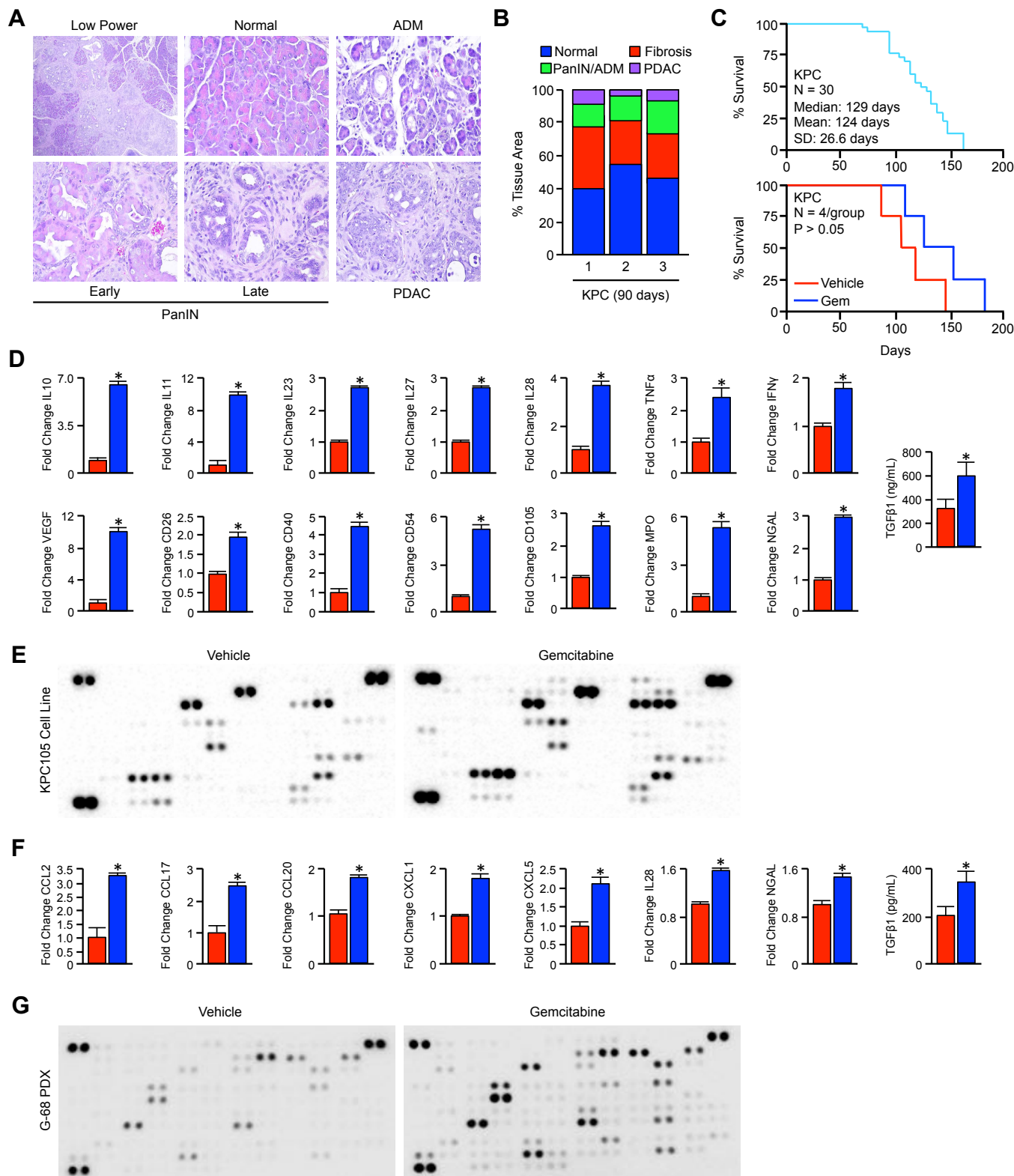


Figure S1. Gemcitabine alters antigen presentation and tumor-derived cytokine production in murine tumor cells

(A,B) *Pdx1-Cre x LSL-Kras^{G12D} x LSL-TP53^{R172H}* (KPC) mice were generated as a model of advanced PDAC. Tissues were collected from three 90-day (~13 weeks) mice and representative histology displayed showing a mix of normal glandular tissue, acinar-to-ductal metaplasia (ADM), early and late PanIN disease, and rare focal areas of PDAC. (C) The Kaplan-Meier curve indicating survival for 30 KPC mice in our colony in the last calendar year, showing a median survival of 124±26 days (N=30). Based on these data, mice were enrolled at 90 days of age and administered twice-weekly intraperitoneal injections of either PBS vehicle or 100mg/kg Gemcitabine. The Kaplan-Meier curve indicating survival for mice across each group is displayed above (N=4/group). (D) Pancreas tissues were collected when the animals were moribund, homogenized, and 200µg of tumor lysate evaluated by a high throughput proteome profiler array (ARY028). Pixel density was evaluated using ImageJ, and samples normalized to the mean intensity of the reference spots for each blot minus the background density. Composite normalized values for Gemcitabine treated mice were divided by those for vehicle treated mice, and are presented as fold change. 20µg of tissue lysate was also subjected to TGF β 1 ELISA (N=3/group, *p < 0.05). (E) KPC105 cells were incubated with 2µM Gemcitabine for 24 hours, after which cells were incubated with a protein transport inhibitor for one hour, lysed, and 200µg of total cell lysate was evaluated by a high throughput proteome profiler array (ARY028). Pixel density was evaluated using ImageJ, and samples normalized to the mean intensity of the reference spots for each blot minus the background density. Composite normalized values for all experimental groups were divided by those for untreated KPC105 cells, and are presented as fold change plus standard deviation. Media was also collected at this time and subjected to TGF β 1 ELISA (N=4, *p < 0.05). (G) G-68 human pancreatic cancer cells were injected subcutaneously into NSG mice, and once tumors reached 100-200 mm³, animals were treated with either a saline vehicle or 40mg/kg Gemcitabine. Animals were sacrificed when moribund or when tumors ulcerated (N=4-5/group). Tumor tissues were harvested, homogenized, and 200µg of tumor lysate evaluated by a high throughput proteome profiler array (ARY022B). Representative blots from each group are displayed above.