Figure S1. EGF affects the viability of cholangiocarcinoma cells. (A and B) Serum-starved (A) SSP-25 and (B) HuCCT1 cells were left unstimulated or stimulated with various concentrations of EGF (10, 20, 60 and 100 ng/ml) for the indicated times. Cells that did not receive stimulation with EGF were instead treated with PBS. After stimulation, the cells were subjected to the Cell Counting Kit-8 cell proliferation assay. The quantitative results were expressed as fold changes by defining the viability of the unstimulated group of each cell line at 0 h as 1. Data are represented as the mean ± SD of triplicate cultures in three independent experiments. $^{*}P<0.05$ and $^{**}P<0.01$ compared with EGF-unstimulated cells at the same time point. EGF, epidermal growth factor.

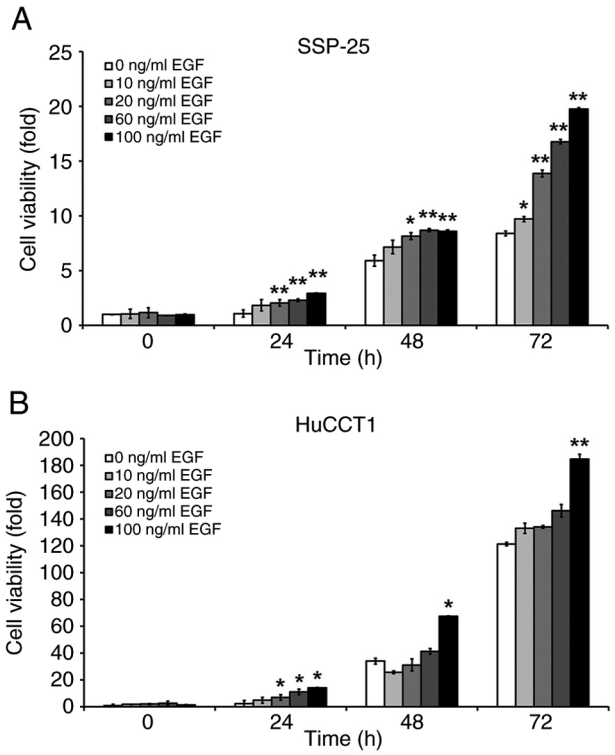

Figure S2. Blocking of PI3K activity affects the expression of p-STAT1, p-STAT3 and PD-L1 in EGF-stimulated cholangiocarcinoma cells. (A and B) Serum-starved (A) SSP-25 and (B) HuCCT1 cells were left unpretreated or pretreated with LY294002 (10 μM) for 1 h and then were left unstimulated or stimulated with 100 ng/ml EGF for 24 h. Cells that did not receive treatment with LY294002 and/or EGF were instead treated with DMSO and/or PBS, respectively. After treatment, the cells were lysed and cell lysates were subjected to western blotting for the detection of the indicated p-AKT, AKT, p-STAT1, STAT1, p-STAT3, STAT3, PD-L1 and GAPDH, as a loading control. Similar results were obtained in three independent experiments. PI3K, phosphoinositide 3-kinase; p-, phosphorylated; STAT, signal transducer and activator of transcription; PD-L1, programmed cell death-ligand 1; EGF, epidermal growth factor.