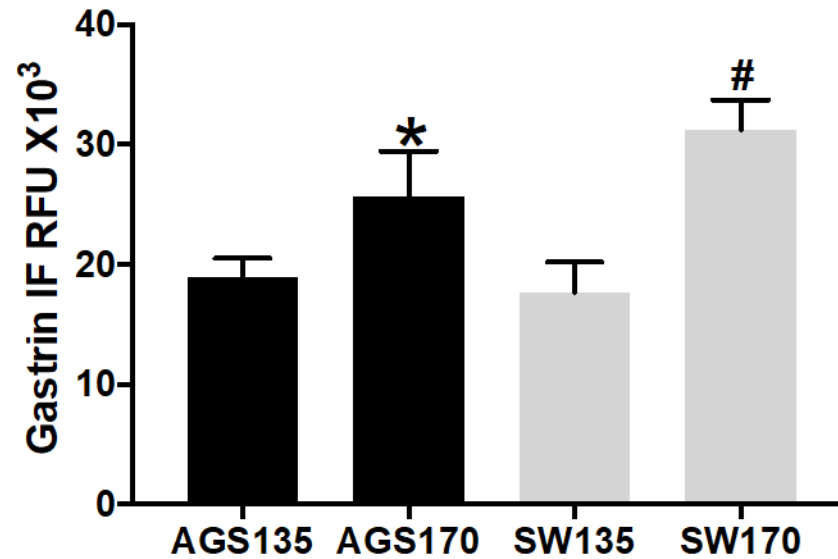
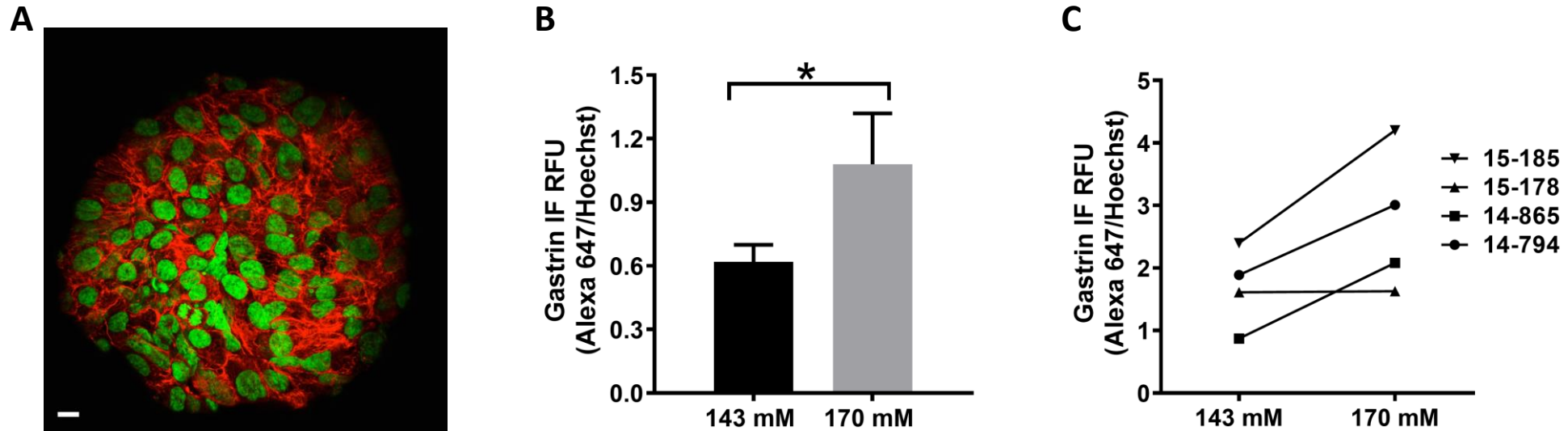


Supplemental Figure 1. Human G and ovarian adenocarcinoma SW626 cells express the appropriate molecular (Mol) size of gastrin mRNA, ~300 base pairs (bp), expected from the primers used. Actin is used for control.



Supplemental Figure 2. Effect of NaCl concentration in the incubation medium on gastrin expression in gastrin-secreting tumor cells, SW626 and AGS. Increasing the sodium concentration in the buffer from 135 to 170 mM NaCl (4 hr incubation) increases gastrin protein expression in both SW626 and AGS cells (*, #, $P < 0.01$ vs 135 mM NaCl, $n = 6/\text{group}$, one-way ANOVA, Holm-Sidak test). IF = immunofluorescence, RFU = relative fluorescence unit



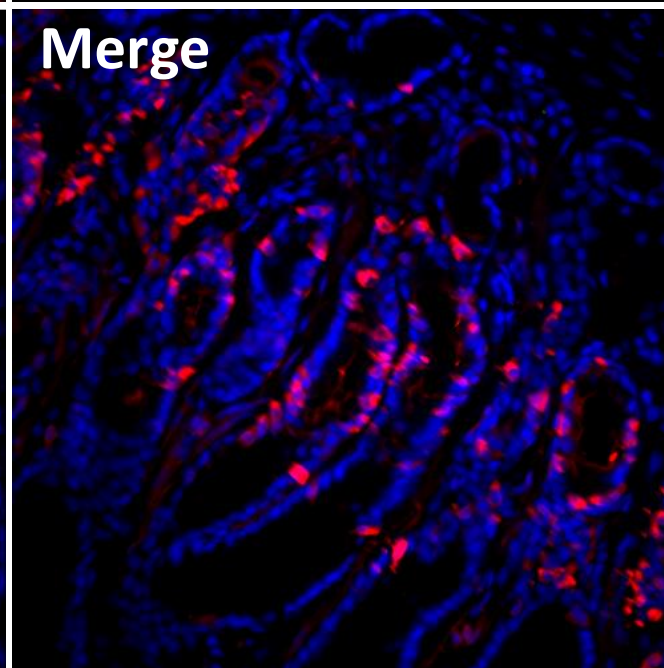
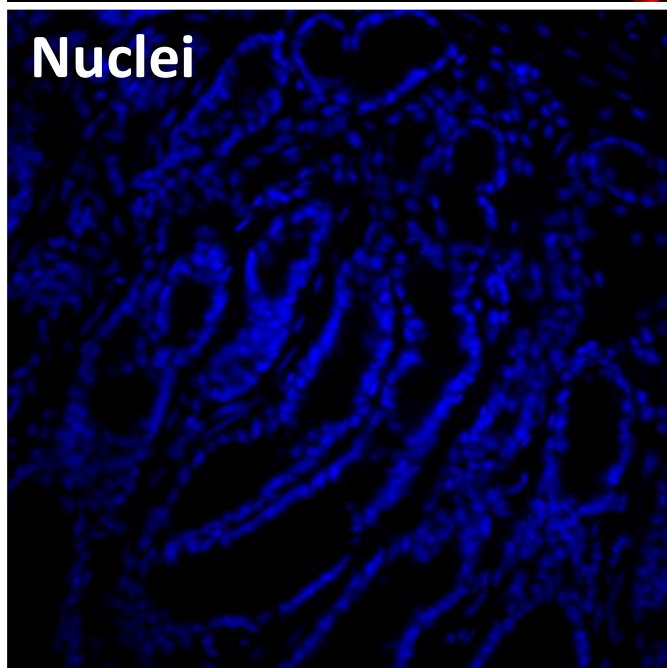
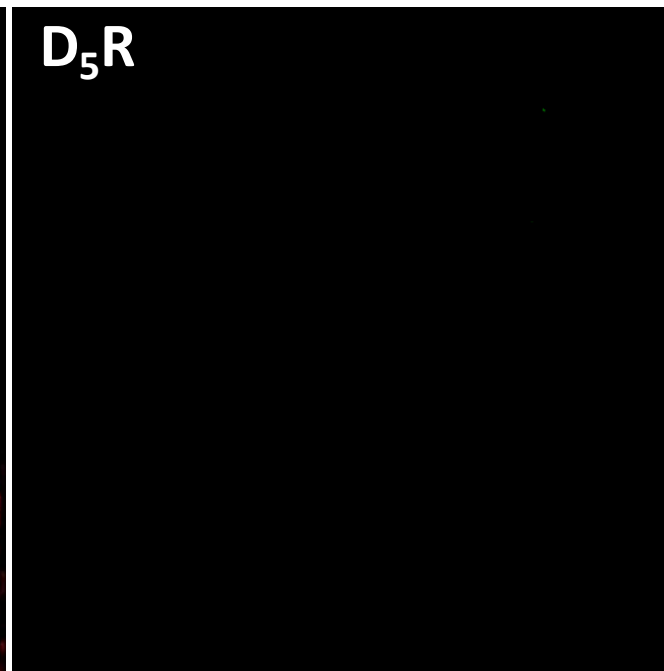
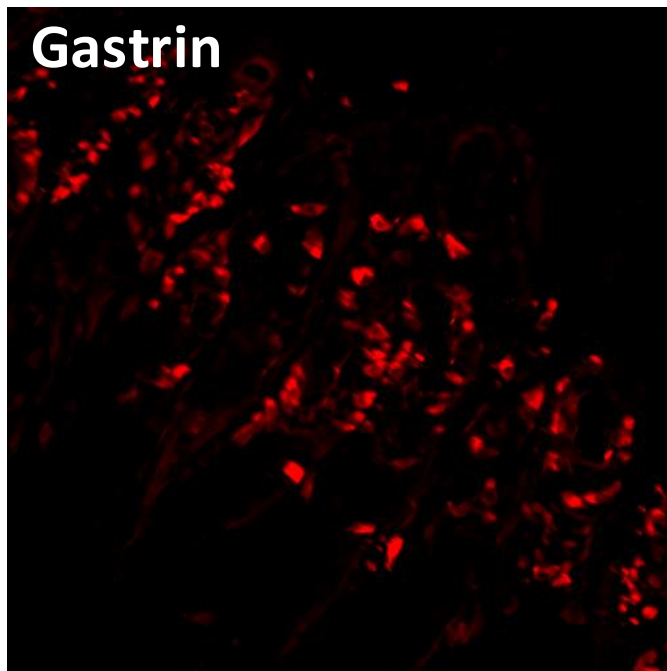
Supplemental Figure 3.

(A) Human G cells were grown on alginate microspheres (Global Cell Solutions). Nuclei were stained with DAPI (green) and F-actin stained with phalloidin (red). This 3D model system enables the study of polarized G cells in monolayer.

(B) G cells, grown as a transporting monolayer in a 3D system, have increased gastrin protein expression under high extracellular sodium concentration (170 mM, 4 hr) ($P < 0.05$, paired t-test, $n = 4$), but not substantially different from conventional cell culture demonstrating that Petri dish cultures achieve optimal results.

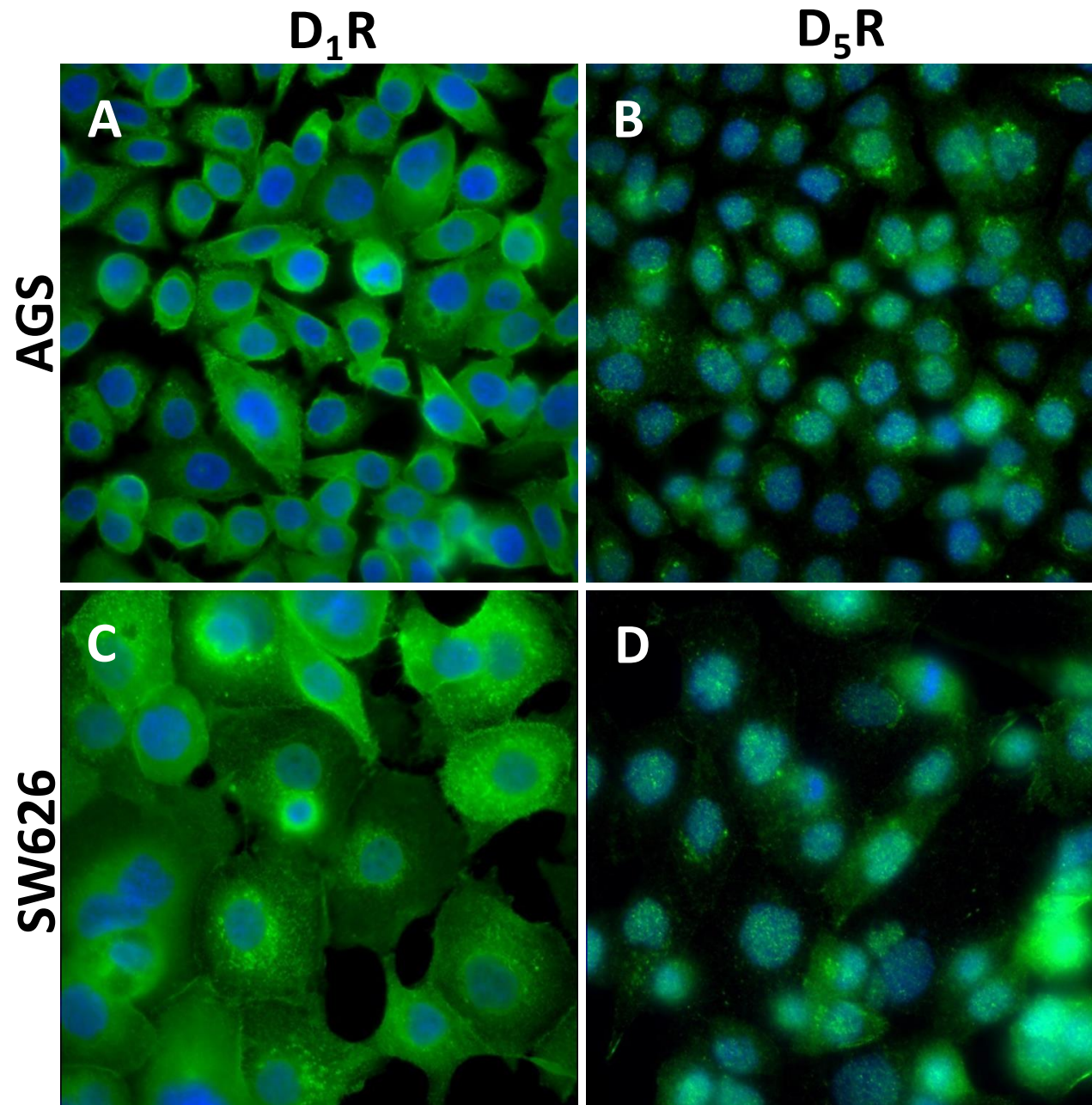
(C) Individual data points for 4 independent cell lines measured in 3 independent assays are shown. The numbers in the graph indicate the assigned cell identification numbers.

IF = immunofluorescence, RFU = relative fluorescence unit



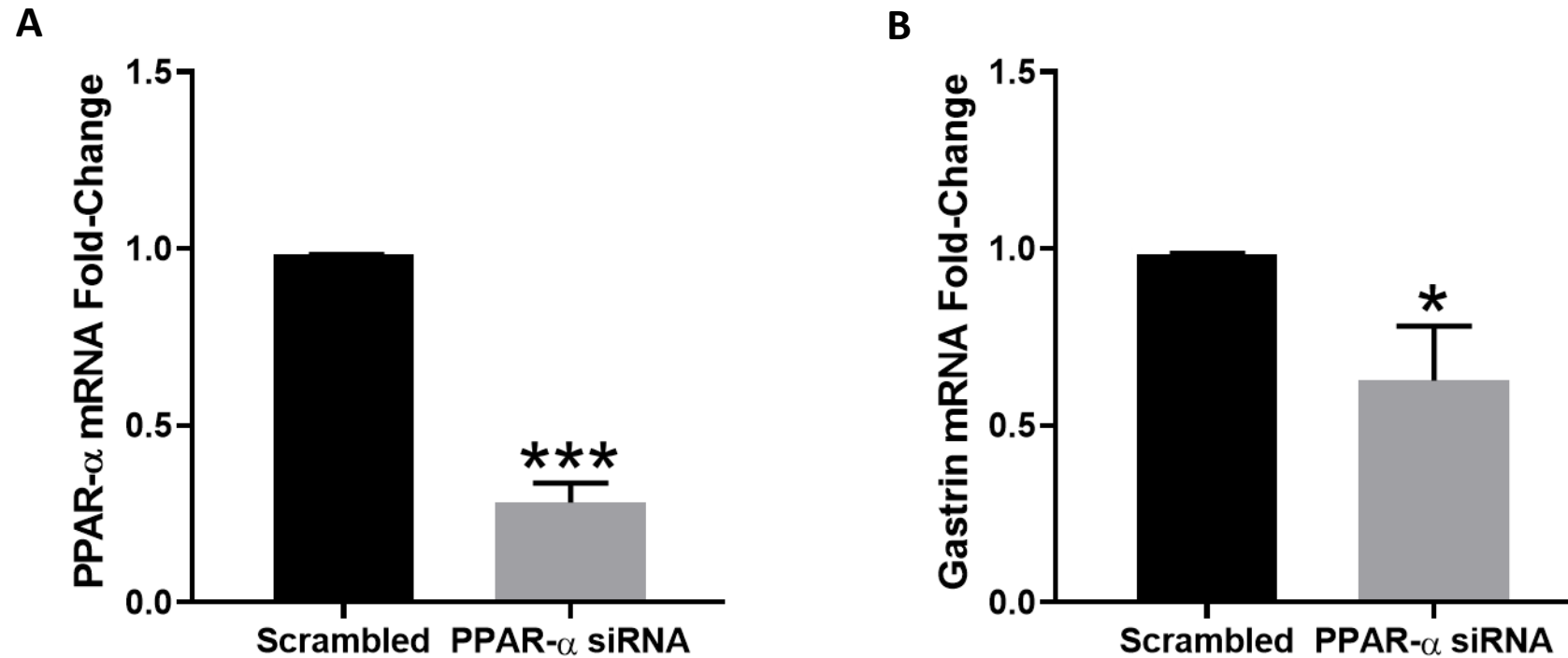
Supplemental Figure 4.

There is no D₅R expression in human stomach antrum. Gastrin is stained red and nucleus stained blue with DAPI.

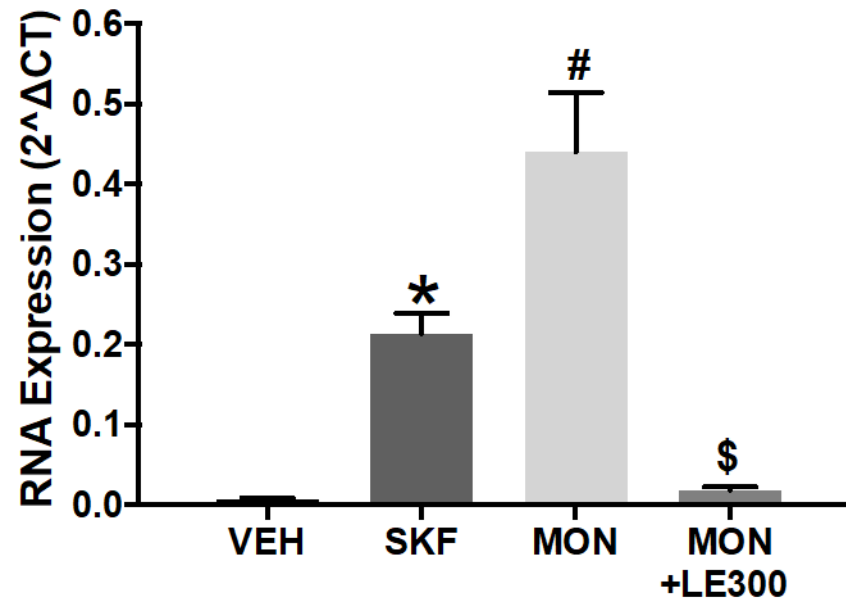


Supplemental Figure 5.

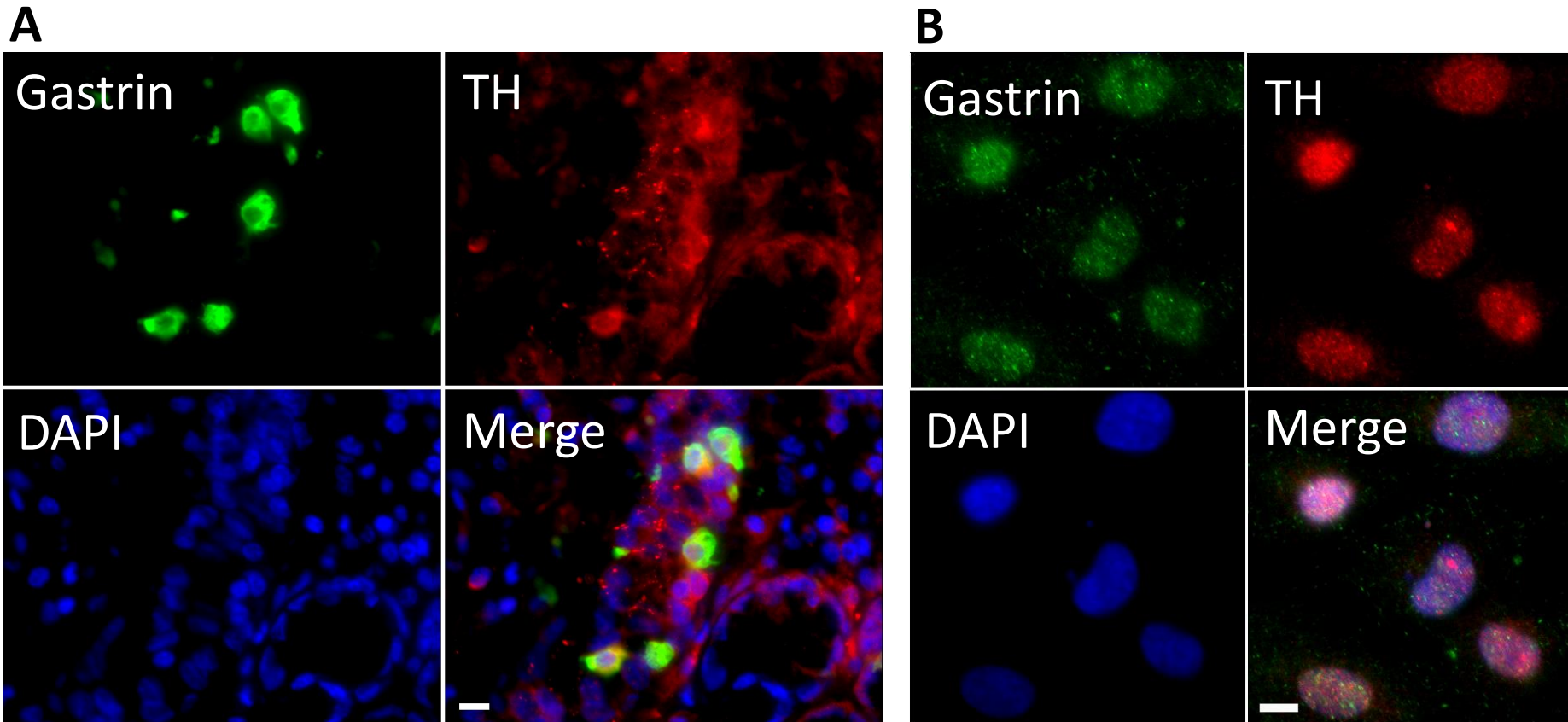
Both D₁R and D₅R are expressed in AGS and SW626 cells.



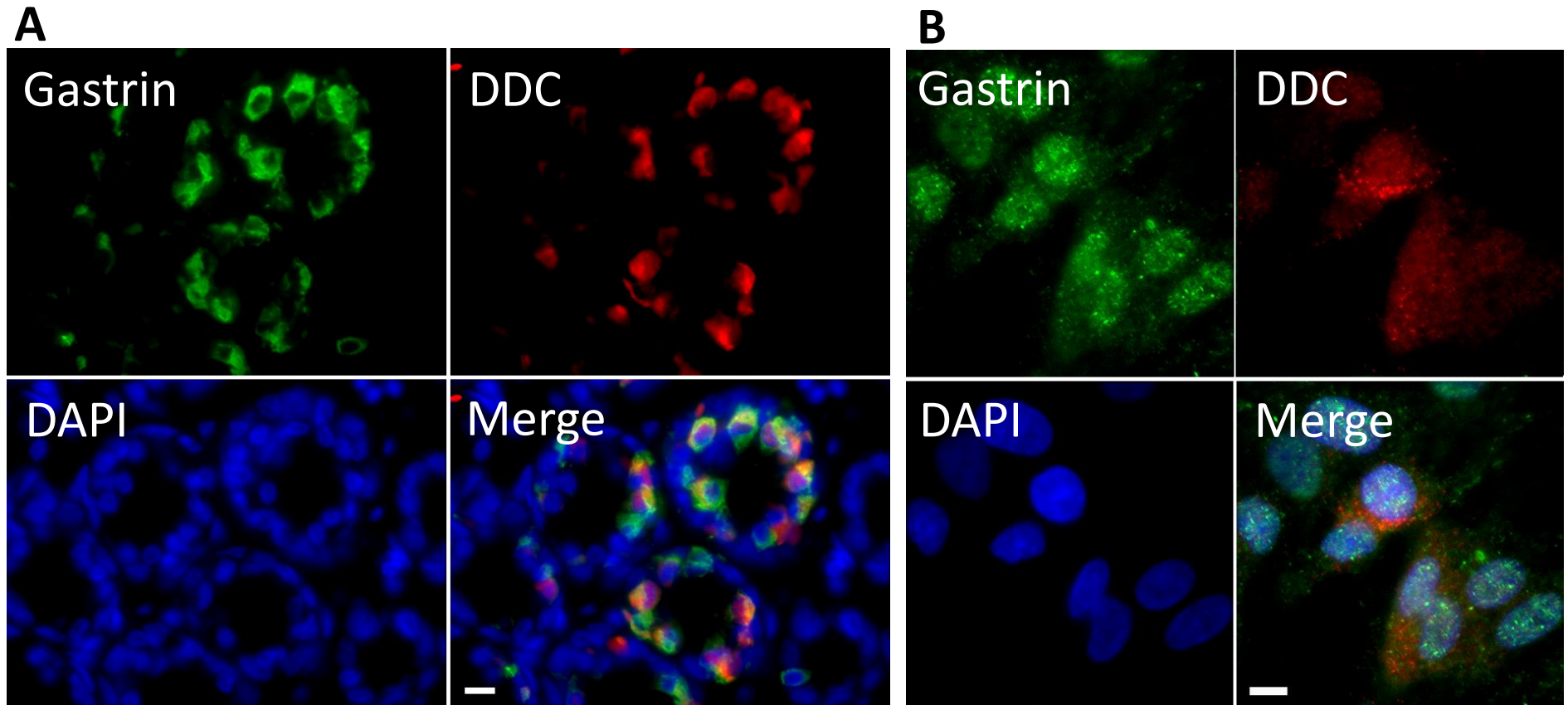
Supplemental Figure 6. PPAR- α silencing in G cells. The transcriptions of intracellular PPAR- α and gastrin mRNAs were measured using qRT-PCR in G cells. The G cells were incubated with PPAR- α siRNA (100 nM/3 d). PPAR- α silencing RNA (siRNA) decreases PPAR- α mRNA to 0.28 ± 0.02 -fold and gastrin mRNA to 0.63 ± 0.08 -fold, relative to scrambled RNA, set at 1.0 (t-test, $p < 0.05$, $n = 4$).



Supplemental Figure 7. Gastrin synthesis, D₁-like receptor, and the PPAR- α pathway in SW626 cells. The transcription of intracellular gastrin mRNA was measured using RT-PCR in SW626 gastric carcinoma cells. Monensin (MON, 1 μ M/24 hr), which increases intracellular Na⁺ (* P<0.001, one-way ANOVA, Tukey test, n=3), and SKF (D₁-like receptor agonist, 10 μ M/24 hr), #P<0.001, one-way ANOVA, Tukey test, n=3) increase intracellular gastrin transcription. However, the D₁-like receptor antagonist, LE300 (10 μ M/24 hr), completely blocks the MON-induced increase in gastrin RNA level (\$P<0.001, one-way ANOVA, Tukey test, n=3).



Supplemental Figure 8. Expression of tyrosine hydroxylase in G cells in stomach antrum and isolated G cells in culture. The D_1R participates in the increase in gastrin expression caused by an increase in extracellular and intracellular sodium. G cells in the human stomach antrum (**Supplemental Figure 8A**) and isolated human G cells in culture (**Supplemental Figure 8B**) express tyrosine hydroxylase (TH) which converts tyrosine to L-DOPA. Gastrin is stained green, TH is stained red, and nucleus is stained blue with DAPI. There is widespread TH red fluorescent staining throughout the crypt cells. The merged images show minimal colocalization of gastrin and TH-staining G cells and not always in the same intracellular site. Nevertheless, these images indicate the presence of the first enzyme (TH) in the synthesis of dopamine in human G cells. Scale bar = 10 μm



Supplemental Figure 9. Expression of DOPA decarboxylase in G cells in stomach antrum and isolated G cells in culture. G cells in the human stomach antrum (**Supplemental Figure 9A**) and isolated human G cells in culture (**Supplemental Figure 9B**) express aromatic L-amino acid decarboxylase (**DDC**) which converts L-DOPA to dopamine. Gastrin is stained green, DDC is stained red, and nucleus is stained blue with DAPI. There is widespread DDC red fluorescent staining throughout the crypt cells. The merged images show minimal colocalization of gastrin and DDC-staining G cells and not always in the same intracellular site. Nevertheless, these images indicate the presence of the second enzyme (DDC) in the synthesis of dopamine in human G cells. Scale bar = 10 μm