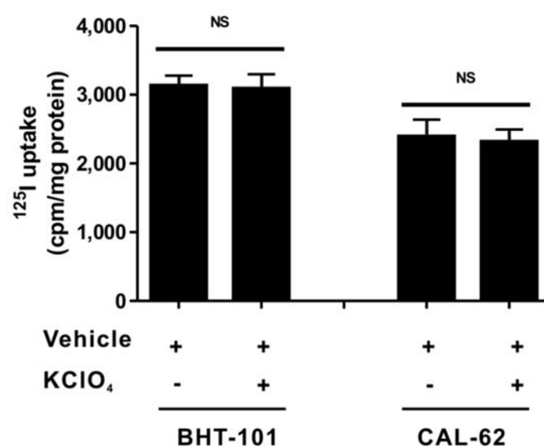
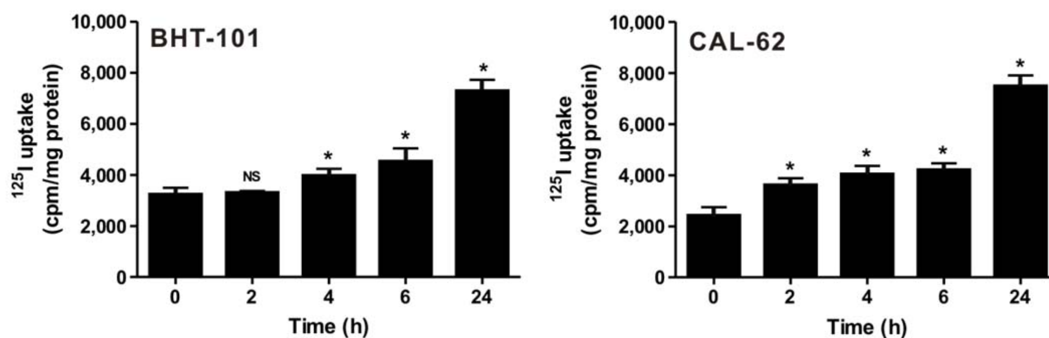
^{125}I Uptake Assay

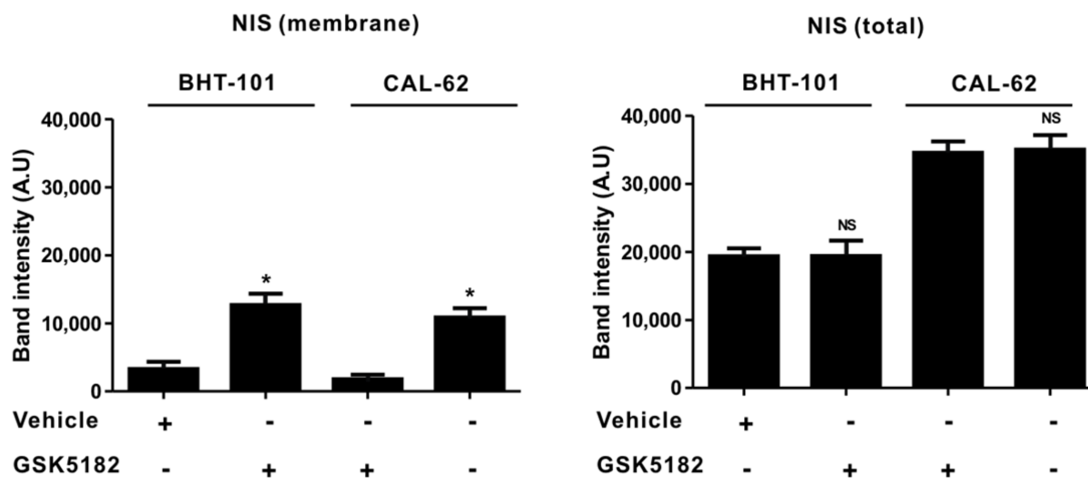
ATC cells were plated in 24-well plates for 24 h. To inhibit the iodide uptake, vehicle-treated ATC cells were preincubated with 300 μM $KClO_4$ (as a specific inhibitor of NIS) for 30 min. After aspiration of the medium, the cells were washed with 1 mL of HBSS and incubated with 500 μL of Hanks balanced salt solution (HBSS) containing 0.5% bovine serum albumin (bHBSS), 3.7 kBq of carrier-free ^{125}I (Perkin-Elmer), and a 10 μmol/L solution of sodium iodide (specific activity of 740 MBq/mmol) at 37℃ for 30 min. The cells were then washed twice with ice-cold bHBSS and were lysed with 500 μL of 2% sodium dodecyl sulfate (SDS). The radioactivity was measured using a γ counter (Cobra II; Canberra Packard, Packard Bioscience). The radioactivity of the cells was normalized using total protein concentrations determined by a BCA kit (Pierce Protein Biology).



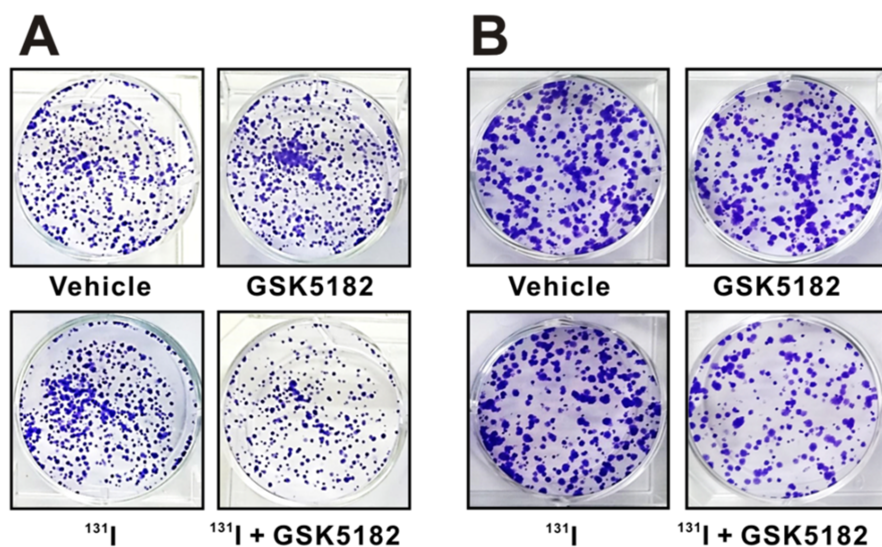
Supplemental Fig 1. Comparison of iodide uptake between vehicle-treated and vehicle + $KClO_4$-treated ATC cells. Each cell was treated with vehicle for 24 h. Before iodide uptake, 300 μM $KClO_4$ was incubated with each cell lines for 30 min; afterward, the iodide uptake levels of both cell lines were confirmed. NS = not significant when compared with vehicle + $KClO_4$ cells. All data are expressed as mean ± SD; $n = 3$.



Supplemental Fig 2. The change in iodide uptake in GSK5182-treated cells according to time course. **P* < 0.05; NS = not significant when compared with untreated cells. All data are expressed as mean ± SD; *n* = 3.



Supplemental Fig 3. Quantitative analysis of membrane and total NIS protein levels by scanning densitometry. Data are mean ± SD of 3 samples per group expressed in arbitrary units. **P* < 0.05, when compared with untreated cells. NS = not significant when compared with vehicle.



Supplemental Fig 4. Representative photographs of plates of CAL-62 (A) and BHT-101 (B) cells after clonogenic assay.