

Figure S1

Figure S1. R-568 inhibits forskolin-induced Cl<sup>-</sup> secretion in T84 cells. A. Short-circuit current ( $I_{sc}$ ) traces showing maximal forskolin (10 μM) response and CFTR<sub>inh</sub>-172 (10 μM) inhibition following 20 min pretreatment with indicated concentrations of R-568. B.  $\Delta$   $I_{sc}$  induced by maximal (10 μM) forskolin at different R-568 concentrations. C.  $\Delta$   $I_{sc}$  following CFTR<sub>inh</sub>-172 (10 μM) treatment at different R-568 concentrations. n=4-5 experiments per group. Mean  $\pm$  S.E.M., one-way analysis of variance with Newman-Keuls multiple comparisons test, \*\*\*p<0.001, ns: not significant.

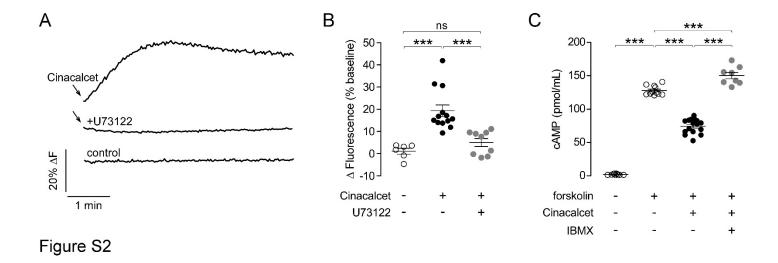


Figure S2. Cinacalcet increases intracellular  $Ca^{+2}$  via phospholipase C and inhibits forskolin-induced cAMP elevation in T84 cells. A. Intracellular  $Ca^{+2}$  traces measured by Fluo-4 NW fluorescence in T84 cells with cinacalcet (30  $\mu$ M, top two traces) and vehicle control (1% DMSO, bottom) treatment at indicated times. In some experiments, T84 cells were pretreated with PLC inhibitor U73122 (10  $\mu$ M) for 5 min before cinacalcet treatment (middle trace). B. Summary of data in A presented as maximum changes in Fluo-4 NW fluorescence as percentage of baseline reading. n=6-13 experiments per group. C. cAMP concentration in T84 cell lysates after maximal forskolin (10  $\mu$ M) treatment with and without 30  $\mu$ M cinacalcet ( $\pm$  500  $\mu$ M IBMX, phosphodiesterase inhibitor) pretreatment. n=8-16 experiments per group. Mean  $\pm$  S.E.M., one-way analysis of variance with Newman-Keuls multiple comparisons test, \*\*\*p<0.001, ns: not significant.