Supplementary information

Figure Legends

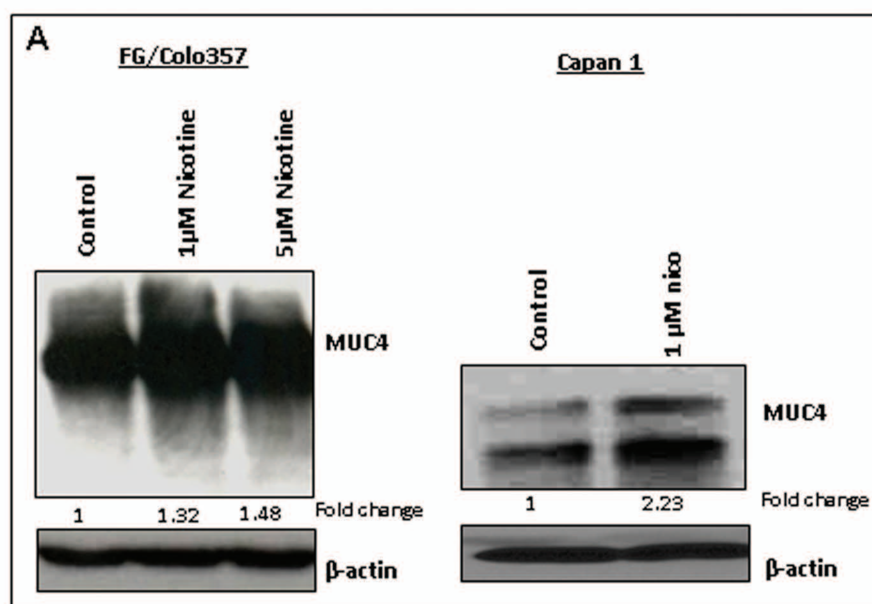
Supplementary Figure 1. Expression levels of MUC4 in PC cells and normal epithelial cells treated with nicotine ***1A.*** MUC4 protein expression was increased in Capan1 PC cells upon treatment with 1μM nicotine and in a dose dependent manner in FG/Colo357 PC cells were treated with different doses of nicotine (1μM, 5μM) for 24 hrs, as compared to the untreated cells. β-actin was used as an internal control. ***1B.*** Human pancreatic ductal epithelial (HPDE) cells were seeded on cover slips and were treated with different doses of nicotine (0.1μM, 5μM) for 24 hrs. The confocal analysis showed no expression levels of MUC4 (FITC: green) in untreated as well as the treated cells. DAPI was used for nuclear counter staining.

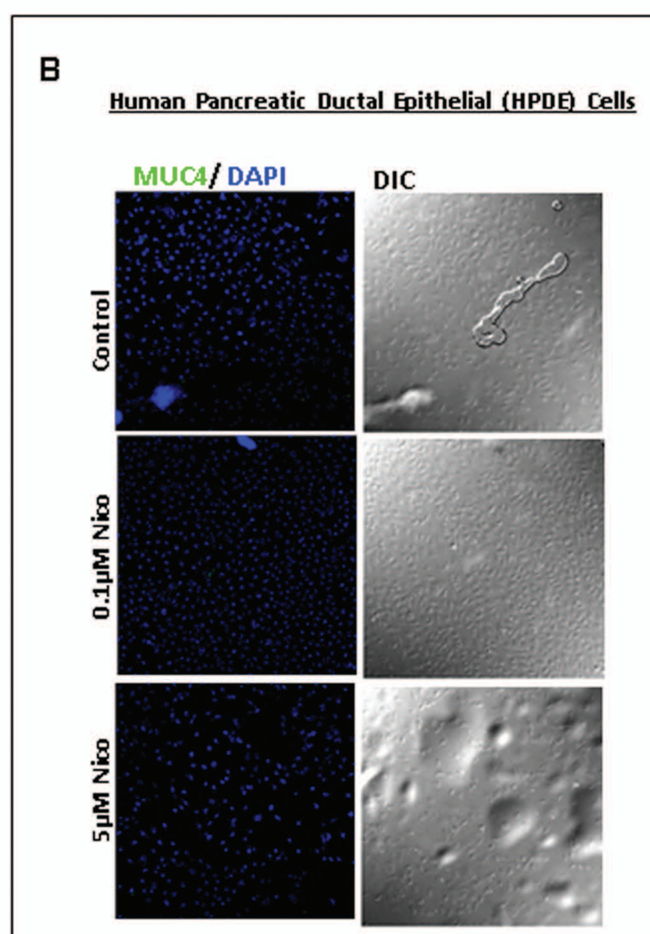
Supplementary Figure 2. Expression levels of α7nAchR in PC cells treated with nicotine and α7nAchR antagonists. ***2A***. Decreased expression levels of α7nAchR subunit and nicotine-induced-MUC4 was observed in Capan1 cells treated with both α-BTX (α-bungarotoxin) and with another α7 nicotine receptor subunit antagonist, mecamylamine, MAA (1μM). ***2B.*** The CD18/HPAF cells were treated with MAA (1μM). The nicotine-induced MUC4 expression was significantly abrogated in MAA-treated-CD18/HPAF cells. ***2C.*** CD18/HPAF cells were treated with calcium channel blocker (Nefedipine) and general antagonists for acetylcholine nicotine receptor (HexamethoniumBrobide) and muscarinic acetylcholine receptor (Atropine). Nifedipine abrogated the expression levels of nicotine-induced MUC4. HexamethoniumBrobide and Atropine demonstrated similar effect on the MUC4 expression levels in nicotine-treated and untreated cells. β-actin was used as an internal control. Western blots were quantified using the

software, ChemiImager4400. The numerical values specified beneath the respective bands of western blots represent the fold change in protein expression as compared to that of control (1.0).

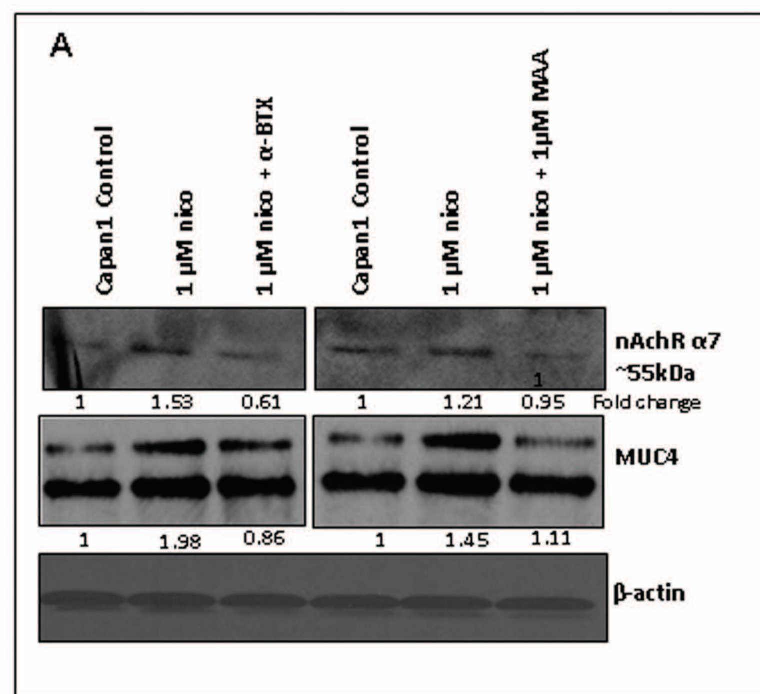
Supplementary Figure 3. Nicotine-mediated increase in the migratory potential of the Capan1 cells through increased activation of FAK. ***3A.*** The wound healing assay showed a gradual increase in the migratory potential of the Capan1 cells in a dose dependent manner (1µM, 5µM of nicotine). ***3B.*** Western blot analysis showed that nicotine treatment increased the phosphorylation of FAK in the Capan1 cells in a dose dependent manner (0.1µM, 1µM of nicotine). The expression levels of total-FAK remained unaltered***.*** ***3C.*** The FAK activation was investigated in MUC4 knockdown Capan1 cells. 1µM nicotine induced high levels of MUC4 expression leading to an increased activation of FAK in Capan1/scr cells as compared to the Capan1/shMUC4 cells. The expression levels of total-FAK remained unaltered.

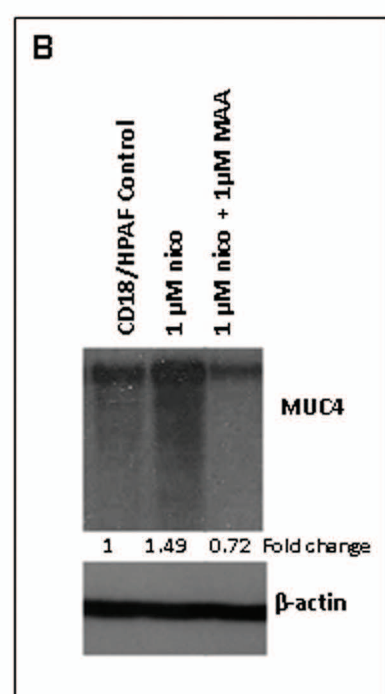
Supplementary Figure 1

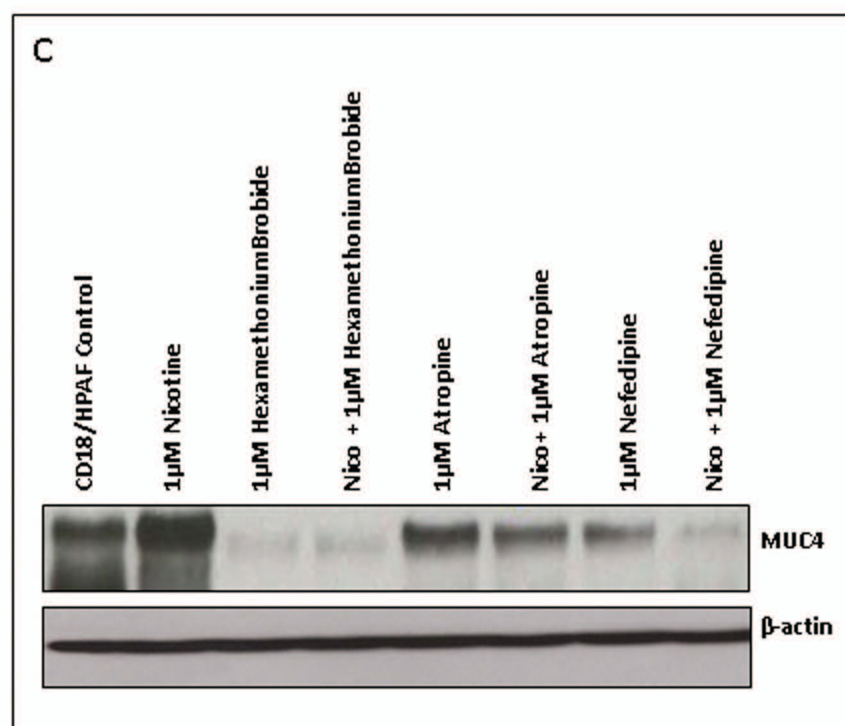




Supplementary Figure 2







Supplementary Figure 3