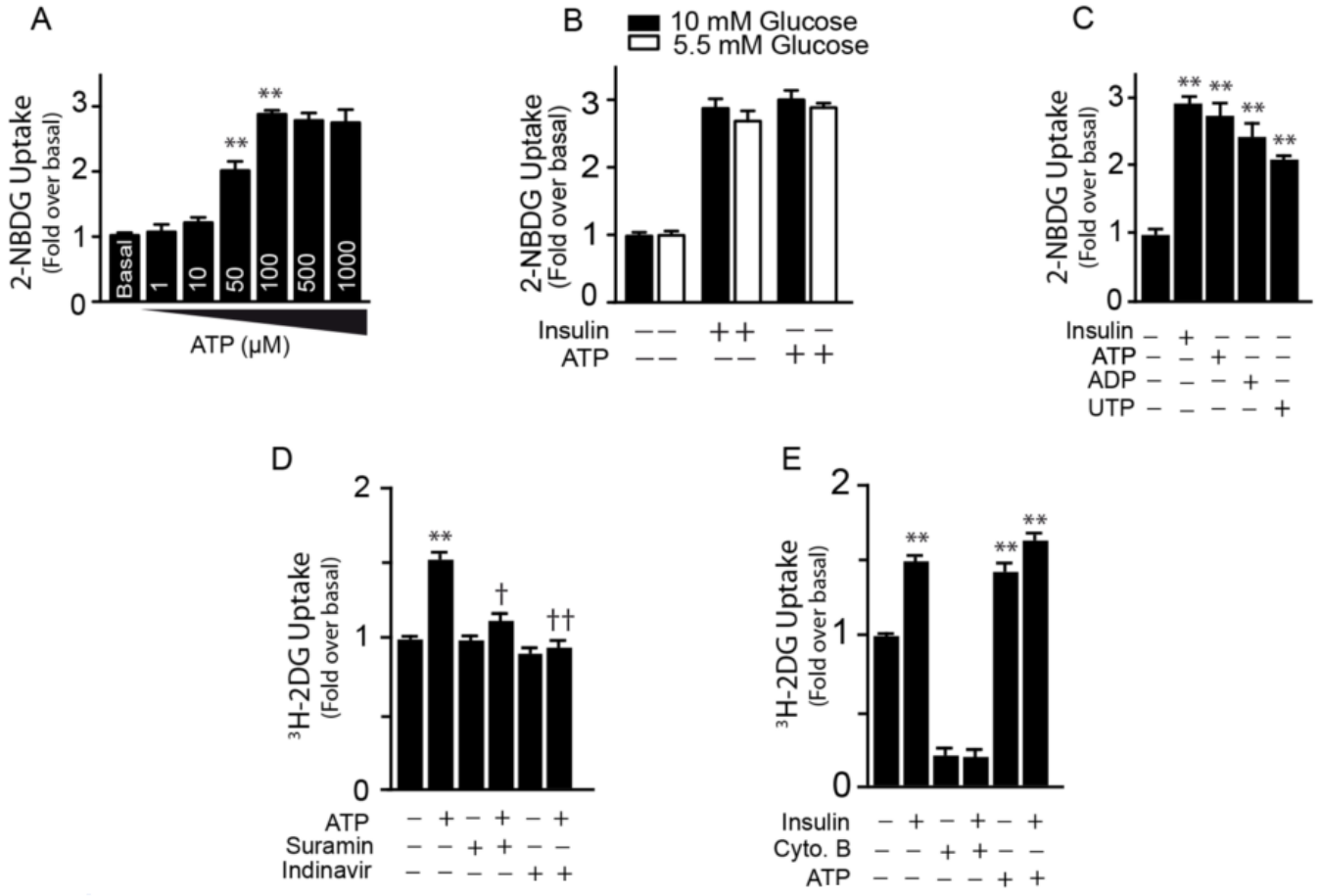
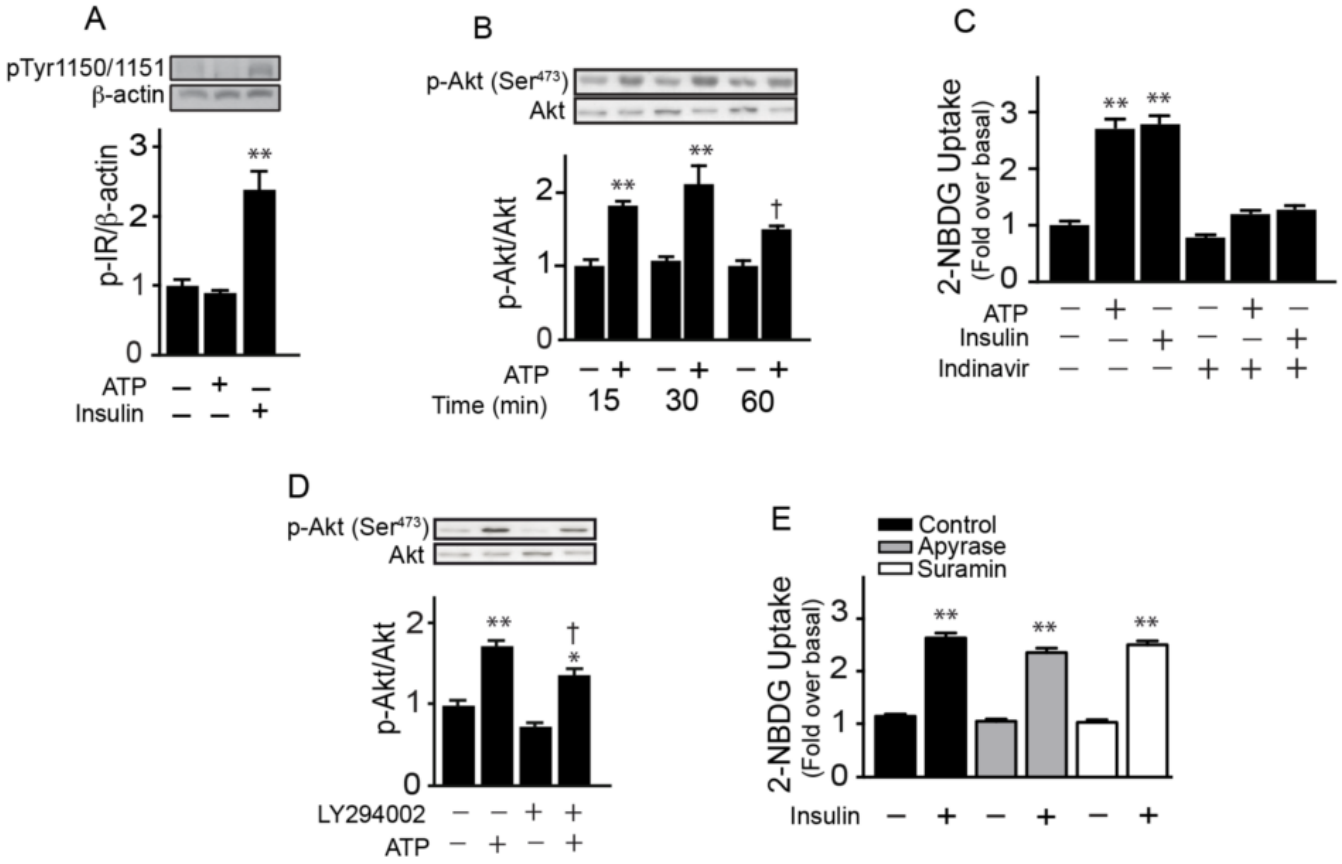
SUPPLEMENTARY DATA

Supplementary Figure 1. (A) ATP dose response for 2-NBDG uptake. **(B)** 100 nM insulin and 100 μM ATP stimulated 2-NBDG uptake were measured in myotubes maintained in DMEM (10 mM glucose) or α-MEM (5.5 mM Glucose) medium. **(C)** Myotubes were stimulated with Insulin (100 nM), ATP (100 μM), ADP (100 μM) or UTP (100 μM), then 2-NBDG uptake was assayed as indicated in *Experimental design and Methods*. Agonists of P2Y receptors ADP and UTP were stimulate 2-NBDG uptake (up 60 myotubes per condition). **(D)** ^{3}H-2DG uptake was measured in myotubes as described in *Experimental design and Methods*, pre-treated with suramin (100 μM) or in the presence of indinavir (100 μM) in the transport solution. **(E)** Insulin (100 nM) and ATP (100 μM) stimulation of ^{3}H-2DG uptake. Cytochalasin B (10 μM) added to the transport solution blocked ^{3}H-2DG uptake in non-stimulated and insulin-stimulated cells. Values are the mean ± SD. ***p< 0.001 vs* basal group, ††*p<0.001,* † p< *0.01 vs* ATP-stimulated condition. Data from at least 5 independent experiments.



SUPPLEMENTARY DATA

Supplementary Figure 2. Myotubes were stimulated with ATP (100 μM), or insulin (100 nM). (**A,B)** Representative immunoblots of pTyr1150/1151 insulin receptor and Akt phosphorylation at Ser 473. (**C)** 2-NBDG uptake assayed as indicated in *Experimental design and Methods* with indinavir (100 μM) present in the uptake solution to inhibit GLUT4. **(D)** Pre-treatment with LY294002 (40 μM) prior and during ATP stimulation followed by Akt phosphorylation in Ser473 phosphorylation. **(E)** Insulin-dependent 2-NBDG uptake was measured in myotubes pre-incubated for 30 min with apirase (2 U/ml) or suramin (100 μM). Values are the mean ± SD. ***p< 0.001, *p<0.01*vsbasal group, †*p<0.01* relative to the ATP-stimulated condition. Data from at least 4 independent experiments, 60 cells analyzed per condition.



SUPPLEMENTARY DATA

Supplementary Figure 3. L6-GLUT4*myc* cells were stimulated with ATP (100 μM) or insulin (100 nM). Extracellular exposure of the *myc* epitope was detected by immunofluorescence in non-permeabilized cells to quantify the exocytosis process as described in *Experimental design and Methods*. ***p<0.001* Insulin or ATP *vs* basal group at the indicated times. Data from at least 3 independent experiments.