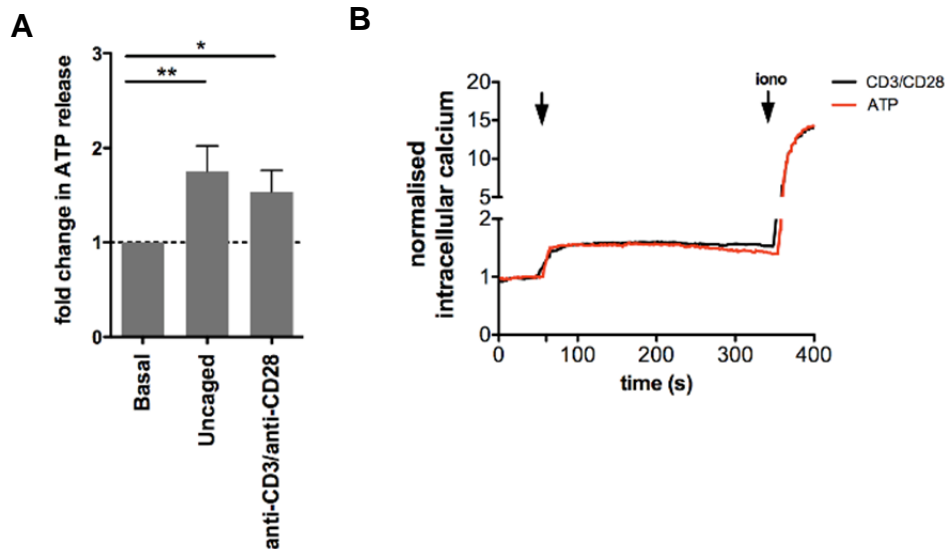


Supplementary Figure S2



Supplementary Figure S2

A. ATP released by 10^6 human peripheral blood $CD4^+$ T cells after photolysis of caged IP3 or stimulation with anti-CD3 (2 $\mu\text{g/ml}$) and anti-CD28 (1 $\mu\text{g/ml}$) antibodies. The level of ATP released after 10 minutes of stimulation was measured (** $P < 0.01$; * $P < 0.05$) (N = 7 different donors for caged-IP3, N = 4 different donors for anti-CD3/antiCD28).

B. Flow cytometry analysis of calcium influx in human $CD4^+$ T cells in response to anti-CD3/anti-CD28 antibody or extracellular ATP. Basal cytosolic calcium level was acquired for 60s prior to the addition of ATP (100 μM) or anti-CD3 antibody (5 $\mu\text{g/ml}$). The changes in calcium level were recorded for the next 5 minutes and ionomycin was added at the end as positive control (second arrow). The ratiometric calcium levels were normalized using the formula (Fluo4/FuraRed ratio)/(mean of the resting Fluo4/FuraRed in the first 60s) to show the fold of change in relation to basal calcium levels. The magnitude of calcium increase was not significantly different for ATP or anti-CD3 antibody stimuli (N = 3 donors; paired t test).