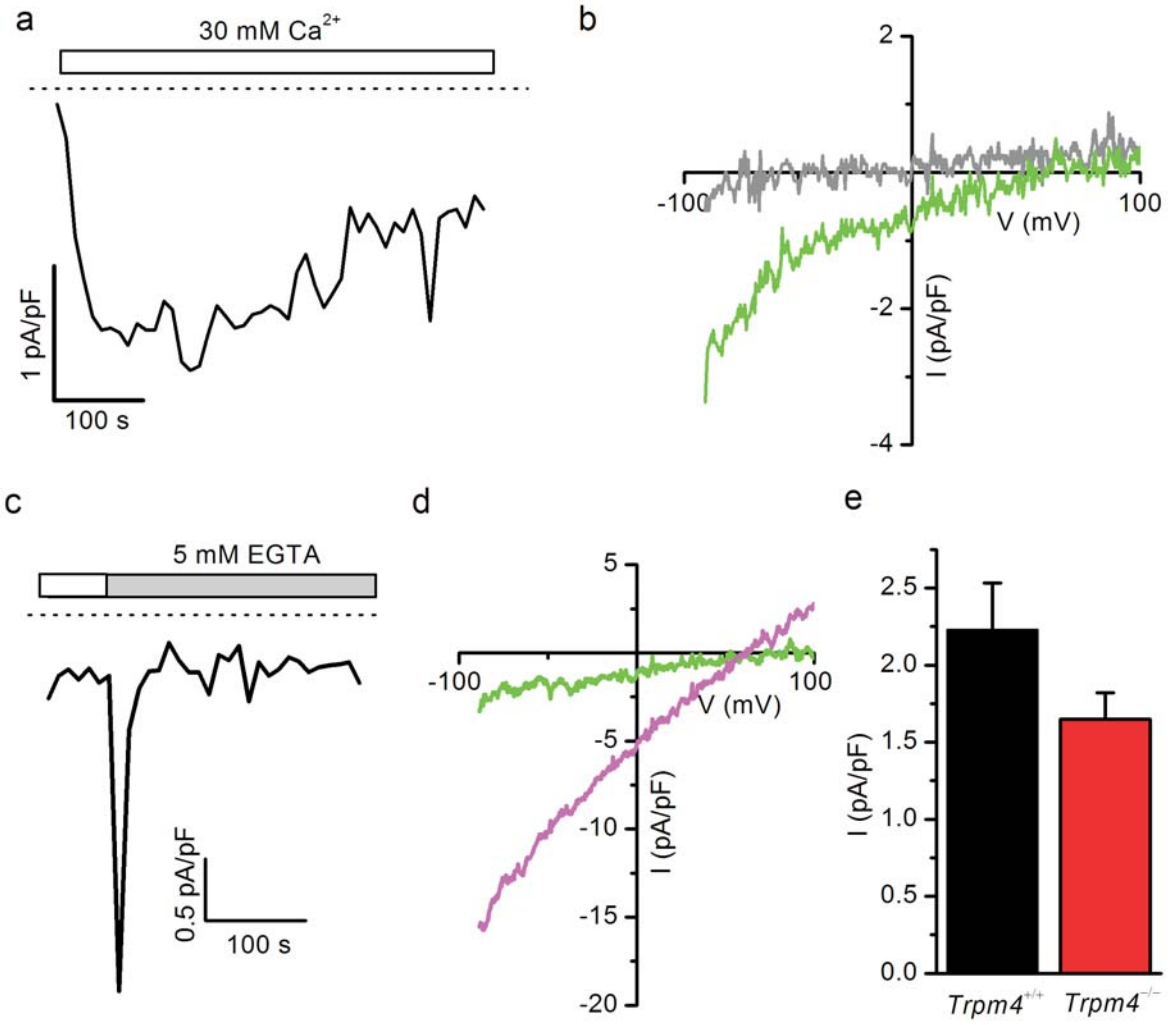
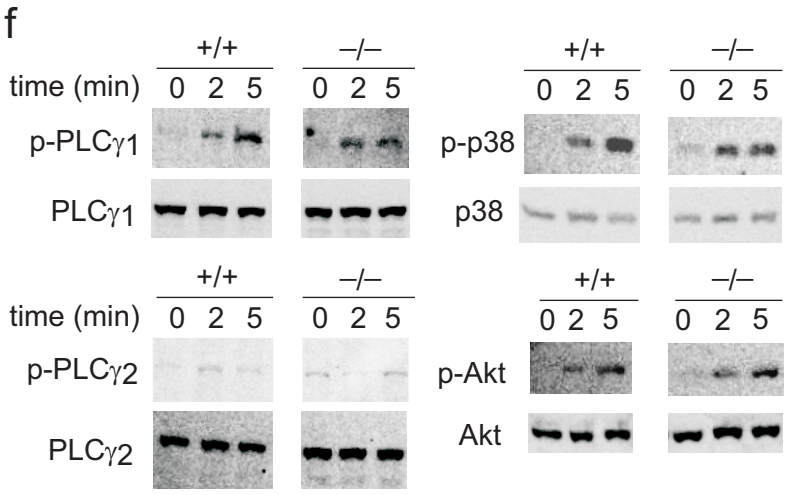

a
30 mM Ca2+
1 pA/pF
100 s
b
2
-100
100
V (mV)
-2
-4
I (pA/pF)
c
5 mM EGTA
0.5 pA/pF
100 s
d
5
-100
100
V (mV)
-5
-10
-15
-20
I (pA/pF)
e
2.5
2.0
1.5
1.0
0.5
0.0
I (pA/pF)
Trpm4+/+
Trpm4−/−


f
+/+
−/−
time (min)
0 2 5
0 2 5
p-PLCγ1
PLCγ1
p-p38
p38
+/+
−/−
time (min)
0 2 5
0 2 5
p-PLCγ2
PLCγ2
p-Akt
Akt

Supplementary Figure 5. (**a-e**) CRAC in *Trpm4*$^{+/+}$ and *Trpm4*$^{-/-}$ BMMCs. (**a**) Time course of a whole cell recording at -80mV with a pipette solution containing 10mM BAPTA and 50µM IP_3 in 120mM CsAsp, 20mM CsCl, 2mM $MgCl_2$, 1mM Na_2ATP, 10mM Hepes, pH7.2. Extracellular solution contained 30mM Ca^{2+}. A representative example is shown for a *Trpm4*$^{+/+}$ cell. (**b**) Representative current traces taken from an experiment as in panel **a**, at the beginning of the experiment (grey trace) and after reaching the plateau (green trace). Currents were corrected for leak currents as previousy described[3]. (**c**) Experiment and pipette solution as in panel **a**. After full activation of I_{CRAC} extracellular solution was switched from 30mM Ca^{2+} (open bar) to 0mM Ca^{2+} (5mM EGTA; grey bar). (**d**) Representative current traces taken from an experiment as in panel **c**, in 30mM Ca^{2+} solution (green trace) and at the peak value after switching to 0mM Ca^{2+} solution (pink trace). The transient increase of the current upon switching to Ca^{2+}-free medium is typical behavior for the I_{CRAC} current[3]. (**e**) Mean values for the current in 30mM Ca^{2+} solution measured at -80mV for *Trpm4*$^{+/+}$ and *Trpm4*$^{-/-}$ BMMCs. *n*= 10 for both genotypes, P > 0.05. (**f**) FcεRI-induced activation of PLCγ1, PLCγ2 , p38 and Akt. *Trpm4*$^{+/+}$ and *Trpm4*$^{-/-}$ BMMCs sensitized with anti-DNP IgE were stimulated with DNP for the indicated time intervals. Total cell lysates were separated by 7 and 10% SDS-PAGE, and lysates were immunoblotted with indicated antibodies. Data are representative of at least three independent experiments and independent cell preparations.