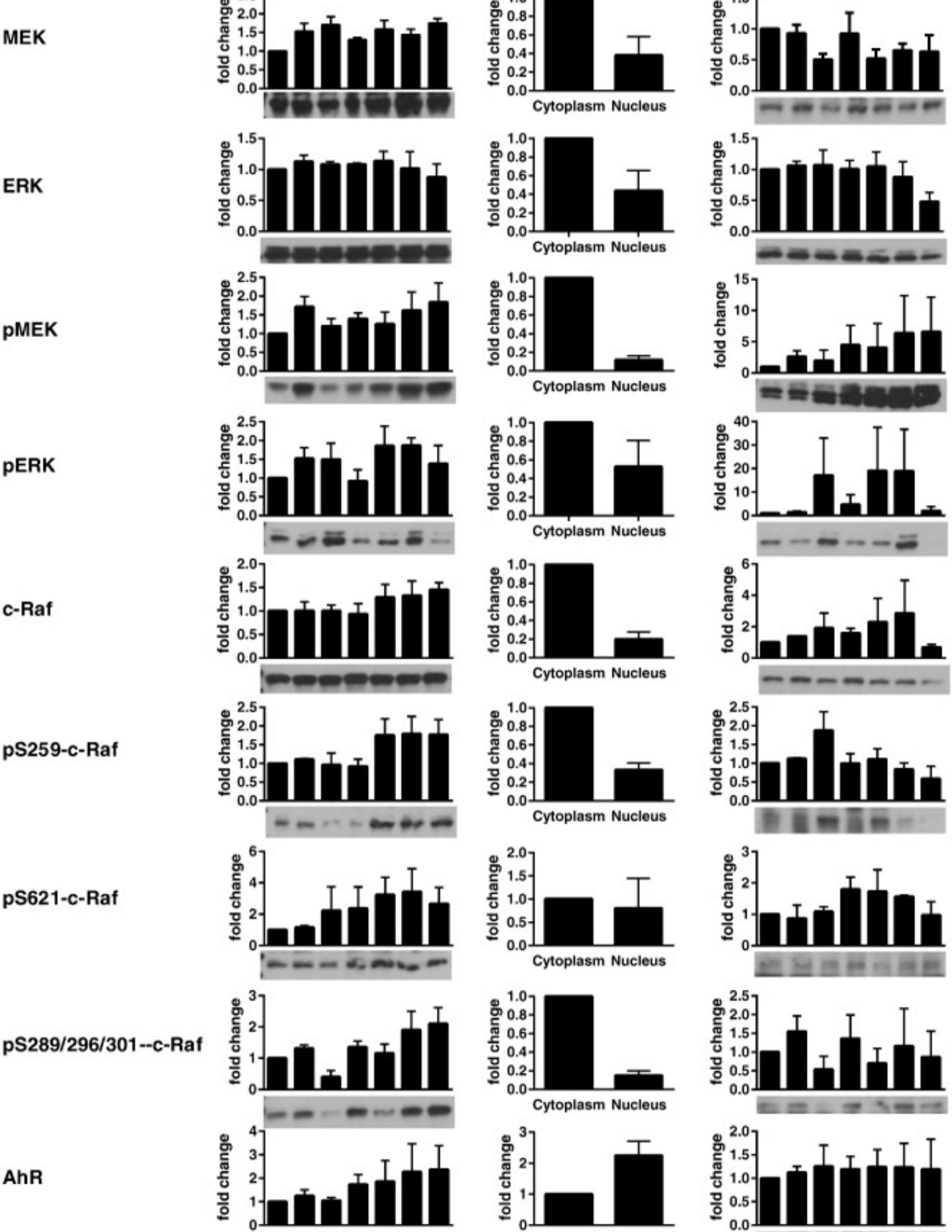

MEK
ERK
pMEK
pERK
c-Raf
pS259-c-Raf
pS621-c-Raf
pS289/296/301--c-Raf
AhR
fold change
Cytoplasm Nucleus

Fig. S1.

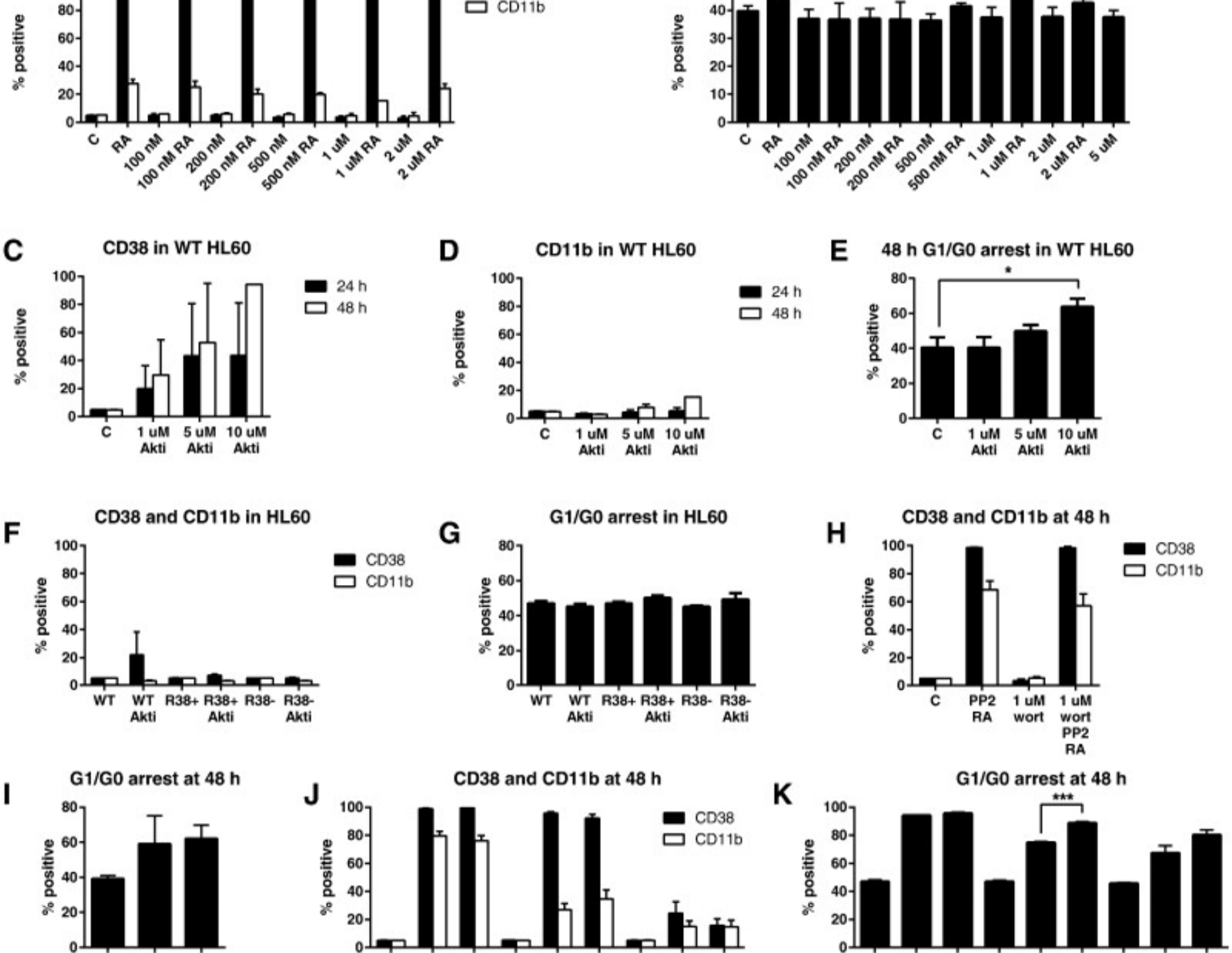
Dose responses to wortmannin and Akti-1/2 in wild-type, R38 + and R38 − HL-60 cells.

Results depict between one and three repeats of dose response investigation. (A–B) Wortmannin doses of 100 nM, 200 nM, 500 nM, 1 μM, and 2 μM were co-treated with RA in wild-type (WT) HL-60 cells. There was no statistically significant difference in CD38 or CD11b expression at 48 h between RA-treated WT HL-60 and combination RA + wortmannin-treated WT HL-60. Additionally, wortmannin doses as high as 5 μM did not statistically affect G1/G0 arrest at 48 h. (C–E) Akti-1/2 doses at 1 μM or 5 μM had no statistically significant effect on CD38 or CD11b expression (24 or 48 h) or G1/G0 arrest (48 h) in WT HL-60. A 10 μM Akti-1/2 dose could increase CD38 and CD11b expressions but statistical analysis (i.e. repeat data) was not possible due to material limitations and diminished cell densities. 10 μM Akti-1/2 treatment did result in statistically significant ($p < 0.05$) G1/G0 arrest in WT HL-60 cells at 48 h. (F–G) Akti-1/2 treated at 1 μM did not significantly rescue CD38 or CD11b expression or G1/G0 arrest in R38 + or R38 − cells at 48 h. (H–I) 1 μM wortmannin combination treatment with PP2 + RA-treated WT HL-60 had no significant effect on CD38 or CD11b expression or G1/G0 arrest at 48 h. (J) Akti-1/2 treated at 1 μM had no significant effect on PP2 + RA-treated WT, R38 + or R38 − HL-60 cells at 48 h. (K) Akti-1/2 treated at 1 μM significantly increased ($p < 0.001$) PP2 + RA-induced G1/G0 arrest at 48 h in R38 + HL-60 cells only.

Fig. S2–7.

Quantified signaling factor expression in wild-type, R38 + and R38 − HL-60 cells.

Quantified Western blot data of at least three repeats for signaling factor expression in cytoplasmic (left graphs) and nuclear (right graphs) compartments. Center graphs depict the comparison between cytoplasmic and nuclear levels. Treatments include RA, PD98059 (PD), GW5074 (GW), wortmannin (Wo), Akti-1/2 (Ai), and PP2. Data is expressed as fold change compared to respective control and error bars represent standard error. Loading control for cytoplasmic (GAPDH) and nuclear (TATA-binding protein) fractions were used to assess even loading (not shown). Since quantified data is estimated from immunoblot images in which signal detection may or may not have been in the linear range, p-value analysis is not applicable to quantified blot data. Instead, a representative blot is shown beneath each graph.


% positive
CD11b
C
RA
100 nM
100 nM RA
200 nM
200 nM RA
500 nM
500 nM RA
1 uM
1 uM RA
2 uM
2 uM RA
5 uM
C
CD38 in WT HL60
24 h
48 h
1 uM Akti
5 uM Akti
10 uM Akti
D
CD11b in WT HL60
E
48 h G1/G0 arrest in WT HL60
*
F
CD38 and CD11b in HL60
CD38
WT
WT Akti
R38+
R38+ Akti
R38-
R38- Akti
G
G1/G0 arrest in HL60
H
CD38 and CD11b at 48 h
PP2 RA
1 uM wort
1 uM wort PP2 RA
I
G1/G0 arrest at 48 h
J
CD38 and CD11b at 48 h
K
G1/G0 arrest at 48 h
***