

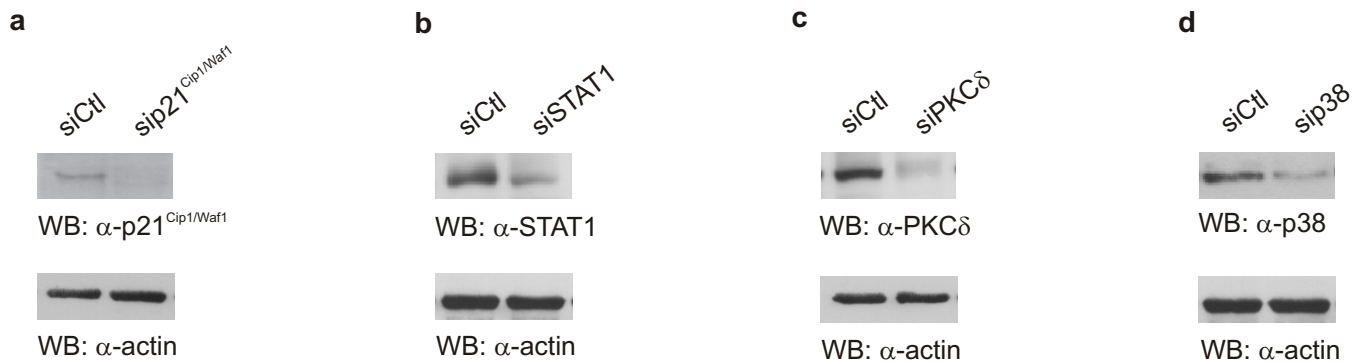
Supplementary Material

IL-12-dependent innate immunity arrests endothelial cells in G0-G1 phase by a p21<sup>Cip1/Waf1</sup>-mediated mechanism

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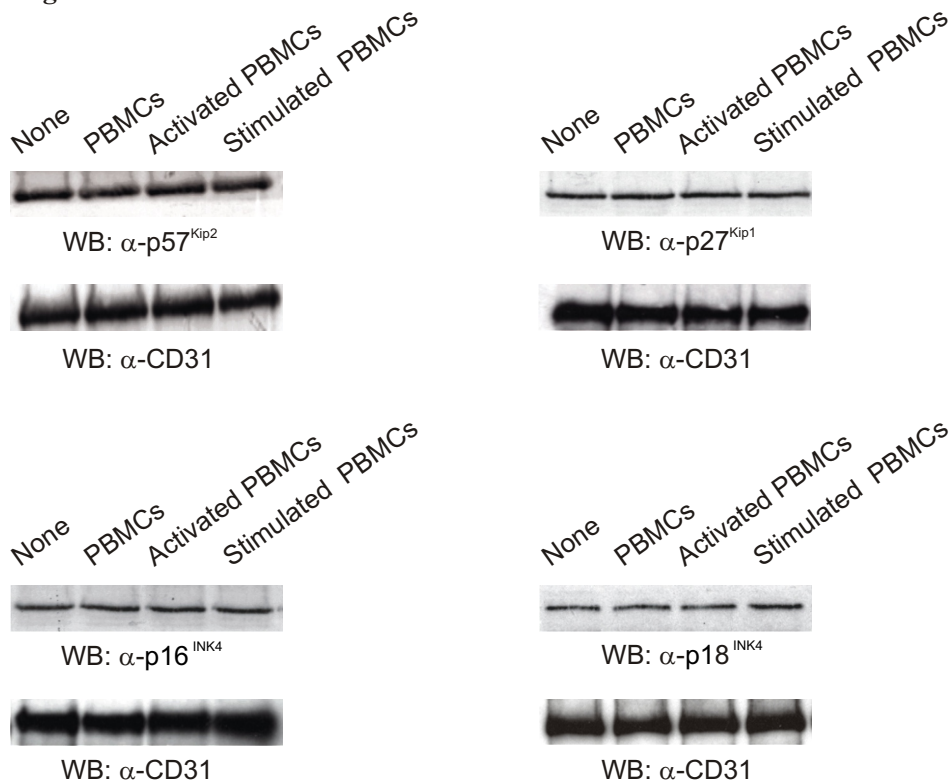
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Figure S1



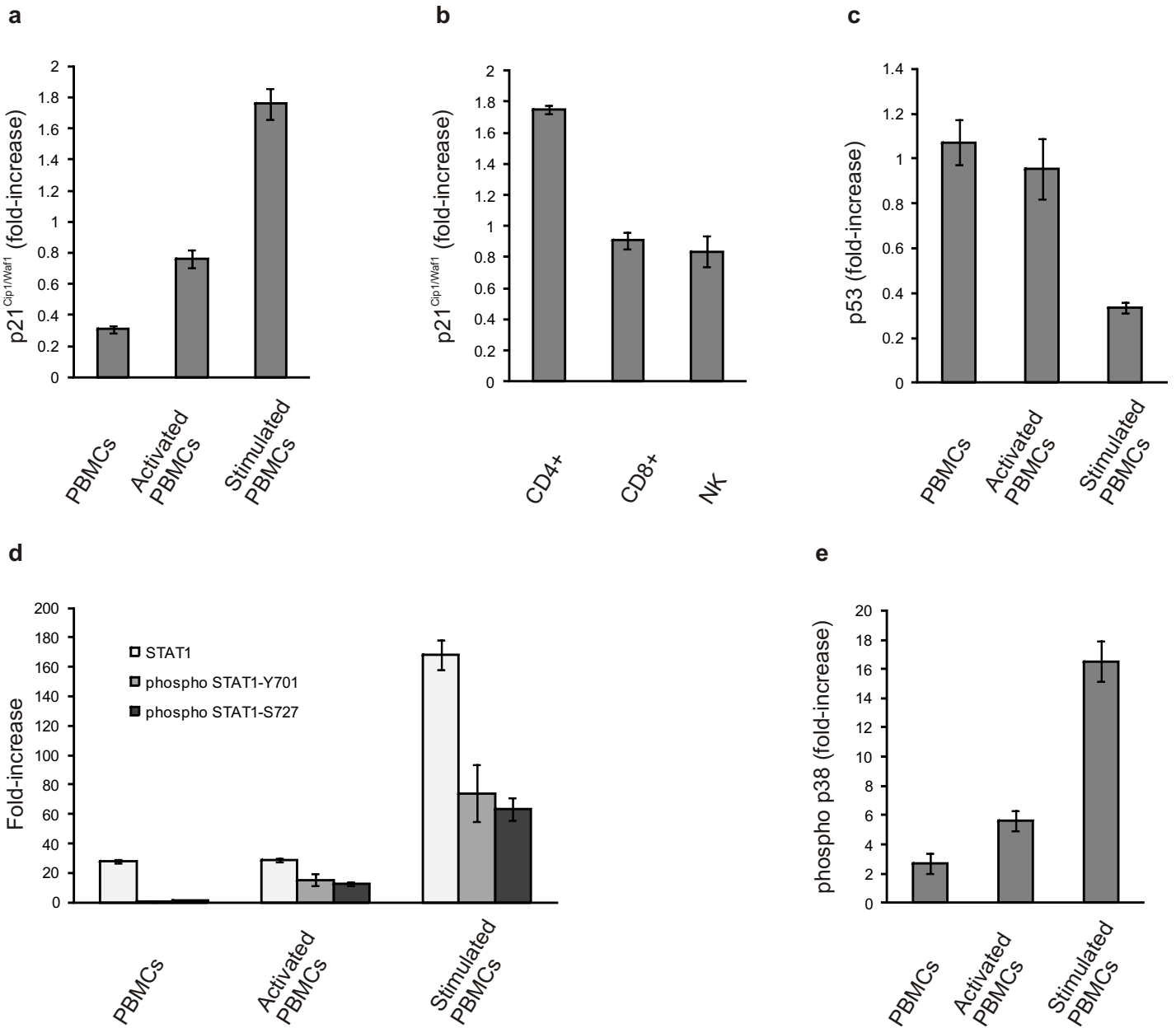
**Fig. S1** Effect of siRNA targeting p21<sup>Cip1/Waf1</sup> (a), STAT1 (b), PKCδ (c) and p38 (d) in ECs. Human ECs were transfected twice with either the specific oligonucleotides or with control non-targeting siRNA. Western blot analysis was performed 24 h after the second transfection. The data shown are exemplificative of at least five experiments

Figure S2



**Fig. S2** Expression of CDK inhibitors in human ECs cultures alone or with PBMCs, activated PBMCs or stimulated PBMCs for 48 h. At the end of incubation EC lysates were divided in four aliquots, separated and immunoblotted as indicated. This picture is representative of one experiment out of three performed

**Figure S3**



**Fig. S3** Densitometric quantification of co-culture induced changes in total and/or phosphorylated protein level of p21<sup>Cip1/Waf1</sup>, p53, STAT1 and p38. Panel **a**, **b**, **c**, **d** and **e** are the densitometric analysis of the results shown in Fig. 2a, Fig. 2b, Fig. 4a, Fig. 5a and Fig. 7a, respectively. Measurement of band intensity was performed as described in Material and Methods (main text). Results are expressed as fold-increase over the basal value in control cells, after normalization for the values of CD31 (**a**, **b**, **c** and **d**) or p38 (**e**). Values shown are the means  $\pm$  SD of three independent experiments

**Table S1** Densitometric quantification of STAT1 protein level in Fig. 5b

<b>Co-culture condition</b>	<b>Cytosol<sup>a</sup></b>	<b>Nucleus<sup>b</sup></b>	<b>Whole cell lysate<sup>c</sup></b>
None	0.14 ± 0.02	0.06 ± 0.02	0.20 ± 0.02
PBMCs	0.33 ± 0.04	0.28 ± 0.04	0.61 ± 0.05
Activated PBMCs	0.86 ± 0.18	0.56 ± 0.06	0.68 ± 0.07
Stimulated PBMCs	2.50 ± 0.29	3.07 ± 0.19	2.52 ± 0.26

Measurement of band intensity was performed as described in Materials and Methods (main text) and calculated as percentage of all the band intensities in each blot. GAPDH<sup>a</sup>, nuclear matrix p84<sup>b</sup> or actin<sup>c</sup> were then used as normalizers. Values are the mean ± SD of three independent experiments

**Table S2** Densitometric quantification of PKCδ protein level in Fig. 6b

<b>Co-culture condition</b>	<b>Cytosol<sup>a</sup></b>	<b>Membrane<sup>b</sup></b>	<b>Whole cell lysate<sup>c</sup></b>
None	1.47 ± 0.01	0.21 ± 0.04	0.99 ± 0.04
Activated PBMCs	0.97 ± 0.12	0.59 ± 0.03	1.04 ± 0.07
Stimulated PBMCs	0.57 ± 0.12	2.25 ± 0.05	0.98 ± 0.08

Measurement of band intensity was performed as described in Materials and Methods (main text) and calculated as percentage of all the band intensities in each blot. GAPDH<sup>a</sup>, CD31<sup>b</sup> or actin<sup>c</sup> were then used as normalizers. Values are the mean ± SD of three independent experiments