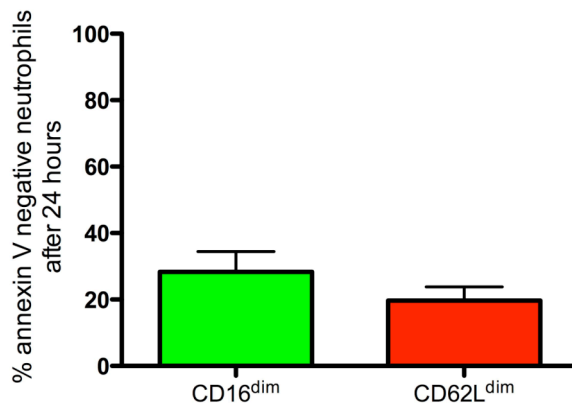


Supplementary Information

Supplemental table 1. Neutrophil phenotype after LPS administration.

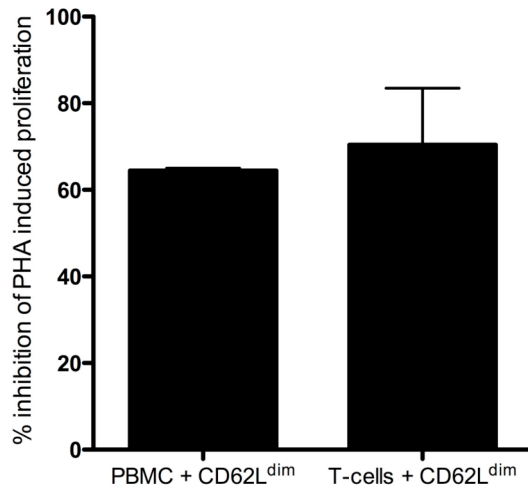
Measurements performed in erythrocyte-lysed whole blood. Leukocytes were stained with CD16, CD62L and different third stains. Neutrophil subsets were gated using CD16 and CD62L. Data are expressed as mean \pm SEM of n=5.

<i>Antigen</i>	<i>CD16^{dim}/ CD62L^{bright}</i>	<i>CD16^{bright}/ CD62L^{bright}</i>	<i>CD16^{bright}/ CD62L^{dim}</i>	<i>Clone</i>	<i>Antibody supplier</i>
CD11b	269.5 \pm 35.6	364.1 \pm 43.2	848.1 \pm 179.4	2LPM19c	DAKO
CD11c	33.6 \pm 10.0	83.4 \pm 23.0	161.1 \pm 3804	s-HCL-3	BD
CD29	43.0 \pm 5.8	54.0 \pm 7.8	60.0 \pm 8.9	4B7R	Serotec
CD49d	1.9 \pm 0.1	1.5 \pm 0.1	3.9 \pm 0.2	9F10	eBioscience
CD32	99.9 \pm 6.0	185.2 \pm 14.2	246.8 \pm 17.7	FL18.26	BD
CD64	16.8 \pm 2.3	13.8 \pm 4.3	12.0 \pm 0.9	10.1	Serotec
CD89	14.1 \pm 3.2	11.2 \pm 2.9	24.2 \pm 4.6	A3	Santa cruz
CD33	24.6 \pm 3.9	18.0 \pm 4.1	34.5 \pm 4.1	WM53	Serotec
CD66b	190.0 \pm 28.6	118.5 \pm 18.8	212.1 \pm 23.5	80H3	Gene Tex
CD63	1.8 \pm 0.1	1.5 \pm 0.1	4.2 \pm 0.2	H5C6	BD
CD14	9.9 \pm 1.2	11.4 \pm 0.9	15.3 \pm 1.1	M5E2	BD
TLR4	8.3 \pm 0.8	9.2 \pm 0.7	11.7 \pm 2.3	HTA 125	Imgenex
CD88	382.0 \pm 169.9	424.1 \pm 182.7	377.4 \pm 165.7	P1 2/1	Serotec
CXCR1	33.7 \pm 15.5	49.1 \pm 22.2	47.7 \pm 20.5	42705	R&D systems
CXCR2	18.8 \pm 4.7	36.8 \pm 8.7	39.4 \pm 10.3	48311	R&D systems
CD54	10.7 \pm 1.6	14.9 \pm 3.4	20.0 \pm 5.4	MEM-111	Invitrogen
CD35	25.4 \pm 3.2	83.0 \pm 22.4	201.0 \pm 21.2	E11	BD
CD95	36.5 \pm 1.1	37.2 \pm 1.1	35.2 \pm 0.9	DX2	DAKO
CD83	2.2 \pm 0.5	2.0 \pm 0.6	5.6 \pm 1.5	HB15a	Santa cruz
CD45	5.5 \pm 0.8	8.8 \pm 1.9	36.4 \pm 2.9	2D1	R&D systems

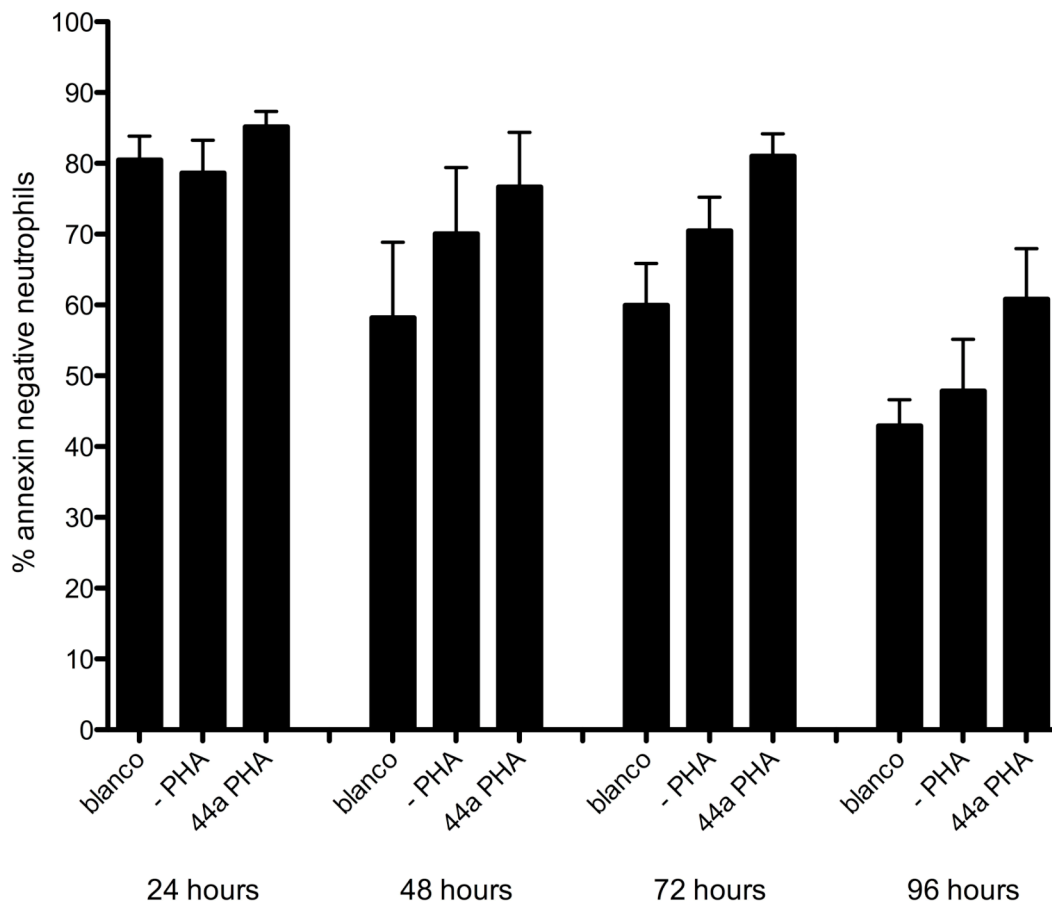


Supplemental figure 1. Neutrophil apoptosis after 24 hours in coculture.

Neutrophils were added to PBMCs in a 2:1 ratio. Culture conditions were as described in the Material and methods. After 24 hours co-culture cells were stained with CD3 FITC, Annexin V PE, 7-AAD and CD16 Alexa647. Neutrophils were gated on forward and sideward scatter and CD16. Survival is defined as the percentage Annexin, 7-AAD negative cells. Data are expressed as mean \pm SEM of n=6.

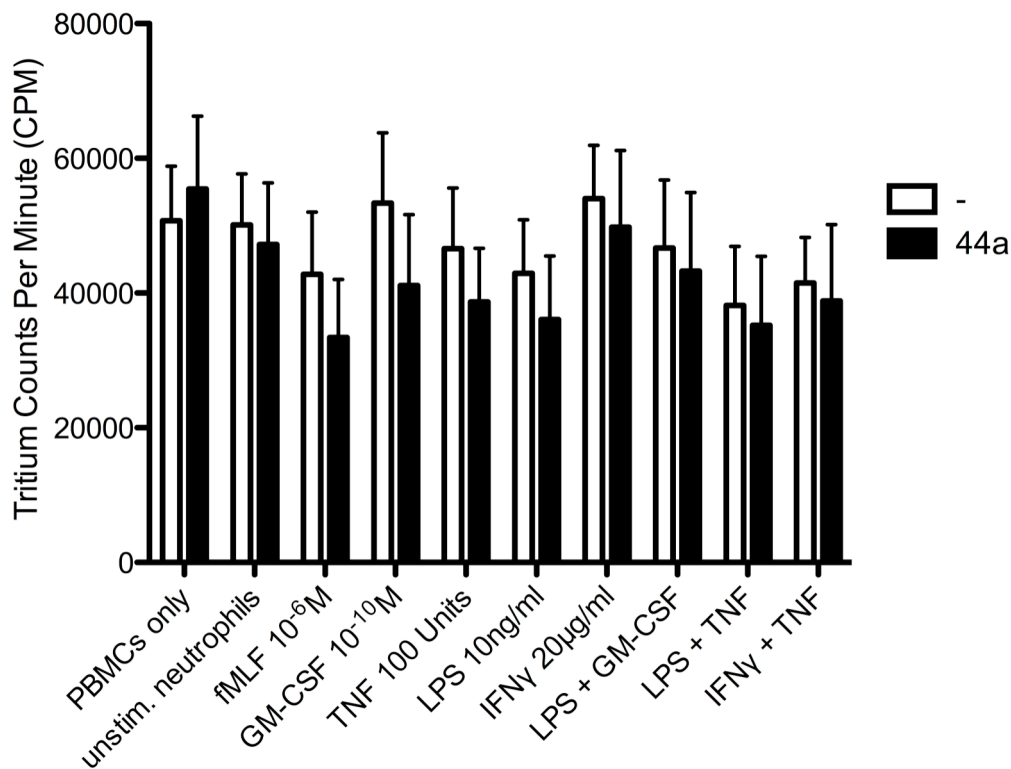


Supplemental figure 2. Inhibition of proliferation after monocyte depletion. Monocytes were depleted by magnetic beads coupled to CD14. A negative selection was performed, which resulted in >95% depletion of monocytes (not shown). Proliferation of T-cells was induced by PHA (10 μ g/ml). CD62L^{dim} neutrophils were added in a 2:1 ratio. Data are expressed as mean \pm SEM of n=3.



Supplemental figure 4. Neutrophil survival in whole blood suppression assay.

Erythrocytes were lysed and total leukocytes were incubated with or without Mac-1 blocking antibody 44a. They were stimulated with PHA. After four days the leukocytes were stained for CD3, CD16, Annexin V, and 7-AAD, neutrophils were gated using forward sideward scatter and CD16. Neutrophil survival was determined as Annexin V negative CD16 positive cells. Data are expressed as mean \pm SEM of n=6.



Supplemental figure 5. T-cell proliferation with in vitro stimulated neutrophils.

Neutrophils of healthy controls were incubated with stimuli for 1 hour at 37°C. After stimulation these neutrophils were added to PBMCs, with or without Mac-1 blocking antibody 44a, for a T-cell proliferation assay. Assays were performed as described in the material and methods. Data are expressed as mean \pm SEM of n=6.

Supplemental movie 1. Time lapse movie of neutrophil- lymphocyte interactions.

CD62L^{dim} sorted neutrophils were stained with CD16 FITC were added in a 2:1 ratio to unlabeled PBMCs stimulated with PHA (10 μ g/ml) were incubated in culture medium containing Amplex Red (20mM) and HRP (200 U/ml). Images were acquired using a Zeiss LSM510 Meta microscope. One representative example is shown of 4 independent experiments.