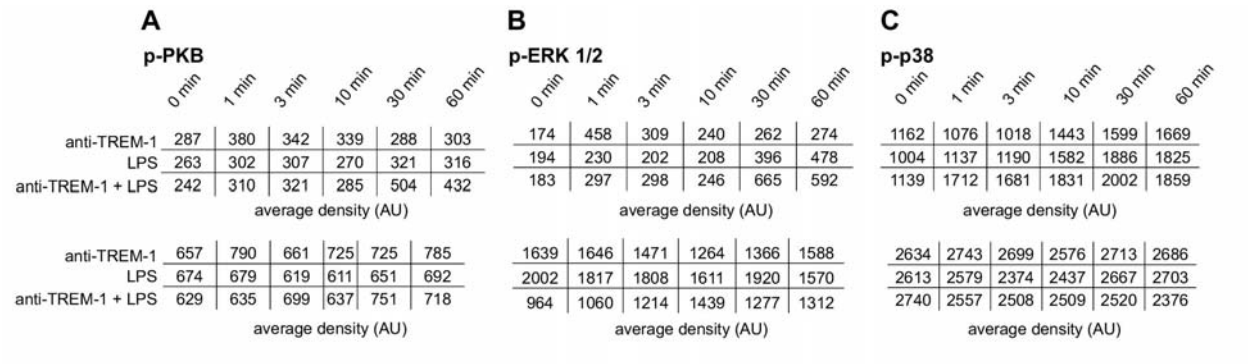


A

p-PKB

	0 min	1 min	3 min	10 min	30 min	60 min
anti-TREM-1	287	380	342	339	288	303
LPS	263	302	307	270	321	316
anti-TREM-1 + LPS	242	310	321	285	504	432

average density (AU)

anti-TREM-1	657	790	661	725	725	785
LPS	674	679	619	611	651	692
anti-TREM-1 + LPS	629	635	699	637	751	718

average density (AU)

B

p-ERK 1/2

0 min	1 min	3 min	10 min	30 min	60 min
174	458	309	240	262	274
194	230	202	208	396	478
183	297	298	246	665	592

average density (AU)

1639	1646	1471	1264	1366	1588
2002	1817	1808	1611	1920	1570
964	1060	1214	1439	1277	1312

average density (AU)

C

p-p38

0 min	1 min	3 min	10 min	30 min	60 min
1162	1076	1018	1443	1599	1669
1004	1137	1190	1582	1886	1825
1139	1712	1681	1831	2002	1859

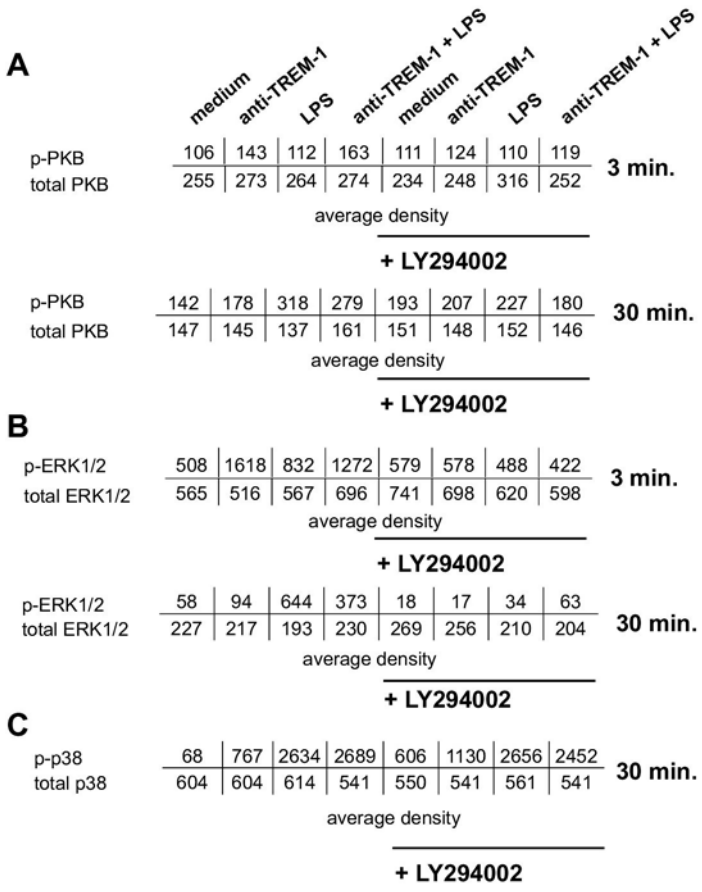
average density (AU)

2634	2743	2699	2576	2713	2686
2613	2579	2374	2437	2667	2703
2740	2557	2508	2509	2520	2376

average density (AU)

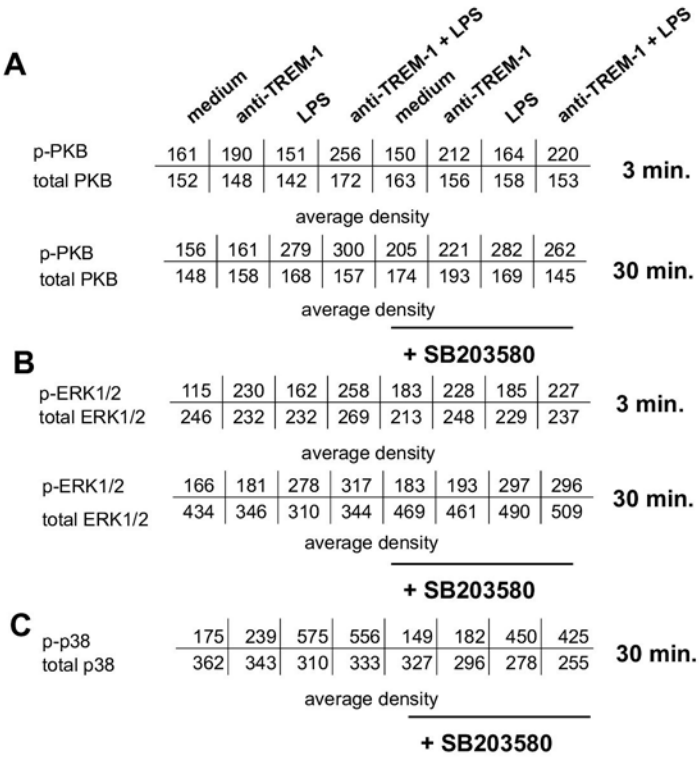
Online supplemental Table S1. Densitometric analysis of PKB, ERK and p38 phosphorylation upon TREM1 and TLR4 ligation. PMN ($2x10^6$/ml) were stimulated with anti-TREM-1 (10 µg/ml and crosslink with goat anti-mouse $F(ab)_2$, 20 µg/ml), LPS (100 ng/ml) or both for the indicated time periods from 0 to 60 min.. Whole cell lysates were prepared and separated by 10 % SDS-PAGE and subsequently analyzed by western blot against total PKB (A), total ERK1/2 (B) and total p38 (C) as protein loading controls. The primary data are depicted in Fig. 2. The blots were subjected to a densitometry analysis as described in the Experimental procedures section. The numbers represent the average density (in arbitrary units) detected by the analysis software.

Table S4: Densitometric analysis of PKB, ERK and p38 phosphorylation upon TREM1 and TLR4 ligation with the PI3K inhibitor LY294002



PMN ($2x10^6$/ml) were stimulated as described in Fig. 2 and analyzed for the phosphorylation of PKB (A), ERK1/2 (B) and p38 (C) in the absence or presence of the PI3K inhibitor LY294002 (5 µM; added 30 min. before stimulation) after stimulation for the indicated time periods. The respective protein loading controls in the presence or absence of LY294002 (5 µM) blotted against total PKB, total ERK1/2 and total p38 showed no differences among the differently activated cells (lower panels). The primary data are depicted in Fig. 3. The blots were subjected to a densitometry analysis as described in the Experimental procedures section. The numbers represent the average density (in arbitrary units) detected by the analysis software.

Table S5: Densitometric analysis of PKB, ERK and p38 phosphorylation upon TREM1 and TLR4 ligation with the p38 inhibitor SB203580



PMN ($2x10^6$/ml) were stimulated as described in Fig. 2 and analyzed for the phosphorylation of PKB (A), ERK1/2 (middle panel) and p38 (lower panel) in the absence or presence of the p38 inhibitor SB203580 (20 nM; added 30 min. before stimulation) after stimulation for the indicated time periods. The respective protein loading controls in the presence or absence of SB203580 (20 nM) blotted against total PKB, total ERK1/2 and total p38 showed no differences among the differently activated cells (lower panels). The primary data are depicted in Fig. 4. The blots were subjected to a densitometry analysis as described in the Experimental procedures section. The numbers represent the average density (in arbitrary units) detected by the analysis software.

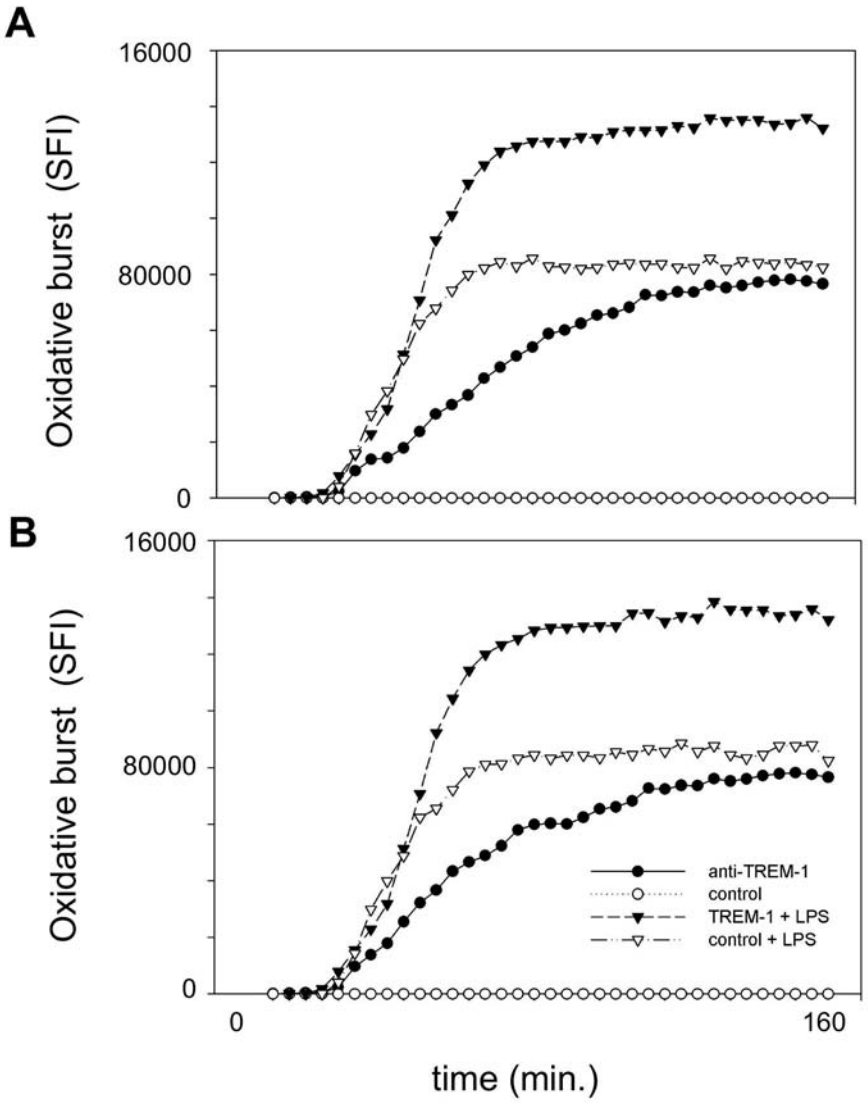
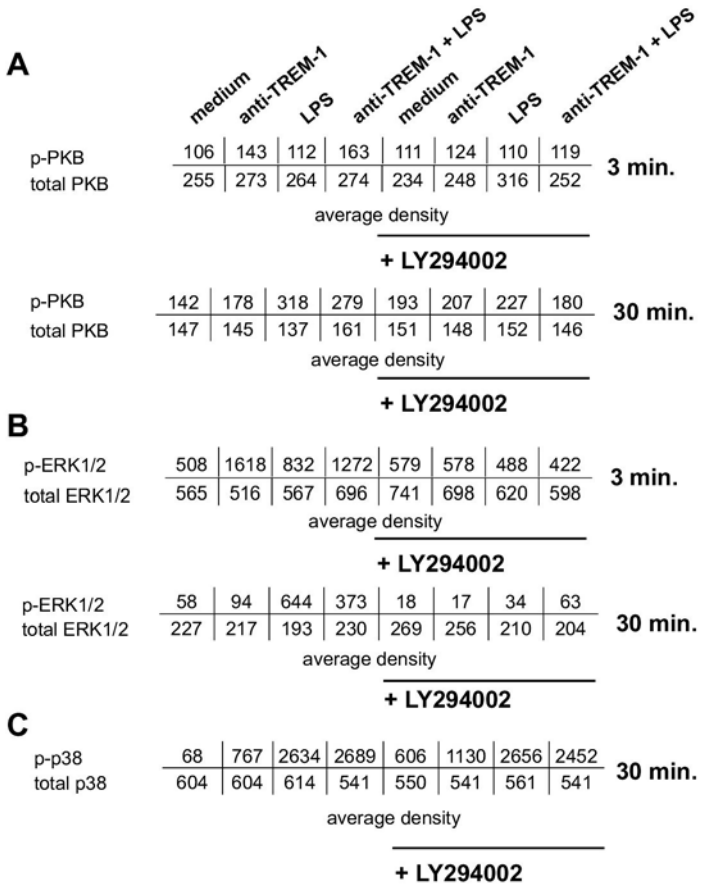


Fig. S6

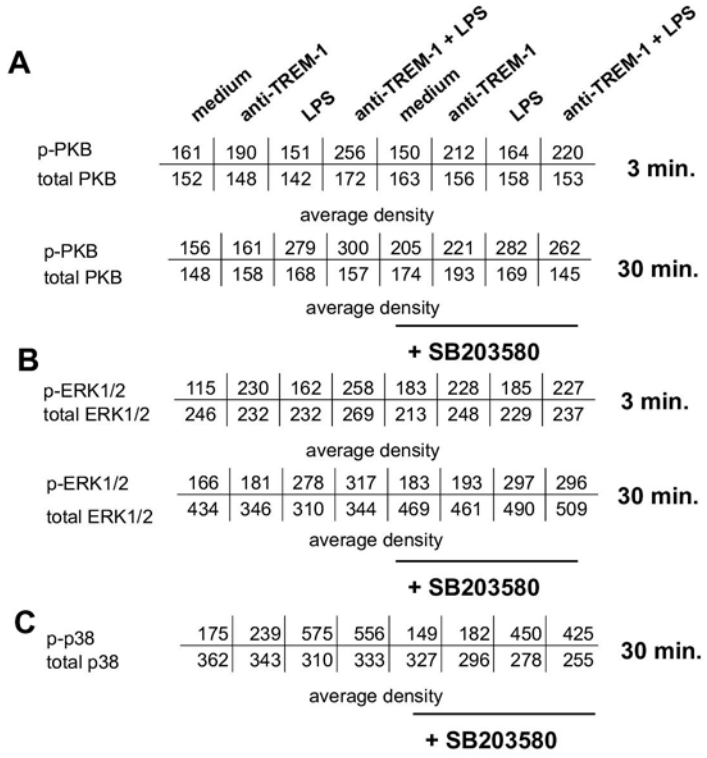
TREM-1 and TLR4 induced oxidative burst after densitiy or magnetic bead purification.

The oxidative burst activity of human PMN ($2x10^5$ per well) was assessed upon stimulation with anti-TREM-1 (10 µg/ml, filled circles), control mAb (control, 10 µg/ml, open circles), control with LPS (100 ng/ml, open triangles) or anti-TREM-1 with LPS (filled triangles). The x axis indicates time (min.) after ligation; y axis indicates the oxidative burst activity as specific fluorescence index [SFI] calculated as described in *Materials and Methods*. (A) Oxidative burst activity by PMN purified by density gradient centrifugation. (B) Oxidative burst activity by PMN purified by magnetic beads.



Online supplemental Table S2. Densitometric analysis of PKB, ERK and p38 phosphorylation upon TREM1 and TLR4 ligation with the PI3K inhibitor LY294002.

PMN ($2x10^6$/ml) were stimulated as described in Fig. 2 and analyzed for the phosphorylation of PKB (A), ERK1/2 (B) and p38 (C) in the absence or presence of the PI3K inhibitor LY294002 (5 µM; added 30 min. before stimulation) after stimulation for the indicated time periods. The respective protein loading controls in the presence or absence of LY294002 (5 µM) blotted against total PKB, total ERK1/2 and total p38 showed no differences among the differently activated cells (lower panels). The primary data are depicted in Fig. 3. The blots were subjected to a densitometry analysis as described in the Experimental procedures section. The numbers represent the average density (in arbitrary units) detected by the analysis software.



Online supplemental Table S3. Densitometric analysis of PKB, ERK and p38 phosphorylation upon TREM1 and TLR4 ligation with the p38 inhibitor SB203580.

PMN ($2x10^6$/ml) were stimulated as described in Fig. 2 and analyzed for the phosphorylation of PKB (A), ERK1/2 (middle panel) and p38 (lower panel) in the absence or presence of the p38 inhibitor SB203580 (20 nM; added 30 min. before stimulation) after stimulation for the indicated time periods. The respective protein loading controls in the presence or absence of SB203580 (20 nM) blotted against total PKB, total ERK1/2 and total p38 showed no differences among the differently activated cells (lower panels). The primary data are depicted in Fig. 4. The blots were subjected to a densitometry analysis as described in the Experimental procedures section. The numbers represent the average density (in arbitrary units) detected by the analysis software.