

1 **Figure S1. Effects of serum from naïve and infected mice on amastigote uptake.** Serum
2 samples were collected from naïve mice or mice infected with *L. amazonensis* for 12 weeks.
3 Serum samples were heat-inactivated for 10 min at 56°C prior to use. CFSE-labeled amastigotes
4 were opsonized in serum at indicated concentrations for 20 min and then co-cultured with
5 neutrophils. At 4 h, cells were stained for flow cytometric analysis and neutrophil carriage of
6 amastigotes was assessed based on CFSE positivity. Neutrophil co-culture with unopsonized
7 amastigotes routinely resulted in approximately 5-15% of neutrophils internalizing parasite (See
8 Fig. 1A).

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10 **Figure S2. Effect of fMLP and amastigotes on NADPH oxidase-derived ROS production in**
11 **neutrophils.** A) Neutrophils were co-cultured with live axenic amastigotes or amastigote lysates
12 (Lys). After 4h, cells were stained with dihydrorhodamine 123, and neutrophil oxidative burst
13 was analyzed by measuring the mean fluorescence intensity (MFI) of the fluorescent oxidation
14 product rhodamine 123 (Rho 123). Shown are the representative results of 1 of 2 independent
15 repeats. B) MFI of Rho 123 in neutrophils treated with fMLP (1 μM), co-cultured with
16 amastigotes for 4 h, or both. Data were pooled from at least 3 independent repeats and shown as
17 the means ± the standard errors. *** ($p < 0.001$) indicates statistically significant differences
18 between groups. C) MFI of Rho 123 in WT and gp47^{-/-} neutrophils treated with fMLP or
19 amastigotes. Shown are representative results from 1 of 2 independent repeats.

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21 **Figure S3. Promastigote-induced neutrophil death.** A) Neutrophils were co-cultured with
22 metacyclic promastigotes (Pm) or axenic amastigotes (AxAm) for 18 h followed by assessment
23 of neutrophil apoptosis. B) Counts of neutrophils remaining after 18 h of culture in medium

24 alone (Med), or after 18 h of co-culture with metacyclic promastigotes or axenic amastigotes.
25 Data are pooled from 2 independent repeats and shown as the means \pm the standard errors. ** (p
26 < 0.01) and *** ($p < 0.001$) indicate statistically significant differences between the groups. NS,
27 not significant.

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29 **Figure S4. Killing of lesion-derived amastigotes by C57BL/6 and BALB/c neutrophils. A)**

30 Clearance of axenic amastigotes and lesion-derived amastigotes by C57BL/6 neutrophils and
31 BALB/c neutrophils. Data are pooled from 2 independent repeats and shown as the means \pm the
32 standard errors.