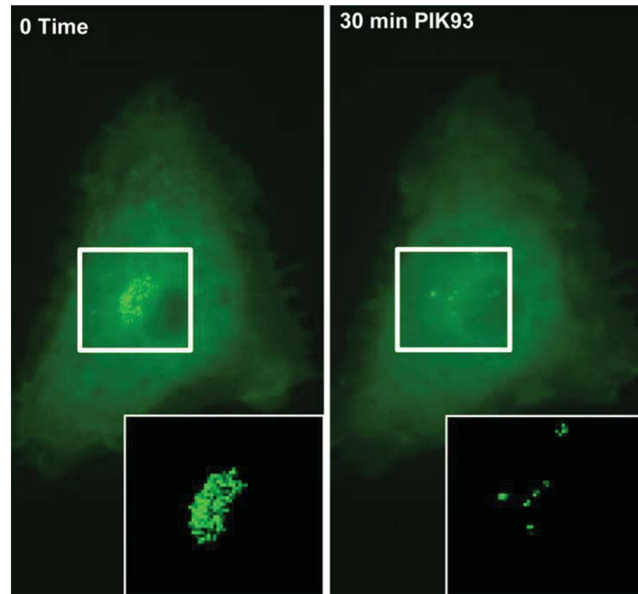

Supplemental Figure 1. PIK93 removes PKC-ε from the Golgi. Z stacks of BMDM expressing PKC-ε-GFP were taken before and after treatment with 0.1 μM PIK93 (30 min, 37°C). The Z stacks were reconstructed and, after thresholding, a 3-D surface rendering of the perinuclear region (inset) was generated and overlayed on a single Z slice. PIK93 treatment substantively reduces Golgi-associated PKC-ε.

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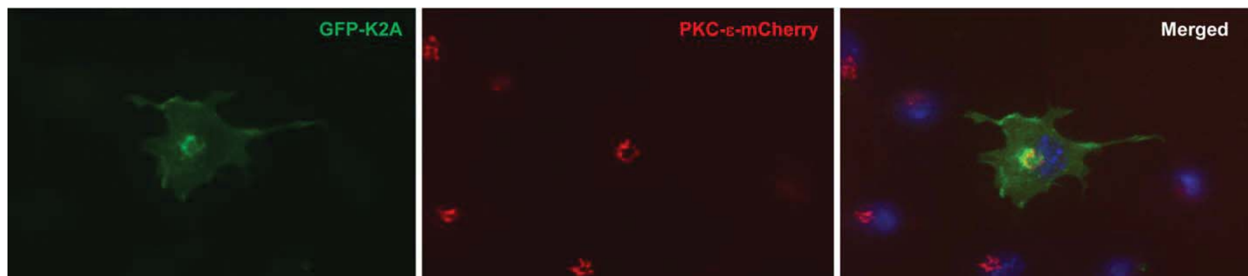

Supplemental Figure 2. Perinuclear PKC-ε co-localizes with the Golgi marker, GM130. RAW 264.7 cells expressing PKC-ε-GFP were fixed and stained for Golgi marker, GM130. A Pearson's correlation of 0.64 (25 cells from 4 independent experiments) corresponds to significant co-localization.

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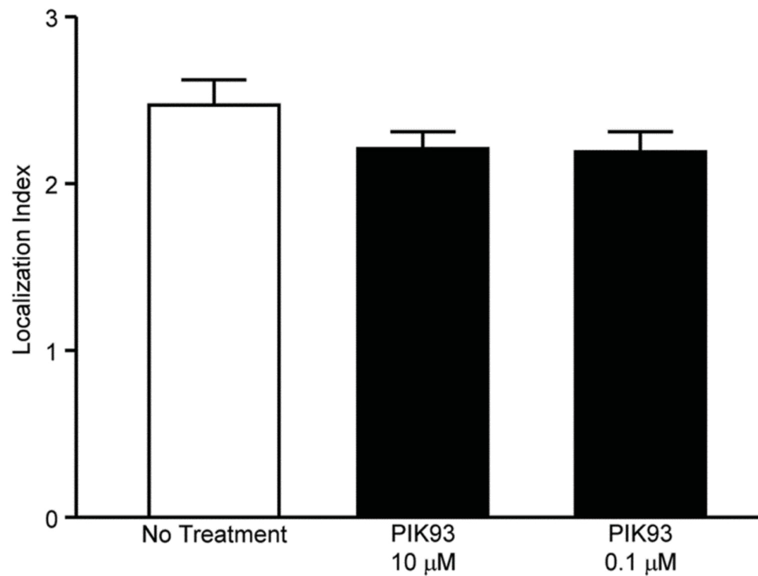

Supplemental Figure 3. PIK93 does not alter PKC-ε concentration at nascent phagosomes. BMDM expressing PKC-ε-GFP were treated with the indicated concentrations of PIK93 (30 min, 37°C) and followed in real time during phagocytosis of IgG-opsonized beads. PKC-ε concentration at the nascent phagosome "flash" (i.e., when the pixel intensity was the highest) was quantified as in Methods. Neither concentration of PIK93 significantly altered the nascent phagosome-associated PKC-ε intensity. Data are presented as mean ± SEM. Significance was tested by ANOVA with Bonferroni's correction, no significance was found. 41-45 events compiled from 3 independent experiments. These are the same cells from which the phagocytic cup intensity was calculated (Fig 4B).

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