FIG. S1

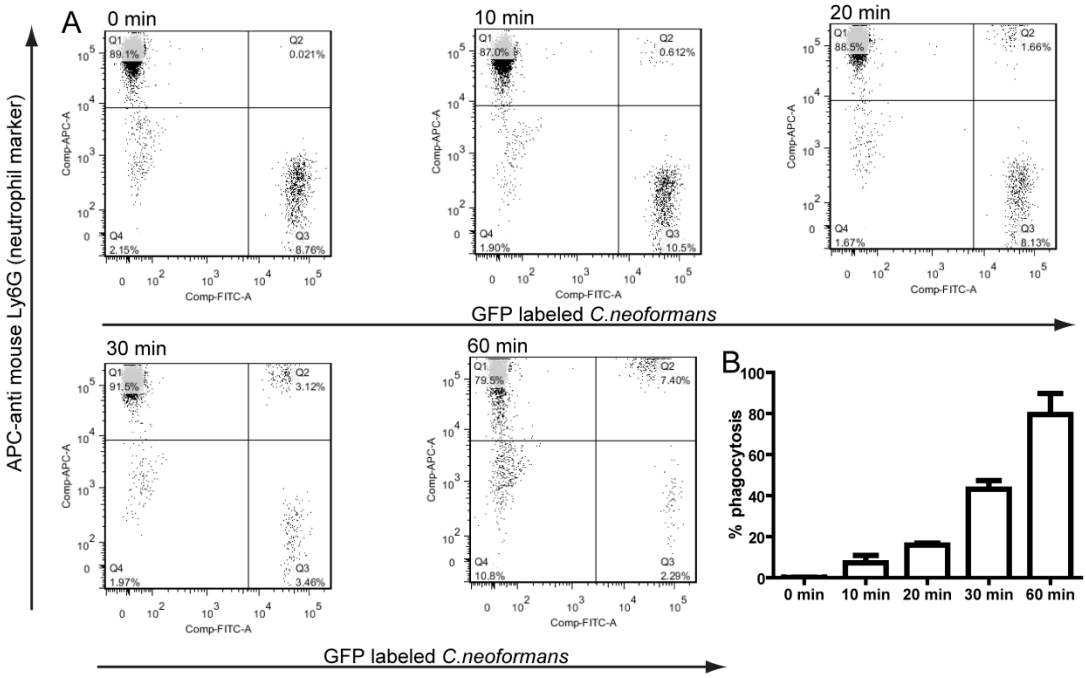

FIG. S1. *C. neoformans* is internalized by or associated with neutrophils in the presence of wild-type mouse serum. (A) Representative dot plots showing the percentage of the yeast cells ingested by or associated with neutrophils (double positive) with time in the presence of mouse serum. (B) A quantification of the yeast cells ingested by or associated with neutrophils with time in the presence of mouse serum. Data are presented as mean ± SEM. Data are representative of 3 independent experiments.

FIG. S2

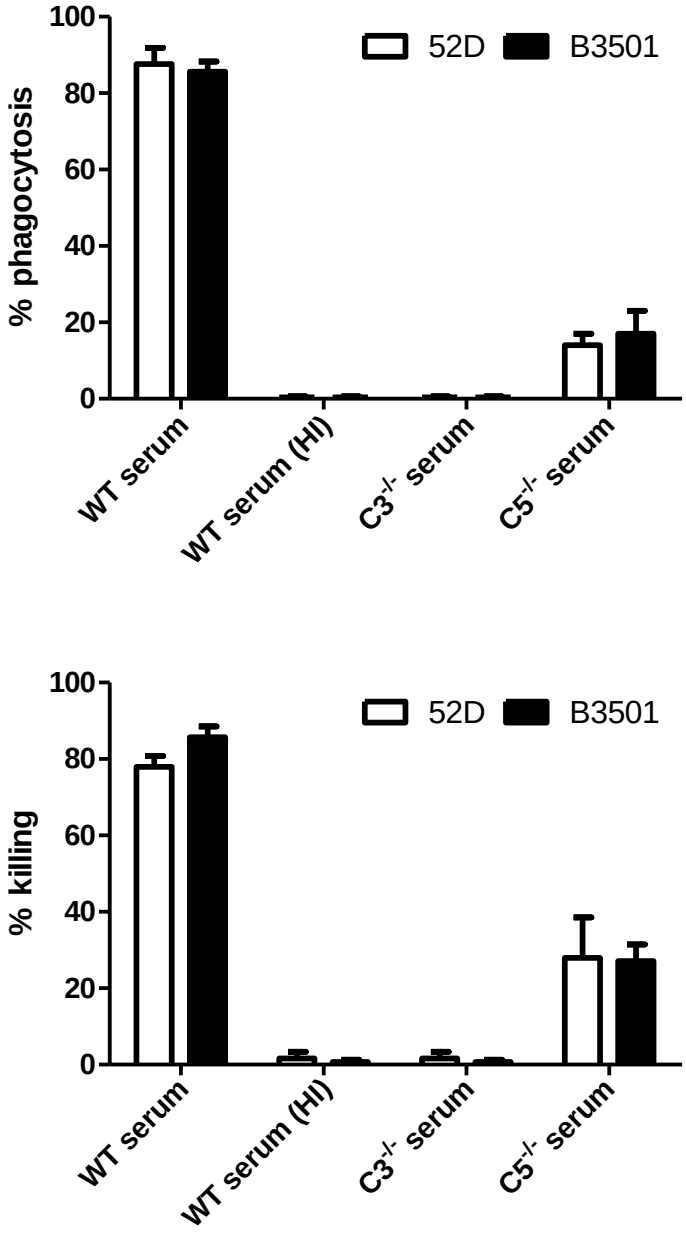

FIG. S2. Percentage of *C. neoformans* 52D (ATCC 24067) and B3501 (ATCC 34873) ingested by neutrophils in the presence of wild-type, $C3^{-/-}$, $C5^{-/-}$ mouse serum or heat-inactivated wild-type mouse serum (upper). Percentage of the yeast cells killed by neutrophils in the presence of wild-type, $C3^{-/-}$, $C5^{-/-}$ mouse serum, or heat-inactivated wild-type mouse serum (lower). Data are presented as mean ± SEM.

FIG. S3

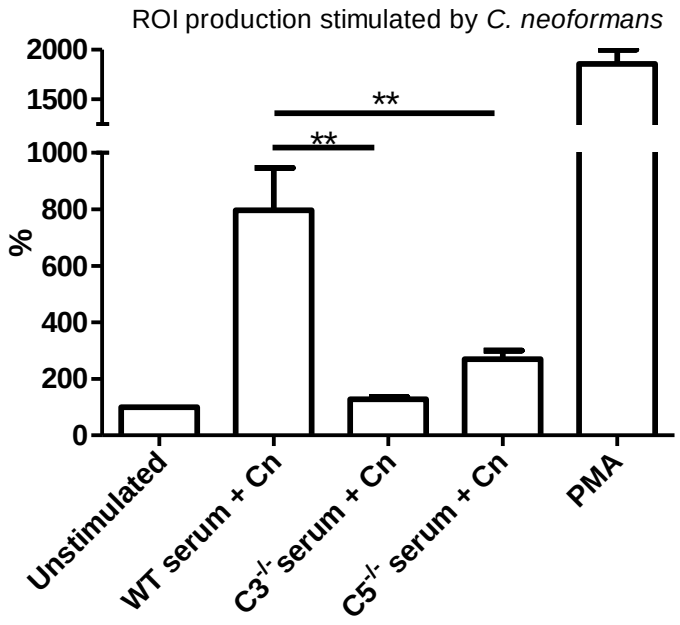

FIG. S3. ROI production of neutrophils stimulated by *C. neoformans* (Cn) is significantly reduced in the presence of $C3^{-/-}$ or $C5^{-/-}$ serum compared to wild-type serum. Data are presented as mean ± SEM. Data are representative of 3 independent experiments. **p<0.01. The oxidative burst of neutrophils was evaluated by quantitative determination of the intracellular generation of reactive oxygen intermediate (ROI). ROI converted fluorogenic substrate dihydrorhodamine (DHR) 123 (Life technology) to rhodamine 123 which was measured by flow cytometry. Neutrophils ($2x10^5$) were suspended in 100 μl RPMI-1640 supplemented with 40% fresh mouse serum and 0.5 μg/ml DHR 123, and then incubated with *C. neoformans* ($2x10^4$) at 37^oC for 15 min. For each assay, neutrophils stimulated with PMA were served as a positive control, and unstimulated neutrophils as a negative control.

FIG. S4

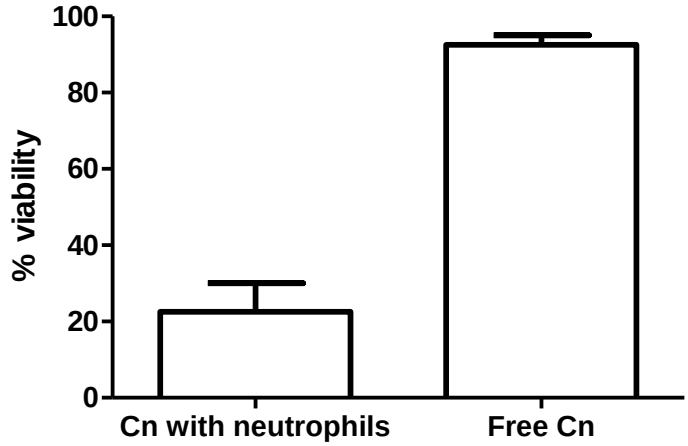

FIG. S4. Killing of *C. neoformans* (Cn) is associated with phagocytosis/binding. Using Flow cytometry, we sorted *C. neoformans* associated with neutrophils and free *C. neoformans* after four hour incubation with mouse neutrophils in the presence of wild-type mouse serum. After serial dilution, CFU was determined and the percentage of viability of *C. neoformans* was calculated as viable yeast cells out of total yeast cells collected. Data are presented as mean ± SEM.

FIG. S5

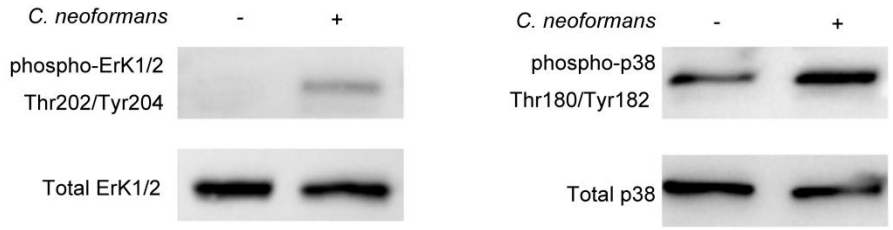

FIG. S5. Co-incubation of neutrophils with *C. neoformans* leads to activation of Erk and p38 pathways. Neutrophils ($1x10^6$) were incubated with or without *C. neoformans* ($5x10^4$) in 100 μl RPMI containing 40% normal mouse serum for 15 min at 37°C. Cells lysates were boiled for 5 min in loading buffer (250 mM Tris-HCl [pH 6.8] with 4% sodium dodecyl sulfate, 20% glycerol, 0.01% bromphenol blue, 6% β-mercaptoethanol) and loaded on 10% SDS-PAGE, electrophoresed, and transferred onto nitrocellulose membrane by semidry equipment for Western Blot. Membrane was blocked in 5% BSA/0.05% Tween 20/tris-buffered saline (TBST) 1 h at room temperature. Phosphotyrosine was detected using rabbit antibodies to phospho-p38 MAPK, phospho-ErK (CST, Beverly, MA, USA). Total protein was determined using mouse antibodies to p38 MAPK, ErK (CST, Beverly, MA, USA). Membrane were incubated with first antibody at 4°C overnight and detected with horseradish peroxidase-conjugated goat anti-mouse IgG or goat anti-rabbit IgG (SouthernBiotech, USA) diluted at 1:10,000 for 1 h at room temperature with gentle agitation and visualized using Bio-rad gel imaging system.

Supplemental Video legends

Video S1. A 3D reconstructive image showing *C. neoformans* (green) was internalized by neutrophils (red) in the presence of wild-type mouse serum.

Video S2. Neutrophils were chasing yeast cells followed by internalization of the organisms in the presence of wild-type mouse serum. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.

Video S3. A neutrophil was chasing a yeast cell in the presence of wild-type mouse serum. The time lapse was recorded at 1 frame per second and exported to video at 5 frames per second.

Video S4. Neutrophils did not chase the yeast cells in the presence of $C3^{-/-}$ mouse serum. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.

Video S5. Neutrophils did not chase the yeast cells in the presence of $C5^{-/-}$ mouse serum. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.

Video S6. Neutrophils travelled actively although they did not chase the yeast cells in

the presence of $C5^{-/-}$ mouse serum with addition of rC5a. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.

Video S7. $CD11b^{-/-}$ neutrophils were chasing the yeast cells in the presence of wild-type mouse serum. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.

Video S8. Neutrophils pretreated with DIDS were chasing the yeast cells followed by attaching the yeast cells without further phagocytosis in the presence of wild-type mouse serum. The time lapse was recorded at 4 frames per minute and exported to video at 5 frames per second.