

Supplementary Methods

Capsular polysaccharide (CPS) purification

Kp strain #20, which has a K1-CPS and has been described previously [16] was inoculated in 1L of LB broth and grown o/n at 37°C. Cells were pelleted by centrifugation and washed with PBS. Cells were resuspended to a 5% (wt/vol) in distilled water and CPS was extracted by phenol-water. CPS, present in the aqueous phase, was precipitated by adding 5 volumes of methanol plus 1% (v/v) of a saturated solution of sodium acetate in methanol and incubated for 1h at 24°C. After dissolving the pellet in water, it was dialyzed against water for 6 hours prior to freeze-drying. The lyophilized polysaccharide was dissolved in 0.8% NaCl, 0.05% NaN₃, 0.1M Tris-HCl (pH 7) to a 10 mg/ml concentration and digested with nucleases (50 mg/ml of DNase I and RNase A) twice for 24h at 37°C. Then proteinase K was added (50 mg/ml) and incubated for 1 h at 55°C and for 24 h at room temperature twice. Polysaccharides were precipitated as stated above and dissolved in water. LPS was removed by ultracentrifugation (105000 × g, 16 h, 4°C) and samples were freeze-dried. CPS was further extracted with phenol following previously published protocol [29]. CPS was further purified by a size-exclusion chromatography on a column S200HR (GE Healthcare Life Sciences) with PBS. Fractions were collected and the presence of polysaccharide was analyzed by the phenol-sulfuric acid method [30].

CPS-PA conjugation

Briefly, CPS was dissolved in NaCl 0.15 M to 10 mg/ml (1ml). At t= 0 s, 100 µl of CDAP (100 mg/ml in acetonitrile) (1 mg CDAP/mg PS) was slowly added while stirring. 30 s later 0.2 M TEA (10 µl/mg PS) was added to raise the pH. At t = 2' 30 s, 0.1M sodium borate pH 9.3 was added to adjust pH to 9 and 10 mg

of PA in 0.15 M NaCl were added. The reaction was incubated o/n at 4°C and then dialyzed against water. PS-protein conjugate product was purified using an S200HR gel filtration column (GE Healthcare Life Sciences). The A_{280nm} in the chromatogram monitored the presence of protein while the PS content was monitored by phenol-sulfuric acid method [31]. Presence of LPS was determined using Limulus Ameobocyte Lysate test and were in acceptable range.

C3c-deposition assays

Briefly, an LB o/n culture was diluted and it grew to reach mid-logarithmic phase. 5×10^7 CFUs were resuspended in 1ml of PBS + 20% Normal Human Serum + 1% BSA. PBS, 20 μ g of IgG Control, 4C5, or 1910 mAb were added and incubated for 15'. Then, cells were washed 2x with PBS + 1% BSA and divided in half. One half was incubated with 1:500 sheep anti-human C3c FITC (Bio-Rad) in PBS + 1% BSA. The other was incubated in the absence of antibody. Samples were incubated for 25 min at 4°C. A sample with bacteria incubated without serum and with secondary antibody was used as a negative control to set the threshold and fluorescence intensity. In addition 2 other binding controls were introduced by incubating bacteria with serum and PBS instead of the anti-C3c antibody and by replacing both the serum and anti-C3c antibody with PBS. All three negative controls gave similar low backgrounds. Intensity higher than 10 was considered positive for C3 binding. Quantification was performed multiplying the percentage of bacteria moving into the gate by the average fluorescence of a defined population (X-mean), to give a Fluorescence Index (FI). Results are expressed by the increment of the signal of mAb treatment with respect to PBS incubation. Data was deemed to be

parametric and analyzed with ANOVA with *post hoc* Tukey's multiple comparisons test.

NETosis experiments

Briefly, peripheral blood was collected from healthy donor volunteers following an approved protocol by the Institutional Review Board of Stony Brook University. Neutrophils were isolated over a Ficoll-Paque PLUS (GE Healthcare) and a dextrin gradient. Wild-type *Candida albicans* (strain SC5314, gift from Dr. Konopka laboratory) was grown overnight in yeast peptone dextrose medium at 37°C. Next day it was subcultured 4h in RPMI medium to induce hyphal morphology for NETosis experiments. 5×10^5 CFUs/ml (5ml) K1-*Kp* were incubated for 1h with either PBS, 1 μ g of either IgG control, 4C5 or 19A10 mAbs in Hank's balanced-salt solution plus Ca^{2+} and Mg^{2+} with 5% of normal human serum. Neutrophils were plated in a 24-well plate at 5×10^4 CFUs/well and preincubated K1-*Kp* samples and *C. albicans* were added at a MOI of 10. After 4h SYTOX (Invitrogen) was added at 500 nM and incubated for 15 min. Then NETs were visualized in at least 7 samples (20X magnification) and quantified and analyzed as described [35]. Data was analyzed with one-way ANOVA with Sidak's multiple comparisons test.

Galleria mellonella virulence assays

10^6 colony forming units/ml (CFUs/ml) (250 μ l) of K1-*Kp* were incubated with either PBS, or 25 μ g of either 4C5 or 19A10 mAbs for 1h. 20 larvae were injected with 10^4 CFUs of K1-*Kp* in 10 μ l PBS. Larvae were kept at 37°C in the dark on sterile Petri plates and survival was assessed for 3 weeks. Analysis of survival curves was performed using log-rank (Mantel-Cox) test corrected for Bonferroni's multiple comparisons test.

Macrophage killing experiments

8×10^4 of J744.16 macrophage cells were plated in a 96-well plate. Next day, 1.6×10^6 K1-Kp CFUs were added. After 30 min of incubation, extracellular bacteria were washed out, and 100 μ g/ml gentamicin was added to kill any remaining extracellular and cell-attached bacteria. Intracellular killing was based on the decrease of viable bacteria 30 min after initial co-incubation relative to time 0.

Intravital microscopy (IVM)

Mice were anesthetized by *i.p.* injection of a mixture of ketamine (120 mg/kg) and xylazine (10 mg/Kg). Tail vein was cannulated for administration of additional anesthetic and experimental reagents as required. A midline laparotomy was performed followed by removal of the skin and abdominal muscle along the costal margin to the mid-axillary line to expose the liver. Mice were placed in the right lateral position and a single liver lobe was exteriorized on a custom-made stage. Exposed tissues were moistened with saline-soaked gauze to prevent dehydration during imaging. Body temperature was maintained with an infrared heat lamp, and the liver was continuously superfused with physiological saline buffer. Intravital microscopy was performed by Examiner Z1 system (Zeiss), time-lapse video was taken at a speed of 1 frame/s. Videos were further analyzed by Zen2012 software (Zeiss). Data from 3-4 mice was analyzed with a one-way or 2-way ANOVA with *post hoc* Tukey's multiple comparisons test.