Diagnostics of Invasive Aspergillosis: From Experimental Models to Clinical Evaluation.

Scott Filler

Laura Najvar

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INVASIVE ASPERGILLOSIS ANIMAL MODELS

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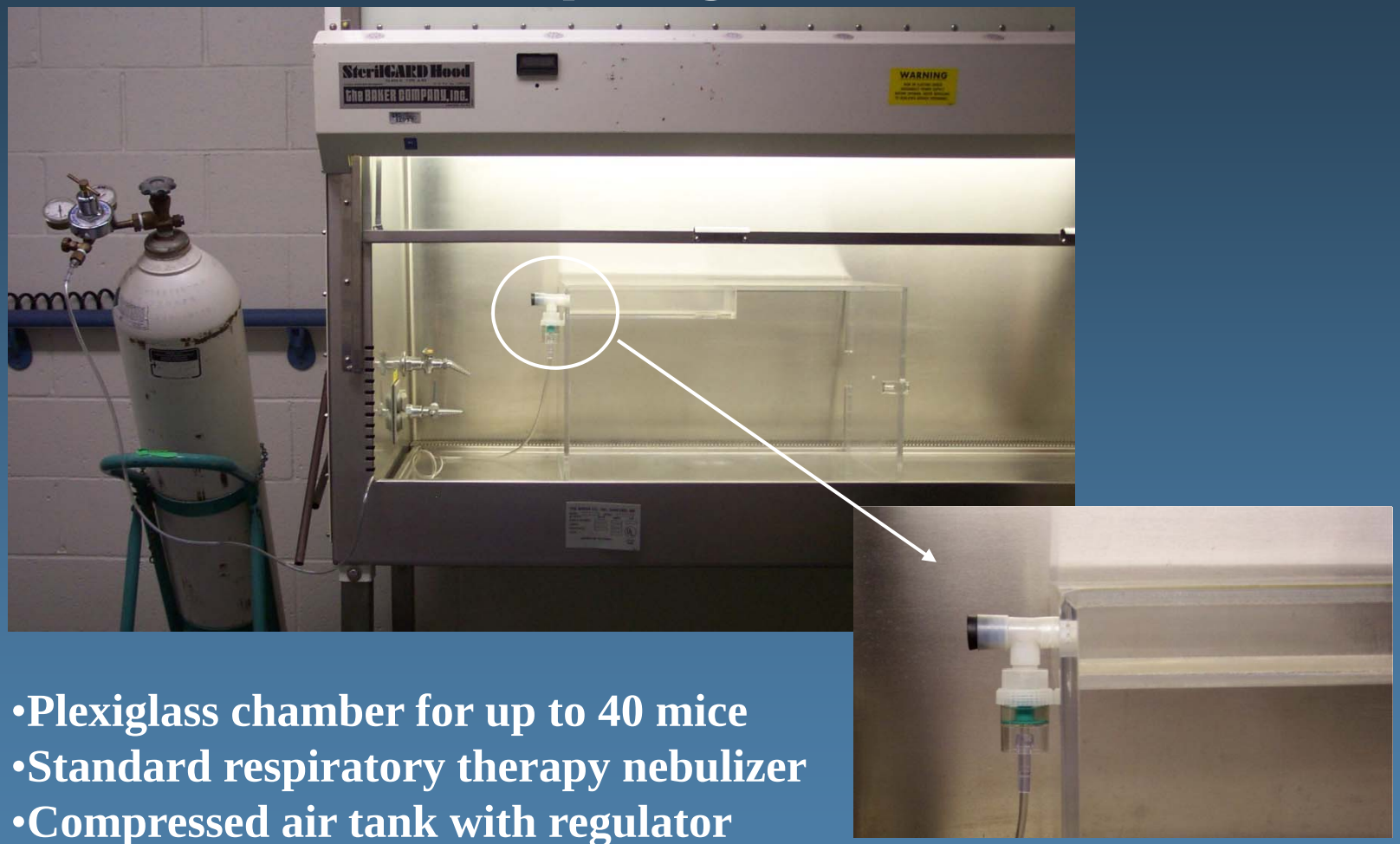
Inhalation Chamber Apparatus for Aspergillosis



- Plexiglass chamber for up to 40 mice
- Standard respiratory therapy nebulizer
- Compressed air tank with regulator

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Inhalational Models Advantages

- Relatively low inoculum reproducibly delivered to alveoli
- Recapitulates human disease
- Late mortality
  - Allows multiple time point sampling without survivor bias
- Mortality complete by 14 days

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Key Question: Model Refinement/Diagnostic Targets Non-neutropenic Model

- Infection in acrylic chamber
  - nebulized conidia of *A. fumigatus* AF293 at 10^9 conidia/ml of fluid for 1 h
  - simultaneous challenge of 50-60 mice
- Immunosuppression:
  - Neutropenic:
    - Cyclophosphamide at 250 mg/kg SC on day -2 and 200 mg/kg SC on day +3 of infection
    - Cortisone acetate at 5 mg/mouse SC on days -2 and +3 of infection
  - Non-neutropenic:
    - Cortisone acetate at 10 mg/mouse SC on days -4, -2, 0, +2, +4 of infection

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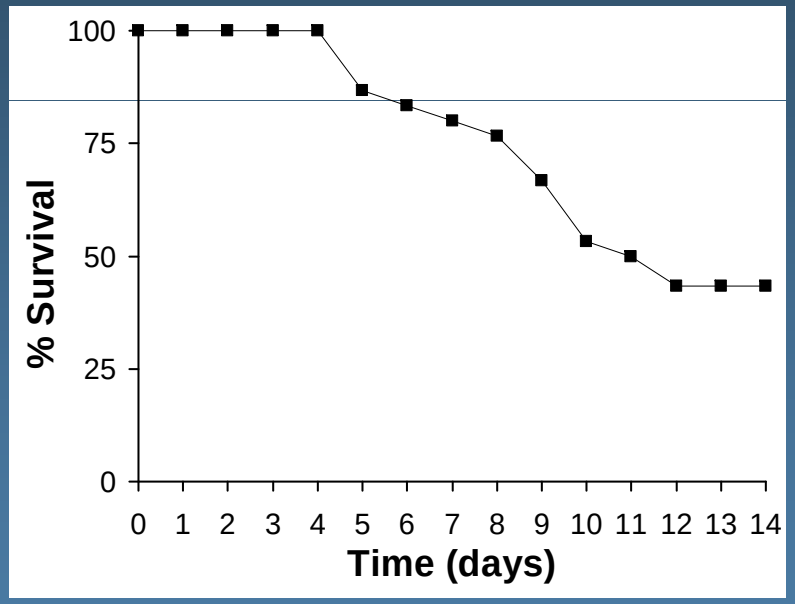
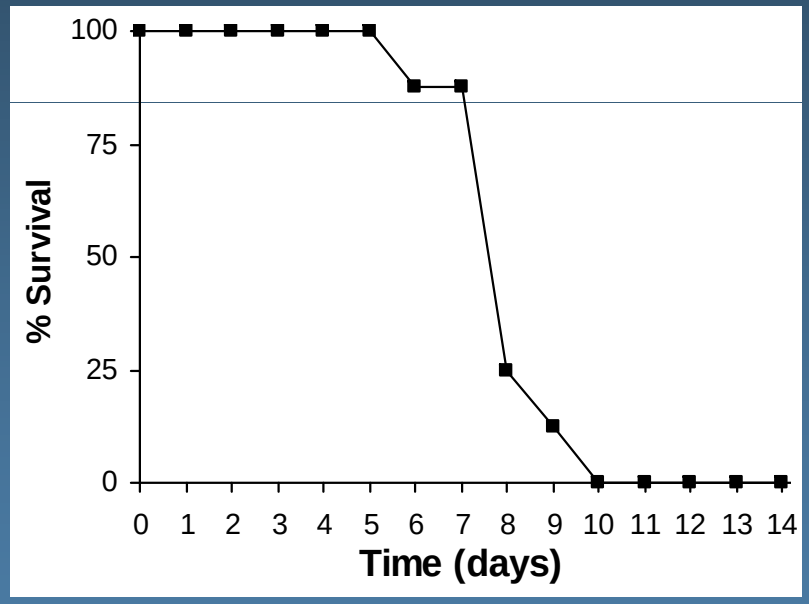
Effects of Different Types of Immunosuppression on Mouse Survival

Neutropenic
(Cortisone Acetate
+ Cyclophosphamide)



Non-neutropenic
(Cortisone Acetate)



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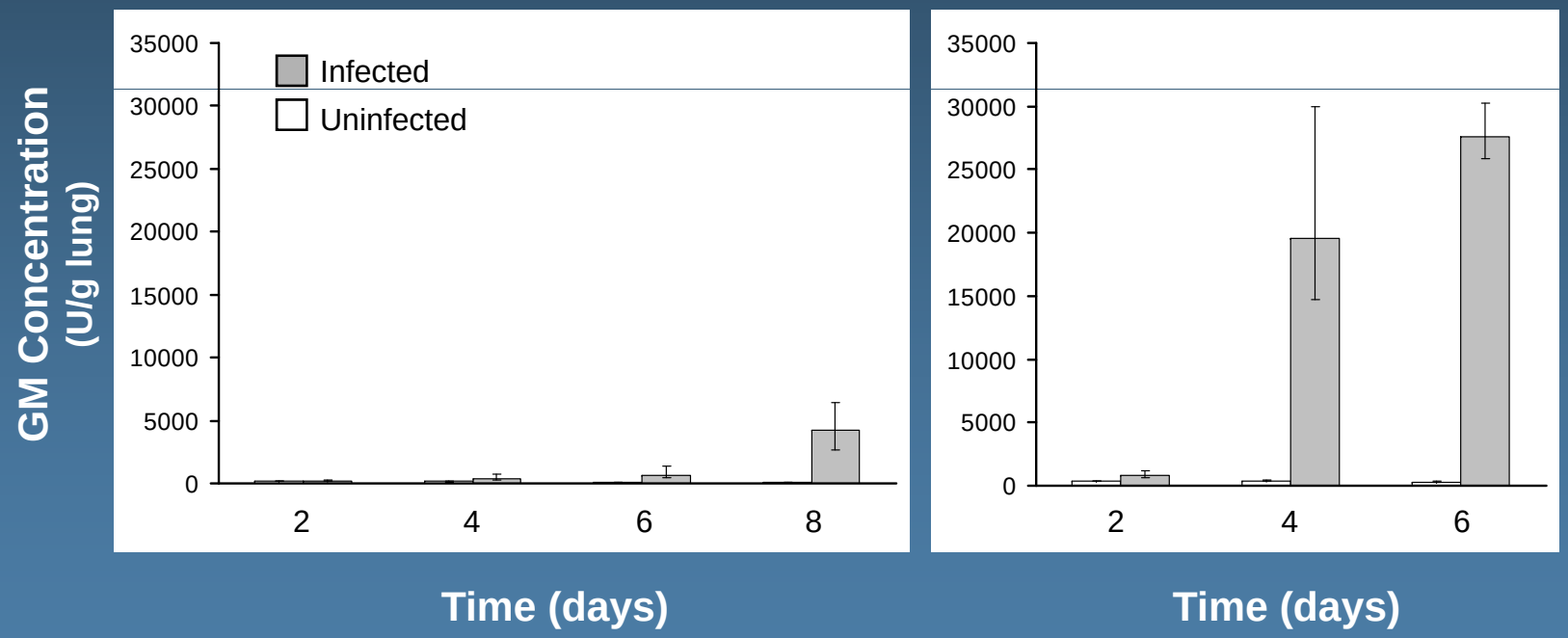
Lung GM Levels are Higher in Non-Neutropenic Mice

Neutropenic
(Cortisone Acetate
+ Cyclophosphamide)

Non-neutropenic
(Cortisone Acetate)



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Serum GM Levels are Higher in Neutropenic Mice

Neutropenic
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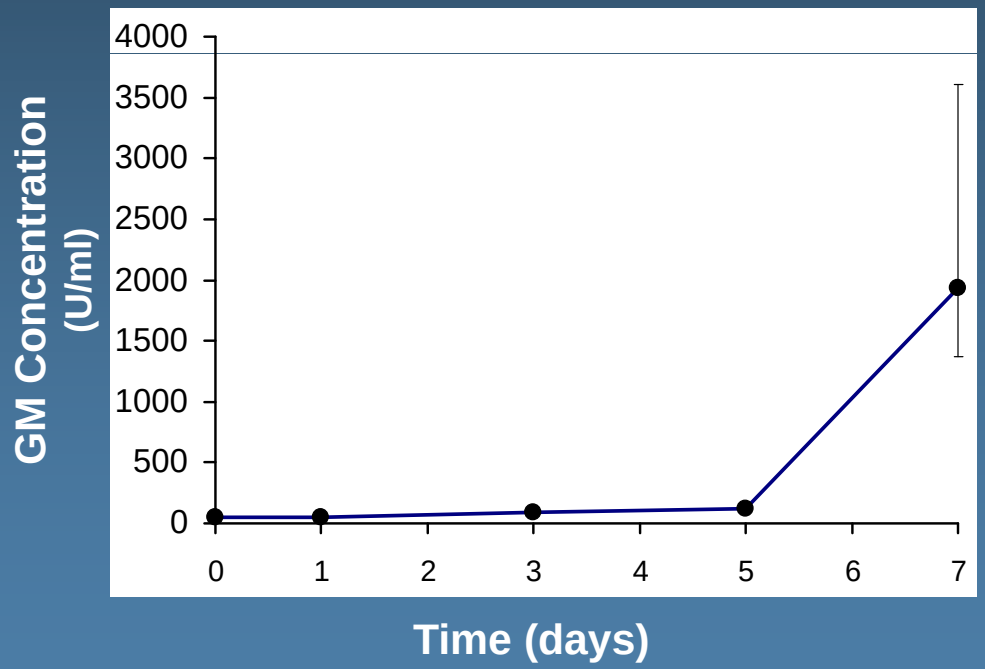
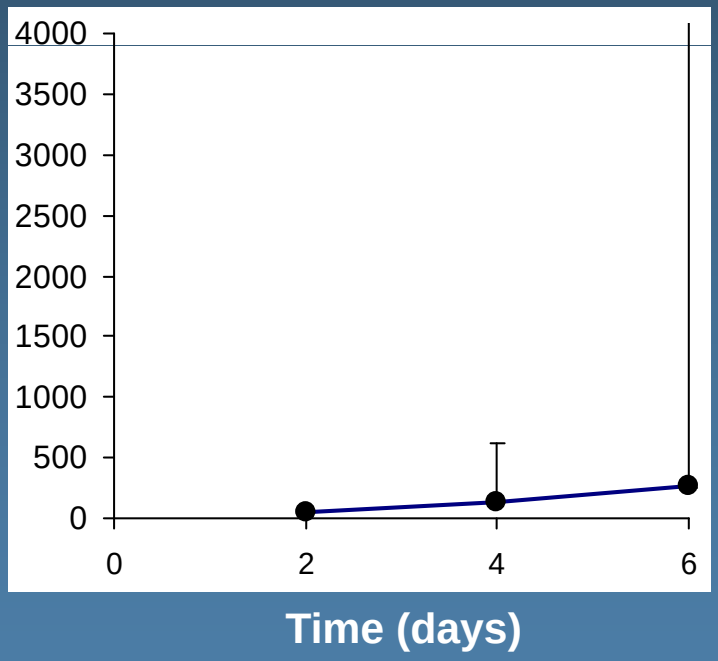
+ Cyclophosphamide)



Non-neutropenic
(Cortisone Acetate)



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Effects of Different Types of Immuno-suppression on Response to Therapy and Impact on Diagnostics

- Therapy:
  - Treatment day 1-7 post infection with amphotericin B (AMB) 3 mg/kg IP, posaconazole (PSC) 40 mg/kg PO, liposomal amphotericin B (LAMB) 10 mg/kg IV, or water IP for Controls.
  - Mice were followed through day 12 post infection.
- Statistics:
  - Log rank test used for survival comparisons.
  - Mann Whitney test used for fungal burden comparisons.
  - $P \geq 0.05$ required for statistical significance.

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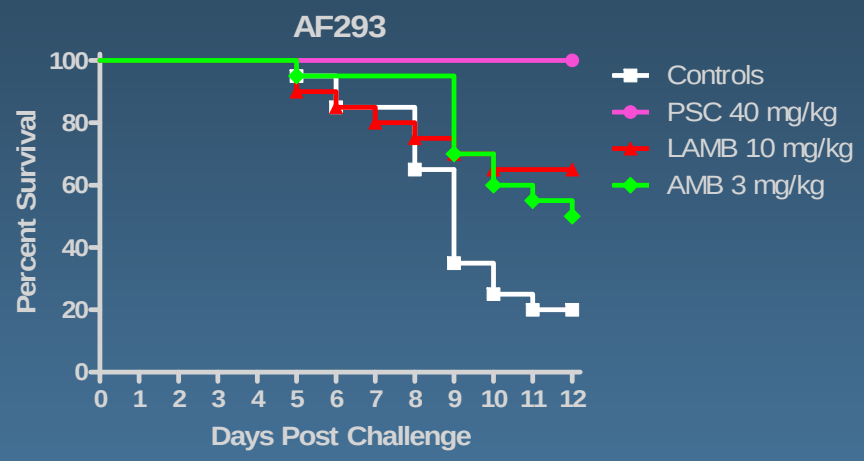
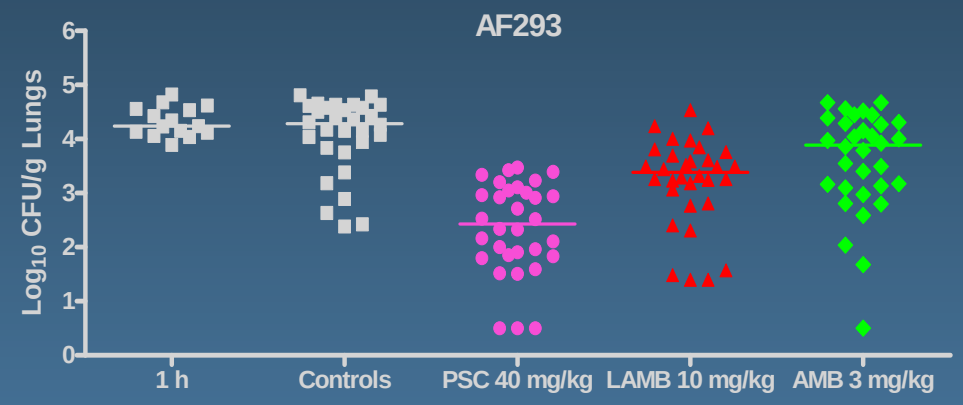
Posaconazole is Highly Efficacious in the Neutropenic Model





N≥18 mice/group. All treatments prolonged survival p≤0.0150.
N= 15 for 1h. N≥30 mice/group. All treatments reduced lung fungal burden compared to controls p≤0.0213.

Najvar LK, et al, ICCAC 2007 (abstract M-1848)

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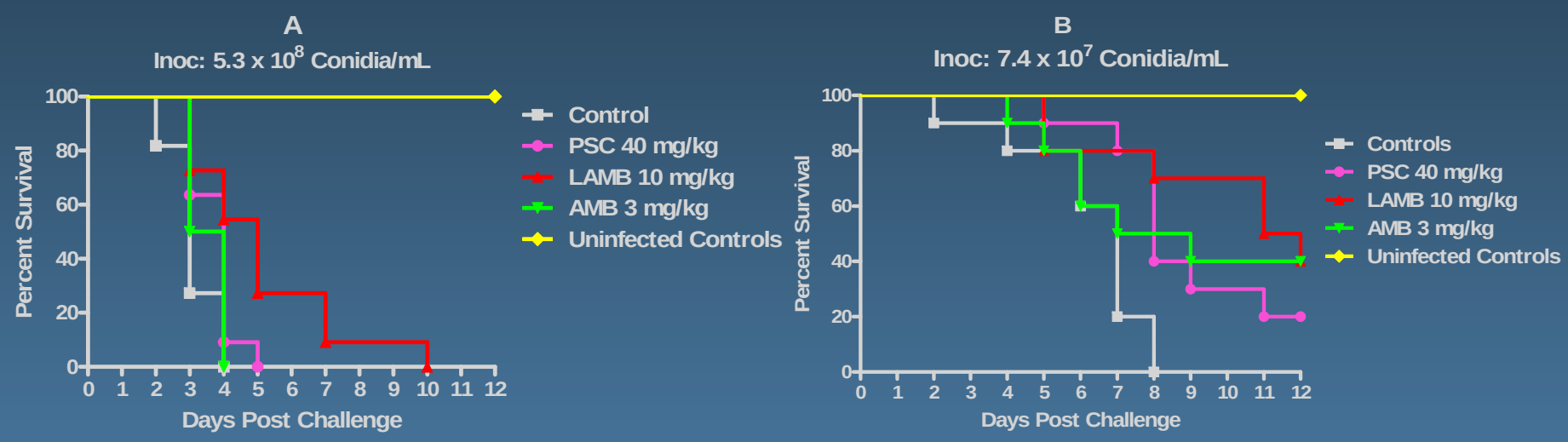
Posaconazole is Less Efficacious in the Non-neutropenic Model



N=10 mice/group. Controls all succumbed by day 4 or 8. The median survival was prolonged for LAMB (p=0.006) in A and both PSC (p=0.007) and LAMB (p=0.009) prolonged survival in B.

Najvar LK, et al, ICCAC/IDSA 2008 (abstract M-1561)

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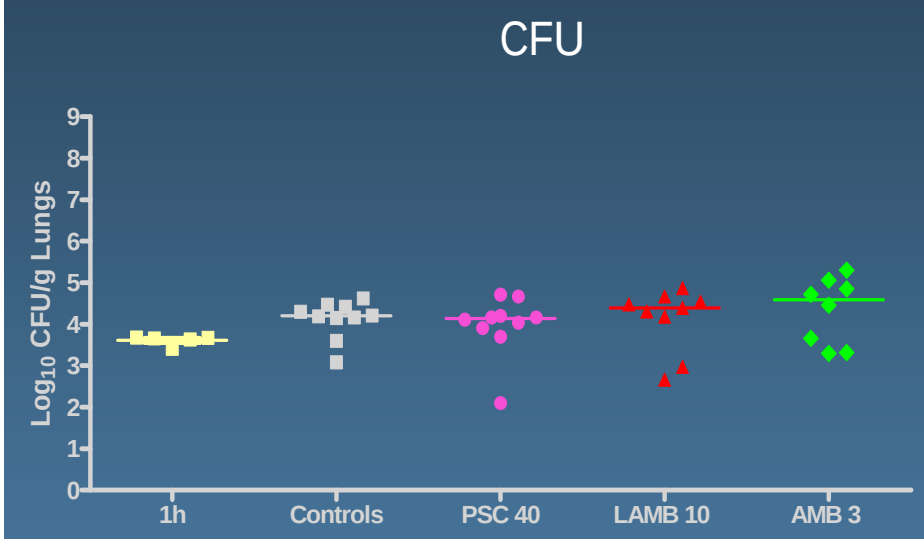
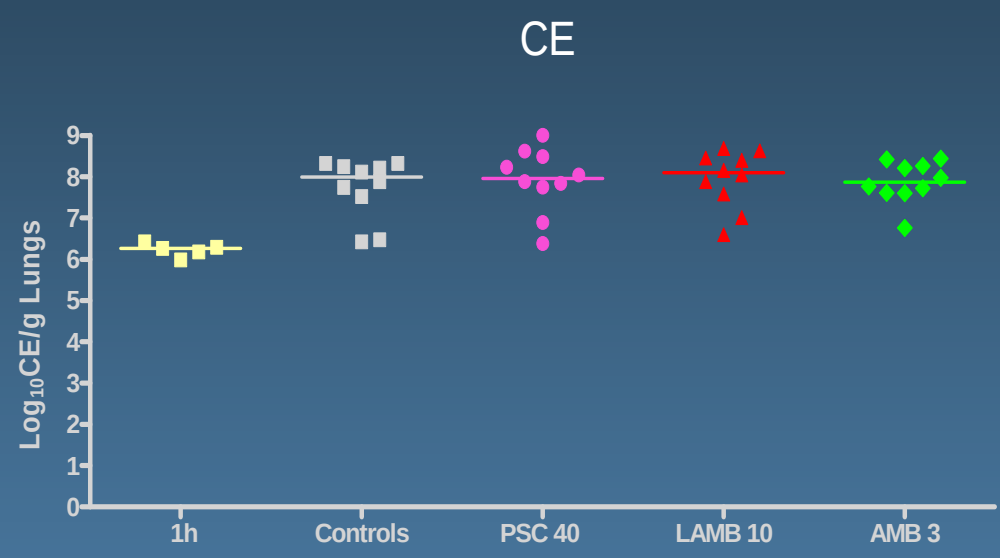
No Therapy Reduced Pulmonary Tissue Burden in the Non-neutropenic Model





Inoculum: 7.4 x 10^7 Conidia/mL. N= 5-10 mice/group. No therapy reduced the fungal burden.

Najvar LK, et al, ICCAC/IDSA 2008 (abstract M-1561)

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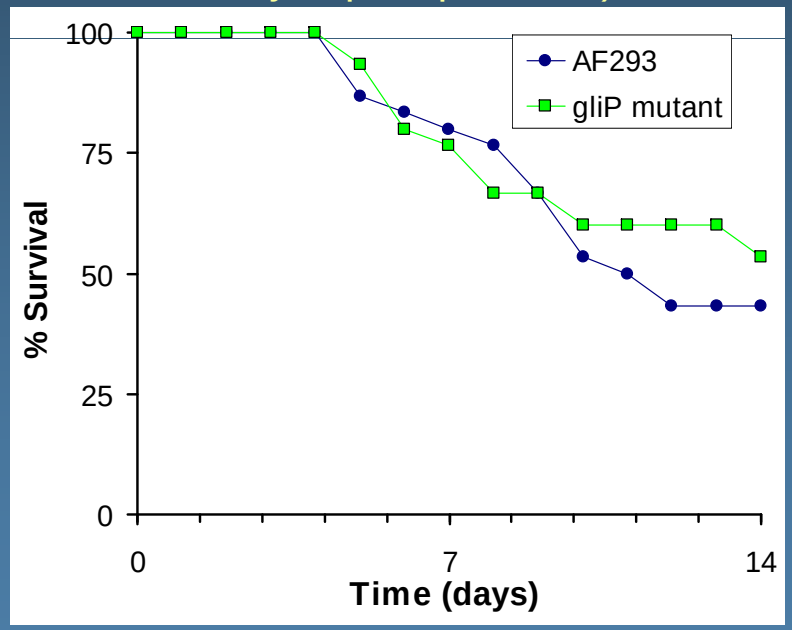
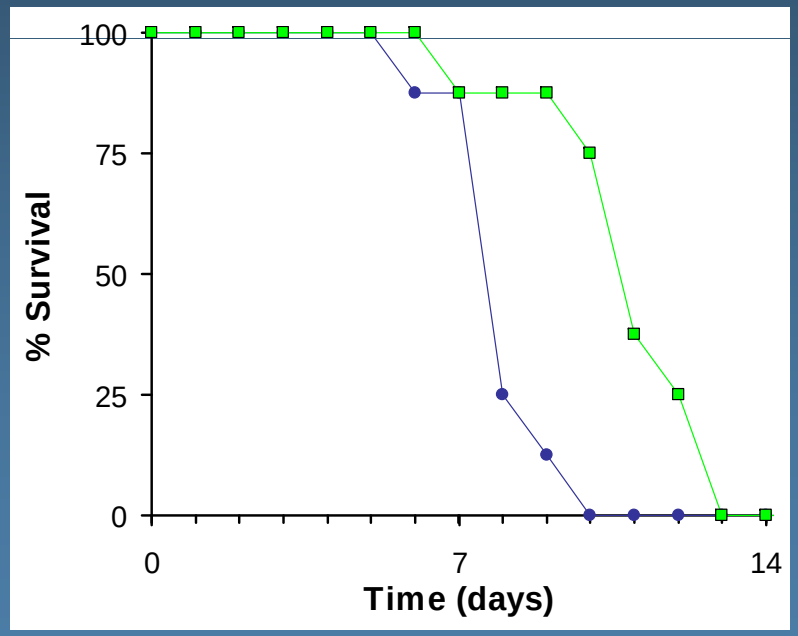
Effects of Different Types of Immuno-suppression on Virulence Mechanisms

Neutropenic
(Cortisone Acetate
+ Cyclophosphamide)



Non-neutropenic
(Cortisone Acetate)



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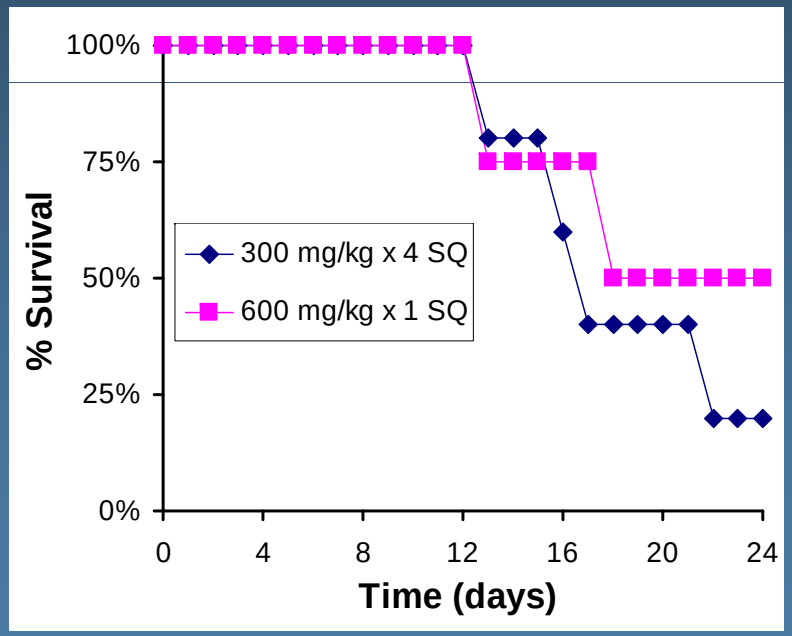
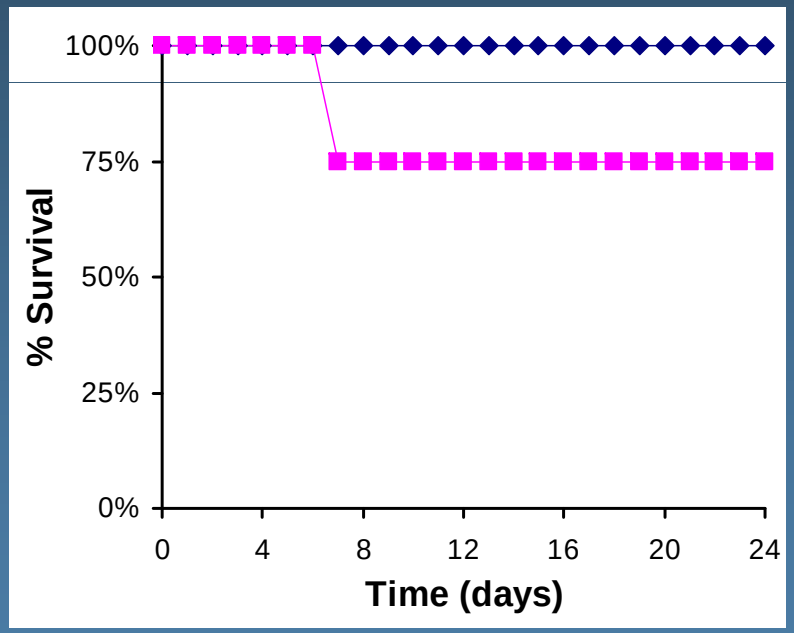
Alternatives to Cortisone Acetate-Depot Medrol

Infected



Uninfected



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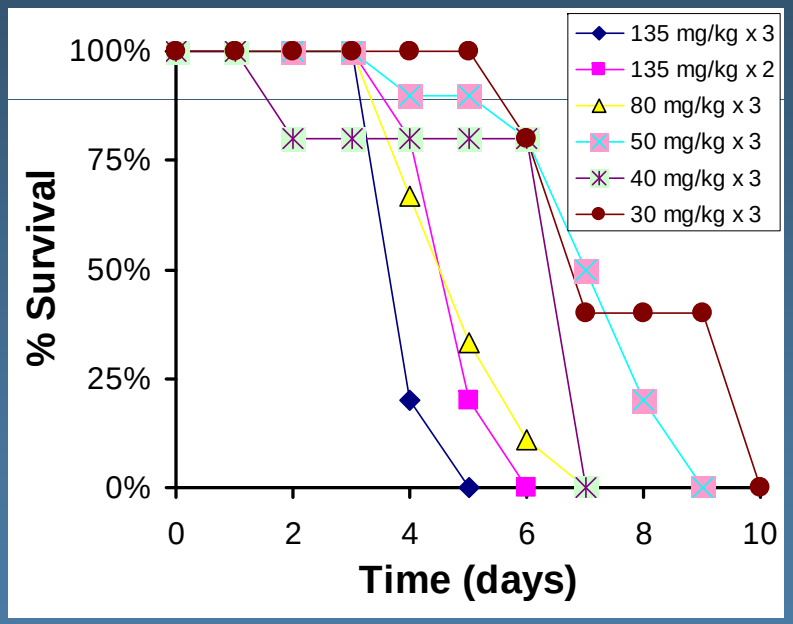
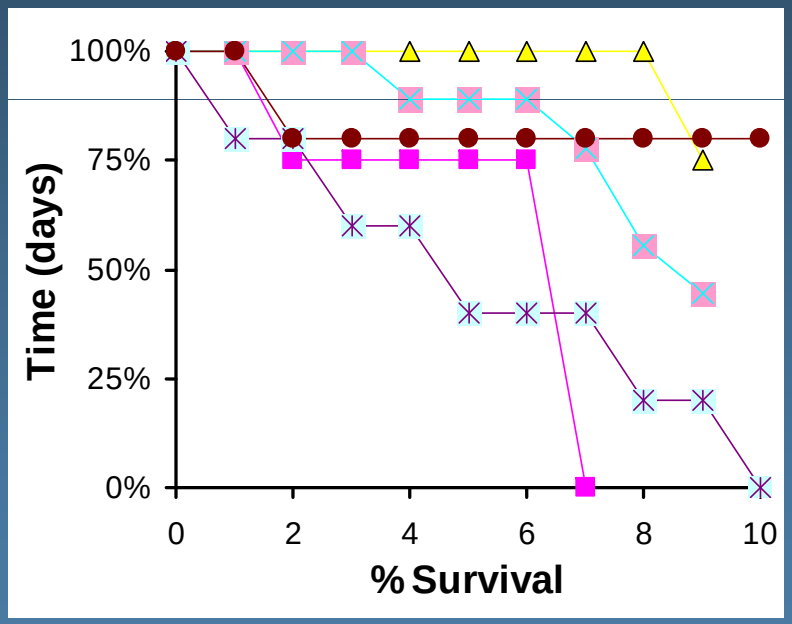
Alternatives to Cortisone Acetate-Triamcinolone

Infected



Uninfected



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