Mucormycosis Axis of Evil: Iron uptake, Invasion and Toxin Production

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George W had no input on the design
or interpretation of the results

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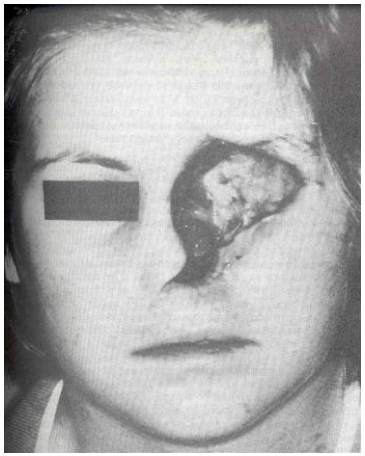
Mucormycosis Hallmarks

- **Three clinical features point to essential attributes of mucormycosis pathogenesis:**

  1. **Angioinvasion → interactions between Mucorales and endothelial cells.**
  2. **The unique predisposition of patients with diabetics in ketoacidosis (DKA), other forms of acidosis, deferoxamine-treated patients → host iron acquisition.**
  3. **The extensive tissue necrosis → potential toxin production.**

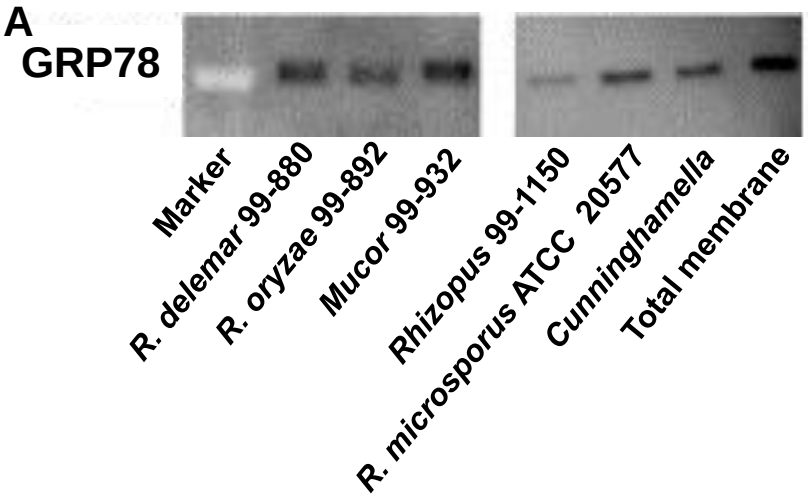


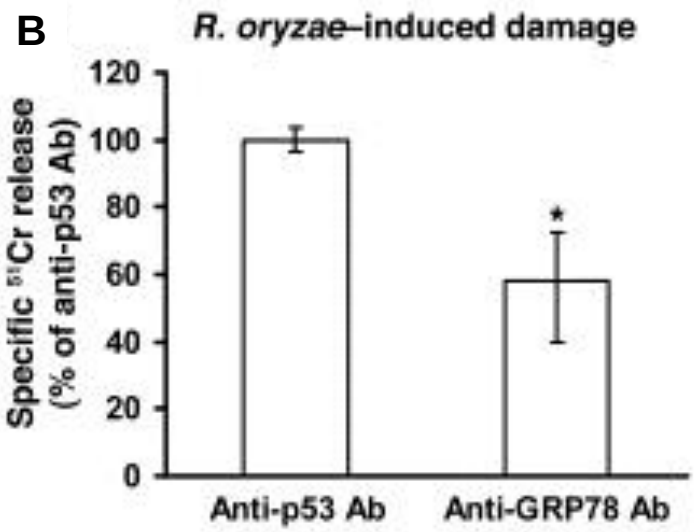
Overview

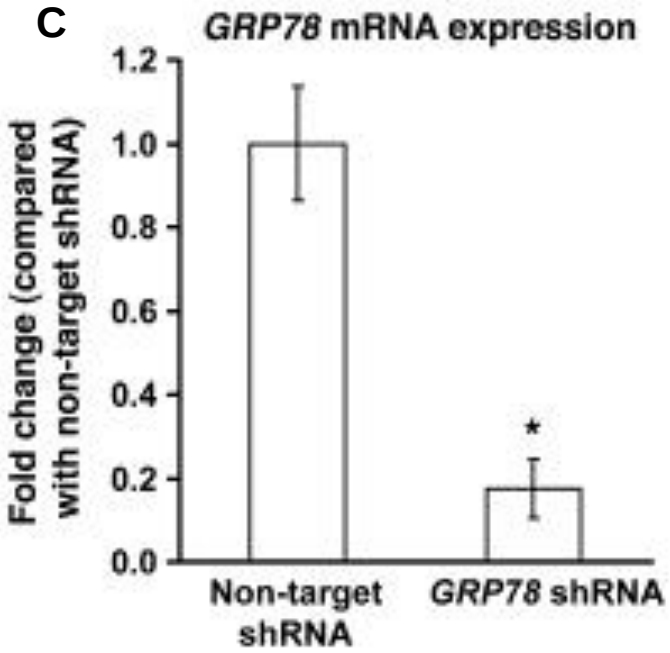
- How does host cell invasion occurs? What host cell receptors and fungal ligands are involved? → Endothelial cell/*Rhizopus* Interactions
- How host factors specific to patients at high risk of acquiring mucormycosis modulate host cell invasion? →Hyperglycemia, acidosis and elevated available serum iron
- What are the mediators of tissue necrosis? → Toxins
- Potential novel therapies for mucormycosis

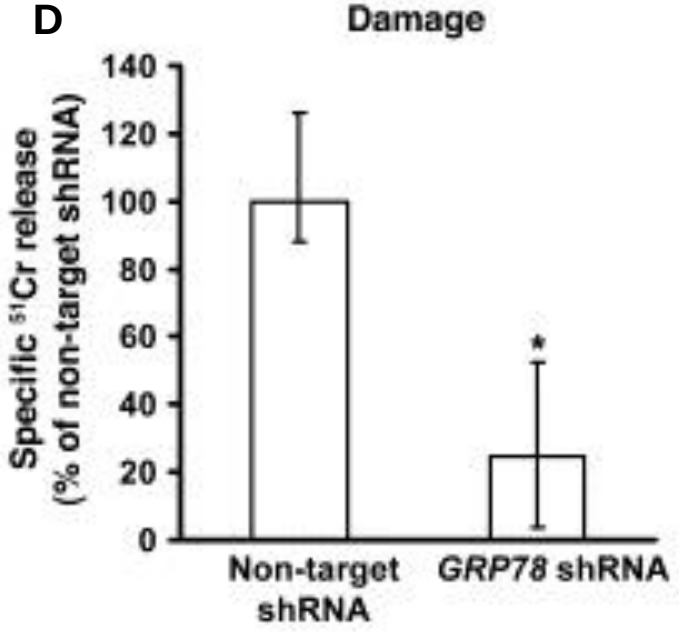
The Heat Shock Protein GRP78 is the Endothelial Cell Receptor to Mucorales

Liu et al. JCI 2010



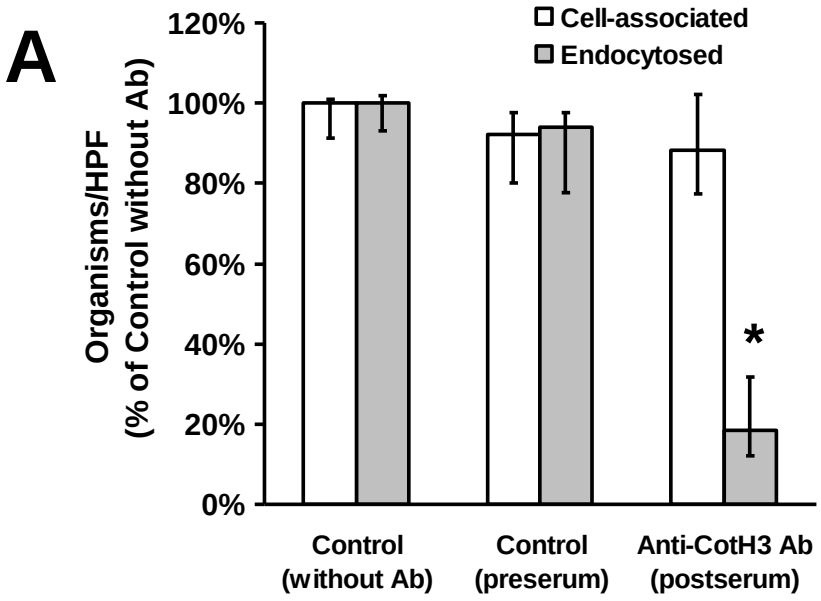


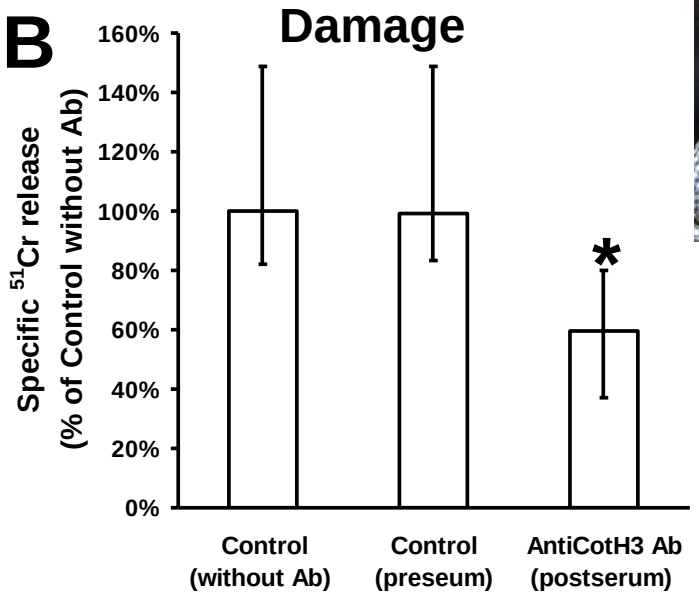


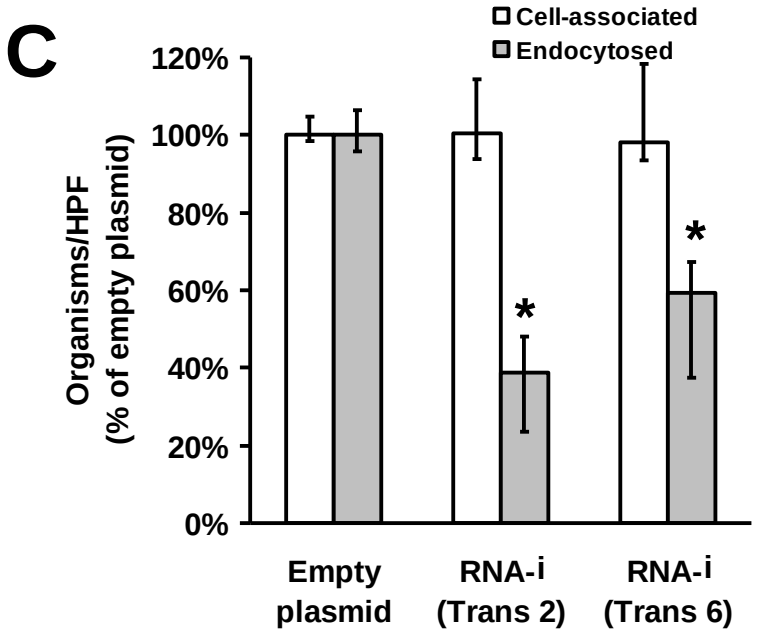


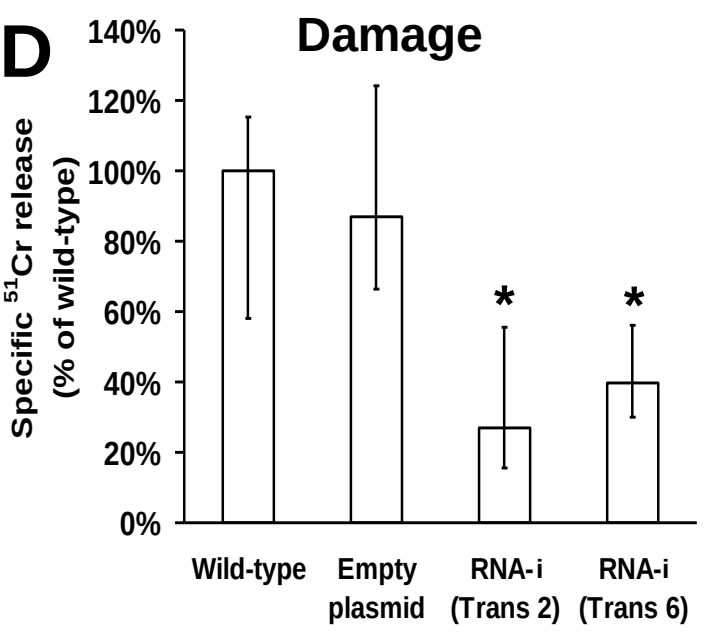
CotH Enables *Rhizopus* to Invade Endothelial Cells

Gebremariam et al. JCI 2014





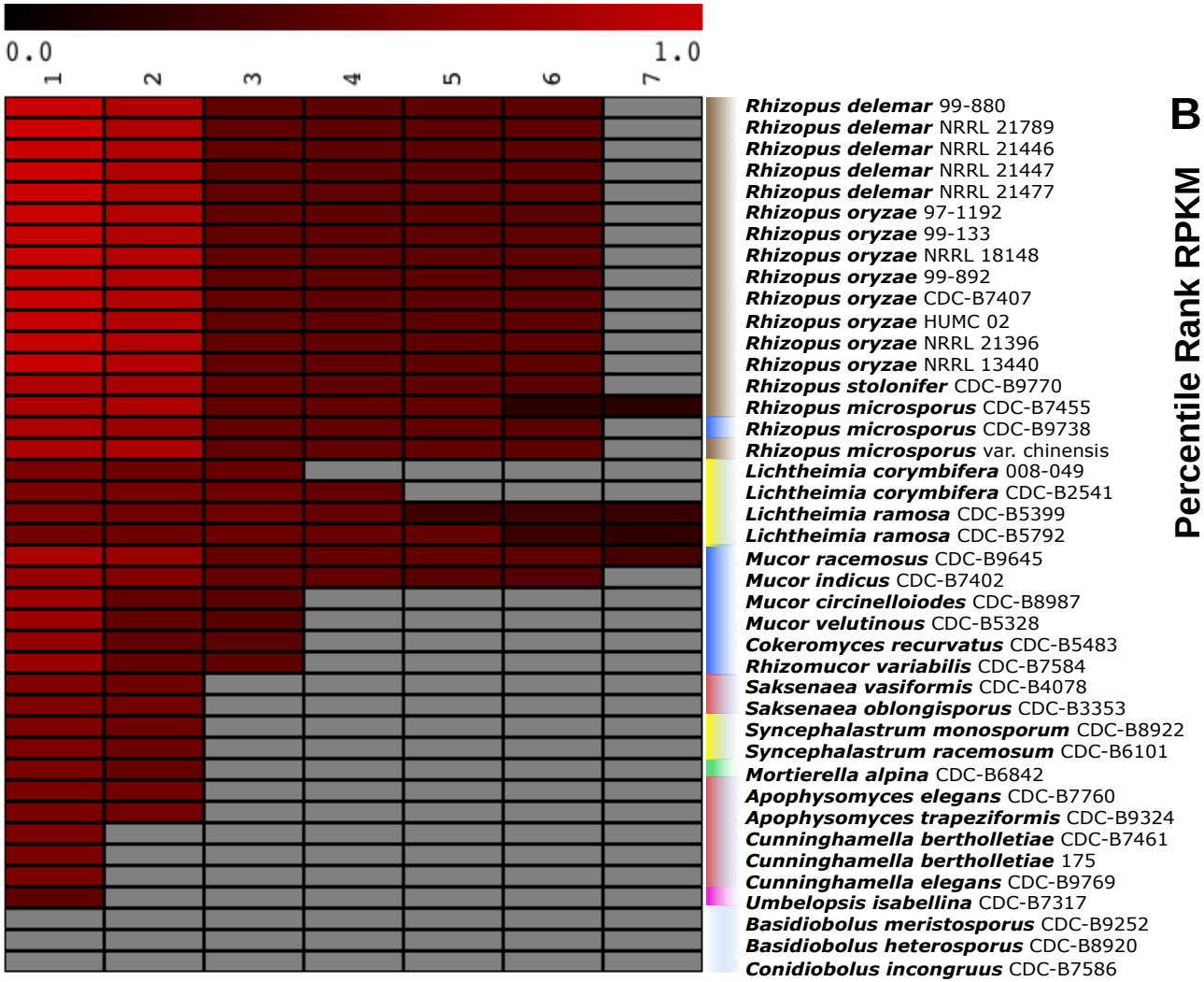




Spore Coating Protein (CotH) are Conserved Among Mucorales

Chibucos et al……Bruno Nature Communications 2016

A.



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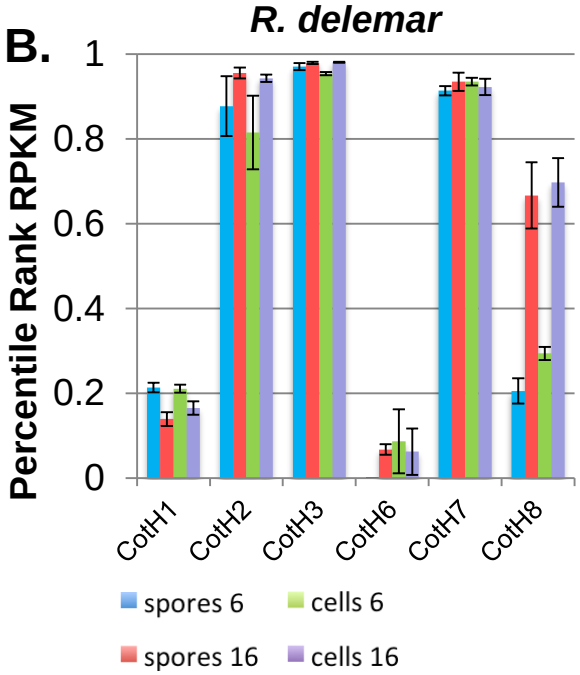
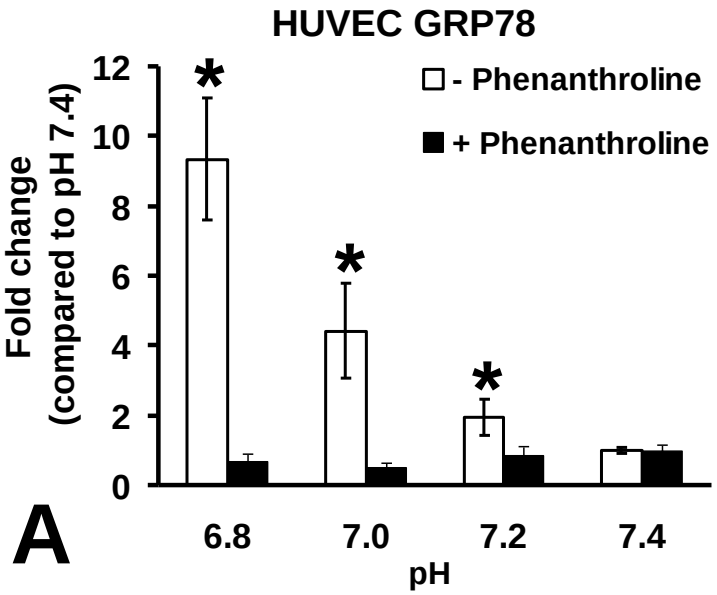


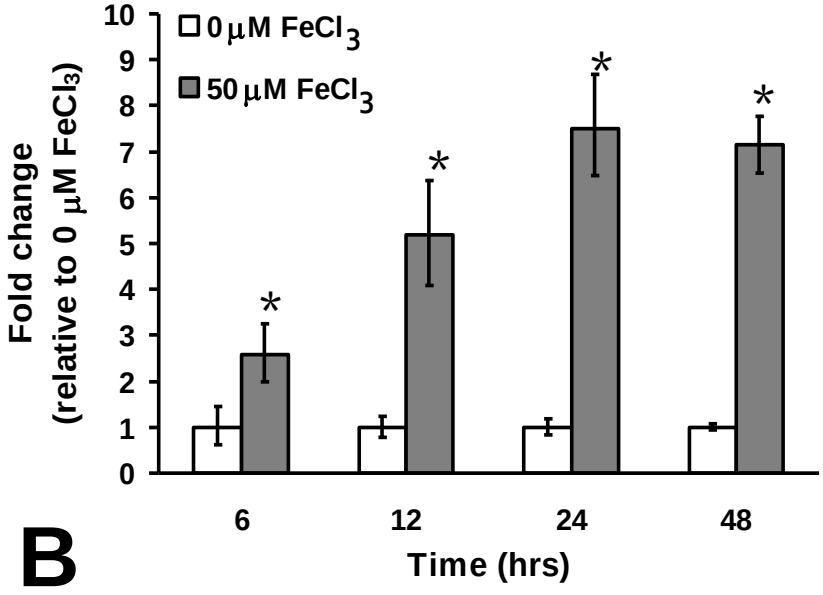
Effect of Hyperglycemia, Acidosis and Elevated Available Serum Iron on GRP78/CotH Interactions

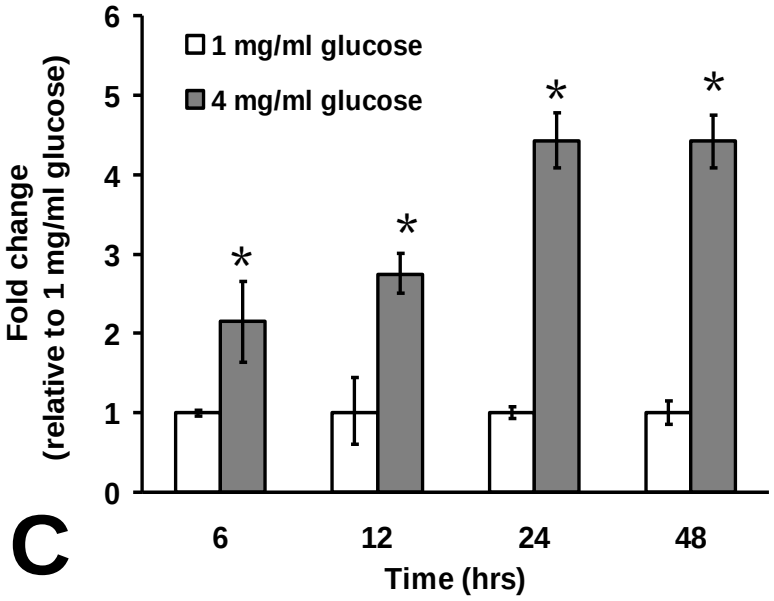
- DKA and deferoxamine-treated patients are uniquely predisposed to mucormycosis.
- These patients suffer from elevated available serum iron
  - Acidosis via compromised ability of transferrin in chelating iron (a protonation mechanism, Artis *et al.* Diabetes 1982).
  - Deferoxamine supplies iron to *Rhizopus* (Boelaret *et al.* JCI 1993).
- Is it possible that DKA patients are uniquely predisposed to mucormycosis because host factors modulate the expression of GRP78/CotH proteins?

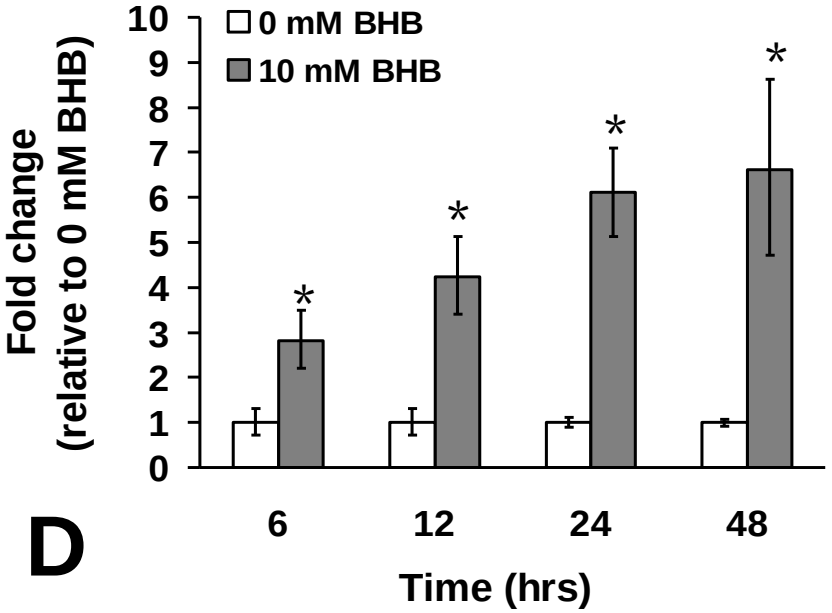
Acidosis, Elevated Available serum, Hyperglycemia, and BHB Induce Expression of Endothelial Cell GRP78

Liu et al. JCI 2010



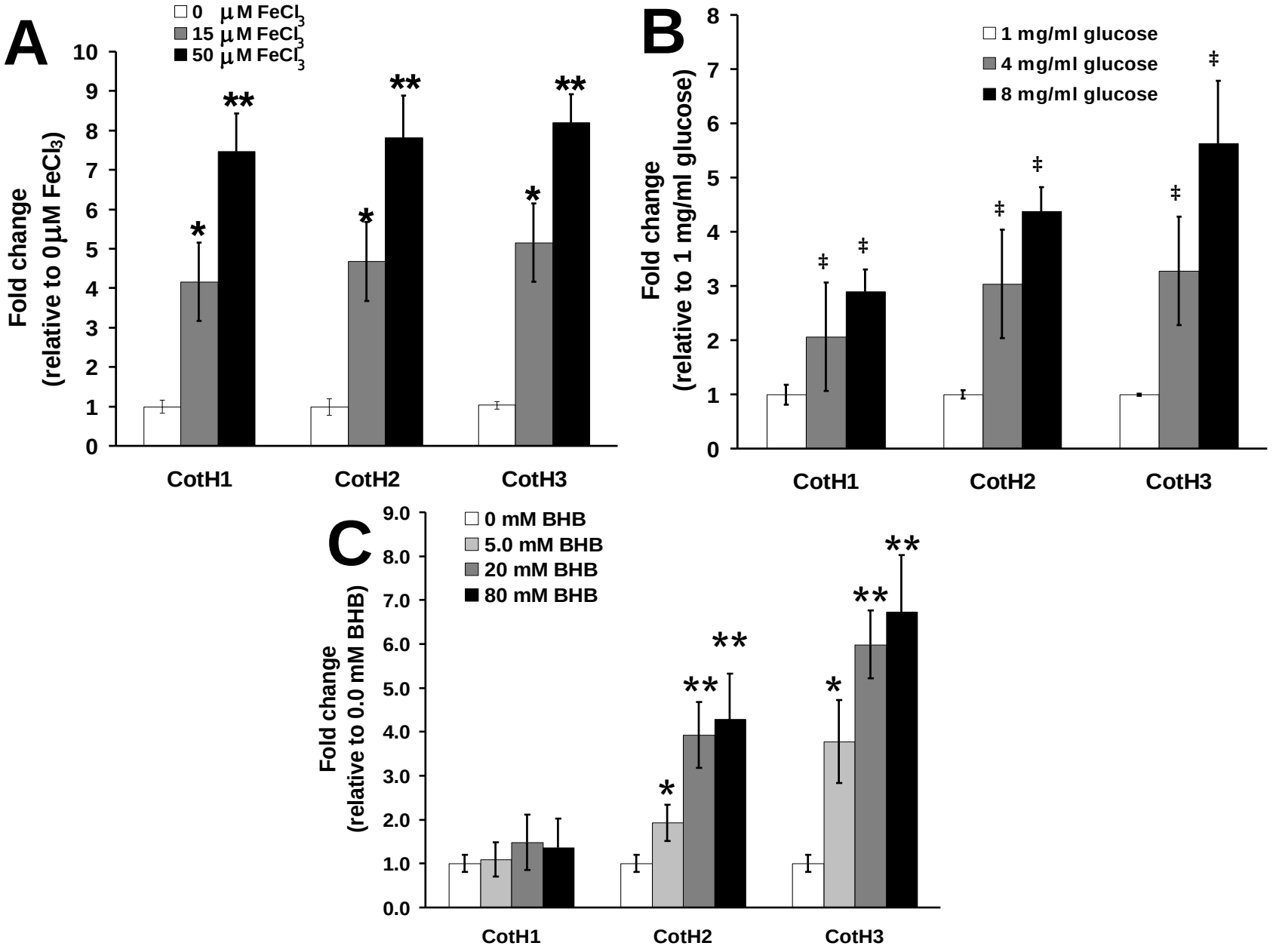






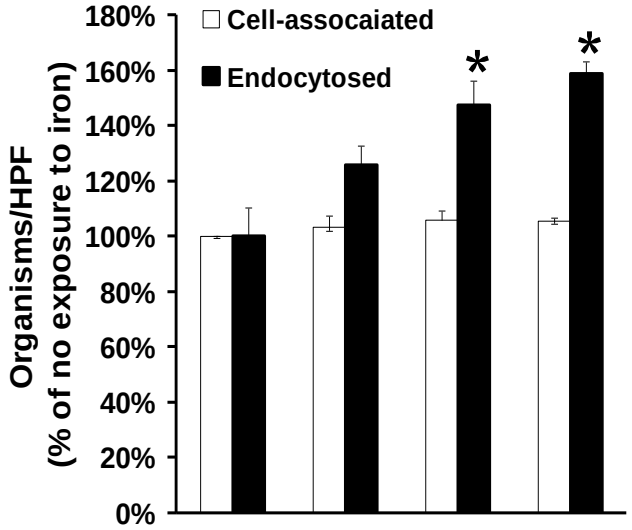
Iron, Glucose, and BHB Induce Expression of *R. delemar CotH*

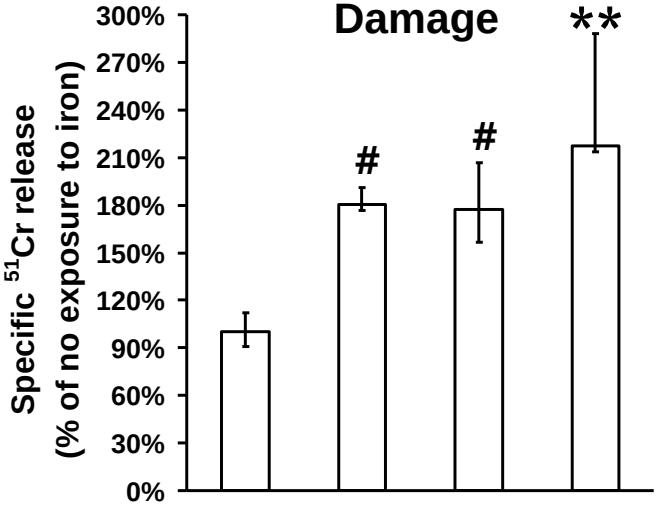
Gebremariam et al. JCI 2016



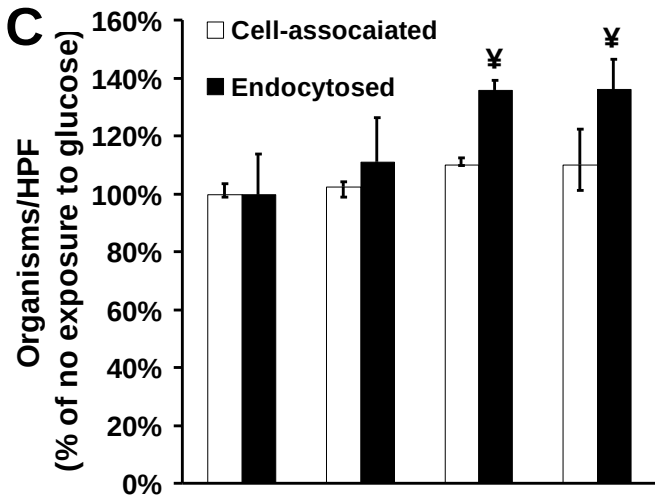
Iron and Glucose Modulate Interactions of *R. delemar* With Endothelial Cells

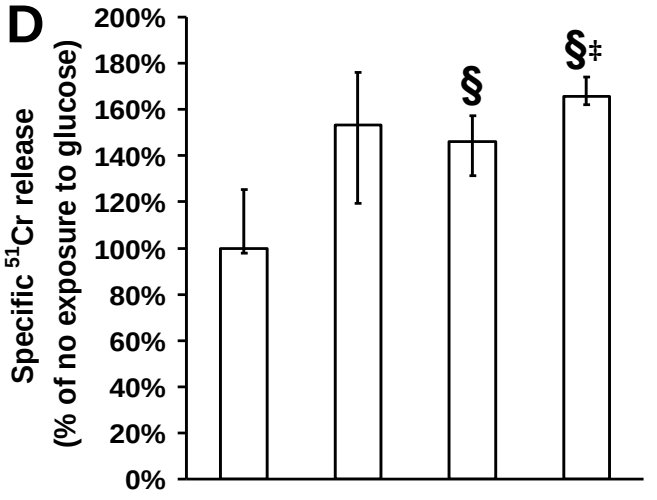
Gebremariam et al. JCI 2016





Endothelial cells + iron	-	-	+	+
R. delemar + iron	-	+	-	+

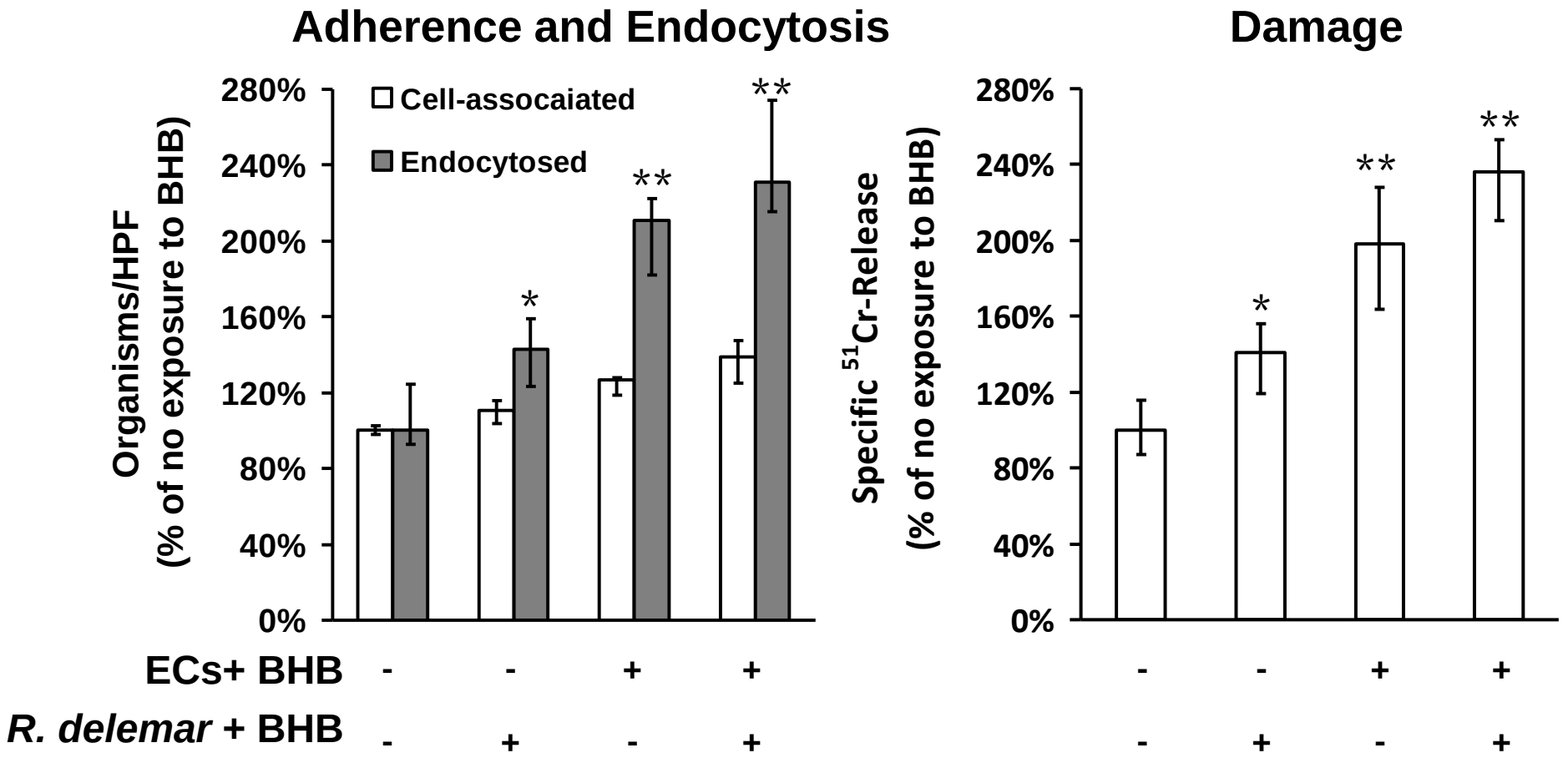




Endothelial cells + glucose	-	-	+	+
R. delemar + glucose	-	+	-	+

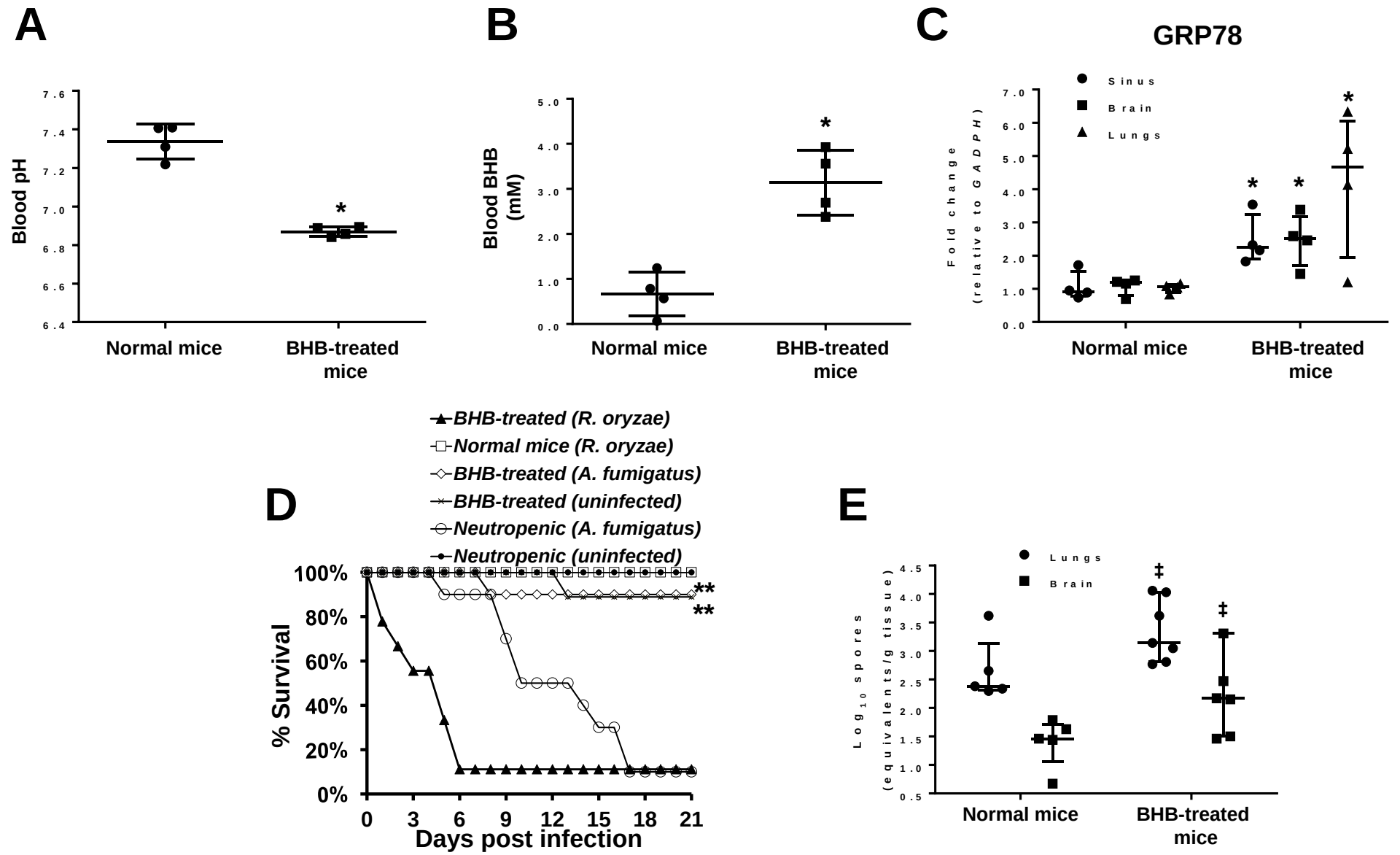
BHB Modulates Interactions of *R. delemar* With Endothelial Cells

Gebremariam et al. JCI 2016



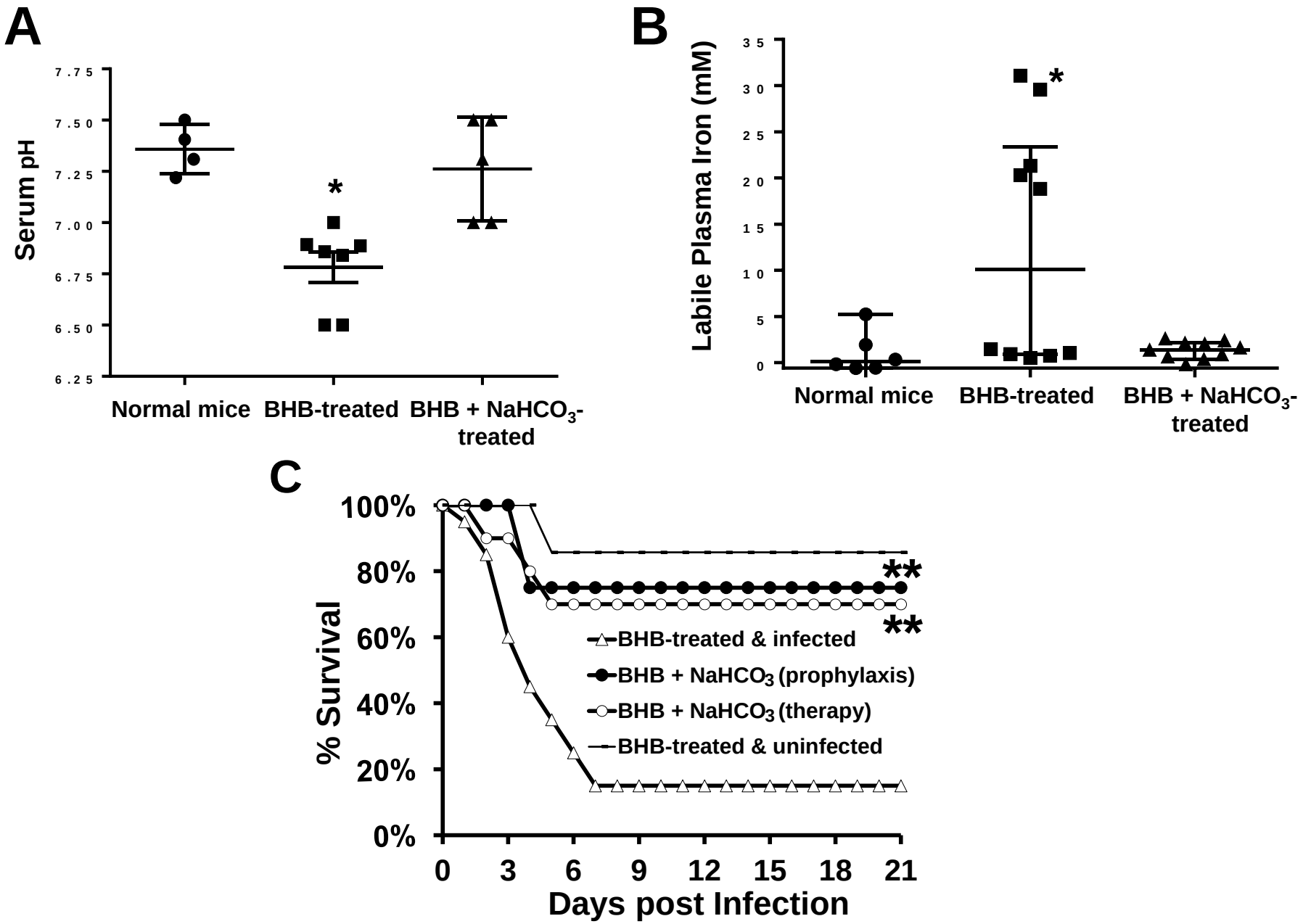
BHB Lowers Mouse Blood pH, Increases GRP78 Expression in Target Organs and Specifically Enhances Susceptibility to Mucormycosis

Gebremariam et al. JCI 2016



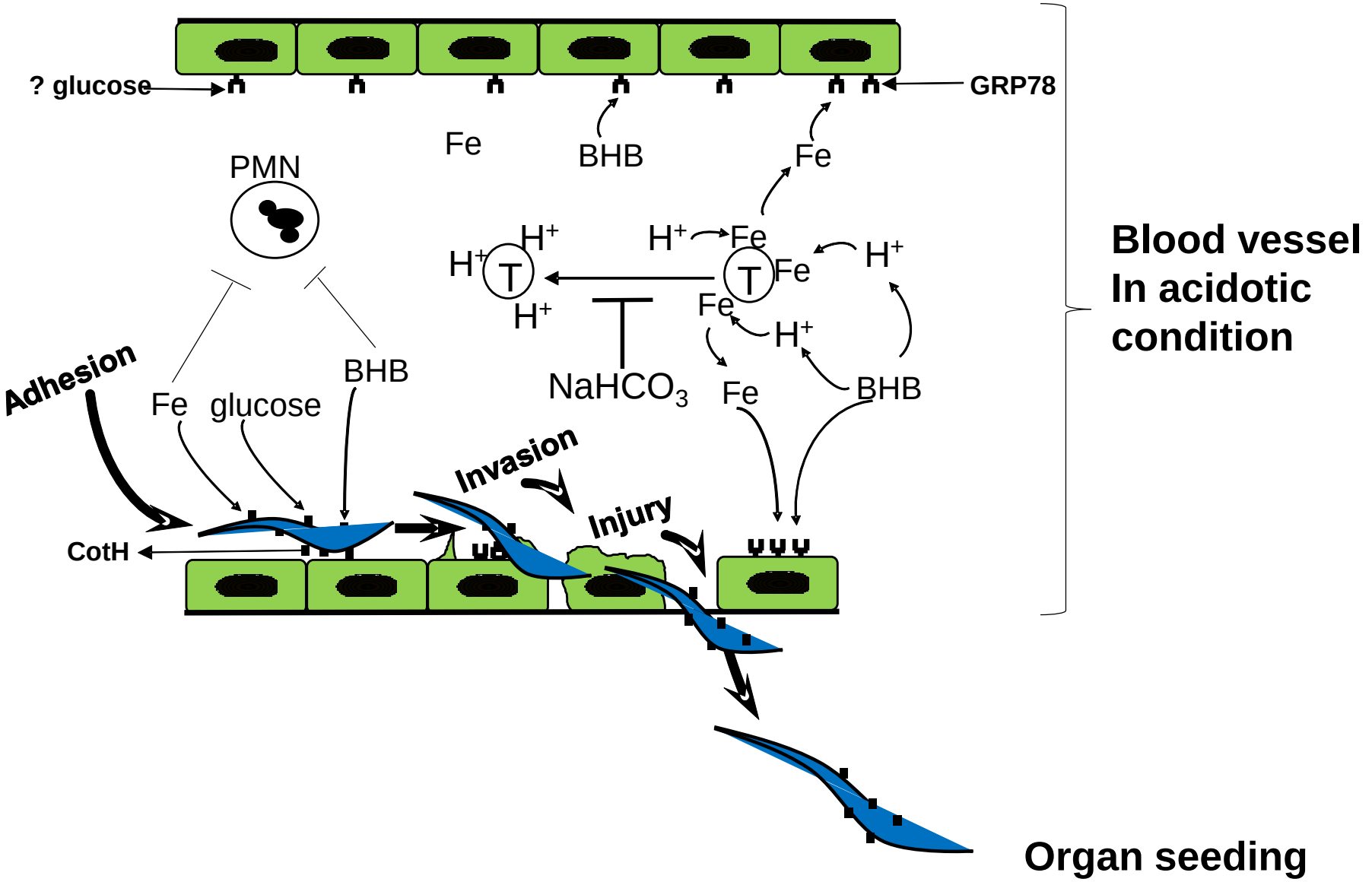
Sodium Bicarbonate Enhances Survival of BHB-Treated Mice with Mucormycosis

Gebremariam et al. JCI 2016



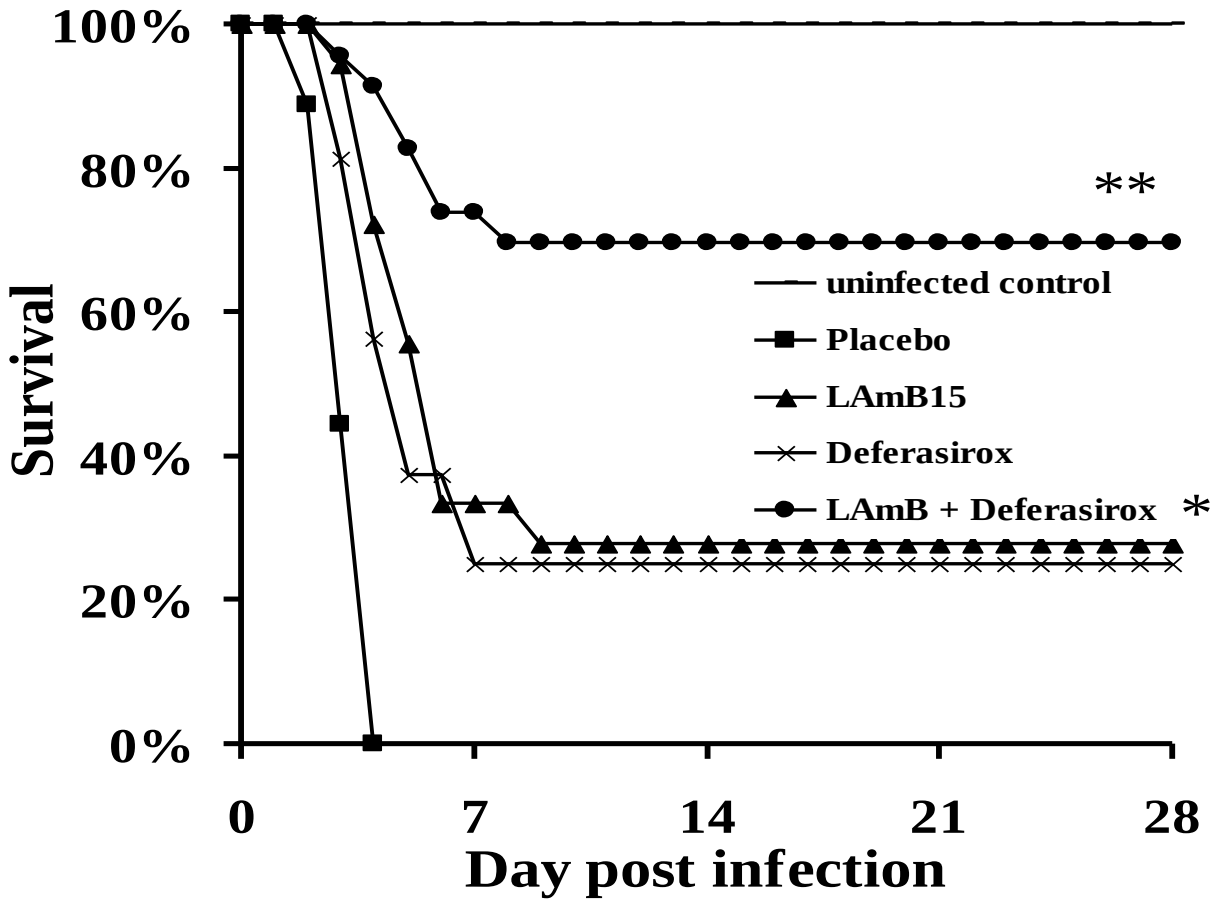
The Model

Ibrahim et al. JCI 2007; Liu et al. JCI 2010; Gebremariam et al. JCI 2014; Gebremariam et al. JCI 2016

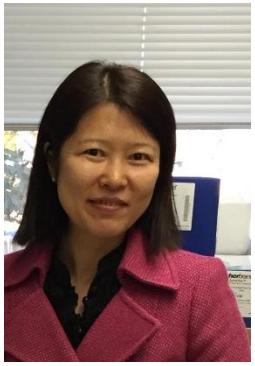


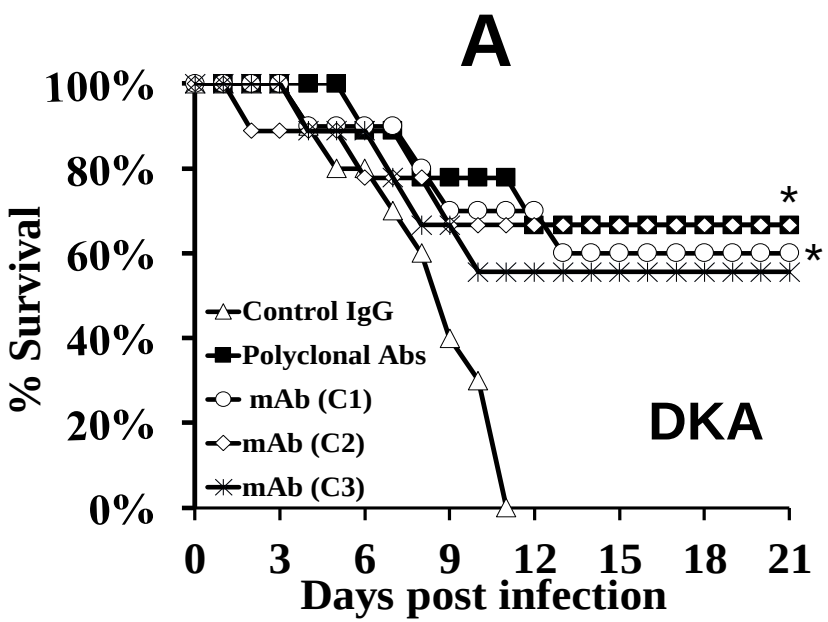
Iron Chelation is Effective Against Mucormycosis in DKA Mice

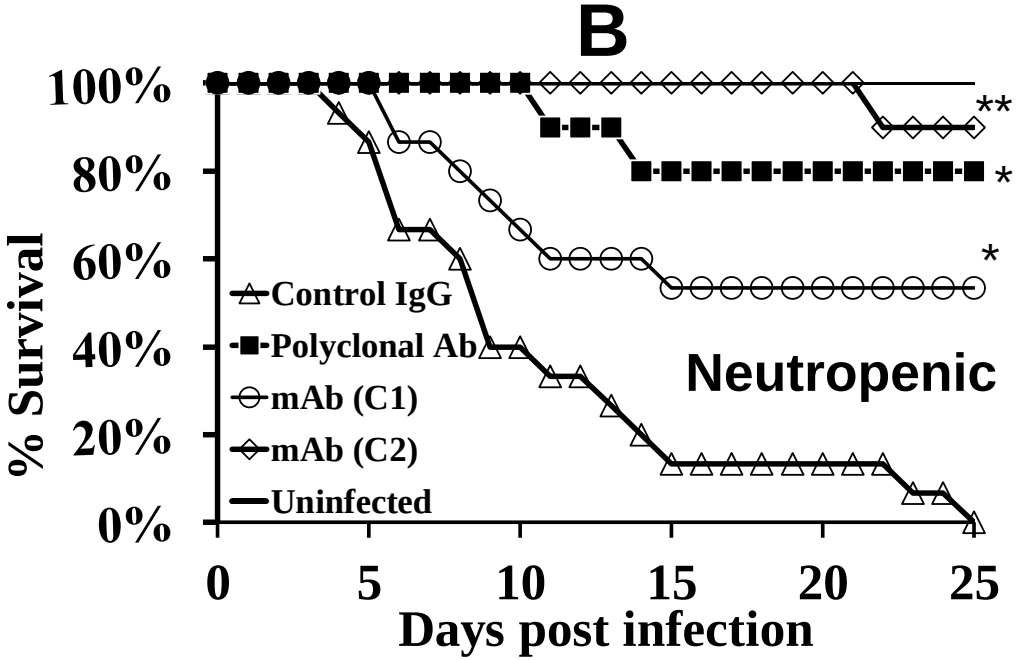
Ibrahim et al. JCI 2007



Targeting GRP78/CotH Interactions to Develop Novel Immunotherapay

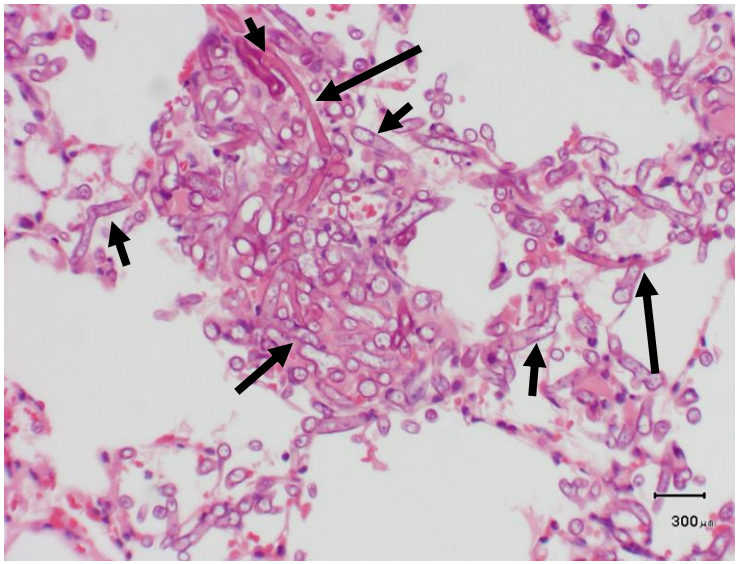
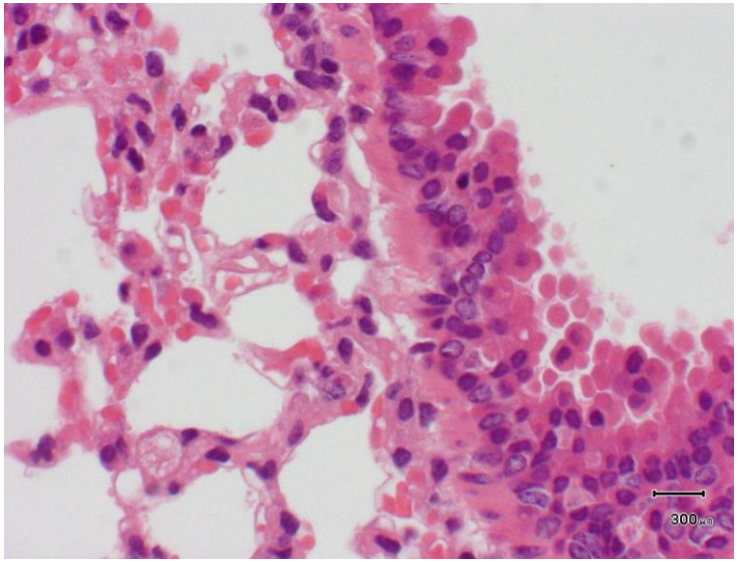
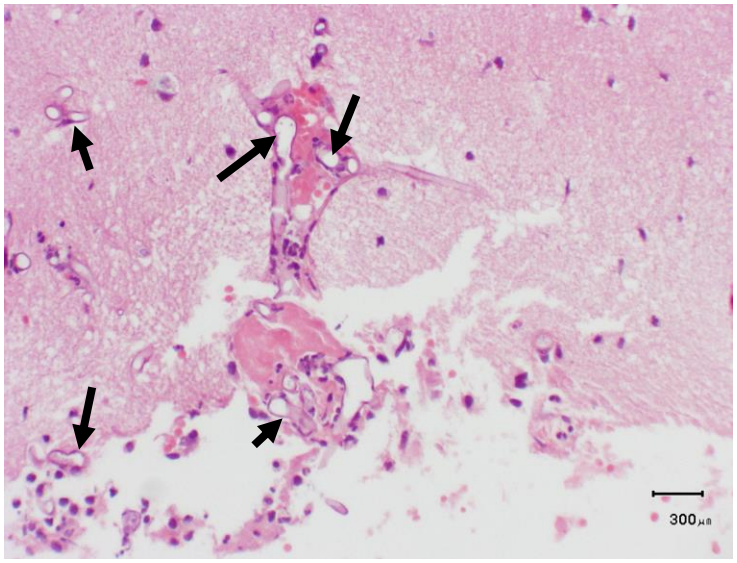
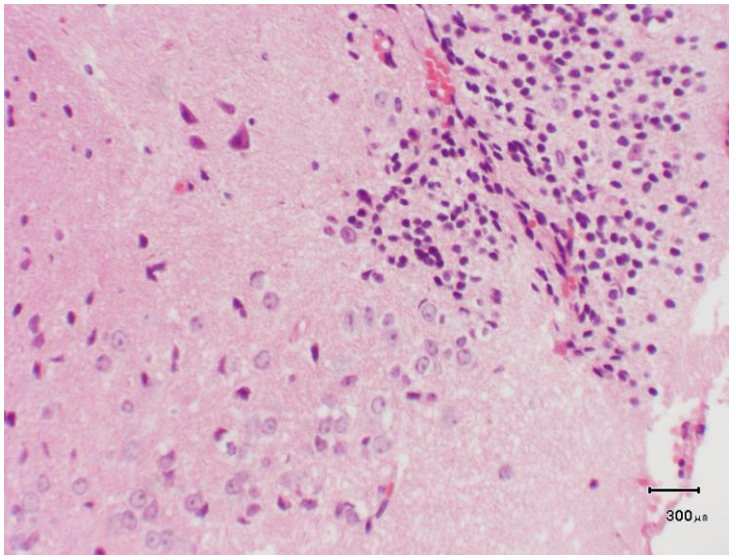






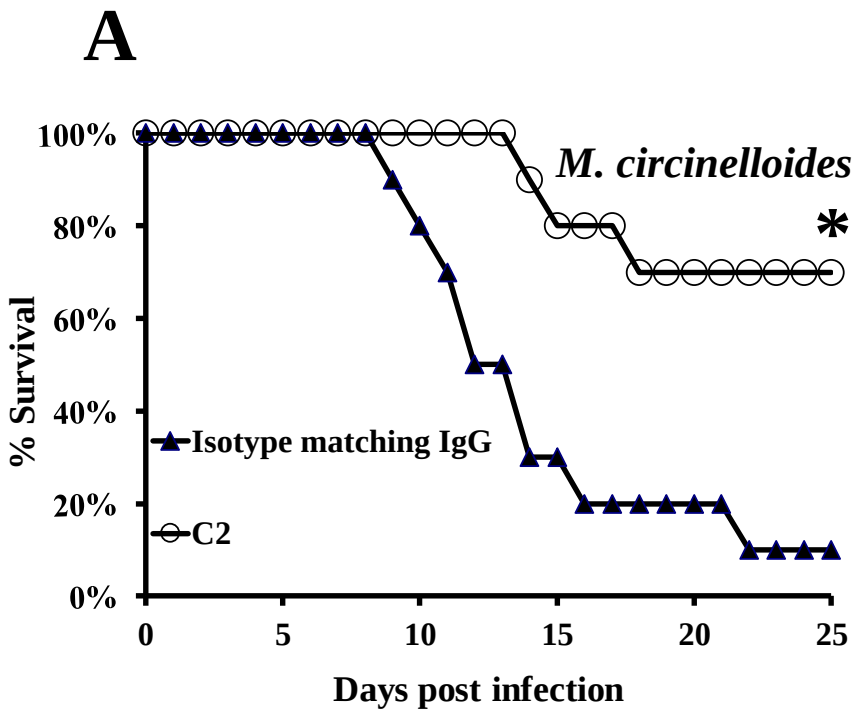
30 µg single dose given therapeutically 24 h post infection

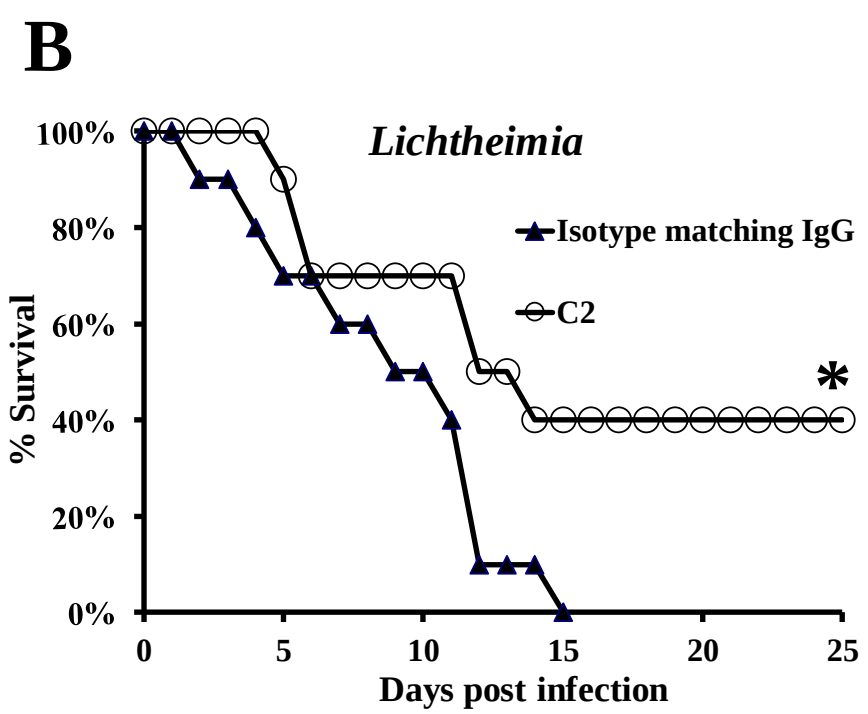
Anti-CotH3 Abs Prevent Angioinvasion in Neutropenic Mice

	Isotype matching IgG	Anti-CotH3 Abs
Lungs		
Brain		

Monoclonal Ab is Protective Against Mucormycosis Caused by Mucorales other than *Rhizopus*

30 μg single dose given therapeutically 24 h post infection



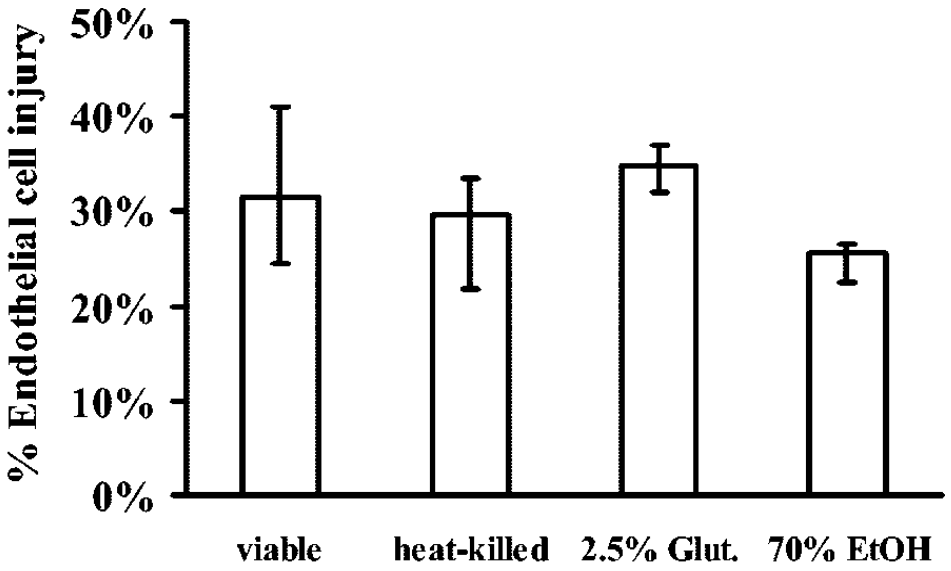


****P* <0.05, N=10 per group**

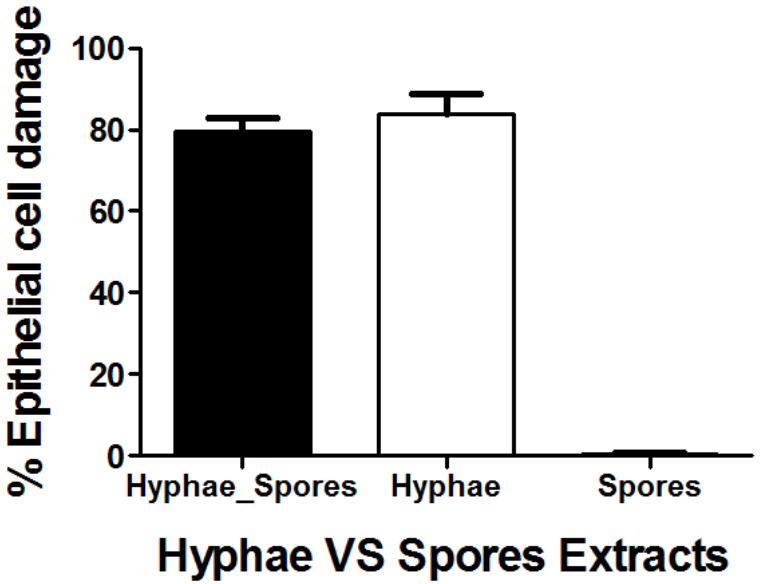
Summary/Future Studies

- GRP78/CotH interactions play a major role in mucormycosis pathogenesis
- Preclinical mucormycosis pathogenesis studies introduced novel therapies that are likely to benefit mucormycosis patients:
  - Sodium bicarbonate for DKA patients suspected of mucormycosis
  - Passive immunization with anti-CotH Abs protects mice from mucormycosis
- Mechanism of protection is reliant on prevention of endothelial cell invasion and inhibition of fungal growth
- We are currently working on humanizing the mAb and developing a viable cell-line

Hyphae Associated Toxin



Ibrahim et al. I&I 2005



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