Updates on *Candida* Immunology

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The Disease Burden of Candidiasis in Humans

- NOT LIFE-THREATENING
  - 75% of all women → vaginitis
  - 25% of antibiotic-treated women → vaginitis
  - 90% of AIDS patients → oral thrush

- LIFE-THREATENING
  - Systemic candidiasis
  - Survival
IL-17 Signaling is Important for Anti-*Candida* Mucosal Host Defense
IL-17 Signaling is Important for Anti-Candida Mucosal Host Defense in Mice and Humans

Chronic Mucocutaneous Candidiasis in Humans with Inborn Errors of Interleukin-17 Immunity
Anne Puel, †Sophie Cypowyj, †Jacinta Bustamante, †8J.F. Wightman, Luyan Liu, Hye Kyung Lim, ‡Mélanie Migaud, Laura Isaac, Maya Chtarbah, ‡Magali Audry, ‡Matthew Gumbleton, ‡Antoine Toulou, ‡Christine Bodemer, ‡Jamila El-Baghadad, ‡Matthew Whitters, ‡Theresa Paradis, ‡Jonathan Brooks, ‡Mary Collins, ‡Neil M. Wolfman, ‡Saleh Al-Muhsein, ‡Miguel Galicchio, ‡Laurent Abel, ‡†Capucine Picard, ‡‡Jean-Laurent Casanova

Inherited IL-17RC deficiency in patients with chronic mucocutaneous candidiasis
Yun Ling, Sophie Cypowyj, Caner Aytekin, Miguel Galicchio, Yildiz Camcioglu, Serdar Nespavov, Aydan Khanoglu, Figen Dogu, Aziz Belgad, Romain Levy, Mélanie Migaud, Bertrand Bosson, Alexandre Brize, Yuval Ilan, Nicolas Goudin, Julien Cottineau, Capucine Picard, Laurent Abel, Jacinta Bustamante, Jean-Laurent Casanova, Anne Puel

IL-17–Mediated Protection via Generation of Anti-Candida Antimicrobial Peptides

Holland, NEJM, 2009; Lionakis & Levitz. Annu Rev Immunol, 2018
The Clinical Observations

Although ICU patients share clinical and microbiological risk factors for candidiasis, only a small minority develops the infection.

Among infected patients, the outcome of candidiasis varies greatly.
The Hypothesis

Variation in immune function-related genes modulates the risk of developing candidiasis and worse outcome after infection

Bench to Bedside Study of Systemic Candidiasis

Can we identify patients with specific host genetics that are at higher risk for the infection?
Phagocytes but not Lymphocytes are Critical in Host Defense against Systemic Candidiasis

- Neutrophils

- Macrophages

MONOCYTES
MACROPHAGES
**Candida Yeasts Are Effectively Picked up by Macrophages Early After Infection in the Kidney**

Lionakis & Levitz. *Annu Rev Immunol.* 2018

2 hrs post-infection

Lionakis *et al.*, *J Clin Invest.* 2013
Macrophages Wrap Around *Candida* Hyphae Early After Infection in the Kidney

2 hrs post-infection

Macrophages (CX₃CR1)
dTomato-*Candida*

Lionakis et al., *J Clin Invest*. 2013

CX3CR1 is Protective against Systemic Candidiasis

WT mice  Cx3cr1 KO mice

Lionakis et al., *J Clin Invest*, 2013
Targeted Immunogenetic Approaches: **CX3CR1-M280**

Cx3cr1 is important for macrophage survival and protects from death after systemic candidiasis (MICE)

The **CX3CR1-M280** mutation impairs human monocyte survival and is a risk factor for systemic candidiasis (HUMANS)

Lionakis et al., *J Clin Invest*, 2013; Collar et al, *JCI Insight*, 2018

**NEUTROPHILS**
Red: dTomato- *C. albicans*

Green: Cx3cr1 (macrophages)

Grey: Ly6G (neutrophils)
~5% of CGD Patients Develop Invasive Candidiasis

Winkelstein et al., Medicine. 2000
~5% of Patients with Complete MPO Deficiency Develop Invasive Candidiasis

Non-oxidative Cytotoxic anti-Candida Mechanisms of Neutrophils are Largely Unknown

LETTERS

Jagunal homolog I is a critical regulator of neutrophil function in fungal host defense

RESEARCH ARTICLE

CXCR1-mediated neutrophil degranulation and fungal killing promote Candida clearance and host survival
Targeted Immunogenetic Approaches: 

**CXCR1-T276**

Cxcr1 is important for neutrophil killing and protects from death after systemic candidiasis (MICE)

The **CXCR1-T276** mutation impairs human neutrophil killing and is a risk factor for systemic candidiasis (HUMANS)

Swamydas et al., Science Transl Med, 2016

Unbiased Approaches: 

**GWAS**

Immunochip SNP array identifies novel genetic variants conferring susceptibility to candidaemia

TAGAP, CD58, LCE4A-C1orf68
Other important examples:

Cytokine Gene Polymorphisms and the Outcome of Invasive Candidiasis: A Prospective Cohort Study

Toll-like Receptor 1 Polymorphisms Increase Susceptibility to Candidemia

Functional genomics identifies type I interferon pathway as central for host defense against *Candida albicans*

**Polymorphisms in Tumor Necrosis Factor-α Increase Susceptibility to Intra-Abdominal *Candida* Infection in High-Risk Surgical ICU Patients**

Agnieszka Wójtowicz, PhD; Frederic Tissot, MD; Frederic Lamothe, MD; Christina Orasch, MD; Philippe Eggimann, MD; Martin Siegemund, MD; Stephan Zimmerli, MD; Ursula Maria Flueckiger, MD; Jacques Bille, MD; Thierry Calandra, MD, PhD; Oscar Marchetti, MD; Pierre-Yves Rochud, MD; and the Fungal Infection Network of Switzerland (FUNGINOS)

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The Clinical Observations

A surge of small molecule inhibitors for treatment of autoimmune and neoplastic conditions

Predictable and non-predictable effects on anti-*Candida* immunity
The Hypothesis

Novel iatrogenic risk factors for candidiasis

- Fostamatinib (SYK inhibitor)
- Avacopan (C5AR1 inhibitor)
- Danirixin (CXCR2 inhibitor)
CARD9 is centrally positioned in antifungal immune pathways

CARD9 deficiency results in decreased neutrophil accumulation in the CSF

The CARD9\(^{-/-}\) Infected CSF is not Chemotactic to Neutrophils *ex vivo*

![Bar graph showing no chemotaxis](image)

*Card9\(^{-/-}\) Mice Develop Uncontrolled Fungal Brain Infection*

![Graph showing fungal brain infection](image)
Card9 is Required for Neutrophil Accumulation in the Fungal-Infected Brain

No defect in neutrophil:
- Production in bone marrow
- Egress in blood
- Survival
- Cell-intrinsic chemotaxis

No defect in the kidney, no defect in staphylococcal brain infection


A Model of Microglial Engagement for CNS Protective Antifungal Immunity

Drummond et al, under revision
Microglia-Mediated Production of IL-1β and CXCL1 is CARD9-Dependent

Frequency of pro-IL-1β/CXCL1 positive microglia determined by intracellular flow cytometry after 4 hour incubation with Brefeldin A +/- additional stimulation.

CXCL1 is not induced in the CARD9-/- infected CSF
SYK-CARD9 are centrally positioned in antifungal immune pathways

39 trials in ClinicalTrials.gov
- RA
- Lymphomas
- Solid tumors
- Autoimmune cytopenias

CXCR2 is Critical for Neutrophil Recruitment and Function During Systemic Candidiasis

10 trials in ClinicalTrials.gov (COPD, severe lung viral infections)
C5AR1 is Critical for Macrophage Killing During Systemic Candidiasis

**Take Home Messages**

**Phagocytes are crucial for immunity during systemic candidiasis**
- Early neutrophil recruitment
- Early monocyte/macrophage-*Candida* contact
- Non-oxidative killing mechanisms

**Individualized host genetics affect the risk of systemic candidiasis and may provide the platform for personalized risk stratification and prognostication strategies in humans**
- Chemokine/cytokine signaling, PRRs, type I interferon genes

**Basic immunology studies may uncover novel iatrogenic risk factors for candidiasis in patients treated with small molecule inhibitors for autoimmunity/malignancies.**

**Future needs: tissue-specific responses, non-*albicans Candida***

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**Randomized Trial of C5a Receptor Inhibitor Avacopan in ANCA-Associated Vasculitis**

6 trials in ClinicalTrials.gov (ANCA vasculitis, HUS, Glomerulonephritis)
66 trials of eculizumab in ClinicalTrials.gov (HUS, Glomerulonephritis, kidney transplantation, PNH, HSCT)
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