Prevention of candidosis

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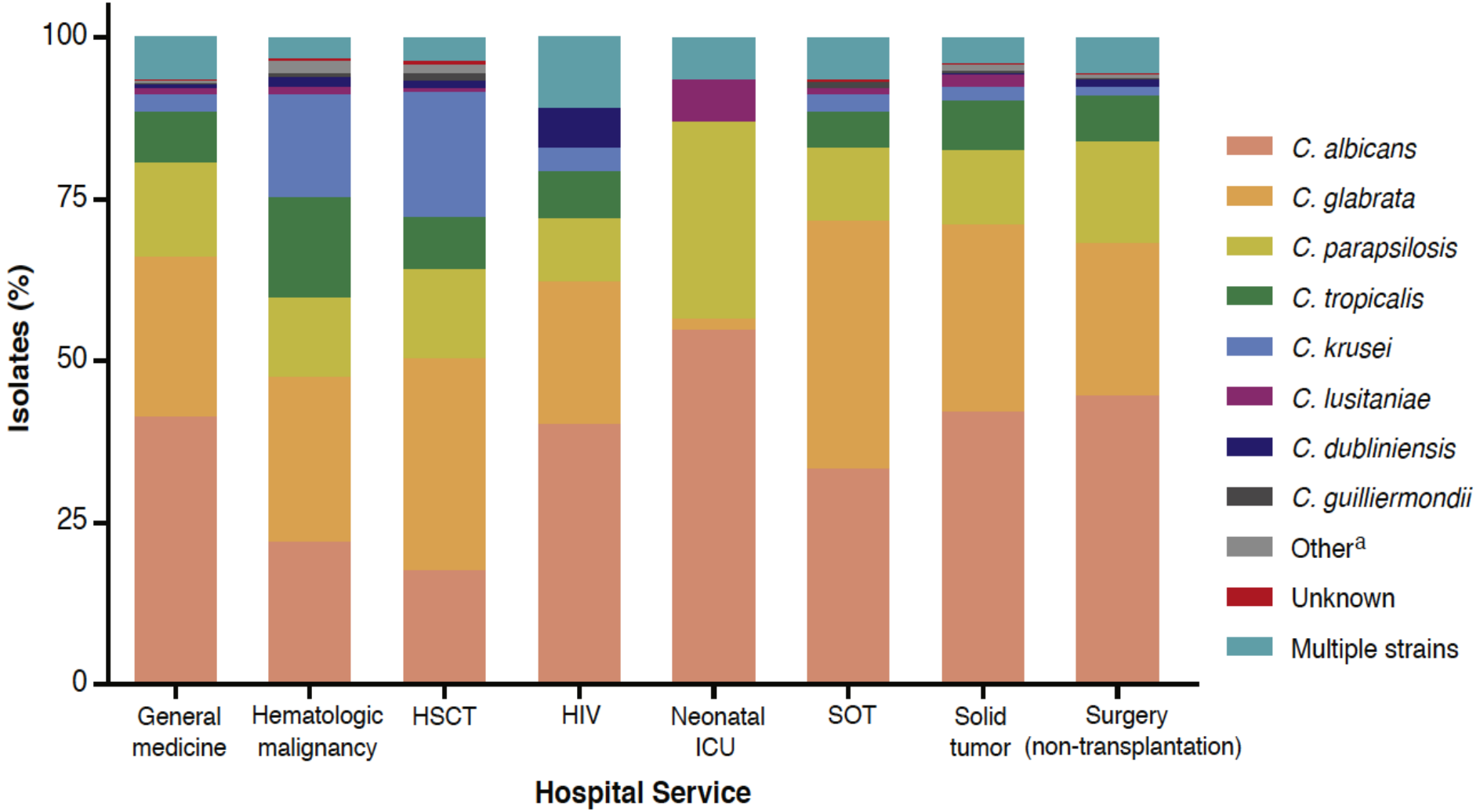
Disclosures

- Speaker from Astellas, Astra Zeneca, Gilead, MSD and Pfizer

- Advisory board membership for Astellas, Astra Zeneca, Cubist, MSD, Pfizer and The Medicines Company

Many units face candida issues

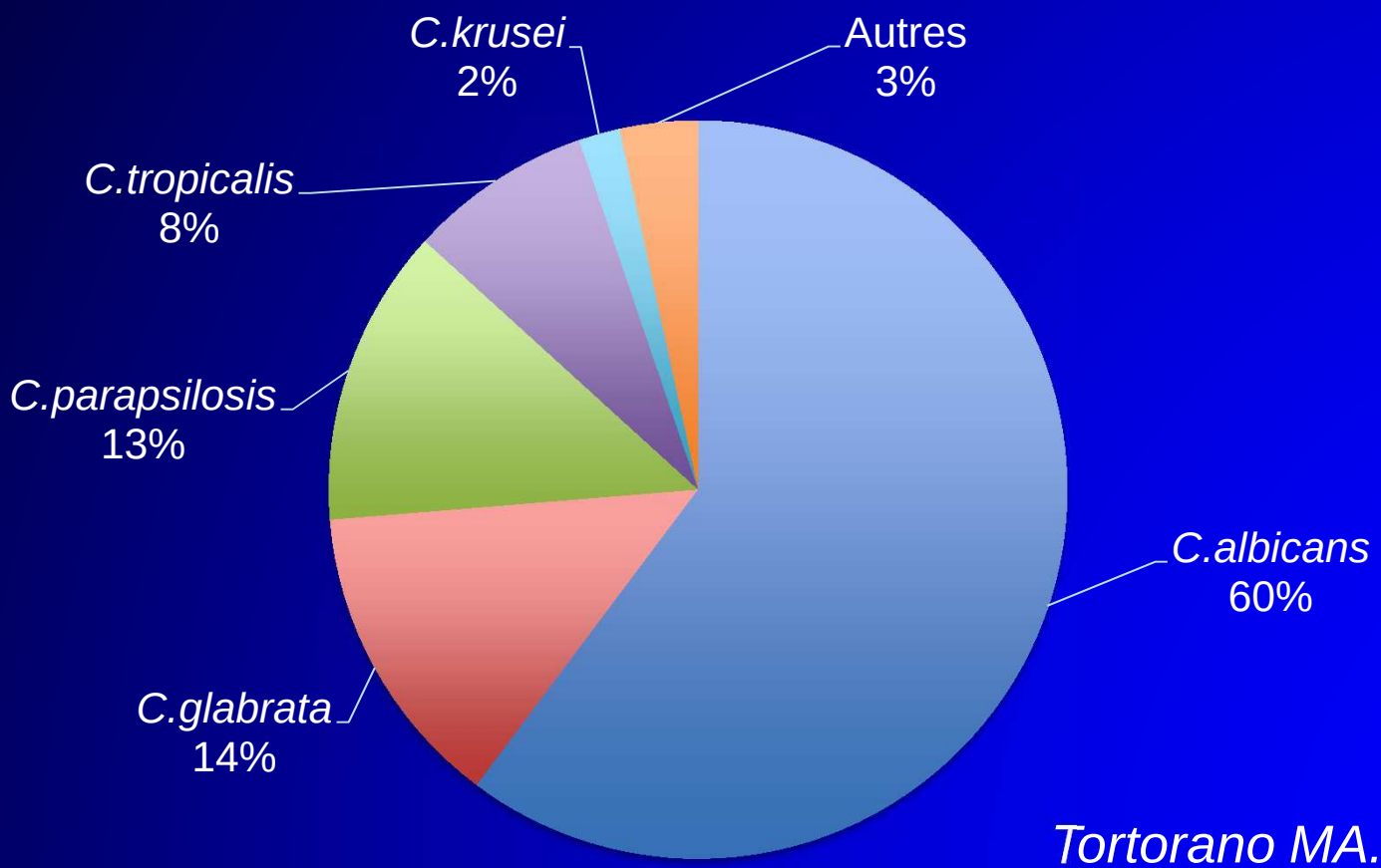


3,648 candidaemias

Pfaller, Diagn Microbiol Infect Dis 2012

ICU as one of the most frequent place

- Multicenter study over 2 years in 38 units
- 384 fungal infections including 318 invasive candidiasis (83%)
- Death rate 46%



Tortorano MA. Mycoses 2012

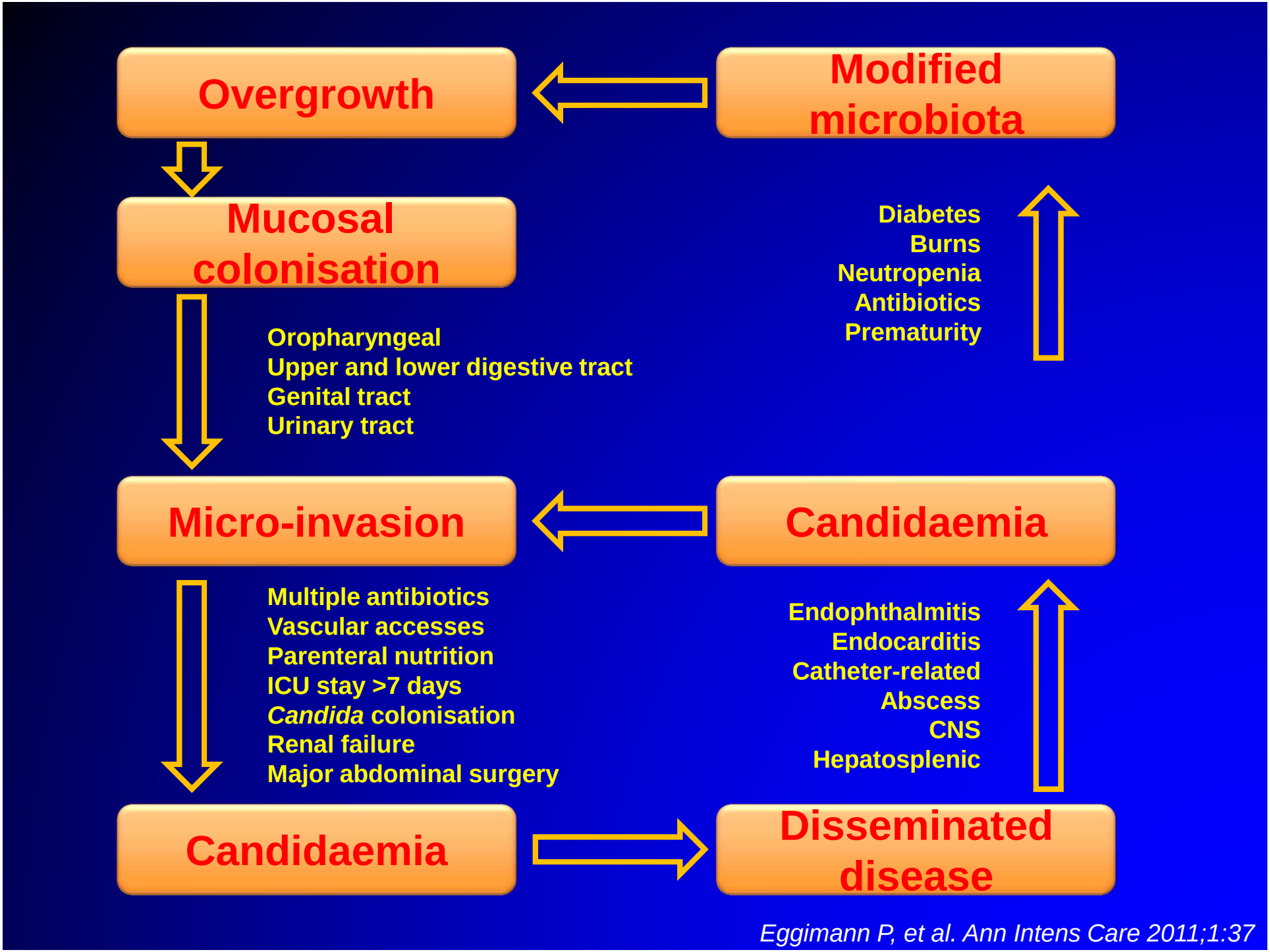
Host factors predisposing to invasive candidiasis

- Candida colonisation
  - Especially if multifocal or heavy
- Exposure to broad spectrum antibiotics

- Central venous catheters
- Total parental nutrition
- Dialysis

- Diabetes
- Steroids
- Chemotherapy

Arendrup MC. Curr Opin Crit Care. 2010;16:445-52



Eggimann P, et al. Ann Intens Care 2011;1:37

Prevention: the act or practice of stopping something bad from happening

Synonyms: averting, forestallment, precluding

Related words: avoidance, circumvention;, negation, neutralization, nullification, baffling, balking, checkmate, crossing…

Merriam Webster Dictionary

= Limiting risk factors

Removing all compromised vascular lines, devices, and implants when possible

Prophylaxis: measures designed to preserve health and prevent the spread of disease **:** protective or preventive treatment

Merriam Webster Dictionary

= Antifungal prophylaxis

Aims of prophylaxis:

…if the risk of a target disease is sharply elevated in a readily identified patient group…

Pappas PG, et al. Clin Infect Dis 2004, 38:161–189

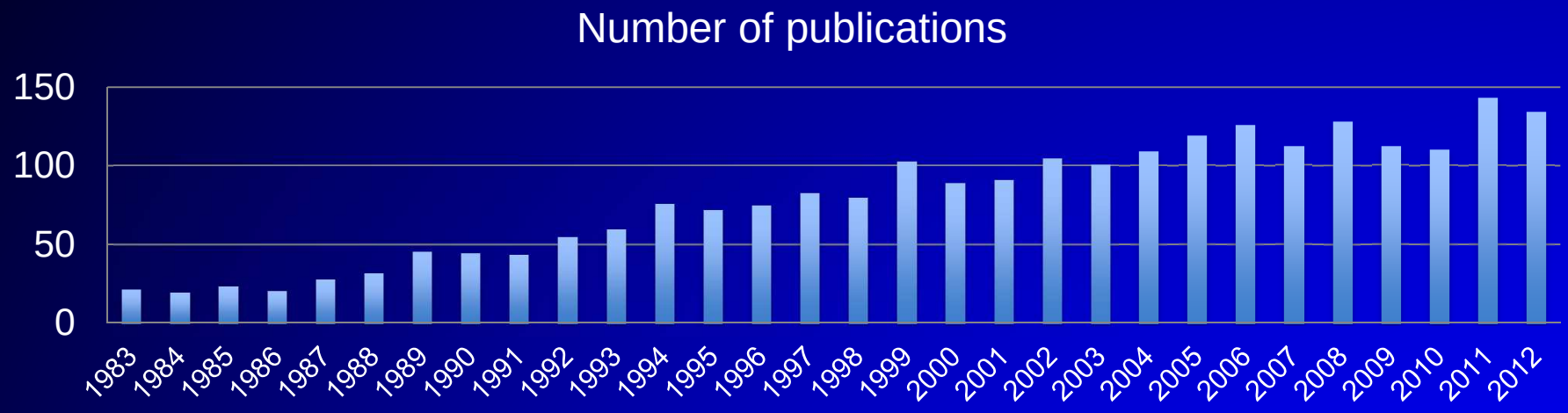
…administering a drug to prevent disease in a high-risk population…

Ostrosky-Zeichner L. Crit Care Med 2006, 34:857-63

…to decrease morbidity and mortality by decreasing fungal burden…

Echeverria PM, et al. Sem Respir Crit Care Med 2011;32:159-73

Candida AND prophylaxis



Overall	2,649 publications (1955-2013)
Cancer patients:	437
Paediatrics:	164
ICU/Crit care patients:	102
Haematology:	80
Guidelines:	97

http://www.ncbi.nlm.nih.gov/pubmed

Clinical Practice Guidelines for the Management of Candidiasis: 2009 Update by the Infectious Diseases Society of America

Pappas PG et al. Clin Infect Dis 2009; 48:503–35

ESCMID* guideline for the diagnosis and management of *Candida* diseases 2012: adults with haematological malignancies and after haematopoietic stem cell transplantation (HCT)

Ullmann AJ et al. Clin Microbiol Infect 2012; 18:53–67

ESCMID* guideline for the diagnosis and management of *Candida* diseases 2012: non-neutropenic adult patients

Cornely OA et al. Clin Microbiol Infect 2012; 18:19–37

ESCMID* guideline for the diagnosis and management of *Candida* diseases 2012: prevention and management of invasive infections in neonates and children caused by *Candida* spp.

Hope WW et al. Clin Microbiol Infect 2012; 18:38–52

And many national guidelines/recommendations…

IDSA ranking recommendations

Table 1. Infectious Diseases Society of America–US Public Health Service Grading System for ranking recommendations in clinical guidelines.

Category, grade	Definition
Strength of recommendation	
A	Good evidence to support a recommendation for or against use
B	Moderate evidence to support a recommendation for or against use
C	Poor evidence to support a recommendation
Quality of evidence	
I	Evidence from ⩾1 properly randomized, controlled trial
II	Evidence from ⩾1 well-designed clinical trial, without randomization; from cohort or case-controlled analytic studies (preferably from >1 center); from multiple time-series; or from dramatic results from uncontrolled experiments
III	Evidence from opinions of respected authorities, based on clinical experience, descriptive studies, or reports of expert committees

NOTE. Adapted from Canadian Task Force on the Periodic Health Examination [15].

IDSA Guidelines. Pappas PG et al. Clin Infect Dis 2009; 48:503–35

ESCMID ranking recommendations

TABLE 1. Definition of the strength of recommendation

Grade	ESCMID EFISG
A	Strongly supports a recommendation for use
B	Moderately supports a recommendation for use
C	Marginally supports a recommendation for use
D	Supports a recommendation against use

TABLE 2. Definition of the quality of evidence

ESCMID EFISG	
Level	
I	Evidence from at least one properly designed randomized, controlled trial
II	Evidence from at least one well-designed clinical trial, without randomization, from cohort or case-controlled analytical studies (preferably from >1 centre); from multiple time series or from dramatic results of uncontrolled experiments
III	Evidence from opinions of respected authorities, based on clinical experience, descriptive case studies or reports of expert committees
Index (for quality of evidence II)	
r	Meta-analysis or systematic review of randomized controlled trials
t	Transferred evidence, that is, results from different patients' cohorts, or similar immune-status situation
h	Comparator group is a historical control
u	Uncontrolled trial
a	Published abstract (presented at an international symposium or meeting)

Ullmann AJ et al. Clin Microbiol Infect 2012; 18:53–67

Prophylaxis in immunosuppressed and cancer patients

Chemotherapy-induced neutropenia

- Fluconazole 400 mg (6 mg/kg) daily (A-I)
- Posaconazole 200 mg 3 times per day (A-I)
- Caspofungin 50 mg daily (B-II)
  during induction chemotherapy for the duration of neutropenia.
- Oral itraconazole 200 mg daily effective alternative (A-I)
  little advantage and is less well tolerated

Stem cell transplant with neutropenia

- Fluconazole 400 mg (6 mg/kg) daily
- Posaconazole 200 mg 3 times daily
- Micafungin 50 mg daily
  during the period of risk of neutropenia (A-I).

IDSA Guidelines. Pappas PG et al. Clin Infect Dis 2009; 48:503–35

Anti-candida prophylaxis for allogeneic haematopoietic stem cell recipients

	Intention: Morbidity reduction		Intention: Survival improvement	
	SoR	QoE	SoR	QoE
Intervention (anti-Candidal prophylaxis) during the neutropenic phase				
Fluconazole 400 mg qd if no prophylaxis is considered	A	I	A	I
Itraconazole* 2.5 mg/kg oral solution tid	B	I	C	I
Posaconazole* 200 mg tid	A	II_t	B	II_t
Voriconazole* 200 mg bid	A	I	C	I
Caspofungin* 70/50 mg qd	C	II_u	C	III
Micafungin* 50 mg qd	A	I	C	I
Anidulafungin	NR	ND	NR	ND
Liposomal amphotericin B 50 mg every other day iv, 100 mg/weekly	B	II	C	III
Intervention (anti-Candidal prophylaxis) during the first 100 days without GVHD and neutrophil recovery				
Fluconazole 400 mg qd	A	I	A	I
Itraconazole* 2.5 mg/kg oral solution tid	B	I	C	I
Posaconazole* 200 mg tid	C	III	C	III
Voriconazole* 200 mg bid	A	I	C	I
Caspofungin* 70/50 mg qd	C	II_u	C	II_u
Micafungin* 50 mg	C	III	C	III
Anidulafungin	NR	ND	NR	ND
Liposomal amphotericin B 50 mg every other day iv, 100 mg/weekly	C	III	C	III
Intervention (anti-Candidal prophylaxis) in GVHD				
Fluconazole 400 mg qd	A	I	C	I
Itraconazole* 2.5 mg/kg oral solution tid	C	I	C	I
Posaconazole* 200 mg tid	A	I	B	I
Voriconazole* 200 mg bid	B	I	C	I

Ullmann AJ et al. Clin Microbiol Infect 2012; 18:53–67

Anti-candida prophylaxis
outside of allogeneic haematopoietic stem cell transplants

(e.g. autologous haematopoietic stem cell transplantation or chemotherapy induced neutropenia)

		Autologous HCT		Severe and prolonged neutropenia	
Intention	**Situation**	**Intervention**	**SoR/QoE**	**Intervention**	**SoR/QoE**
Reduce morbidity and mortality (during and after high dose chemotherapy)		Any prophylaxis	DIII	Any prophylaxis	DIII
Additional antibody treatment (e.g. rituximab)		Any prophylaxis	DIII	Any prophylaxis	DIII
Morbidity reduction or survival advantage*	Neutropenia*	Fluconazole	ND	Fluconazole	CI
		Itraconazole	CII	Itraconazole	CI
		Posaconazole	CIIt	Posaconazole	CIIt
		Voriconazole	ND	Voriconazole	ND
		Anidulafungin	ND	Anidulafungin	ND
		Caspofungin	ND	Caspofungin	CI
		Micafungin	ND	Micafungin	ND
		Nystatin	DIIt	Nystatin	DII
		Any amphotericin B formulation	ND	Any amphotericin B formulation	DI

Ullmann AJ et al. Clin Microbiol Infect 2012; 18:53–67

Prophylaxis
in transplant patients

Solid-organ transplant recipients

- Fluconazole 200–400 mg (3–6 mg/kg) daily
- LAmB 1–2 mg/kg daily

Each for at least 7–14 days
Postoperative prophylaxis
High-risk liver (A-I)
Pancreas (B-II)
Small bowel (B-III) transplant recipients

High-risk liver transplant patients (>2 key risk factors)

- Re-transplantation
- Creatinine level **1**2.0 mg/dL
- Choledocho-jejunostomy
- Intraoperative use of **>**40 U of blood products
- Prolonged intraoperative time (>1 h)
- Fungal colonization detected >2 days before and 3 days after transplantation

IDSA Guidelines. Pappas PG et al. Clin Infect Dis 2009; 48:503–35

Guidelines of the American Society of Transplantation

Infectious Diseases Community of Practice,
Donor-derived fungal infection working group
(guidelines endorsed by the American Society of transplantation)

Prophylaxis for kidney transplant recipients

- When yeast is visualized on stain or Candida species are isolated from the preservation fluid or in recovery of organs from donors with intestinal perforation.
- Fluconazole should be considered as the preferred drug for the treatment or prevention of donor derived candidiasis.

Singh N et al. Am J Transplant 2012; 12: 2414–2428

Guidelines of the American Society of Transplantation

- Liver transplantation

Candida sp identified in the preservation fluid cultures or in pts with surgeries complicated by intestinal contamination during organ recovery empiric AF therapy recommended for 2 weeks

- Pancreas transplantation

If the donor preservation fluid is positive for yeast and in the absence of routine employment of AF prophylaxis, treatment should be initiated as outlined for kidney transplant recipients

- Lung transplantation

AF recommended if donor broncho-pulmonary secretions yield Candida until a repeat bronchoscopy is performed 1 week post-T to evaluate the anastomosis.

Singh N et al. Am J Transplant 2012; 12: 2414–2428

Prevention of fungal disease after lung transplantation

Surveys of antifungal use

37 US centres (66% of all US lung transplantations)
Prophylaxis initiated in 76% of centres
Within 24 hours in 71%, within 1 week in all the centres
Only 59% treated patients colonized with Candida spp.

Dummer JS et al. J Heart Lung Transplant 2004;23:1376-81

International survey
58 centres (USA 45%, Europe 36%)
Prophylaxis within the first 6 months post-T in 34 centers
Universal prophylaxis (given to all recipients immediately postT) directed against Candida spp in 52.9% (voriconazole ± inhaled AmB)

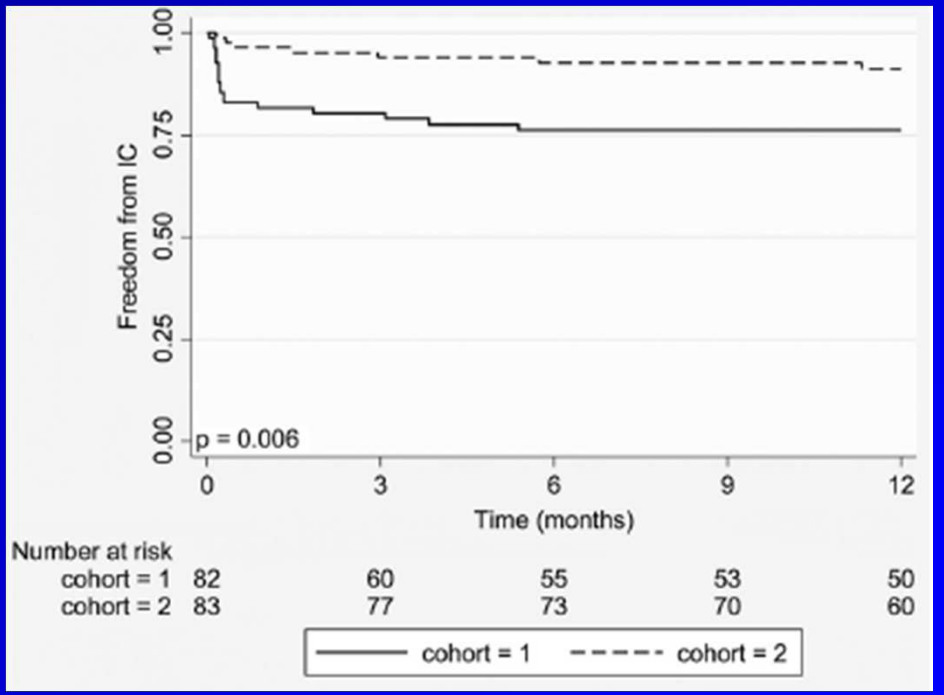
Neoh CF et al. Am J Transplant 2011;11:361-6

Prevention of fungal disease after lung transplantation

Cohort 1: Historic control inhaled AmB 25mg BiD or 20 mg LAmB BiD

Cohort 2: inhaled AmB from Lung transplantation (same policy)
+ 7-10 micafungin 100 mg (BLT)
+ 3-6 months oral AF with yeast or mold in peritransplant cultures or invasive fungal disease on explants

	Cohort 1 (n=82)	Cohort 2 (n=83)
Total IFD cases	29	10
Invasive candidiasis		
Empyema	12[a]	7[b]
Candidemia	4[c]	0
Empyema and candidemia	3[d]	0
Invasive aspergillosis		
Empyema	7[e]	0
Pneumonia	0	1[f]
Tracheobronchitis	1[g]	1[h]
Other IFD	2[i]	1[j]



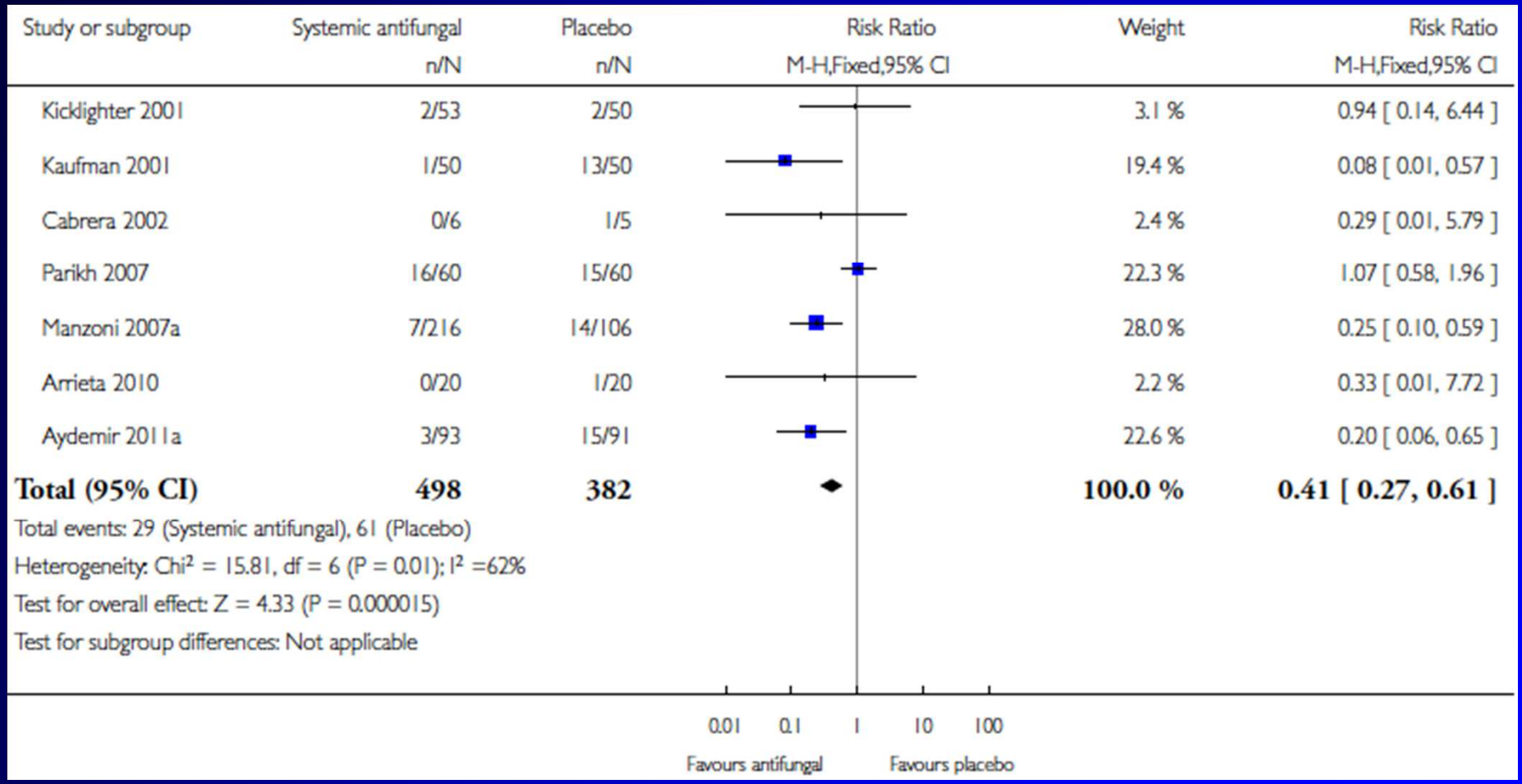
No change in survival between the cohorts within 1 year follow-up

Koo S et al. Transplantation 2012;94:281-6

Prophylaxis in neonates

Prophylactic systemic antifungal agents to prevent mortality and morbidity in very low birth weight infants

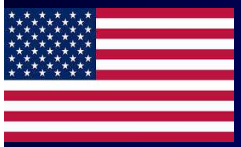
Systemic antifungal agent versus placebo or no drug
Invasive fungal infection



Austin N et al. Cochrane Database of Systematic Reviews 2013, Issue 4. CD003850

In nurseries with high rates of invasive candidiasis, fluconazole prophylaxis may be considered in neonates with birth weights **<**1000 g (A-I).

Antifungal drug resistance, drug-related toxicity, and neurodevelopmental outcomes should be observed (A-III).



IDSA Guidelines. Pappas PG et al. Clin Infect Dis 2009; 48:503–35

Recommendation and grading	Comments
Oral nystatin, 1 mL 100 000 IU Q8 h (B-II)	Reduction in fungal infection, but no change in mortality, potential gut damage & NEC
Miconazole oral gel 15 mg Q8 h (D-II)	Concerns regarding generation of triazole resistance
Lactoferrin 100 mg/day alone or in combination with *Lactobacillus* 10^6 colony-forming units per day from the third day of life until either the end of the sixth week of life or until discharge from the NICU (B-II)	Reduction in fungal infection by *Lactobacillus* and lactoferrin
Fluconazole 3 or 6 mg/kg 2 times per week iv or orally in ALL neonates <1000 g in NICUs with high frequency of IC (A-I)	Reduction in *Candida* colonization, fungal infection, but no change in overall mortality. Concerns for neurodevelopmental toxicity, emergence of resistant species
Fluconazole 3 or 6 mg/kg 2 times per week iv or orally in NICUs with a lower incidence of IC (i.e. <2%) for neonates: (a) with birth weight <1000 g, (b) who have risk factors (i.e. central venous catheters, third-generation cephalosporins and carbapenems) for the development of IC (B-II)	Decision for prophylaxis is on an individual basis

ESCMID Guidelines. Hope WW et al. Clin Microbiol Infect 2012; 18:38–52

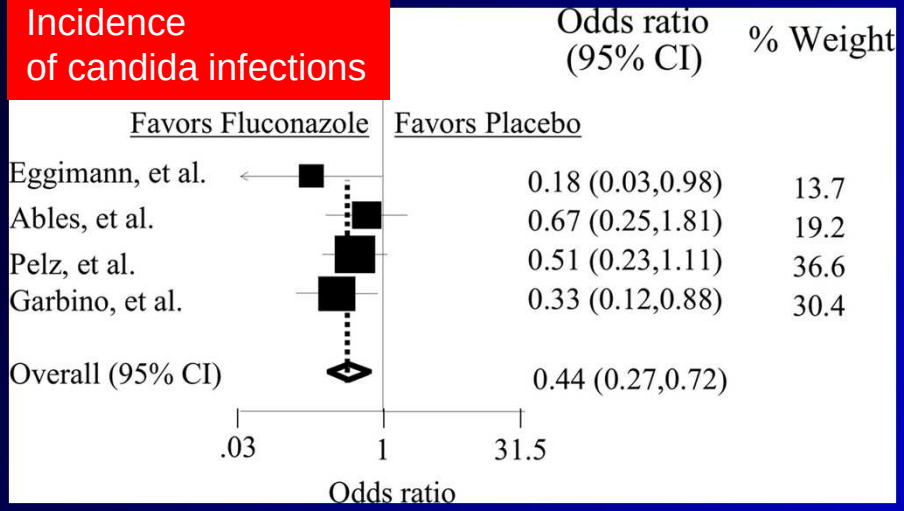
Prophylaxis in ICU patients

Many prophylactic studies in ICU patients

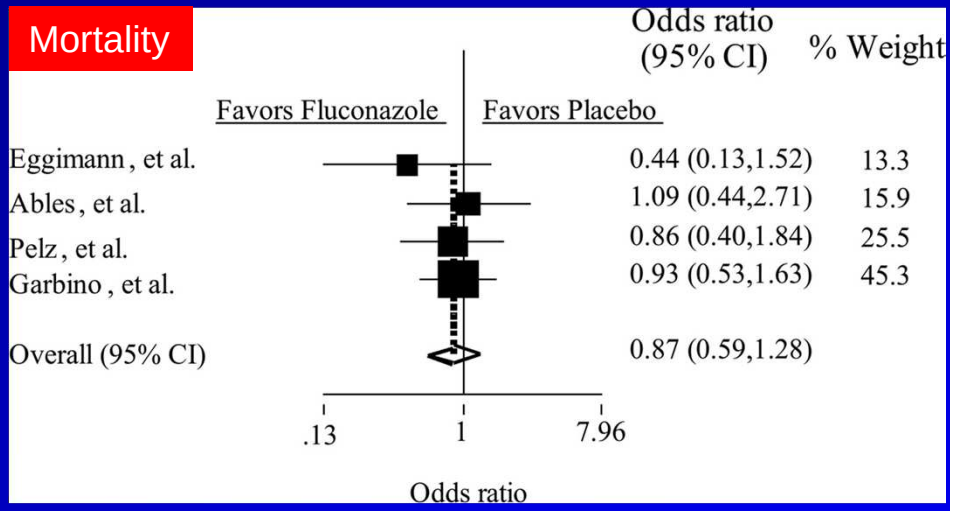
Author	Year	Population	Patients	Colonisation	Number	Treatment	Dose	Duration
Slotman	1987	Surgical ICU	3 risk factors/14	20%	74	ketoconazole	200 mg	ICU LOS
Yu	1993	ICU	Sepsis	NA	56	ketoconazole	200 mg	ICU LOS
Savino	1994	Surgical ICU	ICU LOS ≥ 2d	NA	292	ketoconazole	200 mg	ICU LOS
ARDS Network	2000	Acute lung injury	No risk factors	NA	234	ketoconazole	400 mg	48h after MV
Eggimann	1999	Surgical ICU	Digestive surgery	40%	49	fluconazole	400 mg	16 d
Parizkova	2000	ICU	MV > 48h, ATB > 24h, ICU admi < 5d	NA	38	fluconazole	100 mg	ICU LOS
Pelz	2001	Surgical ICU	ICU LOS ≥ 3d	75%	260	fluconazole	400 mg	ICU LOS
Garbino	2002	ICU	SDD, MV > 48h	48%	220	fluconazole	100 mg	end MV
Sandven	2002	Digestive surgery	Peritonitis	NA	109	fluconazole	400 mg	1 dose
He	2003	Surgical ICU	Acute pancreatitis	NA	75	fluconazole	100 mg	NA
Jacobs	2003	ICU	Septic shock	NA	71	fluconazole	200 mg	Septic shock
Normand	2005	ICU	MV > 48h	NA	98	nystatin	3.10^6 U/d	ICU LOS
Senn	2009	Surgical and ICU	Recurrent g-intest perforation <7d	NA	19	caspofungin	70 mg / 50 mg	Surgical condition

Meta-analyses - Impact of prophylaxis in ICU patients

Incidence of candida infections



Mortality



Shorr A et al. Crit Care Med 2005 ;33:1928-35

Cruciani M et al. Intensive Care Med 2005; 31:1479-87
Playford EG et al. Cochrane Database Syst Rev 2006:CD004920
Vardakas KZ et al. Crit Care Med 2006; 34:1216-24

Meta-analyses – benefits of prophylaxis ?

Authors	Nb studies	Nb patients	Decreased mortality	Decreased candidaemias	Decreased fungal infections
Cruciani	9	1,226	+ (0.61)	+ (0.3)	ND
Shorr	4	626	-	ND	+ (0.44)
Playford	12	1,606	+ (0.76)	ND	+ (0.46)
Vardakas	6	816	-	+ (0.28)	+ (0.26)

Cruciani M et al. Intensive Care Med 2005; 31:1479-87
Shorr A et al. Crit Care Med 2005; 33:1928-35
Playford EG et al. Cochrane Database Syst Rev 2006:CD004920
Vardakas KZ et al. Crit Care Med 2006; 34:1216-24

For ICU patients, fluconazole at a dosage of 400 mg (6 mg/kg) daily is recommended for high-risk patients in adult units with a high incidence of invasive candidiasis (B-I).

IDSA Guidelines. Pappas PG et al. Clin Infect Dis 2009; 48:503–35

Fongiday

SRLF
SFAR
SPILF
INSERM

2047 pts in 169 French ICUs

138 (6.7%) pts received a parenteral antifungal therapy

- 27% Proven invasive candidosis
- 18% Empirical therapy (risk factors of candidaemia)
- 35% Pre-emptive therapy
- 17% Prophylaxis
- 13% Treatment for haemodynamic instability

- 41% Fluconazole
- 24% Caspofungine
- 8% Voriconazole
- 6% LAmB

Azoulay E et al. Crit Care Med 2012;40:813-22

Antifungal prophylaxis has been discussed as a promising approach in ICU patients. At this moment, the optimal target population for antifungal prophylaxis remains unknown, as this question has not been sufficiently addressed in clinical trials. Some special populations though have been enrolled in randomized clinical trials, and recommendations for these can be given.

Recommendations. Fluconazole prophylaxis against invasive candidiasis is recommended in patients who recently underwent abdominal surgery and had recurrent gastrointestinal perforations or anastomotic leakages.

Cornely OA, et al. Clin Microbiol Infect 2012;18(Suppl. 7):19–37

Population	Intention	Intervention	SoR	QoE	
Recent abdominal surgery AND recurrent gastrointestinal perforations or anastomotic leakages	To prevent intraabdominal *Candida* infection	Fluconazole 400 mg/day	B	I	[8]
		Caspofungin 70/50 mg/day	C	II_a	[9]
Critically ill surgical patients with an expected length of ICU stay ≥3 day	To delay the time to fungal infection	Fluconazole 400 mg/day	C	I	[10]
Ventilated for 48 h and expected to be ventilated for another ≥72 h	To prevent invasive candidiasis/candidaemia	Fluconazole 100 mg/day	C	I	[162]
Ventilated, hospitalized for ≥3 day, received antibiotics, CVC, and ≥1 of: parenteral nutrition, dialysis, major surgery, pancreatitis, systemic steroids, immunosuppression	To prevent invasive candidiasis/candidaemia	Caspofungin 50 mg/day	C	II_a	[5]
Surgical ICU patients	To prevent invasive candidiasis/candidaemia	Ketoconazole 200 mg/day	D	I	[22]
Critically ill patients with risk factors for invasive candidiasis/candidaemia	To prevent invasive candidiasis/candidaemia	Itraconazole 400 mg/day	D	I	[21]
Surgical ICU with catabolism	To prevent invasive candidiasis/candidaemia	Nystatin 4 Mio IU/day	D	I	[20]

5 Ostrosky-Zeicher L et al. SHEA annual meeting 2011.
8 Eggimann P et al. Crit Care Med. 1999; 27: 1066-1072.
9 Senn L et al. Intensive Care Med. 2009; 35: 903-908.
10 Pelz RK et al. Ann Surg. 2001; 233:542-548.
20 Cerra FB et al. Arch Surg. 1992; 127: 163-169.
21 Havlicek K et al. Int Surg. 2008; 93: 244-246.
22 Slotman GJ et al. Arch Surg. 1987; 122: 147-151.
162 Garbino J et al. Medicine. 2002; 81: 425-433.

Collateral damages of prophylaxis within ICUs

3 years of routine prophylaxis in ICU
No changes in frequency of *C glabrata* colonization/infection

Magill SS et al. Ann Surg 2009; 249:657-65

9 years survey analysis, decreased non-albicans strains following interruption of fluconazole prophylaxis

Bassetti M et al. J Antimicrob Chemother 2009;64:625-9

Increased incidence of fluconazole resistant candidas strains in ICU following an increased use

Rocco TR et al. Arch Surg 2000; 135:160-5

Risk factors of fluconazole resistant strains : neutropaenia (OR : 4.94; *P*=0.008), chronic renal failure (4.82; *p*=0.01) and previous use of fluconazole (5.09; *p*=0.004).

Garnacho-Montero J et al. Antimicrob Agents Chemother 2010; 54:3149-54

Collateral damages of prophylaxis in other units

Six breakthrough Candida infections in HSCT patients receiving voriconazole prophylaxis, five *C glabrata* and one *C krusei*, all with voriconazole trough levels <2 µg/ml

Trifilio S et al. Bone Marrow Transplant 2007;40:451-66

HSCT or acute myelogenous leukemia patients receiving prophylactic Posaconazole, Itraconazole, or Fluconazole.
Decreased *C albicans* colonization increased *C glabrata* with P and I , and increased *C krusei* with F. Increased MICs more than 4-fold in 40% of *C glabrata* isolates.

Mann PA et al. Antimicrob Agents Chemother 2009; 53:5026-34

Fluconazole resistant strains among 383 invasive Candida sp. isolates from HSCT and SOT :
No significant link in HSCT patients between prophylaxis and fluconazole resistance (any fluconazole use in the 3 months prior to the IFI (OR: 2.66[0.93-7.62], p=0.069).
In SOT recipients, association with any fluconazole use in the 3 months prior to the IFI (OR: 2.65[1.17-5.99]).

Lockhart SR J et al. J Clin Microbiol 2011; 49:2404-10

In summary many questions with prophylaxis

It may be an option in some specific subpopulations

But the conditions of use should be clarified

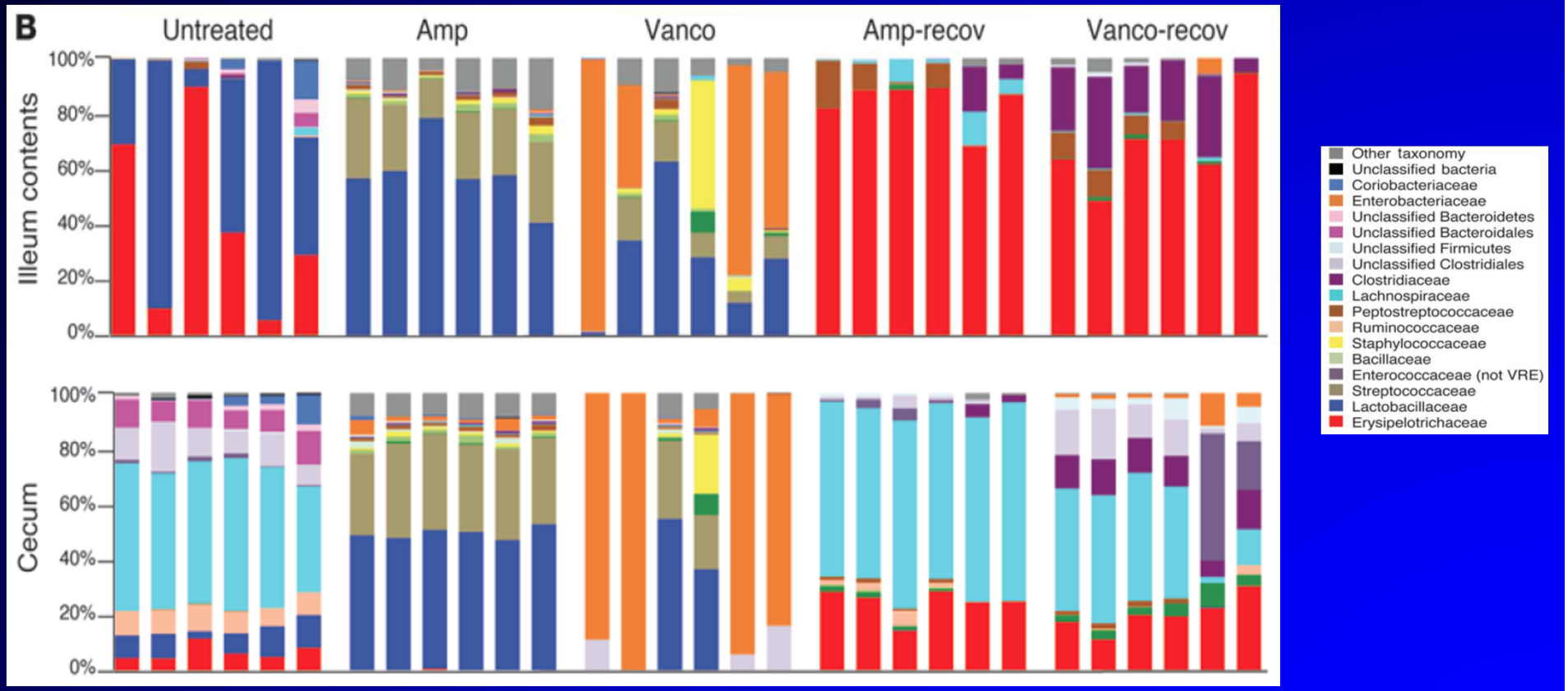
- What is the ecologic price to pay
- For whom, when, how
- What agent, what dose
- What monitoring
- When do we stop
- Is it cost effective
- What should be the role of the laboratories
- Any role of the biomarkers

…

What does the future hold ?

Persistent disturbances following antibiotic therapy

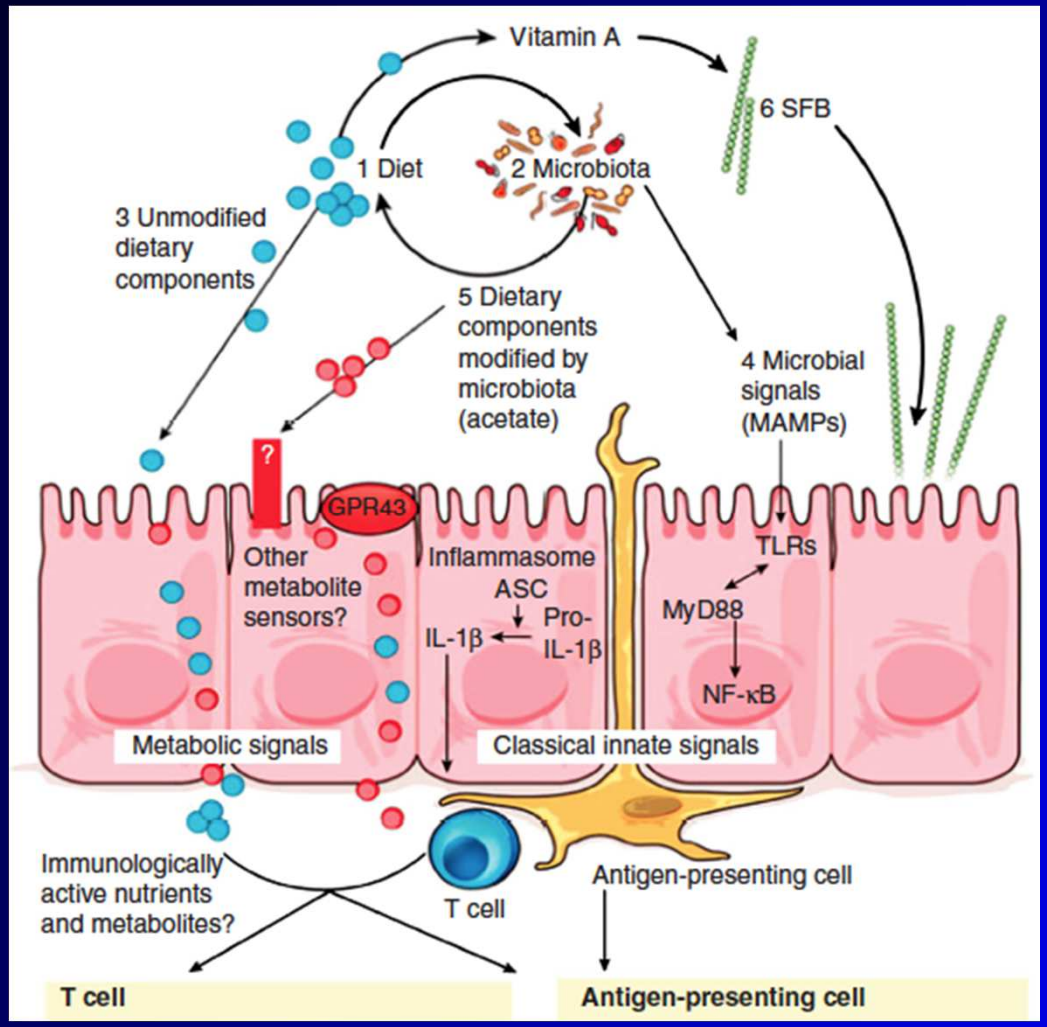
Samples collected from untreated mice, receiving ampicillin or vancomycin or allowed to recover for 2 weeks from antibiotic treatment



Color code for the most predominant bacterial taxa found in the murine intestine
Phylogenetic classification of 16S rDNA frequencies in the ileum or cecum contents

Ubeda C et al. J Clin Invest 2010;120: 4332-4341

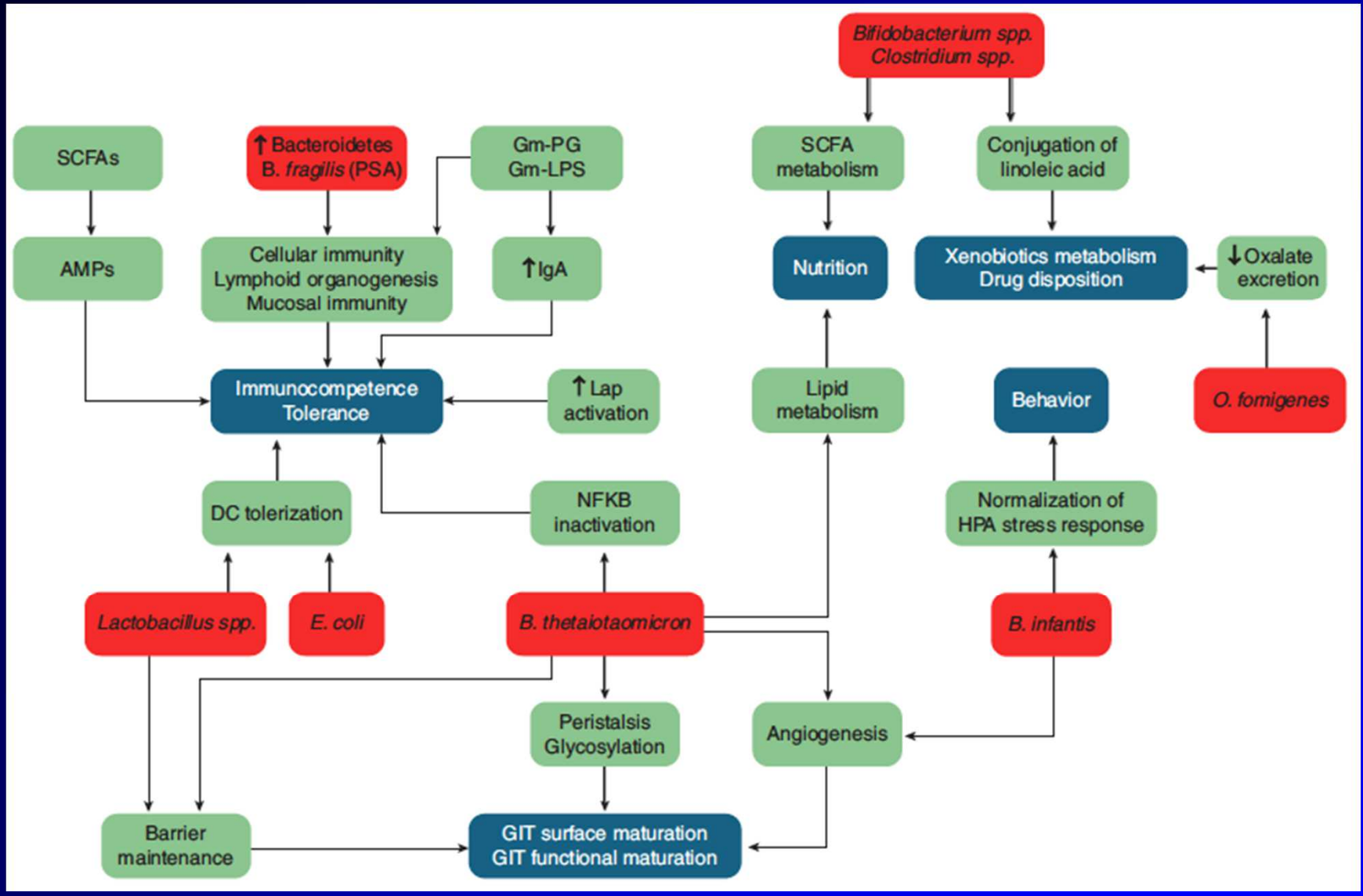
Inter-relationships of nutrients, immune responses and the microbiome



Nutrients (1) influence our microbiota (2) which, in turn, changes the nutritional value of the consumed food. (3) Absorbed dietary components interact with a variety of immune cells. (4) Microbial signals in the form of Microbe Associated Molecular Patterns (MAMPs) also modify local mucosal immune responses through innate signaling pathways, e.g., the inflammasome or Toll-like receptors (TLRs) . (5) Additionally, microbe-modified dietary components (e.g., acetate produced from fermentation of polysaccharides) provide signals by which the immune system can monitor the metabolic activities of the microbiota.
(6) Vitamin A can modify the representation of segmented filamentous bacterium (SFB) in the mouse gut microbiota; SFB induce differentiation of Th17 cells.

Kau AL et al. Nature 2011;474 : 327 – 36

Effects of intestinal microbiota and host physiology



Members of the microbiota in red. Microbial effects on the host in green. Affected host phenotypes in blue. AMP, antimicrobial peptides; DC, dendritic cells; Gm, Gram negative; HPA, hypothalamus-pituitary adrenal; Iap, intestinal alkaline phosphatase; PG, peptidoglycan; PSA, polysaccharide.

Sekirov I et al. Physiol Rev 2010 ; 90 : 859 – 904 .

Ecologic approaches

Probiotics

Live micro-organisms
Administered in adequate amount
To confer a health benefit to the host

Food Agricultural Organization／World Health Organisation (FAO／WHO)

- Non-pathogenic
- Stable in acid and bile
- Able to adhere to human mucosa
- Colonise the gut
- Retain viability during storage and use
- Scientifically demonstrated to be having beneficial physiological effects
- Scientifically demonstrated to be safe

- Used to improve microbial balance
- To confer health benefit to recipients

Floch MH. J Clin Gastroenterol 2010; 44:S19–21.

Probiotics

Many different microorganisms considered as probiotics:

- Lactobacillus sp (*L. acidophillus*, *L. casei*, *L. rhamnosus*)
- Bifidobacterium sp (*B. bifidum*, *B. longum*, *B.lactis*)
- Enterococcus sp (*E. faecalis*, *E. faecium*)
- Saccharomyces (*S. boulardii*, *S. cervesie*)

Most commonly used : Lactobacillus sp and Bifidobacterium sp

Kligler B et al. Am Fam Physician. 2008;78: 1073–8
Kumar S et al. Mycoses. 2013,56:204-211

Probiotics

Prevention/improvement the diseases in which the gut and the disorders of its microbial flora play a major role

Attempts in bacterial colitis including antibiotic-induced diarrhoeas caused by *Clostridium difficile* and ventilator-associated pneumonias

Gu WJ, et al. Chest 2012; 142(4):859-68.
Mihatsch WA et al. Clin Nutr 2012; 31:6–15
Floch MH et al. J Clin Gastroenterol 2011; 45 Suppl:S168–S171

- Competitive context within the intestinal lumen
- Limit the access of yeast to substrates and nutrients

Rehman A et al. BMC Microbiol 2012; 12:47

- Modulate the local/systemic inflammatory response
- Enhance the host antimicrobial defense

Villena J et al. Microbiol Immunol 2011; 55:434–45

- Reduce Candida sp. growth and/or by defending the epithelium against yeast invasion
- Likely to shift the local host cytokine response toward an anti-inflammatory pattern
- Limiting tissue damage and preventing any loss of gut permeability

Arribas B et al. Eur J Nutr 2012; 51:365–74

Probiotics in preterm neonates

Supplementation with oral *Lactobacillus casei* subspecies *rhamosus* GG [Dicoflor 60; Dicofarm spa]; 6.10^9 cfu/day) administered from the third day of life until either the end of the sixth week of life or until discharge from the NICU

Control neonates who were not receiving LGG supplementation

Result or outcome	Neonates receiving LGG supplementation (n = 39)	Control neonates (n = 41)	RR (95% CI)	P
Cultures specimens obtained per neonate, mean no.	8.2	8.6	…	.38
Positive culture results[a]	18/306	59/341	…	.009
Fungal isolates obtained, mean no.				
From each neonate	0.4	1.5	…	.005
From each colonized neonate	1.9	3.1	…	.005
Incidence of gastrointestinal colonization[b]				
Overall, no. (%) of neonates	9 (23.1)	20 (48.8)	0.315 (0.120–0.826)	.01
Incidence of necrotizing enterocolitis, %				
Surgical	0	2.5	…	.51
Stage II	2.5	5	…	.51
Incidence of sepsis, %				
Due to nonfungal agents	37.5	42.5	…	.35
Due to gram-positive bacteria	20	22.5	…	.42
Due to gram-negative bacteria	17.5	20		.55
Incidence of IFI, %				
Overall	10.3	12.2	…	.53
Overall incidence of retinopathy of prematurity, %	40	45	…	.30
Mortality rate before discharge from NICU, %	12.5	15.4	…	.51

Manzoni P et al. Clin Infect Dis 2006; 42:1735–1742

Probiotics in paediatric ICU

3 months to 12 yrs old patients on broad spectrum AB > 48 hrs

- Probiotics EUGI (Wallace pharma, Goa, India) (*Lactobacillus acidophilus* [0.24 billion CFU], *Lactobacillus rhamnosum* [0.24 billion CFU], *Bifidobacterium longum* [0.24 billion CFU], *Bifidobacterium bifidum* [0.24 billion CFU], *Saccharomyces boulardii* [0.05 billion CFU], *Saccharomyces thermophilus* [0.24 billion CFU], fructo-oligosaccharides [300 mg], and lactose as base)
- Placebo

Results	Probiotic Group ($n = 75$)	Placebo Group ($n = 75$)	p[a]
Patients colonized on day 0, *n* (%)	15 (20)	15 (20)	1
Patients colonized on day 7, *n* (%)[b]	19 (27.9)	29 (42.6)	0.07
Patients colonized on day 14, *n* (%)[c]	21 (31.4)	34 (50)	0.02
Patients colonized during study period, *n* (%)	32 (42.6)	45 (60)	0.03
Number of rectal swabs positive for *Candida*[d]	55/210	78/211	0.01
Patients with candiduria, *n* (%)	13 (17.3)	28 (37.3)	0.006
Patients with candidemia, *n* (%)[e]	1 (1.61)	4 (6.35)	0.36
Outcome			
Number that died, *n* (%)	8 (10.7)	7 (9.3)	0.78

Kumar S et al. Crit Care Med 2013; 41:565–572

Synbiotic use

A synbiotic is defined as the combination of

- a probiotic
- a prebiotic: oligosaccharide indigestible by humans but able to be fermented by beneficial gut bacteria such as Lactobacillus and Bifidobacterium spp., therefore promoting their growth

Synbiotic composed of
2×10^{11} freeze dried viable *Bifidobacterium longum* in a gelatin capsule
a sachet containing 6 g of prebiotic fructo-oligosaccharide/inulin mix (Synergy 1; Orafti, Tienen, Belgium)
Administered twice daily for four weeks compared to a placebo

- Treatment of ulcerative colitis
- 18 patients treated one month
- Double blinded randomised controlled trial
- Improvement of the full clinical appearance of chronic inflammation and decreased transcription level of epithelium related immune markers

Furrie E, et al. 2005. Gut 54:242–249.

Effects of synbiotic use on fungal growth

Experimental trial reproducing Percutaneous Endoscopic Gastrostomy tube

Analyses of the biofilm on the surface of the tube

Addition of a synbiotic post-insertion of PEG

Lactobacillus acidophilus DUN-311 (university of Dundee), *Bifidobacterium bifidum* strain BB-02 (Rhodia food, Cranbury, NJ), *Bifidobacterium lactis* BL-01 (Rhodia food)
Prebiotic Synergy 1 (Orafti, Tienen Belgium) fructo-oligosaccharide/inulin (chicory oligofructose produced by partial enzymatic hydolysis of inulin)

- Reduced biofilm formation
- Significant reduction in numbers of *E. coli* and *K. pneumoniae* bacteria

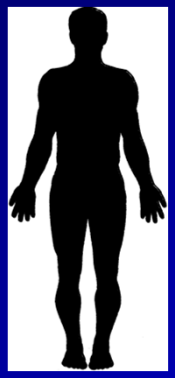
Synbiotic prior to the PEG insertion inhibits colonization by *Candida albicans* and *C. famata*

Smith AR et al. FEMS Microbiol Ecol 2012;80:135-145

Emerging questions raised by probiotics

- Better understanding of the microbiota
- What patients, when, how
- Immunosuppressed and neutropenic patients
- Persisting ileus
- Mixture of probiotics or a single strain
- Adjunction of probiotics in case of invasive candidiasis

Microbiome restauration

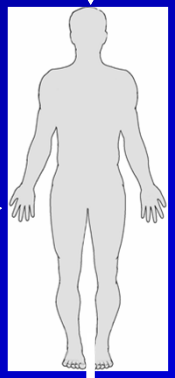


Normal microbiome

Antibiotic pressure on the microbiome

Fecal microbiota banking with auto transplant

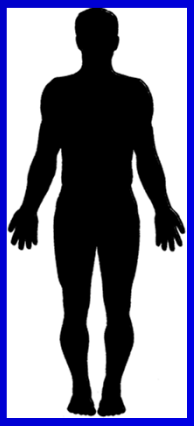
Fecal microbiota with allo transplant



Perturbed microbiome

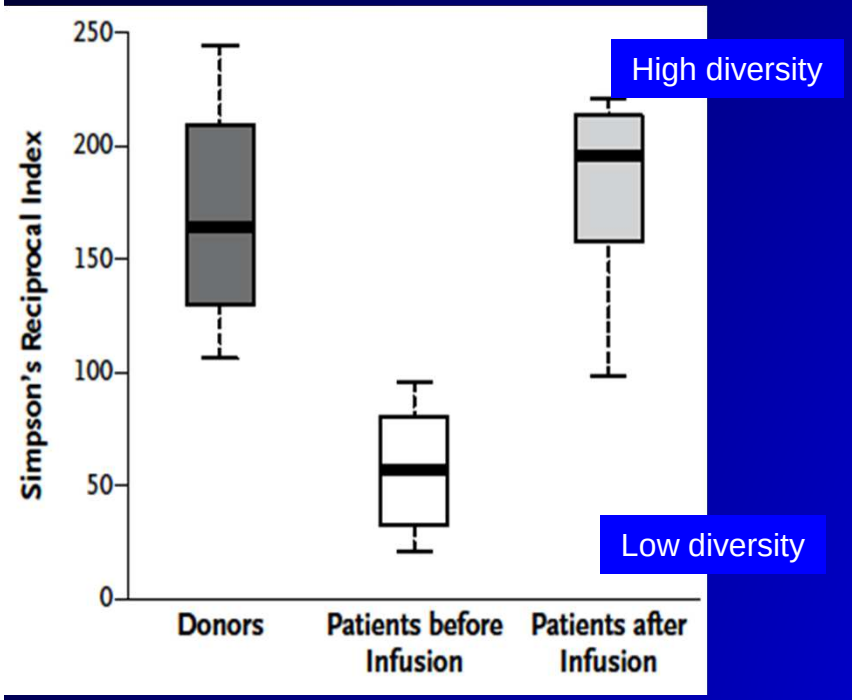
Advanced probiotics

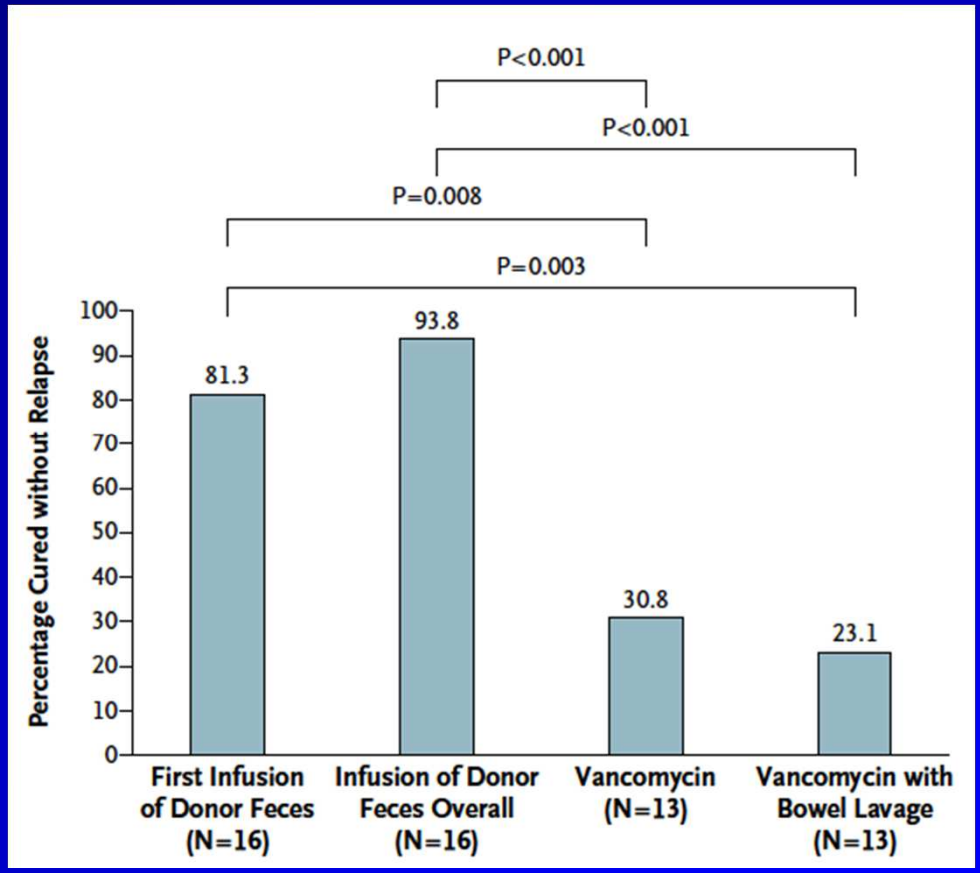
Colonisation and/or clonal expansion prevented



Tosh PK et al. Clin Infect Dis 2012;54:707-13

Duodenal Infusion of Donor Feces for Recurrent *Clostridium difficile*





Van Nood E et al. N Engl J Med 2013;368:407-415

Immunologic approaches

Major candida targets for active and passive immunisation

	Antigens	Underlying immunity
Whole cells and ill-defined cell extracts		
Candidiasis	Strain CA2, live-attenuated	T-helper 1, cell-mediated immunity
	Ribosomal cell fraction	Antibodies and cell-mediated immunity
	Inactivated whole cells	Undefined
Antigen-pulsed cells and T cells		
Candidiasis	Dendritic cell loaded with candida antigens	Cell-mediated immunity, T-helper 1
Subunit and glycoconjucates		
Candidiasis	Agglutinin-like sequences	Cell-mediated immunity
	Secreted aspartic proteinase2	Anti-Sap2 antibodies
	65 kDa mannoprotein	Adhesin-neutralising antibodies
	β-1,3-glucan	Growth-inhibitory and cytocidal antibodies
	β-1,2-mannosides	Antibodies (opsonophagocytic; possibly adherence-blocking)
Idiotypes and mimotopes		
Candidiasis	Killer-toxin neutralising mAb KT4	Fungicidal antibodies
Antibodies		
Candidiasis	Mycograb, anti-Hsp90 peptide†	Unknown
	Anti-β-1,3-glucan mAb 2G8	Growth-inhibitory
	mAb C7 (stess mannoprotein)	Candidacidal
	Single chain fragment variable of anti-idiotypic antibodies	Candidacidal antibodies
	Anti-mannan mAb C6	Opsonophagocytic
	Anti-glycosyl mAb	Candidacidal
	Anti-Sap2 and anti-MP65 domain antibodies	Enzyme and adhesion-neutralising

Cassone A. Lancet Infect Dis 2008;8:114-24

Antibodies used for experimental and clinical passive vaccination

Antibody format	Disease	Setting
Polyclonal	Candidiasis (invasive and mucosal)	Experimental
Monoclonal murine	Candidiasis (invasive and mucosal)	Experimental
Monoclonal human	Candidiasis (invasive)	Experimental and clinical
Single-chain, fragment variable	Candidiasis (invasive and mucosal)	Experimental
Antibody domains	Candidiasis (mucosal)	Experimental
Antibody-derived peptides	Candidiasis (invasive and mucosal)	Experimental

Cassone A. Lancet Infect Dis 2008;8:114-24

Vaccines

Major subunit vaccine candidates against candidiasis infection

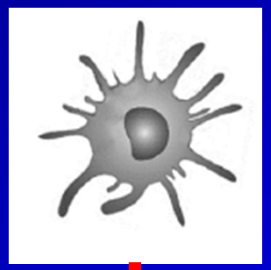
Vaccine	Nature of protective immunity-	References
β-mannan-peptide or protein conjugates	Opsonic, Antibody-mediated	15,20
HyR1	Antibodies neutralizing Candida evasionfrom neutrophil killing	21
Recombinant Als 3* proteins.	Th17-Th1 activity (Abs as surrogate markers or predictors of protection)	22,43,49
Recombinant Sap2* proteins	Antibodies neutralizing Sap activity (enzyme, adhesion and/or others)	16,18
Laminarin-CRM197 conjugate	Anti-beta-glucan Abs with direct anti-Candida activity,	33,44,45

15. Xin H et al. Clin Vaccine Immunol. 2011; 18:1656–67.
16. Sandini S et al. FEMS Immunol Med Microbiol.2011; 62:215–24.
18. De Bernardis et al. Vaccine. submitted.
20. Xin H et al. Proc Natl Acad Sci U S A. 2008;105:13526–31.
21. Luo G et al. J Infect Dis. 2010; 201:1718–28.
22. Spellberg BJ et al. J Infect Dis. 2006; 194:256–60.
33. Torosantucci A, et al. PLoS One. 2009; 4(4):e5392.
43. Lin L et al. FEMS Immunol Med Microbiol. 2009; 55:293–5.
44. Torosantucci A et al. J Exp Med. 2005; 202:597–606.
45. Chiani P et al. Vaccine. 2009; 27:2513–519.
49. Bequin B et al. J Infect Dis. 2010; 201:475–479.

Cassone A et al. Curr Opin Microbiol. 2012; 15(4): 427–433

Th17 cells as a major cellular platform for antifungal defense and vaccination

Pathogens Associated Molecular Patterns (Beta-glucan, mannoproteins, GXM, etc) expressed on fungal surface and/or antigen stimulation of dendritic cells, macrophage pattern recognition receptors (as Toll-like receptors), Dectin-1, mannose receptors and others



Dendritic cell

Intracellular signalling

Cytokines

I-17, IL-22, IL-21

Activation-recruitment of antifungal humoral and cellular effectors antifungal defensins, chemokines, inflammatory cytokines, PMN (neutrophils)

Transcription factors, inflammation activation and cytokines such as type 1-IFN. IL-10, IL-12, IL-23

Cassone A et al. Curr Opin Microbiol. 2012; 15(4): 427–433

Two vaccine antigens of *C. albicans* under clinical trial

Als 3	Sap 2
A cell surface, GPI protein member of Als adhesin family without known enzymatic activity: Interacts with various members of host integrin family	A major member of secreted aspartic proteinase family of *C. albicans* with direct or indirect adhesin activity. May interact and hydrolyze various host immunologically relevant proteins such as complement antibodies and epithelial structural proteins such as E-cadherin
Involved in biofilm formation	No apparent role in biofilm (other members of Sap family may be involved)
Modulates iron acquisition by hyphae	A classical metabolic role as proteinase of *C. albicans*, both in yeast and hyphae
Candida-colonized subjects have both CMI and Ab responses against Als 3	Low or no levels of antibodies and CMI responses in *Candida*- colonized subjects, likely because of Sap2 low immunogenicity in its natural form
Vaccine induces protection through elicitation of Th1 and Th17 cells, then the cohort of antifungal humoral and cellular factors acting locally and promoting inflammation	Vaccine induces protection through elicitation of neutralizing antibody at vaginal level.
Vaccine target: candidiasis systemic and mucosal	Vaccine target: recurrent vulvovaginal candidiasis

Cassone A et al. Curr Opin Microbiol . 2012 August ; 15(4): 427–433

Issues raised by antifungal vaccines

- Need of such vaccines?
- Efficacity in immunodeficient hosts?
- Prevention against disseminated disease?
- Preservation of the role of *C. albicans* as a member of the normal microbiota?
- Prevention or initiation of allergic manifestations?

Mochon AB et al. Med Mycol 2005;43:97–115.
Deepe GS Jr et al. Med Mycol 2005;43:381–389.
Cole GT, et al. Med Mycol 2004;42:189–216.
Deepe GS Jr. Expert Rev Vaccines 2004;3:1–9.
Casadevall A et al. Med Mycol 2005;43:667–680.
Wüthrich M, et al. J Exp Med 2003;197:1405–1416.
Noverr MC, et al.Trends Microbiol 2004;12:562–568.
Noverr MC, et al.Infect Immun 2005;73:30–38.
Noverr MC et al. Infect Immun 2004;72:4996–5003.
Goldman DL, et al. J Infect Dis 2006;193:1178–1186.
Pappagianis D. Fungal Genet Biol 2000;32:1–9.

In summary

- Many unsolved issues
- Prevention based on common sense
  - Remove all unnecessary devices
  - Early interruption of unnecessary antibiotic agents
- Interest of prophylaxis
  - In certain fields
  - To be confirmed in many others
  - Consider collateral effects
- Interesting developments for the future