Gemelli

Fondazione Policlinico Universitario A. Gemelli
Università Cattolica del Sacro Cuore

Update sulla diagnostica micologica e antimicogramma

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Infection	Global burden estimates in main underlying disease groups				
	None	HIV/AIDS	Respiratory	Immune deficit	Critical care
Life-threatening infections					
Invasive aspergillosis			>260,000	>90,000	>100,000
Candida bloodstream infection	50,000			600,000	300,000
Candida peritonitis (intra-abdominal)				42,500	>60,000
Cryptococcal meningitis	10,000's	223,000		10,000's	
Pneumocystis pneumonia		~400,000		>100,000	
Histoplasmosis	>25,000	>25,000	?10,000	>10,000	
Coccidioidomycosis	~150,000	>500		5,000	
Mucormycosis	>5,000			>150,000	>5,000
Talaromycosis (*T. marneffei* infection)		>10,000			
Superficial fungal infections					
Oral thrush		1,900,000	100,000's	millions	
Oesophageal candidasis		537,000		>100,000	
Recurrent Candida vaginitis	~138,000,000				
Candiduria				>200,000	>150,000
Fungal hair infection	200,000,000				
Onychomycosis, ringworm, tinea pedis and other skin fungal infections	>700 million	10,000,000			
Chronic fungal infections					
Chronic pulmonary aspergillosis			3,000,000		
Aspergillus bronchitis			>20,000		
Chronic invasive/granulomotous fungal rhinosinusitis	>200,000				
Paracoccidioidomycosis	~2500				
Allergic fungal infections					
ABPA in asthma and cystic fibrosis			4,800,000		
Severe asthma with fungal sensitisation			>6,500,000		
Fungal rhinosinusitis	~6,000,000				
Fungal NTDs					
Fungal keratitis	>1,000,000				
Sporotrichosis	>40,000				
Mycetoma	>20,000				
Chromoblastomycosis	>25,000				
Totals	>1,044,002,500	~13,095,500	>14,690,000	>2,195,000	>615,000

LEADING INTERNATIONAL FUNGAL EDUCATION

Get The Facts

- Over 300 million people are acutely or chronically infected by fungi, leading to death, long term illness, blindness, psychological problems and reduced work capacity.
- Many recent improvements in diagnostics and treatment have not reached treating clinicians in all countries, and access to appropriate diagnostics and simple antifungal agents is far from universal. **This needs to change**.
- LIFE aims to provide the latest information on the most common human pathological fungi, the diseases they cause and their diagnostics and treatment.
- LIFE is led by Professor David Denning who has been caring for patients with fungal infection for more than 30 years.

"The Fungal Infection Trust. How common are fungal diseases? Fungal Research Trust 20th Anniversary meeting. London June 18th 2011, updated July 2019."

“Classical” cultures, still an important tool for fungal infections diagnosis

Culture approach for fungal infections diagnosis

- Important to use specific fungal media, bacteriological media are less efficient
- At least 3 sputum specimens should be submitted for fungal culture when fungal infections are suspected
- In Invasive Aspergillosis, BAL cultures are positive in ~30% of the samples
- Cultures take 1 to 10 days to grow, so please incubate accordingly in the Lab!
- Identification require time but the MALDI-TOF approach changed completely this scenario

From: Fraczek et al, Mycoses, 2014; Ullmann *et al*., CMI, 2018

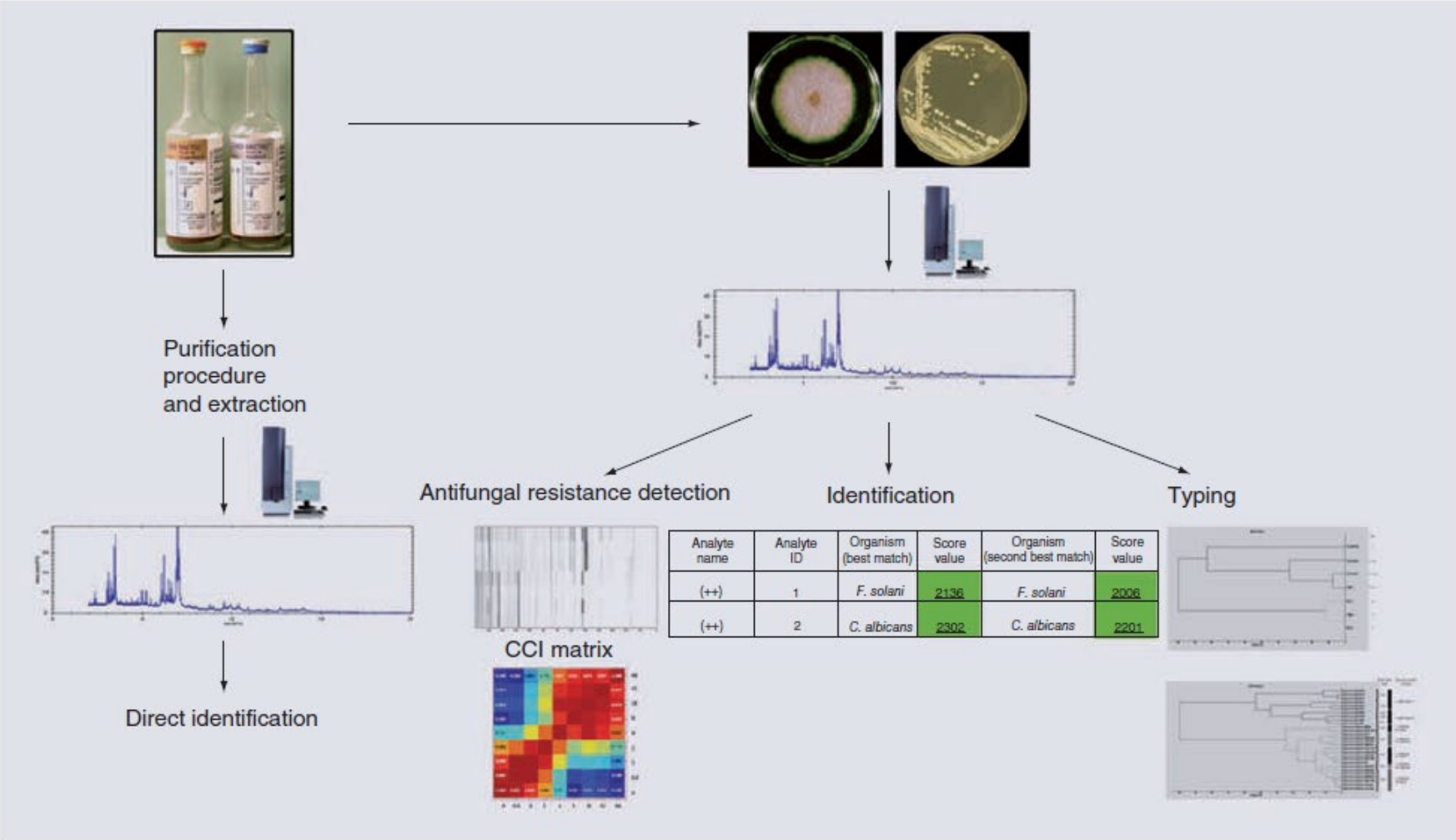


Figure 1. The role of MALDI-TOF mass spectrometry in clinical mycology diagnostics. Principal applications such as identification, typing and antifungal susceptibility testing of yeasts and molds starting from colonies, and direct identification of positive blood cultures are shown.
CCI: Composite correlation index.

From: Posteraro *et al*., Expert Rev. Proteomics, 2013

Centers for Disease Control and Prevention
MMWR
Morbidity and Mortality Weekly Report
Early Release / Vol. 65
November 4, 2016

Investigation of the First Seven Reported Cases of *Candida auris*, a Globally Emerging Invasive, Multidrug-Resistant Fungus — United States, May 2013–August 2016

TABLE. Characteristics of the first seven cases of *Candida auris* identified in the United States—May 2013–August 2016

Patient	Isolation month/ year	State	Site of *C. auris* isolation	Underlying medical condition(s)	Outcome*
1	May 2013	New York	Blood	Respiratory failure requiring high-dose corticosteroids	Died
2	July 2015	New Jersey	Blood	Brain tumor and recent villous adenoma resection	Died
3	April 2016	Maryland	Blood	Hematologic malignancy and bone marrow transplant	Died
4	April 2016	New York	Blood	Hematologic malignancy	Died
5	May 2016	Illinois	Blood	Short gut syndrome requiring total parenteral nutrition and high-dose corticosteroid use	Survived
6	July 2016	Illinois	Urine	Paraplegia with long-term, indwelling Foley catheter	Survived
7	August 2016	New York	Ear	Severe peripheral vascular disease and skull base osteomyelitis	Survived

* Mortality was not necessarily attributable to *C. auris* infection.

Schelenz *et al. Antimicrobial Resistance and Infection Control* (2016) 5:35
DOI 10.1186/s13756-016-0132-5

Antimicrobial Resistance and Infection Control

RESEARCH Open Access

First hospital outbreak of the globally emerging *Candida auris* in a European hospital

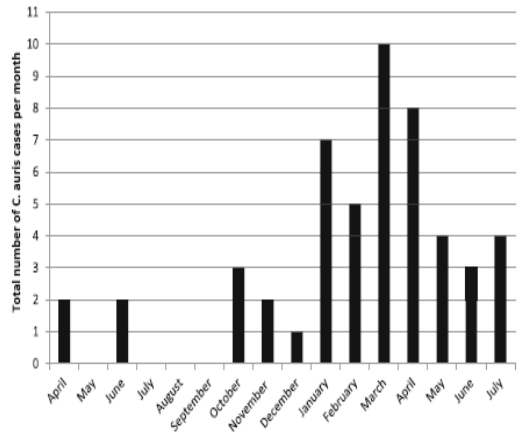


Fig. 1 New cases of *C. auris* per month. Total number of monthly new cases of *C. auris* are listed from the 1 April 2015 to the end of July 2016

Impact of Rapid Organism Identification via Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Combined With Antimicrobial Stewardship Team Intervention in Adult Patients With Bacteremia and Candidemia

Angela M. Huang,[1,2] Duane Newton,[5,6] Anjly Kunapuli,[1,2] Tejal N. Gandhi,[3] Laraine L. Washer,[3,4] Jacqueline Isip,[1,2] Curtis D. Collins,[1,2] and Jerod L. Nagel[1,2]

Received 21 February 2013; accepted 22 July 2013; electronically published 29 July 2013.

Correspondence: Jerod Nagel, PharmD, BCPS (AQID), 1111 E Catherine St, Rm 300, Ann Arbor, MI 48109 (nageljl@umich.edu).

Clinical Infectious Diseases 2013;57(9):1237–45

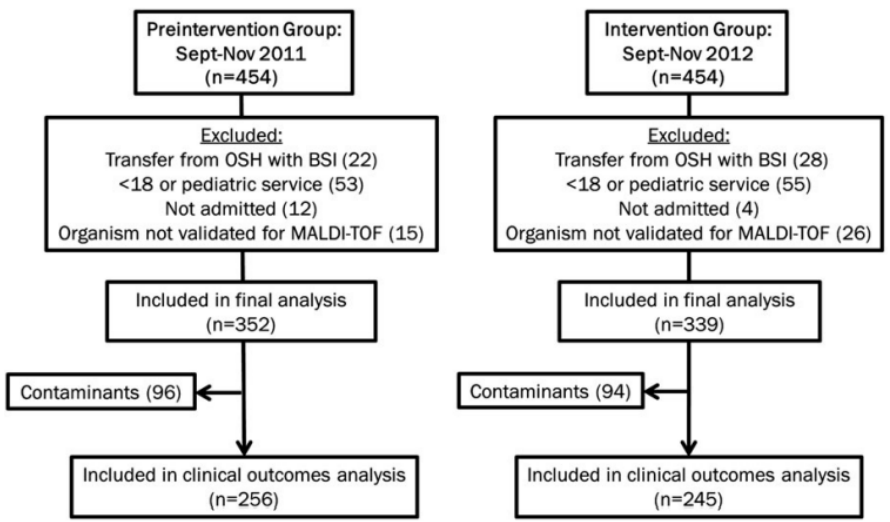


Table 3. Clinical and Treatment-Related Outcomes

Outcome	Total		
	Preintervention (n = 256)	Intervention (n = 245)	*P* Value
Clinical outcomes			
30-day all-cause mortality	52 (20.3)	31 (12.7)	.021
Time to microbiological clearance, d	3.3 ± 4.8	3.3 ± 5.7	.928
Length of hospitalization, d[a]	14.2 ± 20.6	11.4 ± 12.9	.066
Length of ICU stay, d[a]	14.9 ± 24.2	8.3 ± 9.0	.014
Recurrence of same BSI	15 (5.9)	5 (2.0)	.038
30-day readmission with same BSI	9 (3.5)	4 (1.6)	.262
Treatment-related outcomes			
Time to effective therapy, h	30.1 ± 67.7	20.4 ± 20.7	.021
Time to optimal therapy, h	90.3 ± 75.4	47.3 ± 121.5	<.001

Data are No. (%) or mean ± standard deviation.

Abbreviations: BSI, bloodstream infection; ICU, intensive care unit.

[a] Length of hospitalization and ICU stay were defined as time from blood culture positivity to discharge.

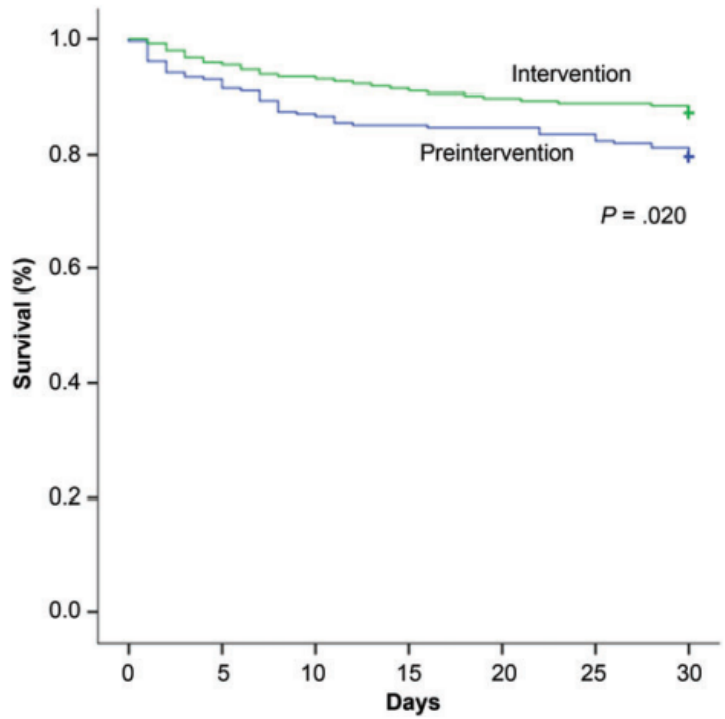
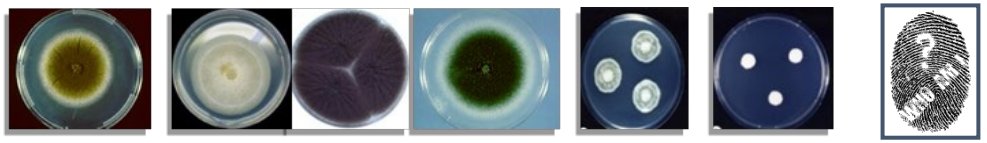


Figure 3. Kaplan-Meier survival analysis: overall survival in both study groups, censored for patients discharged prior to 30 days.

Work-flow for mould identification by MALDI-TOF MS



Unknown mould

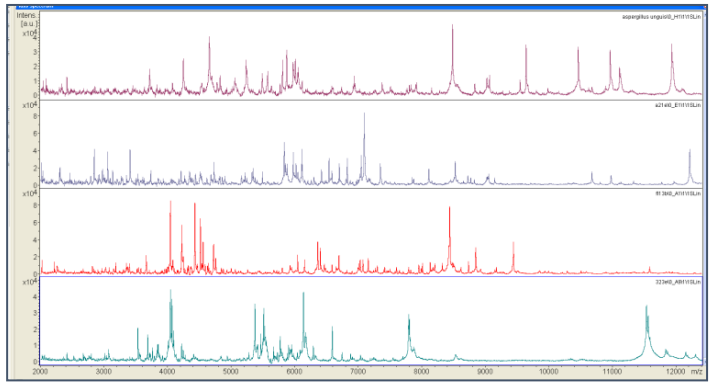
MALDI-TOF MS processing

H_2O

Absolute ethanol

HCCA matrix

MALDI-TOF spectrum profile acqiusition



Match to fungi database

YES

Species identification

Log(score)≥2.0?

NO

Molecular identification

Data entry in fungi database

From: De Carolis *et al.*, CMI, 2012

Rapid diagnosis of fungal infections: a mission impossible?

Commercially available culture-independent diagnostic modalities for detecting *Candida* sp.

IC: invasive candidiasis. IA: intra-abdominal. DS: deep seated

Test Name	Example Commercial Products	Sample Source	TAT	Disadvantages	Sensitivity	Specificity	Notes	Citations
1,3-β-D-glucan (BDG)	Fungitell, Fungitell STAT (Associates of Cape Cod, Inc.) and Fungitec G-MK. (Seikagaku). Wako β-glucan (Fujifilm Wako Chemicals)	Serum	Fungitell STAT (qualitative)40–60 min Fungitell: 24–72 h 120 min	Not specific for Candida (e.g., can be + with invasive aspergillosis, fusariosis, *Pneumocystis jirovecii* infection) High false positives Often run in reference labs Lower sensitivity	IC: 75–80% IA/DS: 56–77%	IC: ~80% IA/DS: 57–83%	FDA approved in 2004, better performance with two consecutive results Available in Europe, does not require batch testing	[8,22,23]
Candida mannan	Pastorex Candida (Bio-Rad) Platelia Candida Ag Plus (Bio-Rad)	Serum or plasma	2 h	May form immune complexes and be rapidly cleared	IC: 58%	IC: 93%	Available in Europe	[8]
Combined mannan/antimannan	Platelia Candida Ag-Plus and Ab-Plus (Bio-Rad) Serion Mannan Kit (Serio GmbH)	Serum or plasma	2 $\frac{1}{2}$ h	Low sensitivity due to rapid clearance and complex formation with antibodies	IC: 83% IA/DS: 40%	IC: 86% IA/DS: 25%	Available in Europe	[8,24,25]
T2 Candida nanodiagnostic panel	T2 Candida (T2 Biosystems)	Whole blood	4.4 +/− 1 h	Identifies limited number of Candida species (only 5 most common) High cost. Needs further validation in IA/DS	IC: 91% IA/DS: 33%	IC: 94% IA/DS: 93%	FDA approved	[26,27]
C. albicans germ tube antibody assays (CAGTA)	CAGTA; Vircell Kit and VirClia IgG Monotest	Serum	~3 h	Lower sensitivity for *C. tropicalis*	IC: 42–96% IA/DS: 53–73%	IC: 54–100% IA/DS: 54–80%	Not FDA approved (used in Europe) Increased accuracy when combined with BDG	[28]
Candida PCR performed directly on clinical specimens	LightCycler. SeptiFast (Roche Diagnostics), SepsiTest (Molzym), Magicplex system (Seegene), or VYOO. (SIRS-Lab),	Whole blood, serum, plasma	Minutes to hours (real-time PCR). Multiplex PCR: 4–12 h	Not standardized or validated in multicenter trials. False negatives (low burden of fungal cells in blood, difficulties with sample preparation and DNA extraction) and false positives (similarities with human DNA, sample contamination)	IC: 73–95% IA/DS: 86–91%	IC: 92–95% IA/DS: 33–97%	None FDA approved Variety of DNA targets including Candida-specific genes or broad range pan-fungal genes	[24,29]

From: Weiss *et al.*, J. Fungi, 2021

De Pascale *et al. Critical Care* (2020) 24:550
https://doi.org/10.1186/s13054-020-03265-y

Critical Care

RESEARCH Open Access

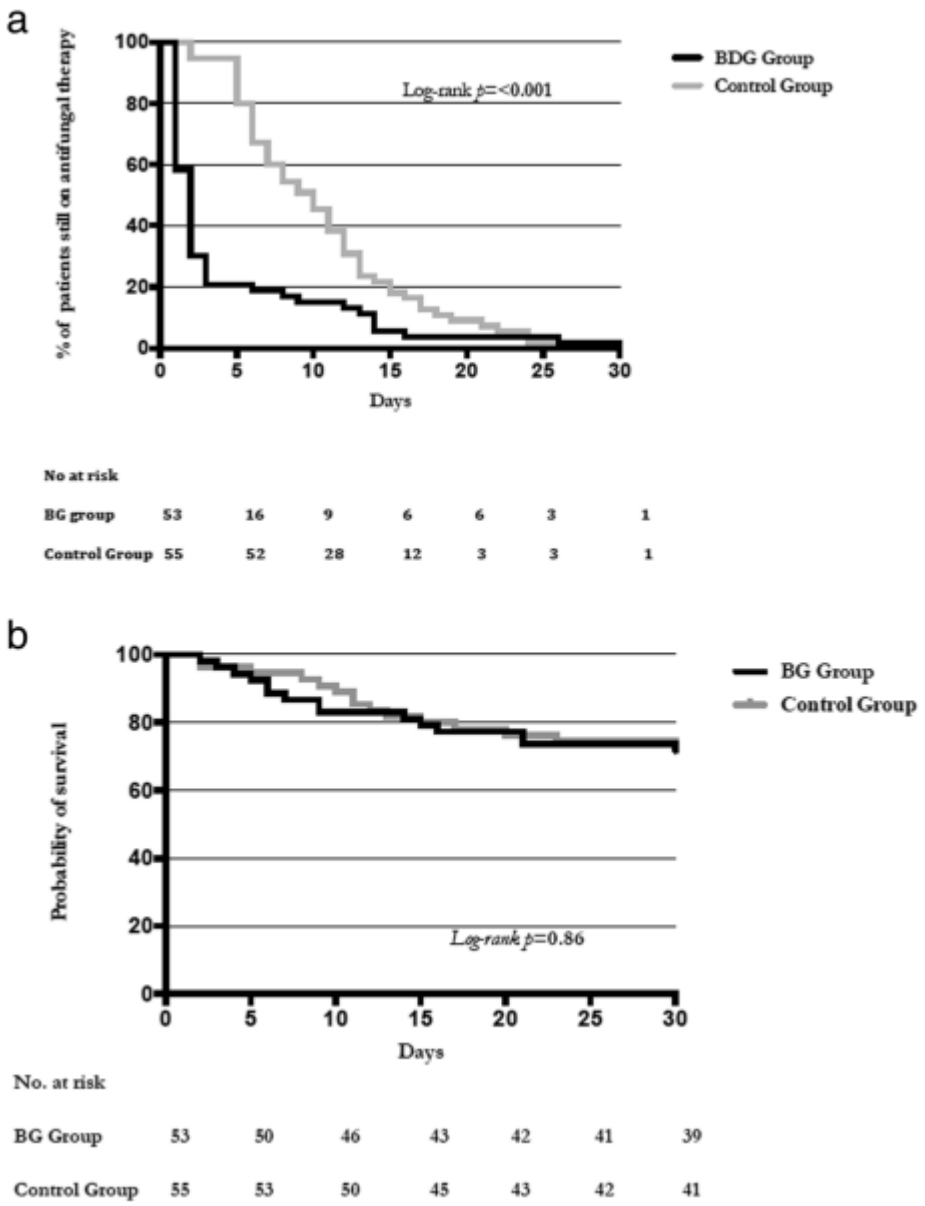
(1,3)-β-D-Glucan-based empirical antifungal interruption in suspected invasive candidiasis: a randomized trial

Gennaro De Pascale[1,2*†], Brunella Posteraro[3,4†], Sonia D'Arrigo[1,2], Giorgia Spinazzola[1,2], Rita Gaspari[1,2], Giuseppe Bello[1,2], Luca Maria Montini[1,2], Salvatore Lucio Cutuli[1,2], Domenico Luca Grieco[1,2], Valentina Di Gravio[1,2], Giulia De Angelis[5], Riccardo Torelli[6], Elena De Carolis[6], Mario Tumbarello[7,8], Maurizio Sanguinetti[5,6] and Massimo Antonelli[1,2]

Table 2 Outcome measures in the (1–3)-β-D-glucan (BDG) and the control groups

Variable	BDG group ($n = 53$)	Control group ($n = 55$)	Between-group absolute difference in means (95% CI)	p value
Primary outcome				
Duration of antifungal therapy, days	2 [1–3]	10 [6–13]	6.29 (3.94 to 8.65)	**< 0.001**
Secondary outcomes				
30-day mortality, *N* (%)	15 (28.3)	15 (27.3)	– 1% (– 16.89 to 18.93)	0.92
ICU mortality, *N* (%)	16 (30.2)	17 (30.9)	0.7% (– 17.7 to 18.97)	0.89
Hospital mortality, *N* (%)	19 (35.9)	18 (32.7)	– 3.2% (– 15.7 to 21.93)	0.88
Subsequent ICI, *N* (%)*	0	2 (3.6)	3.6% (– 3.83 to 12.47)	0.5
Hospital LOS, days	35 [23.75–55.25]	38 [20–59.5]	– 7.41 (– 21.55 to 6.73)	0.87
ICU LOS, days	18 [7.75–24.25]	13 [7–26]	– 0.5 (– 6.95 to 5.95)	0.23
Mechanical ventilation duration, days	9 [4.75–17.25]	9 [3.25–19.75]	3.21 (– 2.05 to 8.46)	0.97
Vasopressors duration, days	4 [0.75–8.25]	3 [0–11]	0.06 (– 2.95 to 3.07)	0.6
Total antifungals costs, €	110 [2.64–708]	113.2 [9.68–1255.6]	318.63 (– 310.1 to 947.3)	0.24
Echinocandins cost, €	708 [185.6–1071.5]	1320 [618.5–30,149.5]	937.05 (– 64.2 to 1938.3)	0.07
BG cost, € mean ± SD	80.8 ± 20.4	–	–	–

Data are presented as median (IQR) and *N* (%). Between-group absolute differences are calculated using the mean values, percentage differences and 95% CIs
BDG (1–3)-β-D-glucan, *ICU* intensive care unit, *ICI* invasive *Candida* infection, *LOS* length of stay, *€* euro, *IQR* interquartile range
*See eTable 1 for further details



ORIGINAL

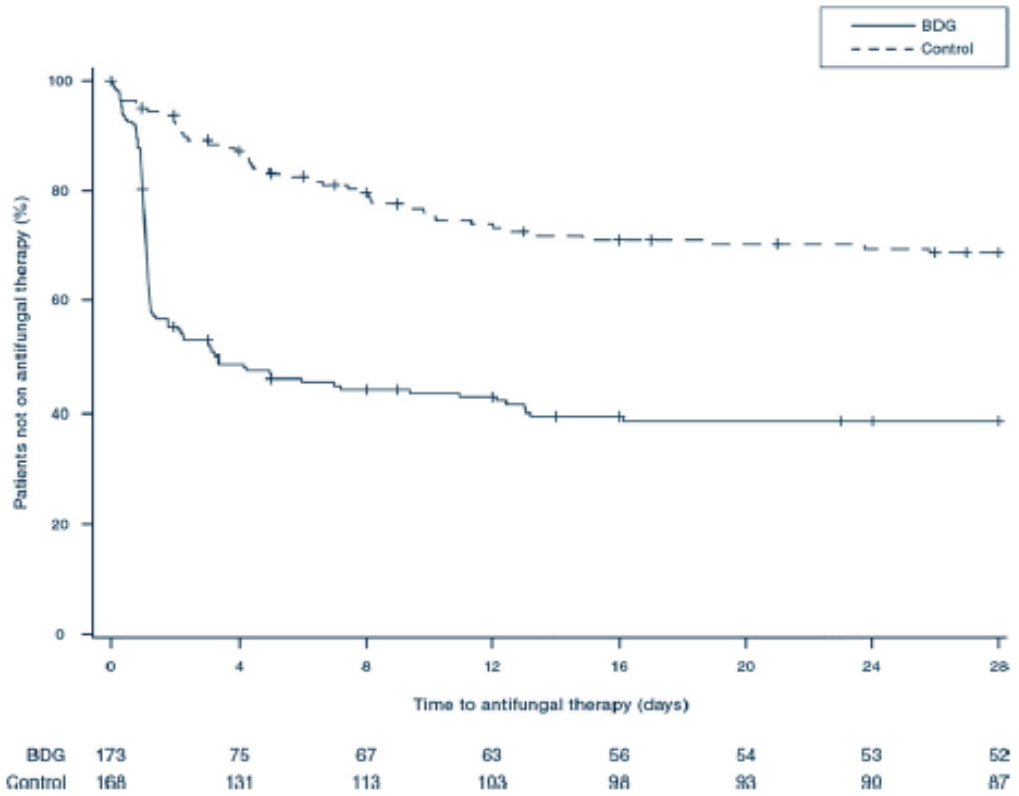
(1 → 3)-β-D-Glucan-guided antifungal therapy in adults with sepsis: the CandiSep randomized clinical trial

Frank Bloos[1,2*], Jürgen Held[3], Stefan Kluge[4], Philipp Simon[5,12], Klaus Kogelmann[6], Geraldine de Heer[4], Sven-Olaf Kuhn[7], Dominik Jarczak[4], Johann Motsch[8], Gunther Hempel[5], Norbert Weiler[9], Andreas Weyland[10], Matthias Drüner[6], Matthias Gründling[7], Patrick Meybohm[11], Daniel Richter[8], Ulrich Jaschinski[12], Onnen Moerer[13], Ulf Günther[14], Dirk Schädler[9], Raphael Weiss[15], Christian Putensen[16], Ixchel Castellanos[17], Oliver Kurzai[18,19], Peter Schlattmann[20], Oliver A. Cornely[21,22,23,24], Michael Bauer[1,2], Daniel Thomas-Rüddel[1,2] on behalf of the SepNet Study Group

Table 2 Primary and secondary outcomes

Outcome	BDG-group *n* = 172	Control-group *n* = 167	Relative risk (95% CI)	*p* value
Primary outcome				
28-Day all-cause mortality—no (%)	58 (33.7)	51 (30.5)	1.1 (0.8–1.51)	0.53
Secondary outcomes				
Hospital mortality—no (%)	59 (34.5)	60 (35.9)	0.96 (0.71–1.29)	0.78
Hospital length of stay—days	25.5 (16–41)	28 (17–48)	NA	0.37
ICU mortality- no (%)	48 (27.7)	47 (27.8)	1 (0.7–1.41)	0.99
ICU length of stay—days	11 (6–20)	11 (4–22)	NA	0.70
Antifungal free survival at day 28—no (%)	52 (30.2)	87 (52.1)	2.97 (2.1–4.2)	< 0.01
Time to antifungal therapy—days	1.1 (1–2.2)	4.4 (2–9.1)	NA	< 0.01
Costs of antifungal therapy—Euro	4451 (1385–6923)	2800 (989–7097)	NA	0.52
Candida Colonization Index				
At randomization	0.20 (0–0.33)	0.2 (0–0.4)	NA	0.69
At day 1	0 (0–0.67)	0 (0–1)	NA	0.66
At day 7	0.25 (0–0.5)	0.25 (0; 0.5)	NA	0.22
At day 14	0.2 (0–0.33)	0.25 (0.08–0.4)	NA	0.14
Total SOFA	10.5 (8.2–14.3)	10.4 (8.2–13.4)	NA	0.42
Vasopressor free days—days	20 (3–25)	20 (3–26)	NA	0.40
Ventilator free days—days	16 (2–25)	15 (2–27)	NA	0.51
Renal replacement free days—days	27 (9–29)	27.5 (9–29)	NA	0.92

NA not applicable, *IQR* interquartile range, *ICU* intensive care unit, *SOFA* sequential organ failure assessment



"In this randomized multicenter clinical trial on sepsis patients we observed an earlier but also immoderate administration of antifungals when therapy was guided by (1 → 3)-β-D-glucan-guidance in comparison to culture guidance. (1 → 3)-β-D-glucan-guidance did not affect mortality, but definite conclusions are hampered by an unexpectedly low rate of invasive Candida infections in this study population"

Received: 8 February 2022 | Accepted: 3 August 2022
DOI: 10.1111/myc.13515

REVIEW ARTICLE

mycoses WILEY

Performance of existing clinical scores and laboratory tests for the diagnosis of invasive candidiasis in critically ill, nonneutropenic, adult patients: A systematic review with qualitative evidence synthesis

Daniele Roberto Giacobbe[1,2] | Erika Asperges[3] | Andrea Cortegiani[4,5] | Cecilia Grecchi[3] | Chiara Rebuffi[6] | Valentina Zuccaro[3] | Luigia Scudeller[7] | Matteo Bassetti[1,2] | the FUNDICU investigators

- Most of the included studies on the use of laboratory tests for the diagnosis of IC in nonneutropenic, critically ill adult patients in ICU evaluated serum beta-D-glucan (BDG).
- The most homogeneous finding in 17 studies was the high NPV (>90% in the majority of studies when using the manufacturers' cut-off), whereas more heterogeneous values were observed for PPV, nonetheless generally higher than that observed for clinical scores

J Antimicrob Chemother 2016; **71**: 2386–2396
doi:10.1093/jac/dkw156 Advance Access publication 12 May 2016

Journal of Antimicrobial Chemotherapy

ECIL guidelines for the diagnosis of *Pneumocystis jirovecii* pneumonia in patients with haematological malignancies and stem cell transplant recipients

Alexandre Alanio[1], Philippe M. Hauser[2], Katrien Lagrou[3], Willem J. G. Melchers[4], Jannik Helweg-Larsen[5], Olga Matos[6], Simone Cesaro[7], Georg Maschmeyer[8], Hermann Einsele[9], J. Peter Donnelly[10], Catherine Cordonnier[11]*, Johan Maertens[12] and Stéphane Bretagne[1] on behalf of the 5th European Conference on Infections in Leukemia (ECIL-5†), a joint venture of The European Group for Blood and Marrow Transplantation (EBMT), The European Organization for Research and Treatment of Cancer (EORTC), the Immunocompromised Host Society (ICHS) and The European LeukemiaNet (ELN)

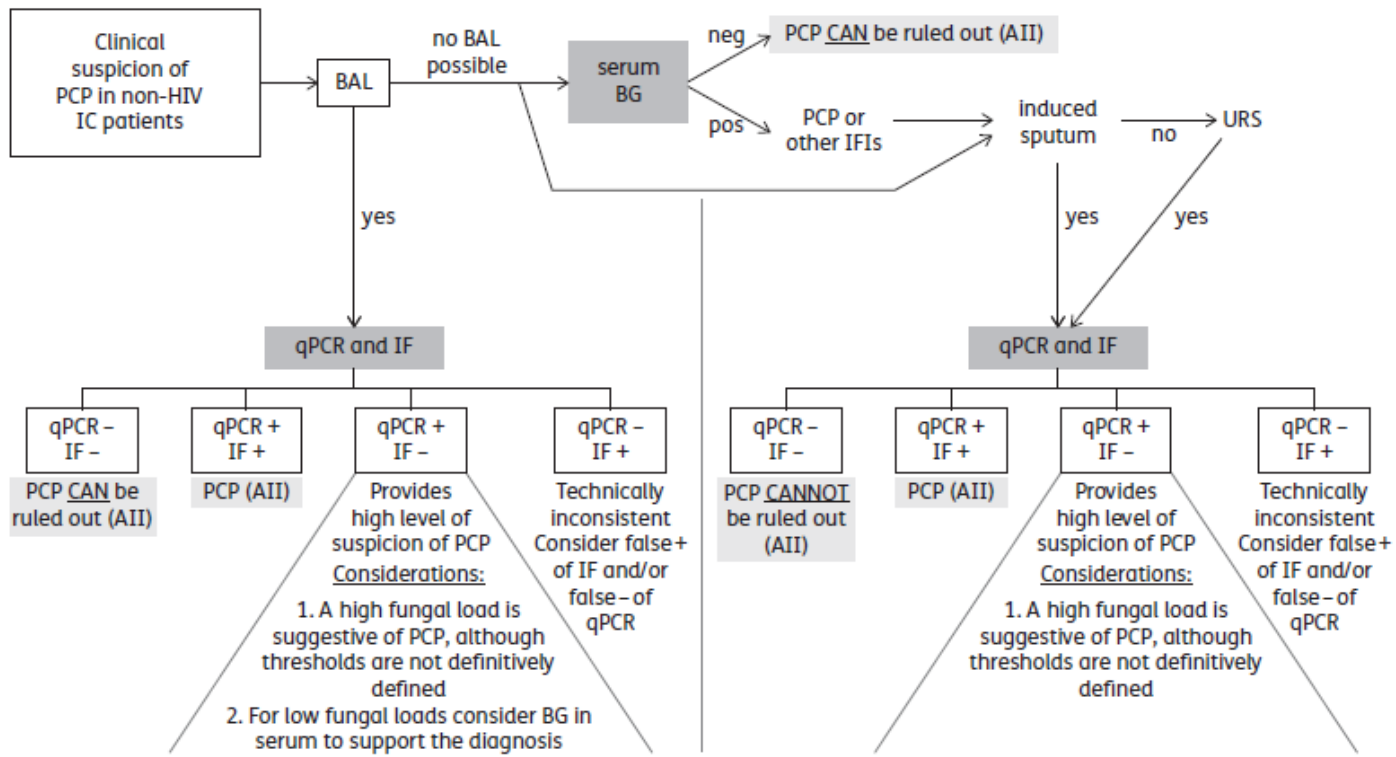


Figure 1. Flow chart for the diagnosis of *Pneumocystis* pneumonia in non-HIV immunocompromised (IC) patients. Biological tests are highlighted in dark grey and recommendations in light grey. BG, β-D-glucan; A-II, level of recommendation; IFI, invasive fungal infection.

T2Dx *Candida* panel

- T2MR demonstrated an overall specificity per assay of 99.4% with a mean time to negative result of 4.2 ± 0.9 hours. The overall sensitivity was found to be 91.1% (96.6% considering also other studies) with a mean time of 4.4 ± 1.0 hours for detection and species identification[1,2]
- Significant reduction of the time to appropriate therapy (from 20 to 28 hours)[3,4]
- Significant reduction of the time to detection of *Candida*[3]
- Significant reduction of the ICU length of stay[3]
- Significant reduction in antifungal consumption[4]
- Strong indicator of complications and poor outcomes[5]

1. Mylonakis E, et al. CID, 2018
2. Clancy C, et al. CID, 2018
3. Wilson et al, ID Week 2016
4. Patch et al., CMI, 2018
5. Munoz et al., CMI, 2018

Commercially available non-culture-based testing for Aspergillosis and *Mucorales*

IC: invasive candidiasis. IA: intra-abdominal. DS: deep seated

Test Name	Example Commercial Product	Sample Source	TAT	Disadvantages	Sensitivity	Specificity	Notes	Citations
1,3-β-D-glucan (BDG)	Fungitell (Associates of Cape Cod, Inc.) and Fungitec G-MK (Seikagaku).	Serum	Fungitell STAT (qualitative): 40–60 min Regular Fungitell: 24–72 h (d)	Cross-reactive with other fungi, False positives frequent. Often run in reference labs.	Fungitell: 33–100% Fungitec: 67–88%	Fungitell: 36–94% Fungitec: 84–85%	FDA approved.	[116]
Galactomannan	Platelia *Aspergillus* EIA/Ag (Bio-Rad)	Serum, BAL (also CSF, pleural fluid)	1–7 days	Cross-reactive with other fungi. False positives frequent.	**Neutropenic/heme malignancy** Serum: 61–79% BALF: 58–90% **Non-neutropenic:** Serum: 38–41% BALF: 65–76%	**Neutropenic/heme malignancy** Serum: 81–95% BALF: 84–96% **Non-neutropenic:** Serum: 87–89% BALF: 81–90%	FDA approved. Serially monitoring can assess treatment response.	[117–121]
Lateral flow devices	AspLFD (OLM Diagnostics) and the Aspergillus galactomannan LFA (IMMY)	Serum, BAL, urine	15–30 min	Serum LFD requires additional preparation steps/pre-treatment. Sensitivity decreased with antifungals.	**AspLFD: Neutropenic/heme malignancy:** Serum: 56–68% BAL: 71–89% **Non-neutropenic:** BAL: 46–69% **LFA: Neutropenic/heme malignancy:** 89–97% **Non-neutropenic:** BALF: 65–69%	**AspLFD: Neutropenic/heme malignancy:** Serum: 87–90% BAL: 88–100% **Non-neutropenic:** BAL: 46–58% **LFA: Neutropenic/heme malignancy:** 88–98% **Non-neutropenic:** BALF: 62–68%	Available in Europe. Urinary GM-like antigen-based test also exists but needs further validation.	[103–107,109]
Aspergillus PCR	MycAssay Aspergillus (real-time PCR) AsperGenius assay (multiplex real-time PCR)	Serum, BAL	12–24 h	Sensitivity decreased by antifungal treatment. Many commercially available assays. Standardization efforts ongoing. Small clinical studies.	**Serum:** 60–79% **BALF:** 77%	**Serum:** 80–95% **BALF:** 94%	Some detect azole-resistant mutations. Independent validation still needed for most.	[90,110–112]
Mucorales PCR	MucorGenius (Pathonostics)	BAL, biopsy fluid	3 h		**90–100%**	**90–99%**		[122,123]

From: Weiss *et al.*, J. Fungi, 2021

AMERICAN SOCIETY FOR MICROBIOLOGY | Journal of Clinical Microbiology®

MYCOLOGY

Aspergillus Lateral Flow Assay with Digital Reader for the Diagnosis of COVID-19-Associated Pulmonary Aspergillosis (CAPA): a Multicenter Study

Brice Autier,[a] Juergen Prattes,[b] P. Lewis White,[c] Maricela Valerio,[d,e] Marina Machado,[d,e] Jessica Price,[c] Matthias Egger,[b] Jean-Pierre Gangneux,[a] Martin Hoenigl[b,f,g]

- This multicenter study evaluated the IMMY Aspergillus Galactomannan Lateral Flow Assay (LFA) with automated reader for diagnosis of pulmonary aspergillosis in patients with COVID-19 associated acute respiratory failure (ARF) requiring intensive care unit (ICU) admission between 03/2020 and 04/2021.
- A total of 196 respiratory samples and 148 serum samples (n=344) from 238 patients were retrospectively included, with a maximum of one of each sample type per patient.
- Cases were retrospectively classified for COVID-19 associated pulmonary aspergillosis (CAPA) status following the 2020 consensus criteria, with the exclusion of LFA results as a mycological criterion.
- At the 1.0 cutoff, sensitivity of LFA for CAPA (proven/probable/possible) was 52%, 80% and 81%, and specificity was 98%, 88% and 67%, for bronchoalveolar lavage fluid (BALF), non-directed bronchoalveolar lavage (NBL), and tracheal aspiration (TA), respectively.
- Sensitivity and specificity of serum LFA were 20% and 93%, respectively, at the 0.5 ODI cutoff.
- Overall, the Aspergillus Galactomannan LFA showed good performances for CAPA diagnosis, when used from respiratory samples at the 1.0 cutoff, while sensitivity from serum was limited

Journal of Infection 81 (2020) 131–146

Contents lists available at ScienceDirect

Journal of Infection

journal homepage: www.elsevier.com/locate/jinf

Review

Performance of existing definitions and tests for the diagnosis of invasive aspergillosis in critically ill, adult patients: A systematic review with qualitative evidence synthesis ☆

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- Sufficient data for evaluating the performance of existing definitions and laboratory tests for the diagnosis of IA in critically ill patients is available only for invasive pulmonary aspergillosis
- Studies on laboratory tests consistently indicated a better diagnostic performance of bronchoalveolar lavage fluid (BALF) galactomannan (GM) than serum GM, and a suboptimal specificity of BALF and serum (1,3)-β-D-glucan

Some of the available commercial methods for antifungal susceptibility testing

Method	Brief Description
[1] SYO ®; TREK	Sensititre colorimetric plate Microdilution plate wells are dosed with the antifungal appropriate dilutions, as well as with the colorimetric indicator. MIC: lowest antifungal concentration showing no color change (no growth) following 24–48 h of incubation.
[2] Etest ®	Gradient concentration methods ®
[3] Biomerieux Liofilchem ®	Based upon a continuous concentration gradient of drug infused on a plastic non-porous strip. The agent diffuses into an agar plate and the MICs are where the growth intersects with the testing strips after 24 to 48 h or until the susceptibility ellipse is created.
[4] Automated Vitek-2 ®	Automated system that spectrophotometrically and simultaneously provides the isolation, identification, and the MIC results of the pathogen.
[5] Neo-Sensitabs tablets ® and disk diffusion	The 9-mm tablets contain the following concentrations: amphotericin B (10 µg), caspofungin (5 µg), fluconazole (25 µg), itraconazole (8 µg), and voriconazole (1 µg). Discs can also be used when available (see CLSI M44 method).
[6] ATB ® fungus panel	This microdilution non-colorimetric panel determine MICs to six antifungal agents.
[7] The Fungitest ®	A microdilution colorimetric breakpoint method for amphotericin B, flucytosine, fluconazole, itraconazole, ketoconazole, and miconazole. Not much since BPs are not available for most of these antifungals.

From: Espinel-Ingroff *et al.*, J. Fungi, 2021

EUCAST/CLSI antifungal breakpoints for *Candida* species

Antifungal agent	MIC breakpoint (mg/L)													
	C. albicans		*C. glabrata*		*C. krusei*		*C. parapsilosis*		*C. tropicalis*		*C. guilliermondii*		Non-species related breakpoints[1]	
	S≤	R>	S≤	R>	S≤	R>	S≤	R>	S≤	R>	S≤	R>	S≤	R>
Amphotericin B														
EUCAST	1	1	1	1	1	1	1	1	1	1	IE	IE	IE	IE
CLSI	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	1	
Anidulafungin														
EUCAST	0.03	0.03	0.06	0.06	0.06	0.06	0.002	4	0.06	0.06	IE[2]	IE[2]	IE	IE
CLSI	0.25	0.5	0.12	0.25	0.25	0.5	2	4	0.25	0.5	2	4	ND	ND
Caspofungin														
EUCAST	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	Note[3]	IE[2]	IE[2]	Note[3]	Note[3]
CLSI	0.25	0.5	0.12	0.25	0.25	0.5	2	4	0.25	0.5	2	4	ND	ND
Fluconazole														
EUCAST	2	4	0.002	32	–	–	2	4	2	4	IE[2]	IE[2]	2	4
CLSI	2	4	0.002	32	–	–	2	4	2	4	ND	ND	ND	ND
Flucytosine														
EUCAST	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
CLSI	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	4	16
Itraconazole														
EUCAST	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP
CLSI	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.125	0.5
Micafungin														
EUCAST	0.016	0.016	0.03	0.03	IE[4]	IE[4]	0.002	2	IE[4]	IE[4]	IE[4]	IE[4]	IE	IE
CLSI	0.25	0.5	0.06	0.125	0.25	0.5	2	4	0.25	0.5	2	4	ND	ND
Posaconazole														
EUCAST	0.06	0.06	IE[2]	IE[2]	IE[2]	IE[2]	0.06	0.06	0.06	0.06	IE[2]	IE[2]	IE	IE
CLSI	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Voriconazole														
EUCAST	0.12[5]	0.12[5]	IE[2]	IE[2]	IE[2]	IE[2]	0.12[5]	0.12[5]	0.12[5]	0.12[5]	IE[2]	IE[2]	IE	IE
CLSI	0.12	0.5	–	–	0.5	1	0.12	0.5	0.12	0.5	ND	ND	ND	ND

ND, not done; IP, in preparation; IE, insufficient evidence.

From: Arendrup et al., Drug Res. Up., 2013

Method-dependent ECVs of three triazoles and three echinocandins for species of *Candida*

Species [3]	Agent/Method-Dependent ECVs (μg/mL) [2]																
	FLU			POS			VOR			AND			CAS		MCA		
	SYO	Etest	CLSI	SYO	Etest	CLSI	SYO	Etest	CLSI	SYO	Etest	CLSI	SYO	Etest	SYO	Etest	CLSI
C. albicans	1	1	0.5	0.06	0.06	0.06	0.01	0.03	0.03	0.12	0.01	0.12	0.25	0.5	0.06	0.03	0.03
C. dubliniensis	1	0.5	0.5	0.12	NA	0.12	0.01	1	0.03	0.25	NA	0.12	0.25	NA	0.12	NA	0.12
C. glabrata	64	64	8	4	NA	1	2	1	0.25	0.12	0.03	0.25	0.25	1	0.03	0.03	0.03
M. guilliemondii	16	16	8	1	NA	0.5	0.5	0.5	0.12	4	NA	8	2	NA	2	NA	2
P. kudriavzevii	128	128	32	1	NA	0.5	1	1	0.5	0.25	0.06	0.25	1	1	0.25	0.25	0.25
C. kefyr	NA	1	1	NA	NA	0.5	NA	NA	NA	NA	NA	0.25	NA	NA	NA	NA	0.12
C. lusitaniae	4	2	1	0.12	NA	0.06	0.03	0.03	NA	0.25	NA	1	1	NA	0.12	NA	0.5
C. parapsilosis SC	2	2	1	0.12	0.12	0.25	0.01	0.25	NA	4	8	4	2	4	4	2	2
C. parapsilosis SS	2	NA	NA	0.25	NA	NA	0.03	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
C. tropicalis	4	4	1	1	0.25	0.12	0.5	0.25	0.12	0.5	0.03	0.12	0.25	1	0.06	0.12	0.06

From: Espinel-Ingroff *et al*., J. Fungi, 2021

Take-home messages Commercial Tests

- Need to check for each drug-bug combination
- BPs can only be adopted if MICs match the reference method!
- Some commercial methods seem to be a good surrogate of reference methods
- Recommendations for echinocandins
  - Anidulafungin Etest with EUCAST BPs, but excellent agreement with micafungin
  - VITEK2 problematic: MIC range doesn't cover the BP for *C. glabrata*

From: Sanguinetti, unpublished

Antimicrobial Agents and Chemotherapy

SUSCEPTIBILITY

Susceptibility Testing of Common and Uncommon *Aspergillus* Species against Posaconazole and Other Mold-Active Antifungal Azoles Using the Sensititre Method

Enrica Mello,[a] Brunella Posteraro,[b] Antonietta Vella,[a] Elena De Carolis,[a] Riccardo Torelli,[a] Tiziana D'Inzeo,[a] Paul E. Verweij,[c] Maurizio Sanguinetti[a]

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TABLE 2 Comparison of *in vitro* activities of posaconazole, voriconazole, and itraconazole tested against *Aspergillus* species by SYO and CLSI methods[a]

Species (no. of isolates tested)[b]	Test method	MIC (μg/ml)								
		PSC			VRC			ITC		
		Range	Mode(s)	% EA	Range	Mode(s)	% EA	Range	Mode(s)	% EA
A. fumigatus (21)	SYO	0.06 to 1	1	100	0.125 to >8	0.25	100	0.125 to >8	0.25	95.2
	CLSI	≤0.03 to 2	1		0.06 to >8	0.125		0.25 to >8	1	
A. flavus (19)	SYO	0.06 to 0.125	0.125	100	0.125 to 1	0.25	100	0.03 to 0.25	0.06	94.7
	CLSI	≤0.03 to 0.25	0.125		0.06 to 0.25	0.125		0.06 to 0.5	0.125	
A. terreus (12)	SYO	0.06 to 0.125	0.06	100	0.06 to 0.25	0.125	100	0.06 to 0.25	0.125	91.7
	CLSI	≤0.03 to 0.25	0.25		0.06 to 0.125	0.06		0.125 to 0.5	0.25	
A. niger (7)	SYO	≤0.03 to 0.25	0.25	85.7	0.25 to 0.5	0.25	100	0.125 to 0.5	0.25	100
	CLSI	0.06 to 0.25	0.25		0.06 to 0.25	0.25		0.5 to 1	0.5	
A. tubingensis (6)	SYO	0.125 to 0.25	0.125	100	0.25 to 0.5	0.5	100	0.25 to 0.5	0.25, 0.5	100
	CLSI	0.125 to 0.5	0.125, 0.25, 0.5		0.125 to 0.5	0.25		0.5 to 2	0.5	
A. nidulans (5)	SYO	0.06 to 0.125	0.06	100	0.125 to 0.25	0.25	100	0.125 to 0.25	0.125	100
	CLSI	≤0.03 to 0.06	0.03		0.06 to 0.25	0.06, 0.125		0.25 to 1	0.5	
A. oryzae (5)	SYO	0.125 to 0.5	0.125, 0.25	80.0	0.5 to >8	0.5, 1	100	0.125 to 0.5	0.125	100
	CLSI	≤0.03 to 1	0.03		0.125 to 2	0.125		0.25 to 1	0.25	
A. lentulus (3)	SYO	0.03 to 0.125	0.03	100	2 to 4	2	100	0.06 to 0.125	0.125	100
	CLSI	≤0.03 to 0.06	0.03		0.5 to 2	ND		0.25 to 0.5	0.25	
A. (Neosartorya) species (3)[c]	SYO	0.125 to 0.25	0.125	66.7	0.5 to 2	ND	100	0.25 to 0.5	0.25	100
	CLSI	≤0.03 to 0.125	0.125		0.5 to 1	1		0.5 to 1	0.5	
A. foetidus (3)	SYO	0.125 to 0.25	0.125	100	0.5 to 1	0.5	100	0.5	0.5	100
	CLSI	≤0.03 to 0.25	ND		0.25 to 0.5	0.25		0.25 to 2	ND	
A. awamori (2)	SYO	0.06	0.06	100	0.25	0.25	100	0.125 to 0.25	ND	100
	CLSI	≤0.03 to 0.125	ND		0.125 to 0.25	ND		0.25 to 0.5	ND	

Antimicrobial Agents and Chemotherapy

SUSCEPTIBILITY

Susceptibility Testing of Common and Uncommon *Aspergillus* Species against Posaconazole and Other Mold-Active Antifungal Azoles Using the Sensititre Method

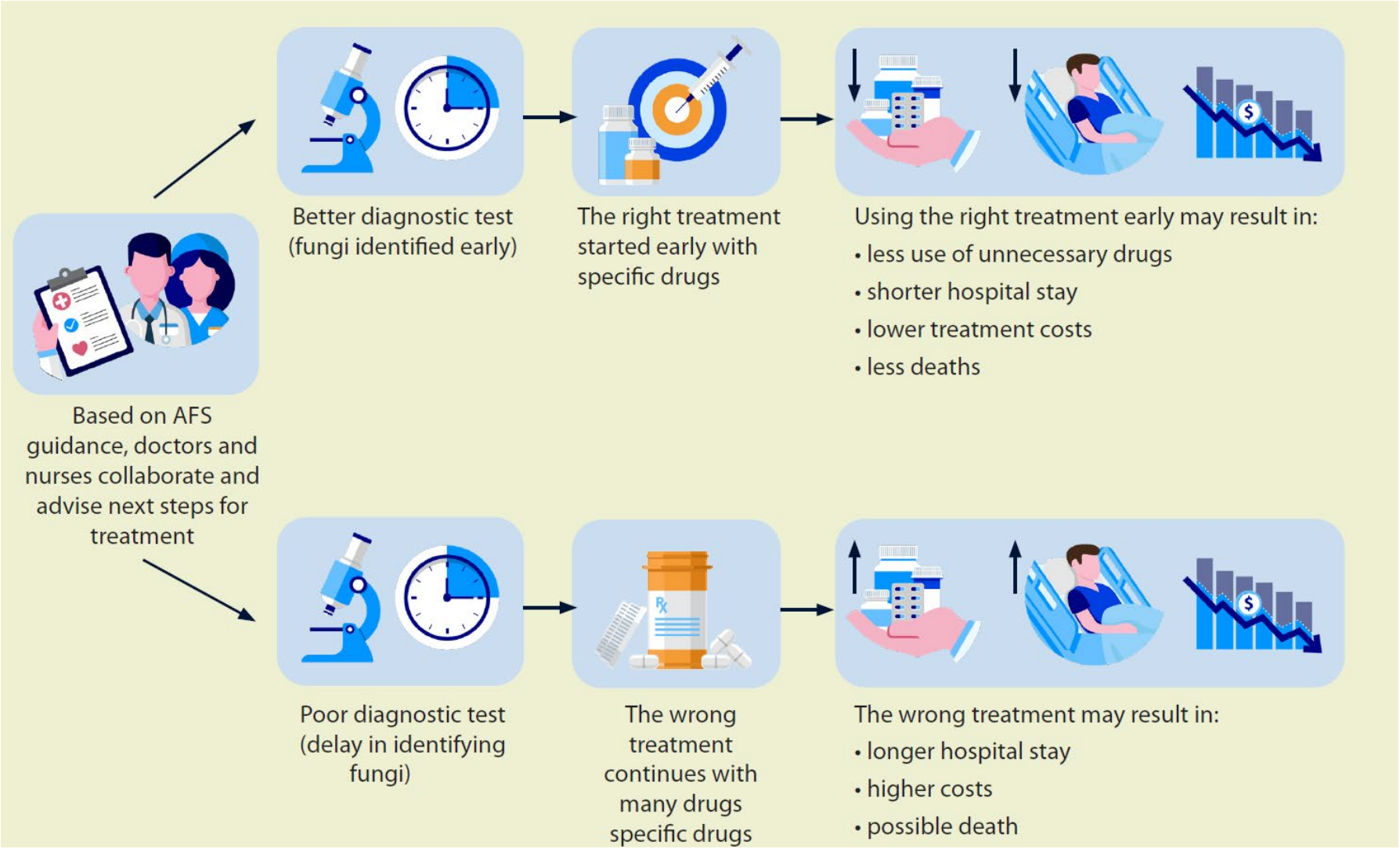
Enrica Mello,[a] Brunella Posteraro,[b] Antonietta Vella,[a] Elena De Carolis,[a] Riccardo Torelli,[a] Tiziana D'Inzeo,[a] Paul E. Verweij,[c] Maurizio Sanguinetti[a]

June 2017 Volume 61 Issue 6

TABLE 3 Triazole MICs for *Aspergillus* species isolates carrying a mutated *cyp51* gene, as determined by SYO and CLSI methods[a]

Organism (designation)	Type of mutation detected[b]	SYO MIC (μg/ml)			CLSI MIC (μg/ml)			Non-wild-type phenotype for each indicated triazole, according to the ECV[c]
		PSC	VRC	ITC	PSC	VRC	ITC	
A. fumigatus (v075-77)	TR_{34}/L98H	**0.5**	1	>16	**1**	1	>16	PSC (by CLSI only), ITC
A. fumigatus (v082-04)	TR_{34}/L98H	1	4	>16	1	2	>16	PSC, VRC, ITC
A. fumigatus (v085-79)	TR_{34}/L98H	1	8	>16	2	4	>16	PSC, VRC, ITC
A. fumigatus (v110-25)	TR_{34}/L98H	1	4	>16	1	4	>16	PSC, VRC, ITC
A. fumigatus (v128-51)	TR_{34}/L98H	**0.5**	**2**	2	**1**	**1**	2	PSC (by CLSI only), VRC (by SYO only), ITC
A. fumigatus (v099-47)	TR_{46}/Y121F T289A	1	>8	1	1	>8	1	PSC, VRC
A. fumigatus (v115-49)	TR_{46}/Y121F T289A	1	>8	**1**	2	>8	**2**	PSC, VRC, ITC (by CLSI only)
A. fumigatus (v116-78)	TR_{46}/Y121F T289A	1	>8	**1**	1	>8	**2**	PSC, VRC, ITC (by CLSI only)
A. fumigatus (v134-70)	TR_{46}/Y121F T289A	1	>8	**1**	1	>8	**2**	PSC, VRC, ITC (by CLSI only)
A. fumigatus (v135-16)	TR_{46}/Y121F T289A	1	8	**0.5**	1	>8	**2**	PSC, VRC, ITC (by CLSI only)
A. oryzae (UCSC-943)	T788G	0.5	>8	0.5	1	2	1	ND

Is antifungal stewardship (AFS) helpful? YES!!!!!!



Some key benefits of AFS studies with diagnostic tests:

– Fungi were identified faster with better tests

– Early start of the right treatment

– Days of treatment before the cause of infection was confirmed decreased

– Use of the wrong drugs stopped

– Less overall use of drugs

– Less treatment cost

– Shorter hospital stays

– Less number of deaths

From: Chakrabarti, OFID, 2022

Gemelli

Fondazione Policlinico Universitario A. Gemelli
Università Cattolica del Sacro Cuore

… and special thanks to

Giulia De Angelis

Flora M. Liotti

Giulia Menchinelli

Riccardo Torelli

Elena De Carolis

Antonella Vella

Tiziana D'Inzeo

Teresa Spanu

Barbara Fiori

Federica Travaglia

and

Brunella Posteraro

The UCSC Fungal and Bacterial Rapid Diagnostic Team