



BELGISCHE VERENIGING VOOR MENSELIJKE EN DIERLIJKE MYCOLOGIE

SOCIÉTÉ BELGE DE MYCOLOGIE HUMAINE ET ANIMALE

FILAMENTOUS FUNGI IN HOSPITAL DISTRIBUTION SYSTEMS: WHAT IS THE RISK?

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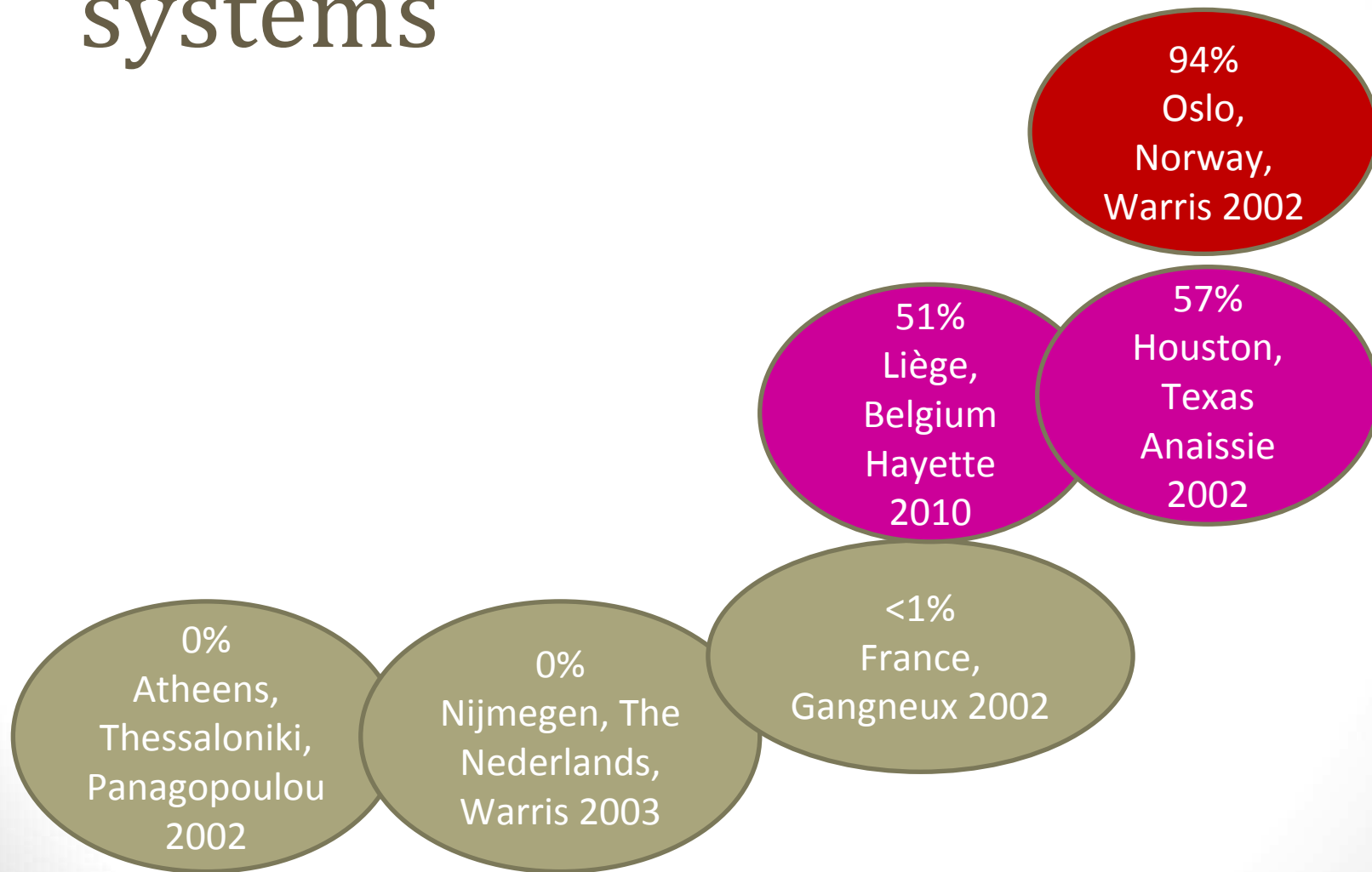


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Presence of fungi in biofilms



Variable situations in hospital water distribution systems



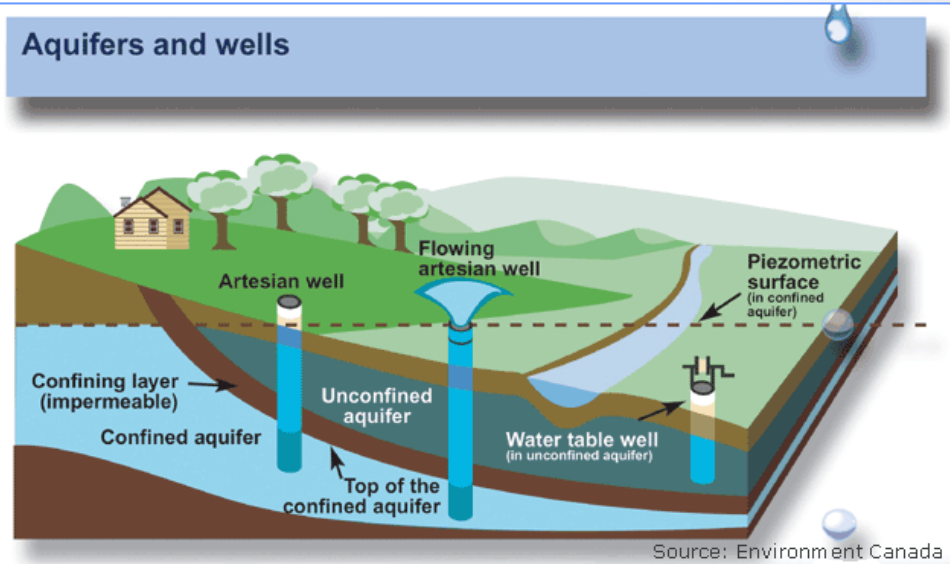
Surveys : 1996-2003

Country, Place, Year	Period of time	Type of water	Main isolation method	Most frequent fungal isolates
United Kingdom, 1996	Autumn and Spring	Surface water and network	Membrane filtration, Direct plating and Bating	<i>Aspergillus</i> , <i>Cladosporium</i> , <i>Epicoccum</i> , <i>Penicillium</i> and <i>Trichoderma</i>
Greece, Thessaloniki, 1998	One collection (126 samples)	Tap water (hospital and community)	Membrane filtration	<i>Penicillium</i> , <i>Aspergillus</i> and <i>Acremonium</i>
Greece, 85 haemodialysis units, 1998	One collection (255 samples)	Municipal water supplies of haemodialysis centres	Membrane filtration	<i>Penicillium</i> and <i>Aspergillus</i>
Germany, North Rhine-Westphalia, 1998/9	12 months	Drinking water	Pour-plating	<i>Acremonium</i> , <i>Exophiala</i> , <i>Penicillium</i> and <i>Phialophora</i>
Norway, 14 networks, 2002/3	December, June and September	Drinking water (surface and groundwater)	Membrane filtration	<i>Penicillium</i> , <i>Trichoderma</i> and <i>Aspergillus</i>

SURVEYS 2004-2010

Country, Place, Year	Period of time	Type of water	Main isolation method	Most frequent fungal isolates
Portugal, Braga, 2003/4	12 months	Tap water	Membrane filtration	<i>Penicillium</i> and <i>Acremonium</i>
Pakistan, Karachi, 2007	One collection (30 samples)	Water (and fruit juice)	Direct plating	<u><i>Aspergillus niger</i></u> and <i>A. clavatus</i>
Australia, Queensland, 2007/8	18 months	Municipal water	Membrane filtration	<i>Cladosporium</i> , <i>Penicillium</i> , <u><i>Aspergillus</i></u> and <i>Fusarium</i>
Brazil, Recife, 2009/10	5 months	Water treatment plant; tap water	Membrane filtration	<i>Penicillium</i> , <u><i>Aspergillus</i></u> and <i>Phoma</i>
Portugal, Lisbon, 2010	4 months	surface water; spring water; groundwater	Membrane filtration	<u><i>Aspergillus</i></u> , <i>Cladosporium</i> , <i>Penicillium</i>
Belgium, Liège	4 months	Tap water+MDS	Membrane filtration	<i>Fusarium</i> , <u><i>Aspergillus</i></u> , <i>Penicillium</i> , <i>Paecilomyces</i>

Origin of the water



Water
storage tanks

Highest concentration in autumn

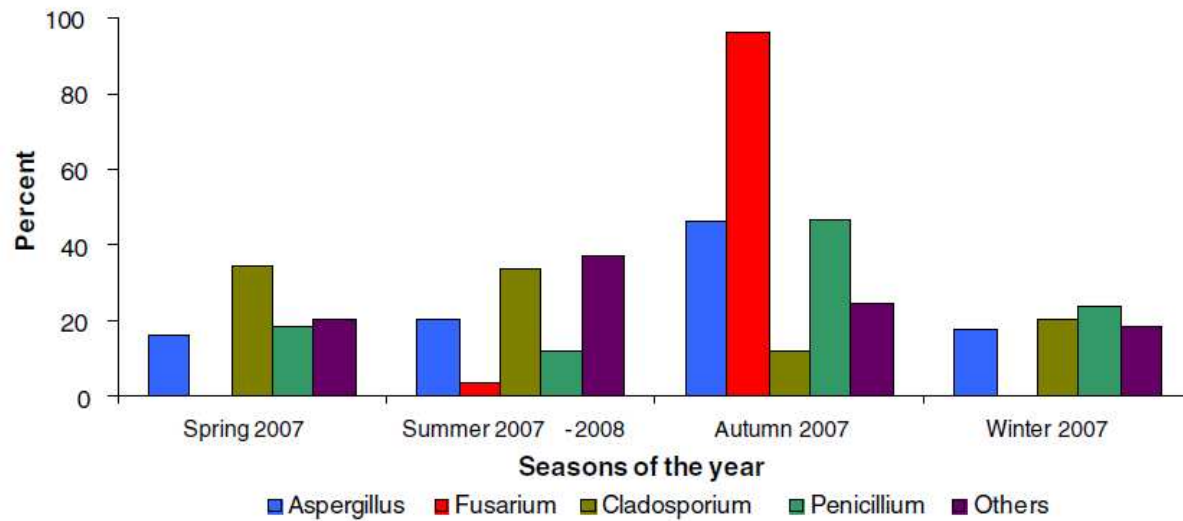


Figure 2 Distribution of fungal propagules in water samples collected from 4 different seasons of the year.

Fusariosis Associated with Pathogenic *Fusarium* Species Colonization of a Hospital Water System: A New Paradigm for the Epidemiology of Opportunistic Mold Infections

Elias J. Anaissie,¹ Robert T. Kuchar,² John H. Rex,² Andrea Francesconi,¹ Miki Kasai,¹ Frank-Michael C. Müller,¹ Mario Lozano-Chiu,² Richard C. Summerbell,³ M. Cecilia Dignani,¹ Stephen J. Chanock,⁴ and Thomas J. Walsh¹

Houston university Hospital, Texas, 2001
Numerous cases of *Fusariosis*
over a 10- year period

- 162/283 (57%) *Fusarium* sp. in water samples
- 18 strains of *F. solani* from patients/17 *F. solani* from environment

Table 2. Molecular biotyping profiles of related strains of *Fusarium solani* isolated from patient and environmental samples from a hospital in Houston, Texas.

Type of matched isolate, by source; isolate no. ^a	Pattern score, by laboratory and testing method ^b				Relatedness ^c
	Laboratory A			Laboratory B: RAPD	
	RAPD	RFLP	IR-PCR		
Patient-environment					
1381, 1370	Highly probable	Probable	Probable	Highly probable	Probably related
1379, 1369	Probable	Highly probable	Probable	Probable	Possibly related
Patient-patient					
1328, 1379	Probable	Highly probable	Highly probable	Probable	Probably related
1242, 1319	Highly probable	Probable	Probable	Highly probable	Probably related
1317, 1377	Highly probable	Probable	Probable	Highly probable	Probably related
Environment-environment					
1368, 1370	Probable	Highly probable	Probable	Probable	Possibly related

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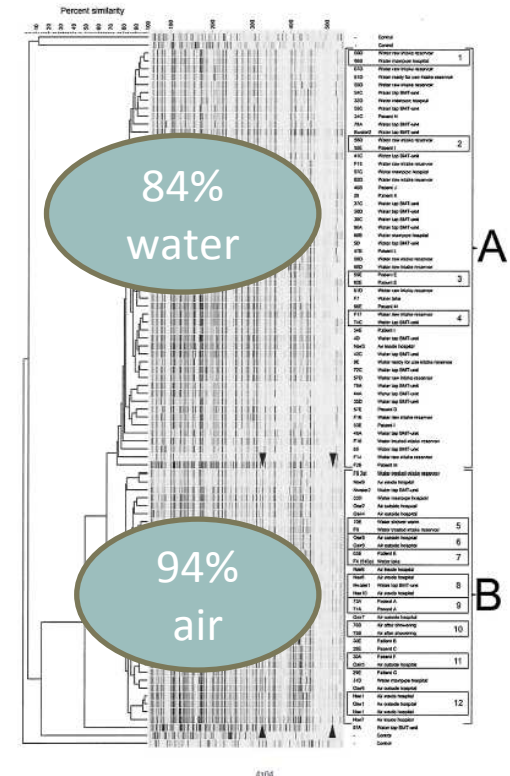
Houston university Hospital, Texas, 2001
Numerous cases of *Fusariosis*
over a 10- year period

1. First study that demonstrates that water is a **reservoir** for opportunistic fungi
2. Genetically diverse strains of *F. solani* can contaminate the water system and persist for years
3. WDS can disseminate the fungi by way of aerosols from shower and sink
4. Isolates of *Fusarium* can cause nosocomial infections

Molecular Epidemiology of *Aspergillus fumigatus* Isolates Recovered from Water, Air, and Patients Shows Two Clusters of Genetically Distinct Strains

Adilia Warris,^{1,2,3*} Corné H. W. Klaassen,⁴ Jacques F. G. M. Meis,⁴ Maaïke T. de Ruiter,⁴ Hanneke A. de Valk,⁴ Tore G. Abrahamsen,⁵ Peter Gaustad,³ and Paul E. Verweij^{2,6}

- University Hospital of Oslo, Norway_18 months
- Sampling of water, air, patients with IA
- Genotypic study



1. *A. fumigatus* in strains were clustered in 2 different genetic groups
2. Intake reservoir is the source of *A. fumigatus* strains found in tap water inside the hospital

Pathogenic *Aspergillus* Species Recovered from a Hospital Water System: A 3-Year Prospective Study

Elias J. Anaissie,¹ Shawna L. Stratton,¹ M. Cecilia Dignani,¹ Richard C. Summerbell,³ John H. Rex,⁴ Thomas P. Monson,² Trey Spencer,¹ Miki Kasai,⁵ Andrea Francesconi,⁵ and Thomas J. Walsh⁵

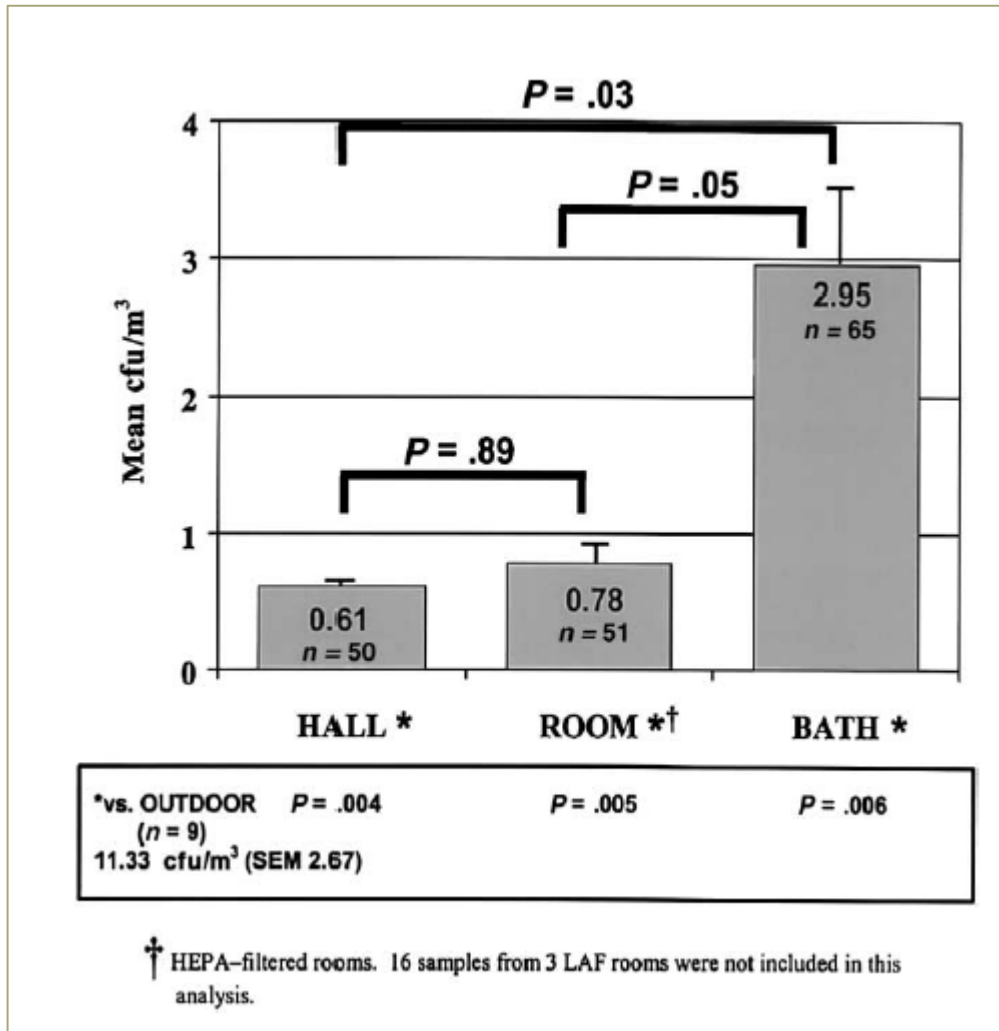
Study conducted in
the Hospital of
Little Rock,
Arkansas

- Comparison of genotypic profile of environmental strains and patients isolates

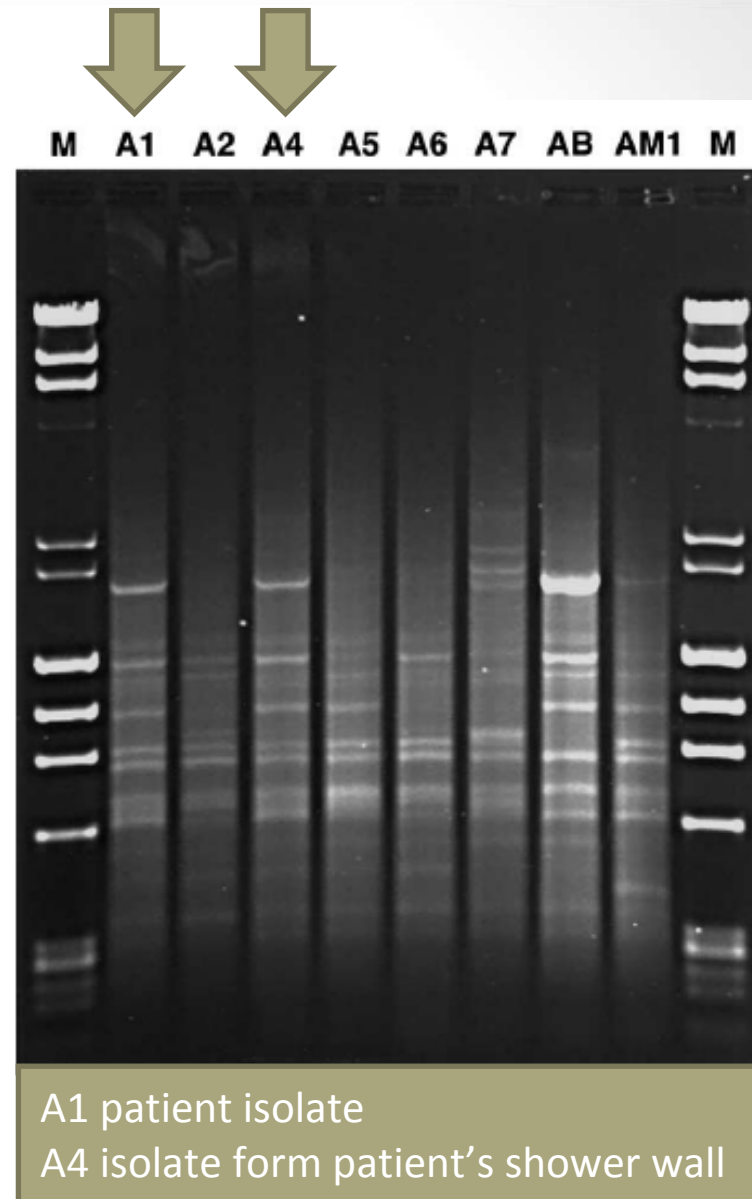
1. 21% *Aspergillus* positive water samples from patients care areas
2. Significantly higher concentration of air-borne propagules were found in bathrooms

{ 11 }

Higher rates in the shower



3. An isolate of *A. fumigatus* of a patient with IPA **was genotypically identical to an isolate recovered from the shower wall** of patient's room



Molecular analyses of *Fusarium* isolates recovered from a cluster of invasive mold infections in a Brazilian hospital

Christina M Scheel^{1*†}, Steven F Hurst^{1†}, Gloria Barreiros^{2†}, Tiyomi Akiti^{2†}, Marcio Nucci^{2†} and S Arunmozhi Balajee^{3†}



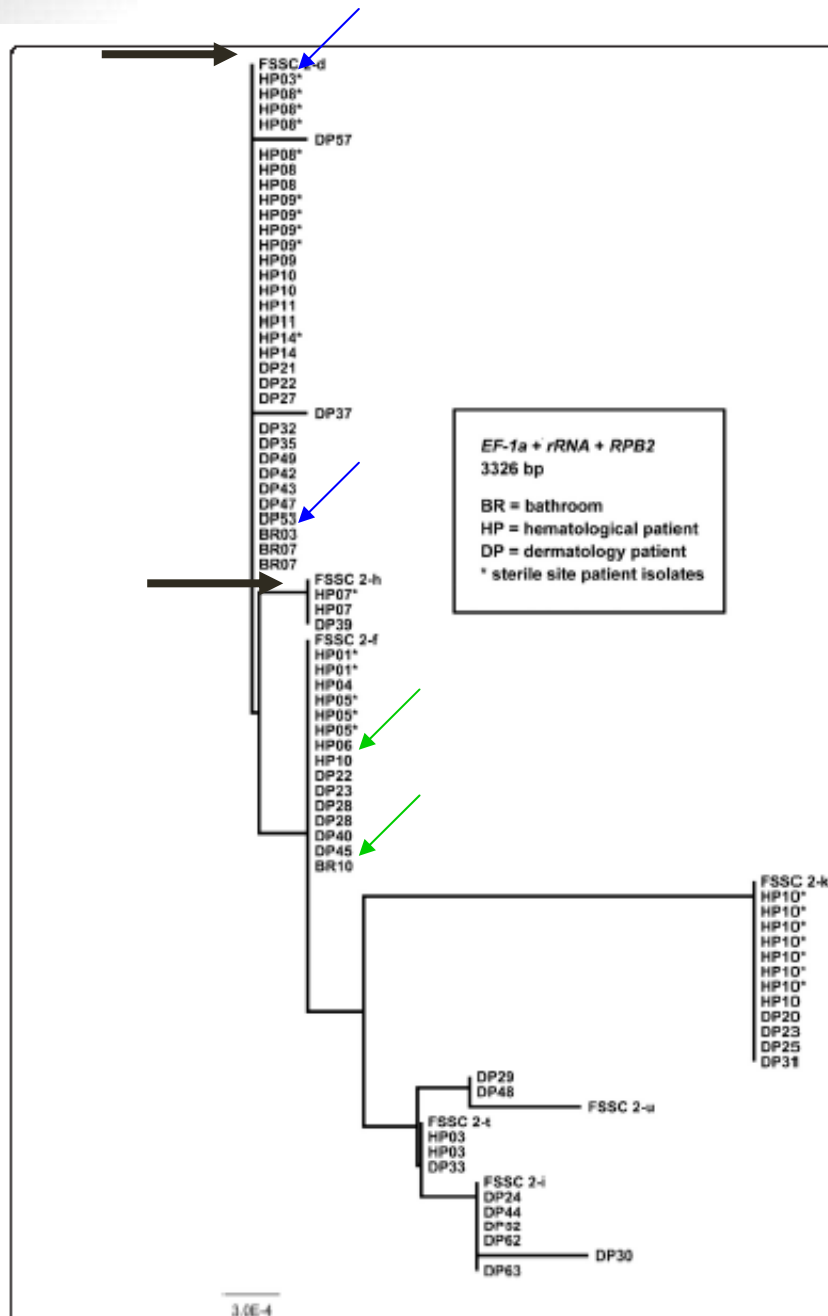
2005, University Hospital
of Rio de Janeiro
Increase of cases of
fusariosis

SEPT 2005 → OCT 2009
Patients isolates
Sampling of air-water-
water related surfaces

BONE MARROW
TRANSPLANT UNIT

DERMATOLOGIC
CLINIC

{ 14 }



Total: 166 *Fusarium isolates*

→ PATIENTS: 68% *F. solani* species complex (FSSC) in clinical samples (BMTU, dermatology clinic outpatients)

→ ENVIRONMENT: 50% *F. oxysporum* species complex FOSC

Two identical profiles between one strain from a BMTU patient and the bathroom but no temporal association

Figure 2 Neighbor-joining phylogeny of *Fusarium solani* species complex (FSSC) species 2 isolates ($n = 74$). Bootstrap values were 100% between individual taxa (1000 iterations, data not shown). Included are previously described FSSC species 2 voucher sequences (labeled FSSC 2-d, FSSC 2-f, FSSC 2-h, FSSC 2-i, FSSC 2-k, and FSSC 2-u) that were retrieved from the FUNGI ID database [8].

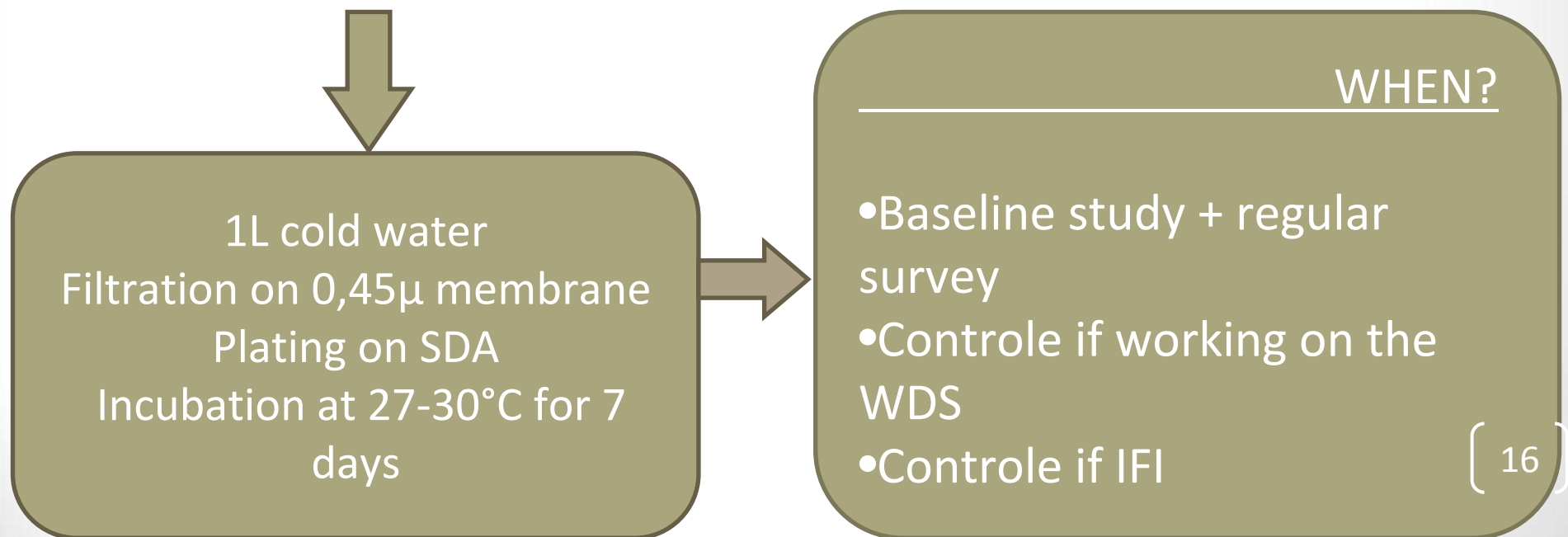
Sampling methodology:

« guidelines? »



Kauffmann-Lacroix C, BMC Infect Dis 2013

- French multicentric study from Feb 2004 → March 2005
- → no difference in colonisation between cold and hot water
- → No *Aspergillus fumigatus* but dematiaceous fungi +++



Sampling methodologies: media

	Glucose (g)	pH	Antibiotiques
CMA/2 Corn meal agar half streng	0	5.8-6.2	No
CZ Czapec agar	30	6.0-6.4	No
DG18 Dichloran 18% glycerol agar			
DRBC Dichloran RoseBengale Chloramphenicol agar			
NGRBA Neopeptone glucose rose Bengale aureomycine	10	6.3-6.7	Aureomycine
PDA Potato dextrose agar	20	5.4-5.8	No
MEA Malt extract agar	20	5.0-5.5	No
SDA Sabouraud dextrose agar	40	5.4-5.8	No

DG18: recommended medium: 2 avantages

→characteristic colony appearance

→inhibits overgrowth of fast growing fungi (*Trichoderma*, mucorales)

Study at the University Hospital of Liège



- CHU Liège: 955 Beds, 3 sites, Surface and underground water
- Filtration + chlorination before to enter the hospital and $>65^{\circ}\text{C}$ for hot water.
- Study during 4 months from Feb 2005- March 2006
- Methodology: 197 sampling points
 - 500 ml cold and 500 ml hot water
 - filtration on $0,45\mu$ Millipore membranes
 - Sabouraud agar medium (+ATB)
 - Incubation at 30°C for 1 month

Results

- contamination rate: 51%

Site 1

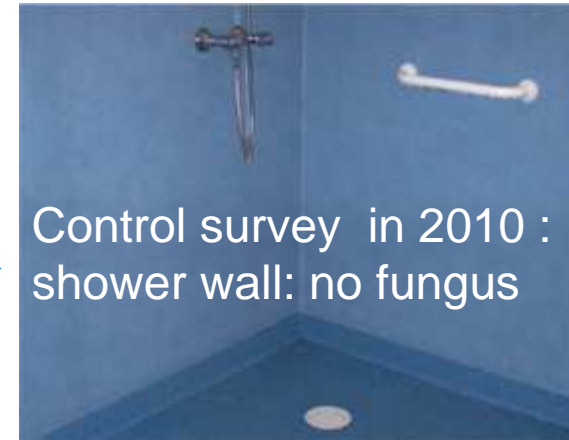
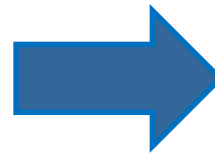
Site 2

Site 3

Filamentous fungi	Sampling sites				P (ST, OA, NDB)
	Three sites (N = 197) n (%)	ST (N = 107) n (%)	OA (N = 40) n (%)	NDB (N = 50) n (%)	
<i>Acremonium</i> spp.	3 (1.5)	1 (0.9)	1 (2.5)	1 (2)	NS
<i>Alternaria</i> spp.	1 (0.5)	1 (0.9)	–	–	NS
<i>Aspergillus</i> spp.	12 (6)	8 (7.4)	4 (10)	–	NS
<i>A. flavus</i>	2 (1)	2 (1)	–	–	NS
<i>A. fumigatus</i>	4 (2)	2 (1)	2 (5)	–	NS
<i>A. nidulans</i>	2 (1)	2 (1)	–	–	NS
<i>A. niger</i>	2 (1)	–	2 (1)	–	NS
<i>A. sydowii</i>	2 (1)	2 (1)	–	–	NS
<i>Cladosporium</i> spp.	6 (3)	4 (3.7)	1 (2.5)	1 (2)	NS
<i>Fusarium</i> spp.	23 (11.6)	3 (2.8)	6 (15)	14 (28)	≤0.001
<i>Monilia</i> spp.	7 (3.5)	2 (1.8)	3 (7.5)	2 (4)	NS
<i>Paecilomyces</i> spp.	14 (7)	6 (5.6)	–	8 (16)	≤0.05
<i>Penicillium</i> spp.	22 (11.2)	9 (8.4)	6 (15)	7 (14)	NS
Sterile mycelia	10 (5)	5 (4.6)	1 (2.5)	4 (8)	NS
<i>Trichoderma</i> spp.	4 (2)	1 (0.9)	2 (5)	1 (2)	NS
Contamination rates	102 (51)	40 (37.3)	24 (60)	38 (76)	≤0.01

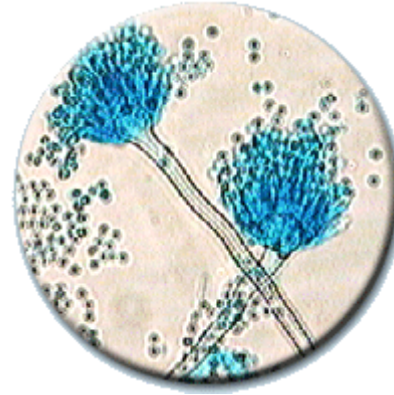
ST, Sart Tilman; OA, Ourthe-Amblève; NDB, Notre-Dame des Bruyères; N, total number of tested samples; n, number of samples; NS, not significant; (–), no observation.

Conclusion of the study and preventive measures



- Implementation: quite easy
- Regular replacement more difficult to implement !!!

Conclusion



- There is a need for guidelines
- Every hospital with severe immunosuppressed patients should be aware of the potential danger
- →sampling tap water and shower walls
- →implement point-of-use filtration systems and organize replacement
- →avoid showering during severe immunosuppression
- →replace shower by baths

