



# Etablierung einer real-time PCR für *Rasamsonia argillacea* Komplex bei Patienten mit Mukoviszidose

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## Occurrence and relevance of filamentous fungi in respiratory secretions of patients with cystic fibrosis – a review

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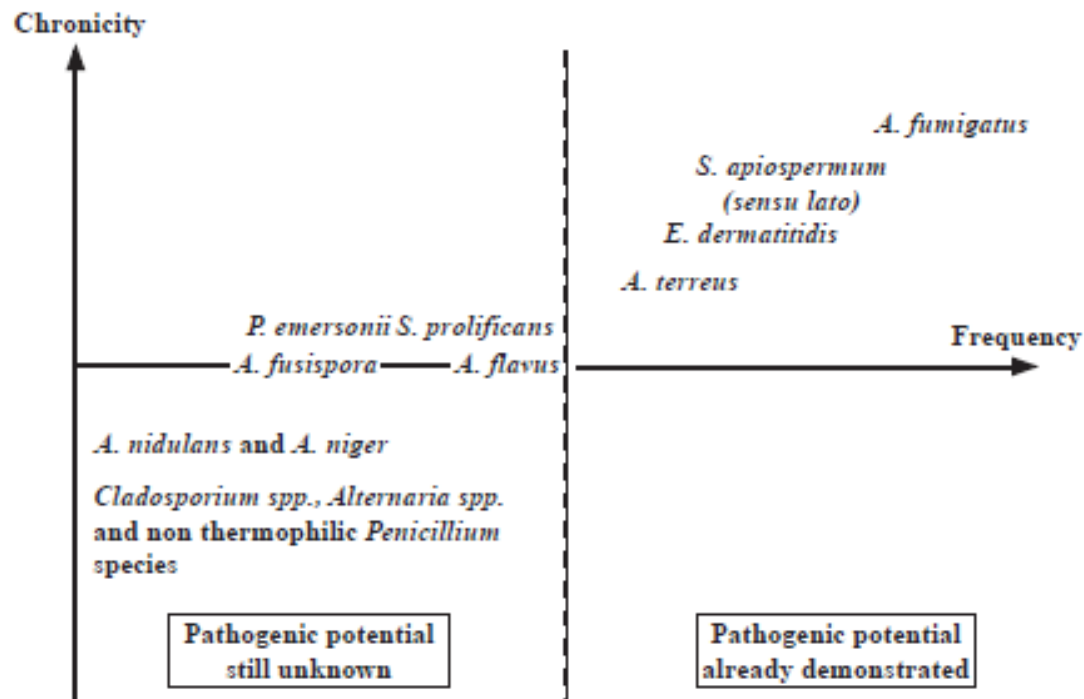


Fig. 2 Filamentous fungi associated with CF according to their frequency in this clinical context and to their capacity to chronically colonize the airways.

# Rasamsonia Taxonomy

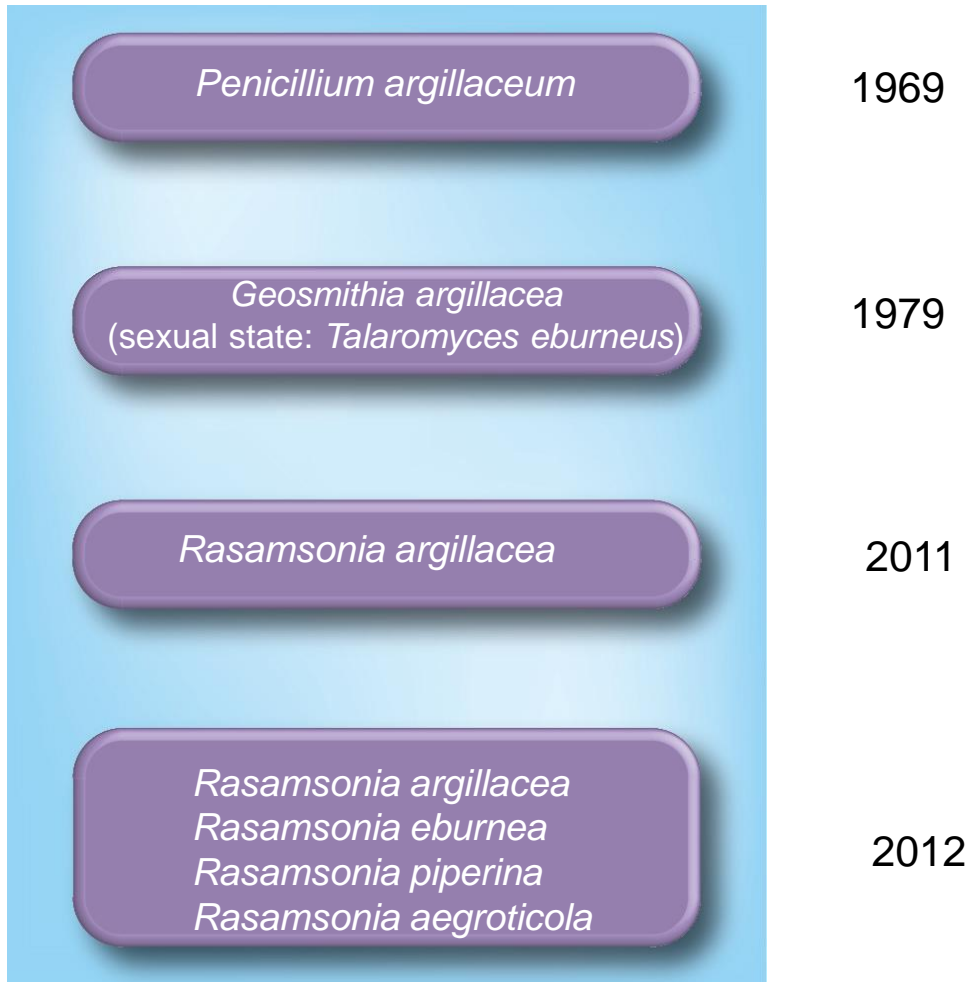


Figure 1. Taxonomic changes in the *Rasamsonia argillacea* species complex.

# Phenotypic characteristics

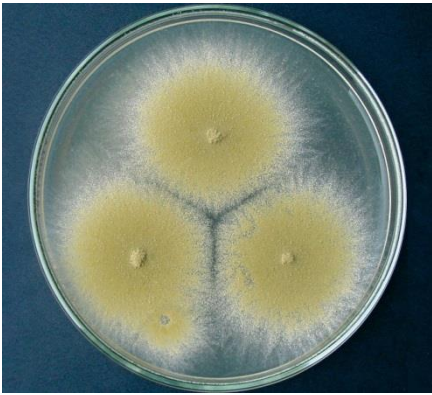
TABLE 2 Differential characteristics of *Rasamsonia* species<sup>a</sup>

Species	Colony diam (mm), growth on CYA (7 days, 37°C)	Shape and size of conidia	Length/width ratio of conidia	Reverse color on MEA
<i>R. aegroticola</i>	(20–) 25–40, good growth	Variable; predominantly cylindrical or ellipsoid, 2.5–3.5 × 1.8–2.5 μm	1.3–1.6:1	Grayish brown
<i>R. argillacea</i>	30–40, good growth	Cylindrical or ovoid, (3–) 3.5–4.5 (–5.0) × 1.5–2.0 (–2.3) μm	1.8–2.3:1	(Light) brown
<i>R. brevistipitata</i>	11–17, good growth	Ellipsoidal or ovoid, (2.0–) 2.5–3.0 (–3.5) × 1.7–2.1 μm	1.3–1.5:1	(Light) brown
<i>R. cylindrospora</i>	5–10, good growth	Cylindrical, 4.0–5.0 × 1.6–2.1 μm	2.1–2.5:1	Blackish brown
<i>R. eburnea</i>	30–40, good growth	Variable, predominantly ellipsoidal or ovoid but also cylindrical, 2.5–3.5 (–4) × 1.8–2.5 μm	1.1–1.4:1	Dark brown or blackish brown
<i>R. piperina</i>	CYA (10–)15–25, moderate growth	Ellipsoidal or cylindrical, 2.0–3.5 × 1.7–2.5 μm	1.3–1.7:1	Brown or grayish brown

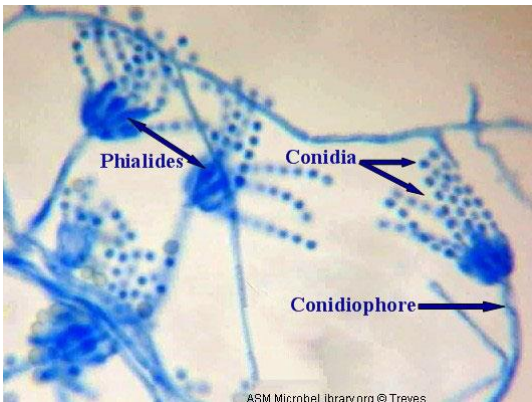
<sup>a</sup> The phenotypically similar species *R. cylindrospora* and *R. brevistipitata* are included, while the ascoma- and ascospore-producing species *R. bysochlamydoides*, *R. composticola*, and *R. emersonii* are excluded.



# Similarities to *Paecilomyces* spp. and *Penicillium* spp.



*Paecilomyces* spp.



*Penicillium* spp.

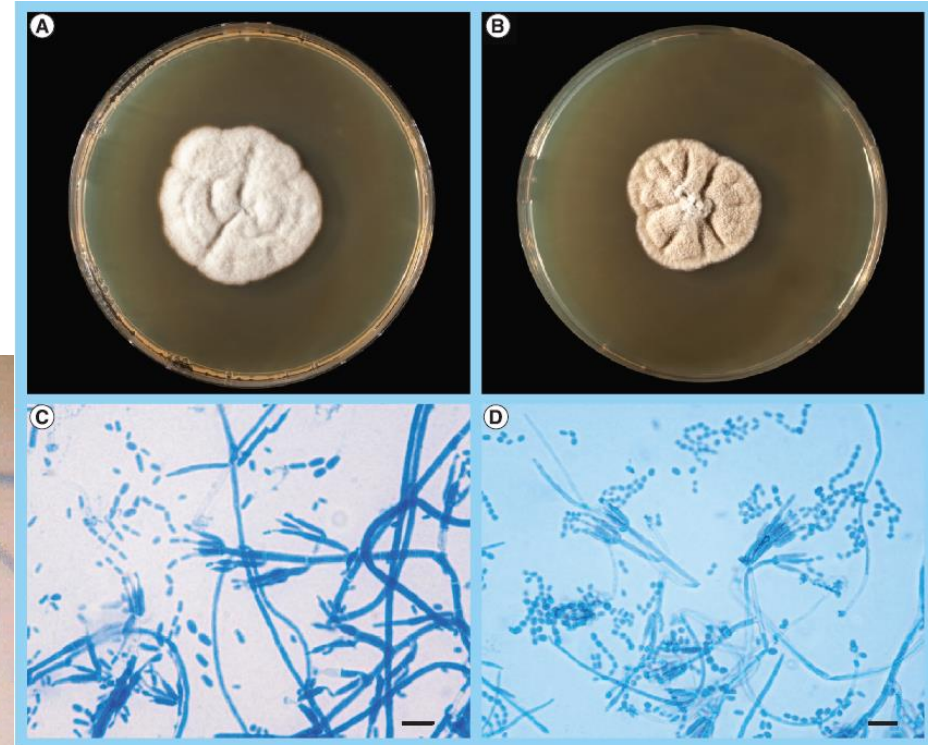


Figure 2. Morphological features of *Rasamsonia argillacea* and *Rasamsonia aegroticola*. (A & C) *Rasamsonia argillacea* and (B & D) *Rasamsonia aegroticola*. (A & B) Seven-day-old cultures at 37°C on yeast extract-peptone-dextrose-agar medium. (C & D) Microscopic examination with lactic blue stain. Scale bars: 10 µm.

*Rasamsonia* spp.

# Susceptibility against antifungal agents

TABLE 3 Susceptibility results of *Rasamsonia* species, including GM and MIC distributions by species and antifungal agent<sup>a</sup>

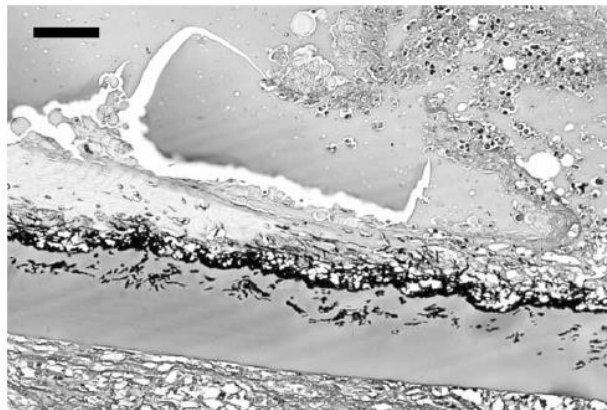
Species	No. of isolates	GM MIC (mg/liter) (range)				
		AMB	ITZ	PCZ	CAS	VCZ
<i>R. aegroticola</i>	5	2.00 (0.5–2)	1.32 (1–2)	1.74 (1–4)	0.19 (0.06–0.5)	>16
<i>R. argillacea</i>	6	2.00 (0.125–2)	5.04 (1–32)	3.17 (1–4)	0.28 (0.125–0.5)	>16
<i>R. brevistipitata</i>	2	0.09 (0.03–0.25)	0.12 (0.06–0.25)	0.06 (0.03–0.12)	0.35 (0.25–0.5)	0.12 (0.06–0.25)
<i>R. cylindrospora</i>	2	1.41 (1–2)	1.41 (1–2)	2.83 (1–8)	0.13	>16
<i>R. eburnea</i>	4	2.00 (1–4)	2.38 (1–4)	2.00 (1–4)	0.35 (0.125–0.5)	>16
<i>R. piperina</i>	11	1.22 (0.25–2)	1.00 (0.5–1)	1.41 (0.06–2)	0.25 (0.03–0.5)	19.33 (8–>16)

<sup>a</sup> The phenotypically similar species *R. cylindrospora* and *R. brevistipitata* are included in this analysis. Given are the geometric mean MICs, and the range in MICs, if observed, is given in parentheses. AMB, amphotericin B; ITZ, itraconazole; PCZ, posaconazole; CAS, caspofungin; VCZ, voriconazole.

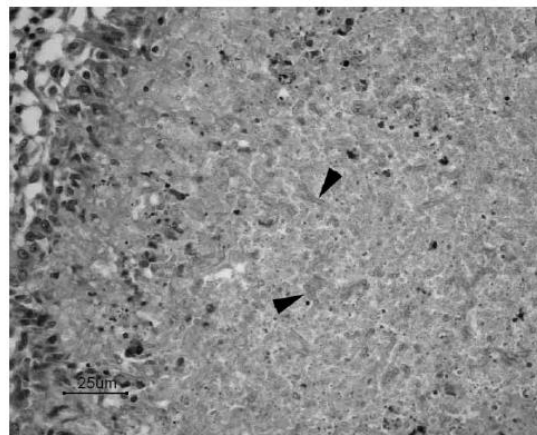
# Disseminated *Geosmithia argillacea* infection in a German Shepherd dog

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**Fig. 1** GMS stain, eye, (bar equals 50 microns). Multiple septate hyphae invading the anterior lens capsule and lens cortical material.



**Fig. 4** H&E, kidney (bar equals 25 microns). The centers of granulomas are necrotic and contain poorly staining septate, dichotomous branching fungal hyphae (arrowheads) with bulbous endings.



# Rasamsonia bei septische Granulomatose

JOURNAL OF CLINICAL MICROBIOLOGY, Apr. 2011, p. 1681–1683

0095-1137/11/\$12.00 doi:10.1128/JCM.02456-10

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## Emergence of Disseminated Infections Due to *Geosmithia argillacea* in Patients with Chronic Granulomatous Disease Receiving Long-Term Azole Antifungal Prophylaxis<sup>∇</sup>

Marie Machouart,<sup>1</sup> Dea Garcia-Hermoso,<sup>2,3</sup> Alexandre Rivier,<sup>1</sup> Noura Hassouni,<sup>6</sup>  
Emilie Catherinot,<sup>4</sup> Alexandra Salmon,<sup>5</sup> Anne Debourgogne,<sup>1</sup> H el ene Coignard,<sup>4</sup>  
Marc Lecuit,<sup>4</sup> Marie-Elisabeth Bougnoux,<sup>6</sup> St ephane Blanche,<sup>7</sup>  
and Olivier Lortholary<sup>2,3,4\*</sup>

**We report two cases of invasive infections due to *Geosmithia argillacea* an emerging mold, in patients with chronic granulomatous disease receiving prolonged azole antifungal prophylaxis. One patient died despite receiving a combination of four antifungals, and the other developed cerebral and medullary lesions under a combination of caspofungin, posaconazole, terbinafine, and gamma interferon.**

MAJOR ARTICLE

7 patients, 3 died

## *Geosmithia argillacea*: An Emerging Cause of Invasive Mycosis in Human Chronic Granulomatous Disease

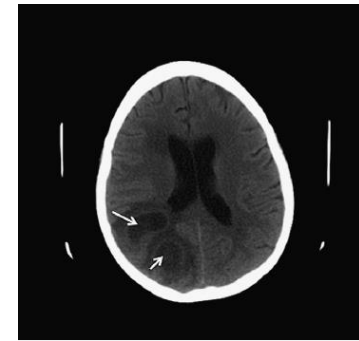
Suk See De Ravin,<sup>1</sup> Malliswari Challipalli,<sup>2</sup> Victoria Anderson,<sup>3</sup> Yvonne R. Shea,<sup>4</sup> Beatriz Marciano,<sup>3</sup> Dianne Hilligoss,<sup>1</sup> Martha Marquesen,<sup>1</sup> Rosamma DeCastro,<sup>1</sup> Yen-chun Liu,<sup>5</sup> Deanna A. Sutton,<sup>6</sup> Brian L. Wickes,<sup>6</sup> Patricia L. Kammeyer,<sup>2</sup> Lynne Sigler,<sup>7</sup> Kathleen Sullivan,<sup>8</sup> Elizabeth M. Kang,<sup>1</sup> Harry L. Malech,<sup>1</sup> Steven M. Holland,<sup>3</sup> and Adrian M. Zelazny<sup>4</sup>

Clinical Infectious Diseases 2011;52(6):e136–e143

Published by Oxford University Press on behalf of the Infectious Diseases Society of America 2011.

1058-4838/2011/526-0001\$37.00

DOI: 10.1093/cid/ciq250





## Disseminated *Geosmithia argillacea* infection in a patient with gastrointestinal GvHD

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I Zollner-Schwetz<sup>1</sup>, M Hoenigl<sup>1</sup>, HJF Salzer<sup>1</sup>, R Krause<sup>1</sup>  
and W Buzina<sup>3</sup>

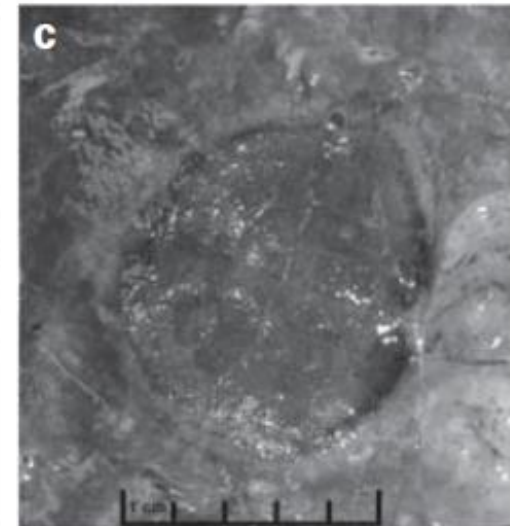
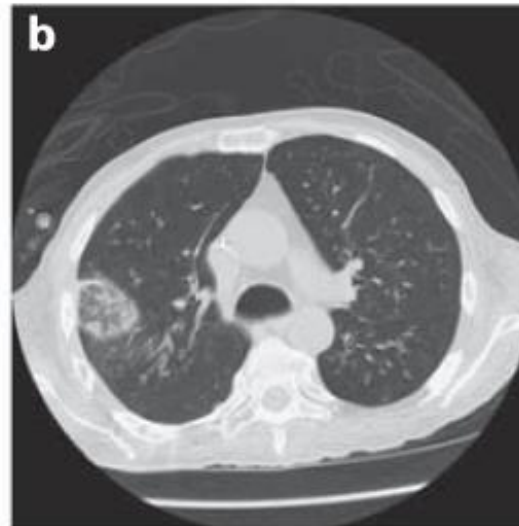
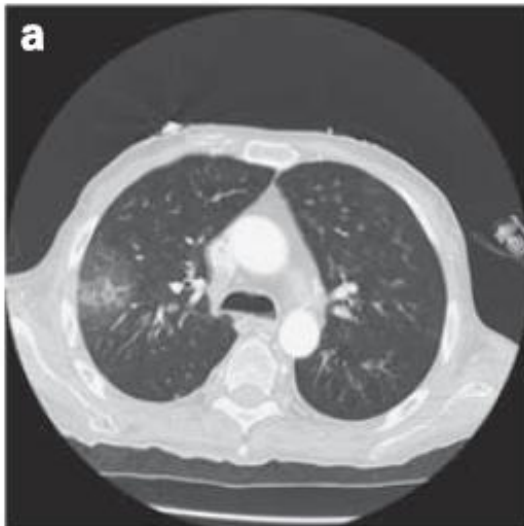
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## Case Report

Clinical Microbiology

Ann Lab Med 2013;33:136-140

<http://dx.doi.org/10.3343/alm.2013.33.2.136>

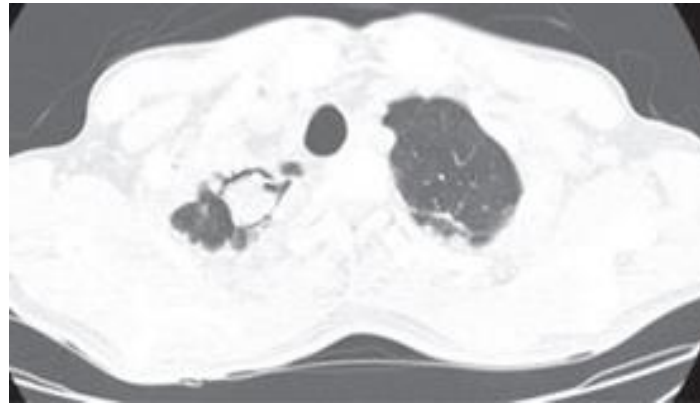
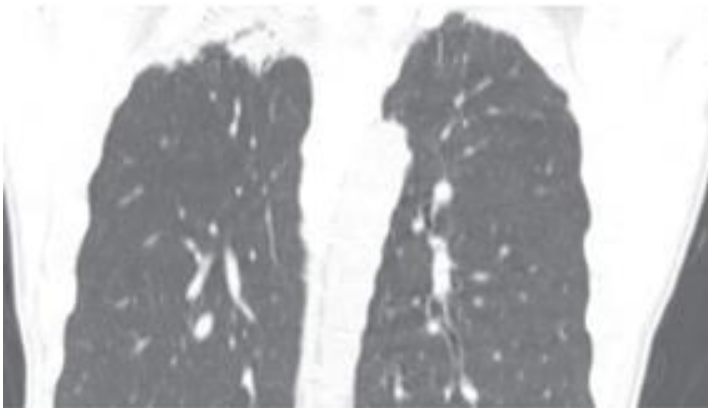
ISSN 2234-3806 • eISSN 2234-3814

# ANNALS OF LABORATORY MEDICINE

## Isolation and Identification of *Geosmithia argillacea* from a Fungal Ball in the Lung of a Tuberculosis Patient

Ji Yeon Sohn, M.D., Mi-Ae Jang, M.D., Jang Ho Lee, M.T., Kyung Sun Park, M.D., Chang-Seok Ki, M.D.,  
and Nam Yong Lee, M.D.

Department of Laboratory Medicine and Genetics, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea



## *Rasamsonia argillacea* Pulmonary and Aortic Graft Infection in an Immune-Competent Patient

Jeffrey B. Doyon,<sup>a</sup> Deanna A. Sutton,<sup>b</sup> Pierre Theodore,<sup>c</sup> Gurmohan Dhillon,<sup>d</sup> Kirk D. Jones,<sup>e</sup> Elizabeth H. Thompson,<sup>b</sup> Jianmin Fu,<sup>f</sup> Brian L. Wickes,<sup>f</sup> Jane E. Koehler,<sup>g</sup> Brian S. Schwartz<sup>g</sup>

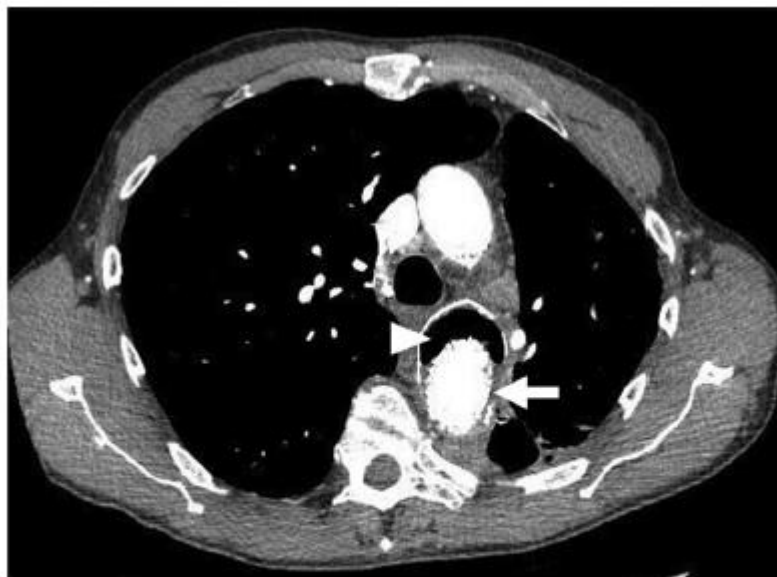


FIG 2 Selected axial image from a contrast-enhanced study of the thorax, with arterial phase timing at the proximal descending thoracic aorta. The arrow identifies an endovascular stent graft for treatment of an aortic aneurysm. The arrowhead indicates gas within the residual aneurysm sac.

# Rasamsonia in cystic fibrosis

JOURNAL OF CLINICAL MICROBIOLOGY, July 2010, p. 2381–2386  
0095-1137/10/\$12.00 doi:10.1128/JCM.00047-10  
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## *Geosmithia argillacea*: an Emerging Pathogen in Patients with Cystic Fibrosis<sup>∇</sup>

Sandrine Giraud,<sup>1\*</sup> Marc Pihet,<sup>1,2</sup> Bienvenue Razafimandimby,<sup>1</sup> Jacqueline Carrère,<sup>3</sup> Nicolas Degand,<sup>3</sup> Laurent Mely,<sup>4</sup> Loïc Favennec,<sup>5</sup> Eric Dannaoui,<sup>6,7</sup> Jean-Philippe Bouchara,<sup>1,2</sup> and Alphonse Calenda<sup>1</sup>

TABLE 2. Clinical characteristics of CF patients colonized by *Geosmithia argillacea*

Patient	Hospital location	Sex <sup>a</sup>	Age (yr) at CF diagnosis	Relevant <i>CFTR</i> mutation	Age (yr) at first isolation of <i>Geosmithia argillacea</i>
A	Angers	F	14	F508del/C.622-248-4del20pb	23
B	Angers	M	Birth	F508del/F508del	8
C	Giens	F	2	F508del/F508del	7
D	Giens	M	Birth	F508del/F508del	6
E	Giens	M	20	F508del/E92K	36
F	Giens	M	4	F508del/F508del	17
G	Rouen	F	Birth	F508del/F508del	13
H	Rouen	F	14	F508del/I336K	48
I <sup>b</sup>	Giens	M	Birth	F508del/F508del	12

<sup>a</sup> F, female; M, male.

<sup>b</sup> This patient was reported previously (6) as having chronic airway colonization by *Penicillium emersonii*.

# Rasamsonia in cystic fibrosis

JOURNAL OF CLINICAL MICROBIOLOGY, July 2010, p. 2615–2617  
 0095-1137/10/\$12.00 doi:10.1128/JCM.00184-10  
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## Isolation of the Fungus *Geosmithia argillacea* in Sputum of People with Cystic Fibrosis<sup>∇</sup>

R. C. Barton,<sup>1\*</sup> A. M. Borman,<sup>4</sup> E. M. Johnson,<sup>4</sup> J. Houbraken,<sup>5</sup> R. P. Hobson,<sup>1</sup> M. Denton,<sup>2</sup>  
 S. P. Conway,<sup>3</sup> K. G. Brownlee,<sup>3</sup> D. Peckham,<sup>3</sup> and T. W. R. Lee<sup>3</sup>

TABLE 1. Number of sputum samples and sputum samples positive for *G. argillacea*, clinical details, and other culture results

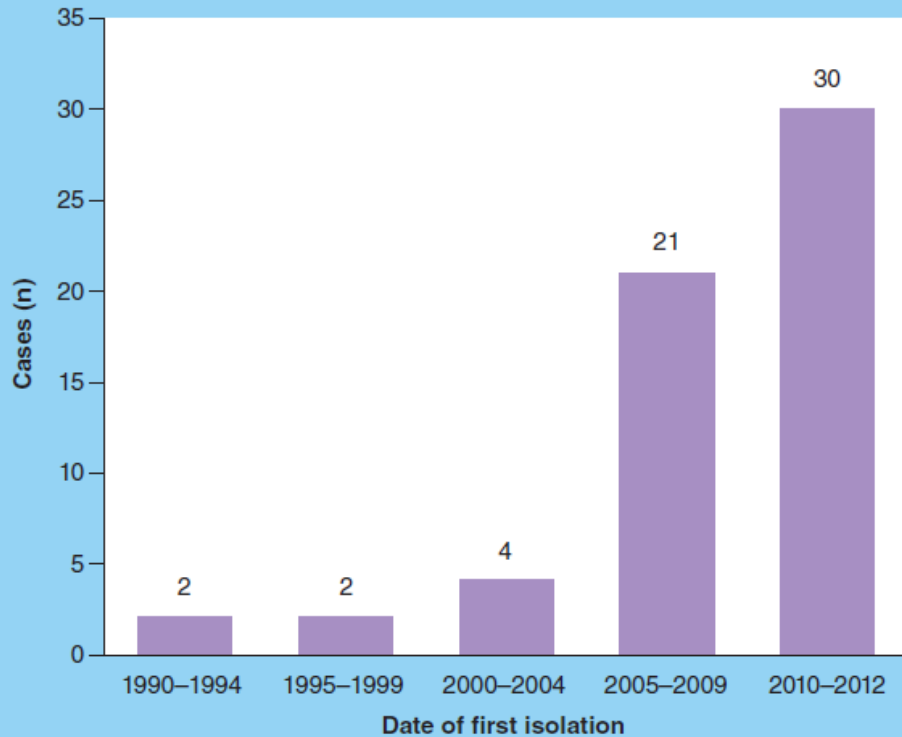
Characteristic	Value/description for subject:							
	1	2	3	4	5	6	7	8
No. of sputum samples	79	73	68	13	11	26	78	91
No. of positive samples (%)	29 (36.7)	1 (1.4)	11 (16.8)	9 (69)	15 (73.3)	2 (7.7)	1 (1.3)	29 (31.9)
Age at first isolation (yr)	26	24	27	21	6	19	8	14
CF genotype	DF508 DF508	DF508 DF508	DF508 2184delA	DF508 Q493X	DF508 DF508	DF508 DF508	DF508 1717-1(G>T)	DF508 DF508
FEV1 at first isolation (% predicted) <sup>c</sup>	15	82	17	55	73	58	58	27
Other organisms <sup>a</sup>	Af, As, Mv	Af, Sa, MRSA, Pa, Mc	Bccm	Af, Sa, Pa	Af, Sm	Af, Pa, Mc	Bccc, Pa, Af, Ma	Af, Psp, Pa, Ps, Ax, As, Mv
Any associated decline? Comment(s) <sup>b</sup>	No Listed for lung transplantation prior to isolation	No Two episodes of hemoptysis within 2 months of isolation, not; to be related	No Subsequently listed for lung transplantation	No	No CT chest shows plugged dilated bronchus and nodularity in right upper lobe	No	No	No Listed for lung transplantation prior to isolation thought has subsequently undergone transplantation

<sup>a</sup> Af, *Aspergillus fumigatus*; As, *Alcaligenes* sp.; Ma, *Mycobacterium abscessus*; Sa, *Staphylococcus aureus*; MRSA, methicillin-resistant *S. aureus*; Pa, *Pseudomonas aeruginosa*; Mc, *Mycobacterium chelonae*; Bccm, *Burkholderia cepacia* complex sp. *B. multivorans*; Sm, *Streptococcus milleri*; Bccc, *Burkholderia cepacia* complex sp. *B. cenocepacia*; Psp, *Paecilomyces* sp.; Ps, *Pseudoxanthomonas* sp.; Ax, *Achromobacter xylosoxidans*; Mv, *Mycobacterium avium*; Hf, *Haemophilus influenzae*.

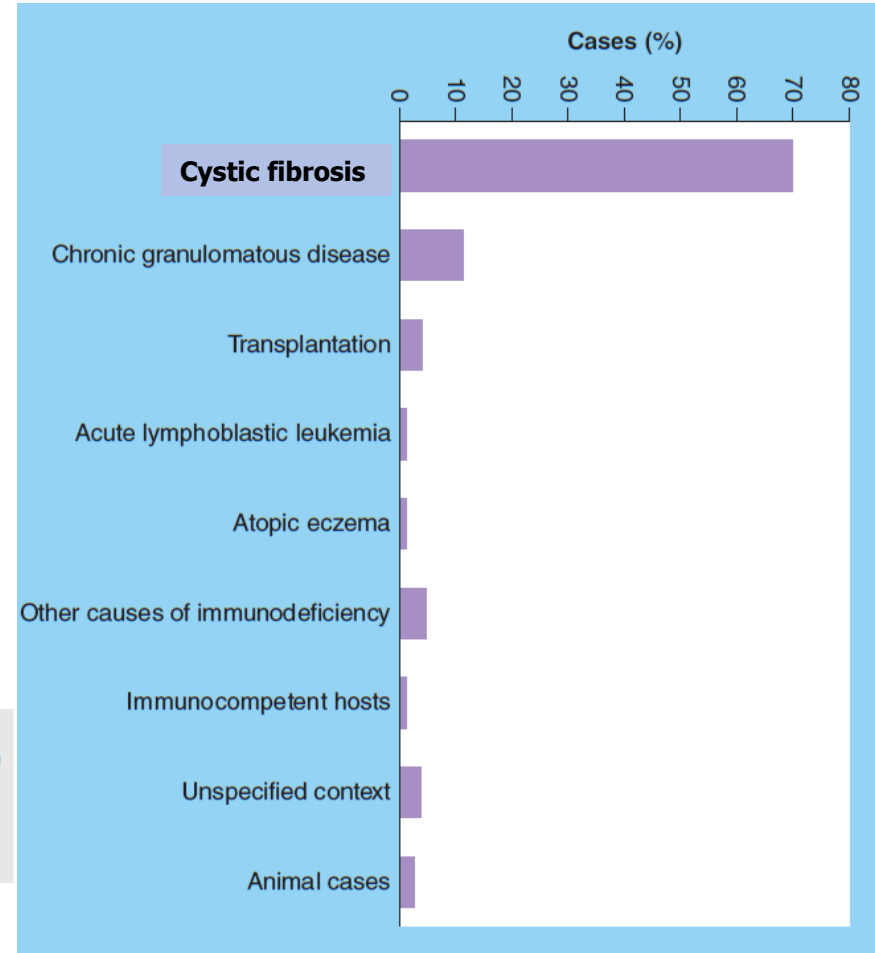
<sup>b</sup> CT, computed tomography.

<sup>c</sup> FEV1, forced expiratory volume in 1 s.

# Prevalence and clinical context



**Figure 3. Distribution of the cases of *Rasamsonia argillacea* colonization/infection by 5-year periods.** All the cases of *Rasamsonia argillacea* colonizations/infections (80) reported in the medical literature or in congresses were analyzed. Records of isolation were available for 59 of them and a classification of these cases by 5-year periods was performed.

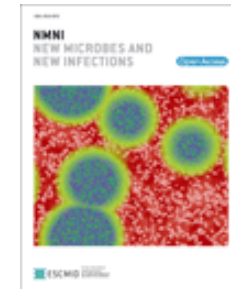


**Figure 4. Predisposing factors for *Rasamsonia argillacea* colonizations/infections.**

## Validation of a novel real-time PCR for detecting *Rasamsonia argillacea* species complex in respiratory secretions from cystic fibrosis patients

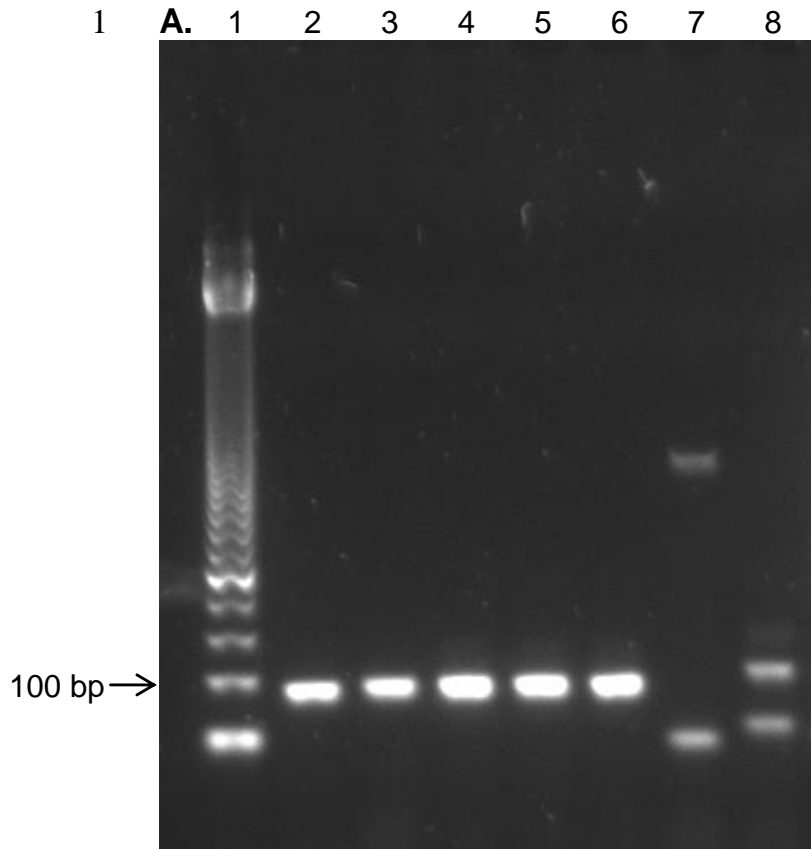
J. Steinmann<sup>1</sup>, S. Giraud<sup>2</sup>, D. Schmidt<sup>1</sup>, L. Sedlacek<sup>3</sup>,  
A. Hamprecht<sup>4</sup>, J. Houbraken<sup>5</sup>, J. F. Meis<sup>6,7</sup>, J. P. Bouchara<sup>2,8</sup>,  
J. Buer<sup>1</sup> and P.-M. Rath<sup>1</sup>

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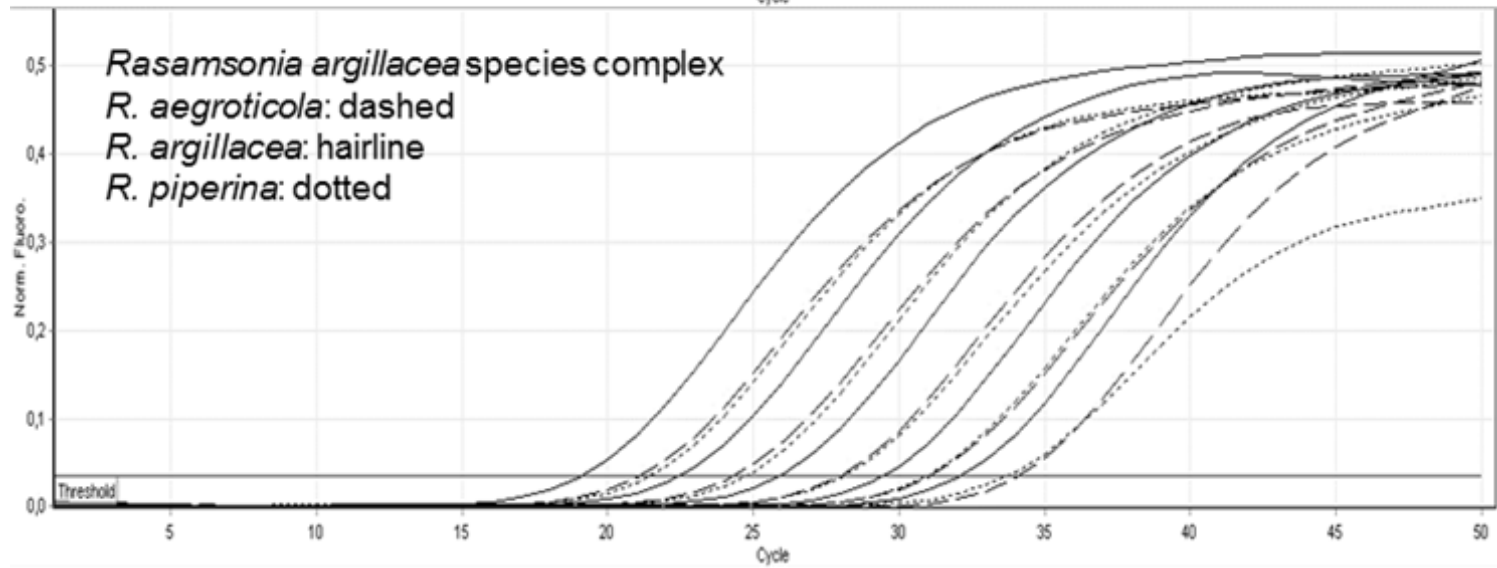
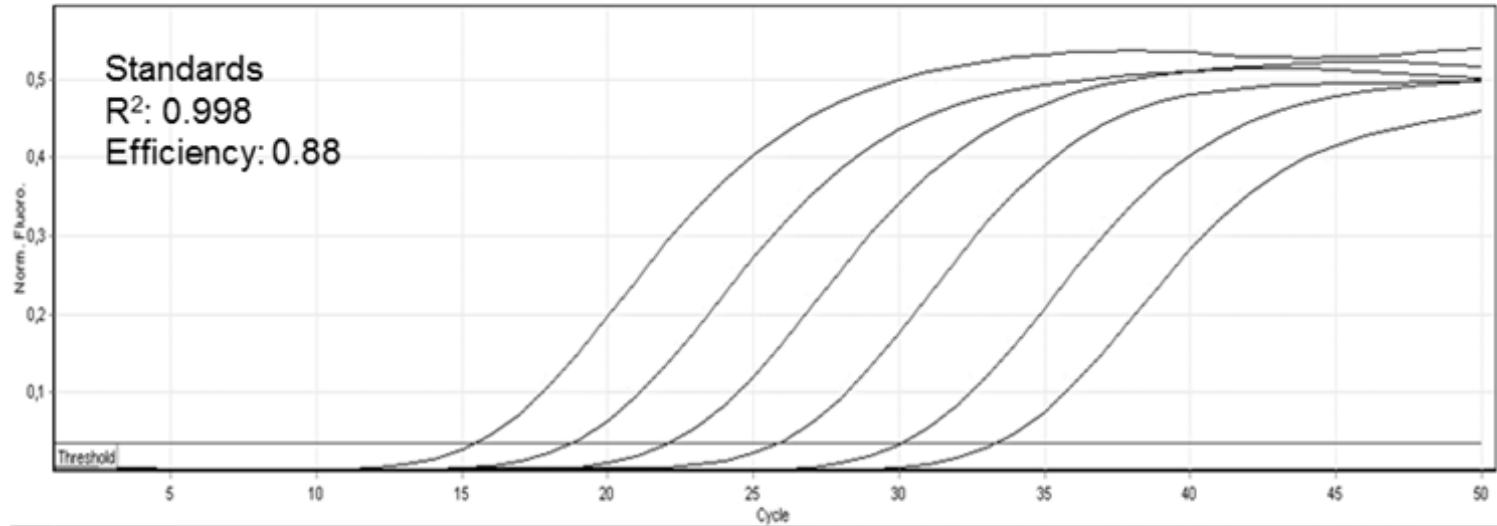
# Primer



**Figure 1.** Results of the real-time PCR assay. (A) Results obtained with DNA extracts from pure cultures of reference strains: Lane 1, 50-bp DNA marker; Lane 2, *R. argillacea* reference strain; Lane 3, *R. eburnea*; Lane 4, *R. aegroticola*; Lane 5, *R. piperina*; Lane 6, *R. argillacea*; Lane 7, *R. cyclindrospora*; Lane 8, *R. brevistipitata*; Lane 9, 50-bp DNA marker.



# Probes



**TABLE 2. *Rasamsonia* culture isolates (n = 35) used for validation of the *Rasamsonia argillacea* species complex PCR**

Species	Strain no.	Source of isolation	Origin
<i>Rasamsonia</i> reference strains (n = 22)			
<i>R. aegroticola</i>	DTO 049D4	Sputum from CF patient	UK
<i>R. aegroticola</i>	DTO 137A8 CBS 132819	Respiratory secretion from CF patient	France
<i>R. argillacea</i>	CBS 128787	Heat-treated fruit concentrate	The Netherlands
<i>R. argillacea</i>	DTO 097E4	Mine tip	UK
<i>R. argillacea</i>	DTO 097E5	Air	UK
<i>R. argillacea</i>	DTO 097E7	Unknown source	UK
<i>R. brevistipitata</i>	DTO 25H2	Indoor environment of school	Germany
<i>R. brevistipitata</i>	DTO 26B1	Indoor environment of school	Germany
<i>R. cylindrospora</i>	DTO 138F7	Sputum	The Netherlands
<i>R. cylindrospora</i>	DTO 138F8	Culture contaminant	UK
<i>R. eburnea</i>	DTO 045I3	Contaminant of blood culture	UK
<i>R. eburnea</i>	DTO 049D9	Peritoneal dialysis fluid	UK
<i>R. piperina</i>	DTO 076F1	Seed of <i>Piper nigrum</i>	Spain
<i>R. piperina</i>	DTO 097E6	Wood chips of <i>Picea abies</i>	Sweden
<i>R. piperina</i>	DTO 097E9	Bronchial washing	Canada
<i>R. piperina</i>	DTO 138F5	Necropsy thoracic vertebra of dog	USA
<i>R. piperina</i>	DTO 138F6	Necropsy thoracic vertebra of dog	USA
<i>R. piperina</i>	DTO 139F9	Air	Germany
<i>R. piperina</i>	DTO 138G1	Wood chips of <i>Picea abies</i>	Sweden
<i>R. piperina</i>	DTO 138G2	Wood chips of <i>Picea abies</i>	Sweden
<i>R. piperina</i>	DTO 138G3	Seed of <i>Piper nigrum</i>	Spain
<i>R. piperina</i>	CBS 128034	Necropsy ex vertebra of canine	USA
<i>Rasamsonia</i> clinical isolates (n = 13)			
<i>R. aegroticola</i>	IMMI 1419	BAL from lung transplant recipient	Germany
<i>R. aegroticola</i>	IMMI 1603	Bronchial secretion from lung transplant recipient	Germany
<i>R. aegroticola</i>	IMMI 1869	BAL from lung transplant recipient	Germany
<i>R. aegroticola</i>	IMMI 1896	Sputum from CF patient	Germany
<i>R. aegroticola</i>	IMMI 2824	Sputum from CF patient	Germany
<i>R. argillacea</i>	IMMI 1764	BAL from bone marrow transplant recipient	Germany
<i>R. argillacea</i>	IMMI 1862	Sputum from CF patient	Germany
<i>R. argillacea</i>	IMMI 1870	Eczema of integument	Germany
<i>R. argillacea</i>	IMMI 1881	Sputum from CF patient	Germany
<i>R. argillacea</i>	IMMI 1893	Sputum from CF patient	Germany
<i>R. argillacea</i>	IMMI 1894	Sputum from CF patient	Germany
<i>R. argillacea</i>	IMMI 1895	Sputum from CF patient	Germany
<i>R. piperina</i>	IMMI 1464	Sputum from CF patient	Germany

**TABLE 1. Non-*Rasamsonia* strains (n = 39) tested for cross-reactivity in the *Rasamsonia argillacea* species complex PCR**

Species	Strain number
Fungi	
<i>Aspergillus cf. tamarii</i>	ATCC 64841
<i>Aspergillus flavus</i>	CBS 113.49
<i>Aspergillus fumigatus</i>	NCPF2140
<i>A. fumigatus</i>	CBS 133.61
<i>A. fumigatus</i>	CBS 154.89
<i>Aspergillus nidulans</i>	CBS 100.20
<i>Aspergillus niger</i>	CBS 112.30
<i>Aspergillus brasiliensis</i>	CBS 733.88
<i>Aspergillus terreus</i>	CBS 469.81
<i>Aspergillus versicolor</i>	IMMI F81
<i>Candida albicans</i>	ATCC 44374
<i>Candida glabrata</i>	DSM 70614
<i>Candida guilliermondii</i>	ATCC 90877
<i>Candida kefyr</i>	DSMI 195
<i>Candida krusei</i>	DSM 70075
<i>Candida parapsilosis</i>	ATCC 22019
<i>Candida tropicalis</i>	ATCC 750
<i>Exophiala dermatitidis</i>	CBS 120550
<i>Fusarium solani</i>	IMMI 1650
<i>Paecilomyces lilacinus</i>	IMMI 1900
<i>Penicillium citrinum</i>	IMMI 1945
<i>Penicillium notatum</i>	IMMI 2013
<i>Pseudallescheria boydii</i>	FMR 84
<i>Rhizomucor microsporus</i>	IMMI 1672
<i>Rhizomucor pusillus</i>	IMMI 1671
<i>Scedosporium apiaspermum</i>	IMMI F71
<i>Scedosporium prolificans</i>	IMMI F78
Bacteria	
<i>Enterococcus faecalis</i>	ATCC 29212
<i>Enterococcus faecium</i>	DSM 13590
<i>Staphylococcus aureus</i>	ATCC 43300
<i>Staphylococcus epidermidis</i>	DSM 1789
<i>Streptococcus pyogenes</i>	DSM 11728
<i>Acinetobacter baumannii</i>	IMMI 150
<i>Enterobacter cloacae</i>	IMMI 253
<i>Haemophilus influenzae</i>	DSM 9999
<i>Klebsiella pneumoniae</i>	IMMI 251
<i>Legionella pneumophila</i>	ATCC 33152
<i>Proteus mirabilis</i>	DSM 4479
<i>Serratia marcescens</i>	ATCC 13880
<i>Mycobacterium tuberculosis</i>	ATCC 27294
<i>Mycobacterium avium</i>	DSM 44156

**TABLE 3.** Characteristics of the cystic fibrosis (CF) populations tested by *Rasamsonia argillacea* species complex PCR and culture

CF population	University Hospital, Essen, Germany (n = 138)	University Hospital Angers, France (n = 15)
Mean age $\pm$ SD, years (range)	26.6 $\pm$ 10.1 (4–47)	17.1 $\pm$ 9.3 (3–33)
Female (%)	61 (44.2)	10 (66.7)
Number of specimens	214	20
Specimens per CF patient (range)	1.6 (1–4)	1.25 (1–4)
Specimens positive for the <i>R. argillacea</i> species complex		
By the real-time PCR assay	4	3
By cultures	3	2
By both PCR and cultures	3	2

**TABLE 4. Clinical and microbiological data of cystic fibrosis (CF) patients with DNA detection of *Rasamsonia argillacea* species complex**

	Patient 1	Patient 2	Patient 3	Patient 4
Age, years	18	37	23	7
Sex	Male	Female	Female	Female
F508 del mutation	Homozygous	Homozygous	Homozygous	Heterozygous (W1204X)
Pancreatic insufficiency	No	No	Yes	Yes
CF-related diabetes	No	No	No	No
Listed for lung transplantation	No	No	No	No
FEV1	41%	30%	63%	80%
Clinical specimen tested	Sputum	Sputum	Sputum	Sputum
Cultures				
<i>Pseudomonas aeruginosa</i>	Yes	Yes	No	Yes
<i>Aspergillus fumigatus</i>	Yes	No	Yes	Yes
<i>R. argillacea</i> complex	<i>R. argillacea</i>	<i>R. aegroticola</i>	Yes	Yes

CF, cystic fibrosis; FEV1, forced expiratory volume in 1 seconds.

# Schlussfolgerungen

- Identifizierung von *Rasamsonia* spp. mit molekularbiologischen Methoden ratsam
- Prävalenz von *Rasamsonia* spp. bei CF und anderen Erkrankungen scheint zu steigen
- Sensitive und spezifische PCR für *R. argillacea* Komplex etabliert
- Assay geeignet für
  - Schnellen Nachweis
  - Studien (Häufigkeit, Bedeutung)
  - Herkunftsanalysen

# Acknowledgment

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### AG Mukoviszidose

Peter-Michael Rath

Pedrina Goncalves-Vidigal

Andrea Hain

David Killengray

Dirk Schmidt

Silke Dittmer

Jan Buer



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## **Jean-Philippe Bouchara**

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L'UNAM Universite, Universite d Angers

