



Nouveautés 2013

Infections Pulmonaires de l'immunodéprimé

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Nice

Multicenter Comparison of Serum and Whole-Blood Specimens for Detection of *Aspergillus* DNA in High-Risk Hematological Patients

Jan Springer,^a C. O. Morton,^{b*} Michael Perry,^c Werner J. Heinz,^a Melinda Paholcsek,^{a*} Mona Alzheimer,^a T. R. Rogers,^b Rosemary A. Barnes,^d Hermann Einsele,^a Juergen Loeffler,^a P. Lewis White^c

Assay	Positivity results ^a				P value ^b
	Cases (n = 47)		Controls (n = 31)		
	No. positive	% positive (95% CI)	No. positive	% positive (95% CI)	
GM	38	80.9 (67.5–89.6)	1	3.2 (0.6–16.2)	0.0001
WB PCR	40	85.1 (72.3–92.6)	11*	35.5 (21.1–53.1)	0.0001
Serum PCR	37	78.7 (65.1–88.0)	5*	16.1 (7.1–32.6)	0.0001

* 1 FP concordant

Assay	Performance value (% [95% CI]) ^a	
	Sensitivity	Specificity
GM	80.9 (67.5–89.6)	96.8 (83.8–99.4)
WB PCR	85.1 (72.3–92.6)	64.5 (47.0–78.9)
Serum PCR	78.7 (65.1–88.0)	83.9 (67.4–92.9)

◆ Sensibilités: ≈ ◆ PCR: ↗ FP ◆ 3 tests : VPN 100%

Assay	Performance value (% [95% CI]) ^a			
	Sensitivity	Specificity	PPV	NPV
GM	80.9 (67.5–89.6)	96.8 (83.8–99.4)	97.4 (86.8–99.6)	76.9 (61.7–87.4)
WB PCR	85.1 (72.3–92.6)	64.5 (47.0–78.9)	78.4 (65.4–87.5)	74.1 (65.4–87.5)
Serum PCR	78.7 (65.1–88.0)	83.9 (67.4–92.9)	88.1 (75.0–94.8)	72.2 (56.0–84.2)
Combination testing ^d				
GM/GM	48.9 (35.3–62.8)	100 (89.0–100)	100 (85.7–100)	56.4 (43.3–68.7)
GM/WB	68.1 (53.8–79.6)	100 (89.0–100)	100 (89.3–100)	67.4 (53.0–79.1)
GM/serum	59.6 (45.3–72.4)	100 (89.0–100)	100 (87.9–100)	62.0 (48.2–74.1)
WB/WB	46.8 (33.3–60.8)	93.5 (79.3–98.2)	91.7 (74.2–97.7)	53.7 (40.6–66.3)
WB/serum	57.4 (43.3–70.5)	96.8 (83.8–99.4)	96.4 (82.3–99.4)	60.0 (46.2–72.4)
Serum/serum	53.2 (39.2–66.7)	100 (89.0–100)	100 (86.7–100)	58.5 (45.1–70.7)

Intérêt de l'association de 2 techniques

- ↗ **VPP**: confirmation GM+ par PCR +++
- ↗ **Spécificité** 100%
- NB: **VPN** sous estimées par incidence AI 60%
- **Précocité** : 70% avant culture, biopsies ou TDM

- 
- **Performances** AgGM confirmé / PCR
 - Persistance FN
 - Intérêt: ↗ spécificité ↗ VPP ↗ Précocité

PCR Aspergillus **sang total / serum ?**

PCR sang total

- ↗ sensibilité (NS)
- ↗ précocité (36 vs 15j)



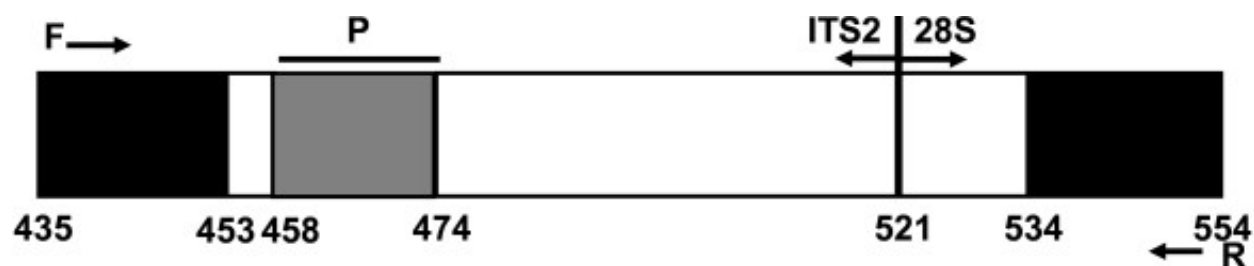
PCR serum

- rapidité
- faisabilité
- même prlvt GM

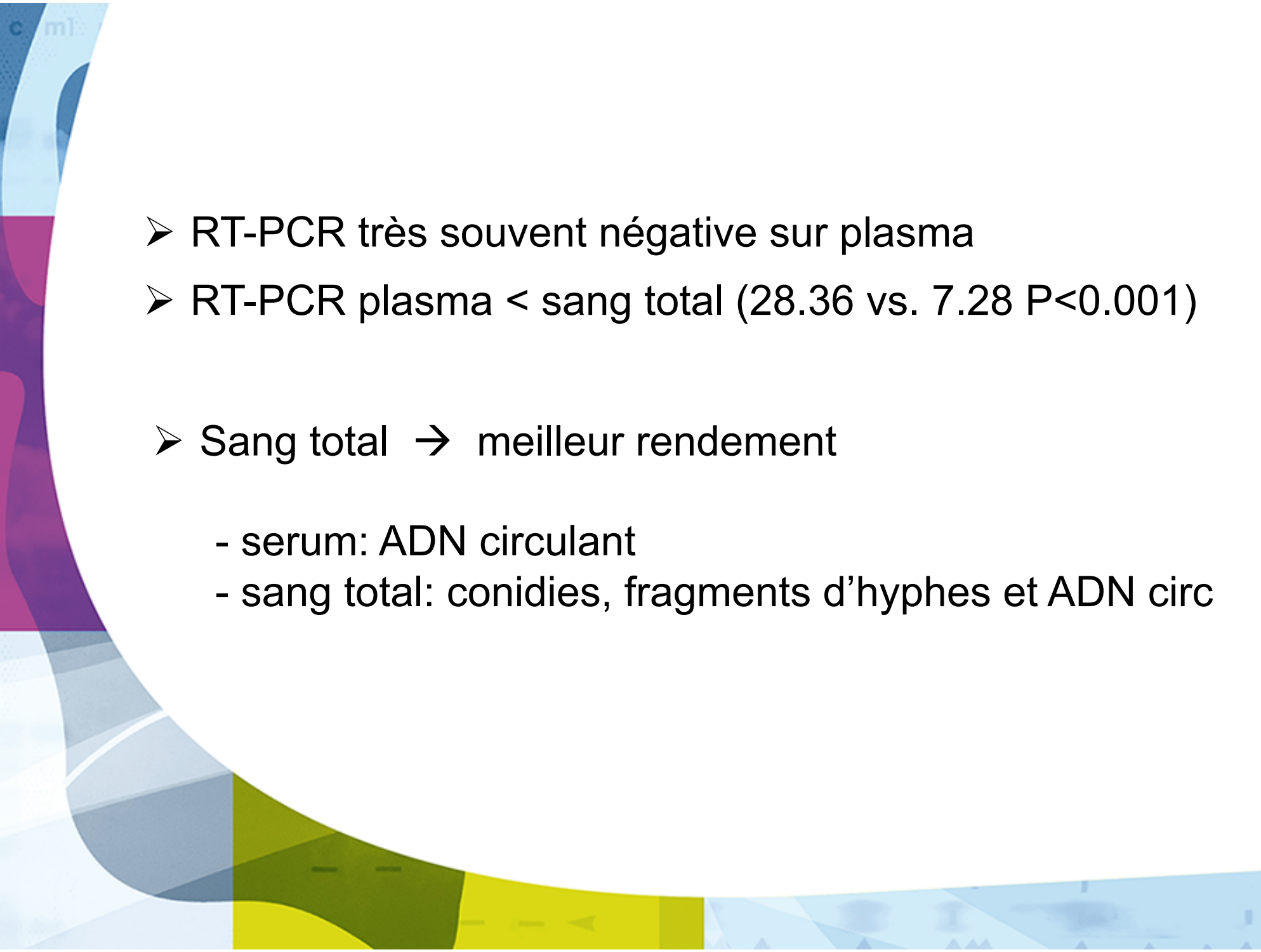
Establishment and application of real-time quantitative PCR for diagnosing invasive Aspergillosis via the blood in hematological patients: targeting a specific sequence of *Aspergillus* 28S-ITS2

Yan Li[†], Li Gao[†], Yi Ding, Yuanyuan Xu, Minhong Zhou, Wenrong Huang, Yu Jing, Honghua Li, Lili Wang and Li Yu^{*}

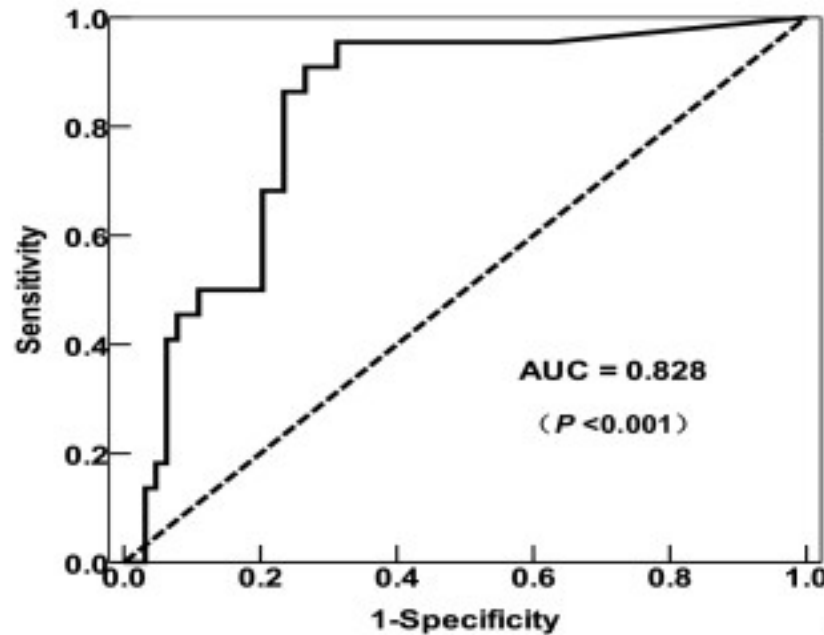
Evaluation d'une cible PCR différente



72 patients à Haut Risque fébriles, 4 non fébriles
10 contrôles sains
→ 41 AI prouvées probable

- 
- RT-PCR très souvent négative sur plasma
 - RT-PCR plasma < sang total (28.36 vs. 7.28 P<0.001)
 - Sang total → meilleur rendement
 - serum: ADN circulant
 - sang total: conidies, fragments d'hyphes et ADN circ

Quantitative Real-Time PCR



PCR (copies/ μ l)	Sensitivity	Specificity	Youden
20.48	0.955	0.656	0.611
20.87	0.955	0.672	0.627
21.34	0.955	0.688	0.643
24.37	0.909	0.703	0.612
24.47	0.909	0.719	0.628
25.24	0.909	0.734	0.643
26.08	0.864	0.766	0.630

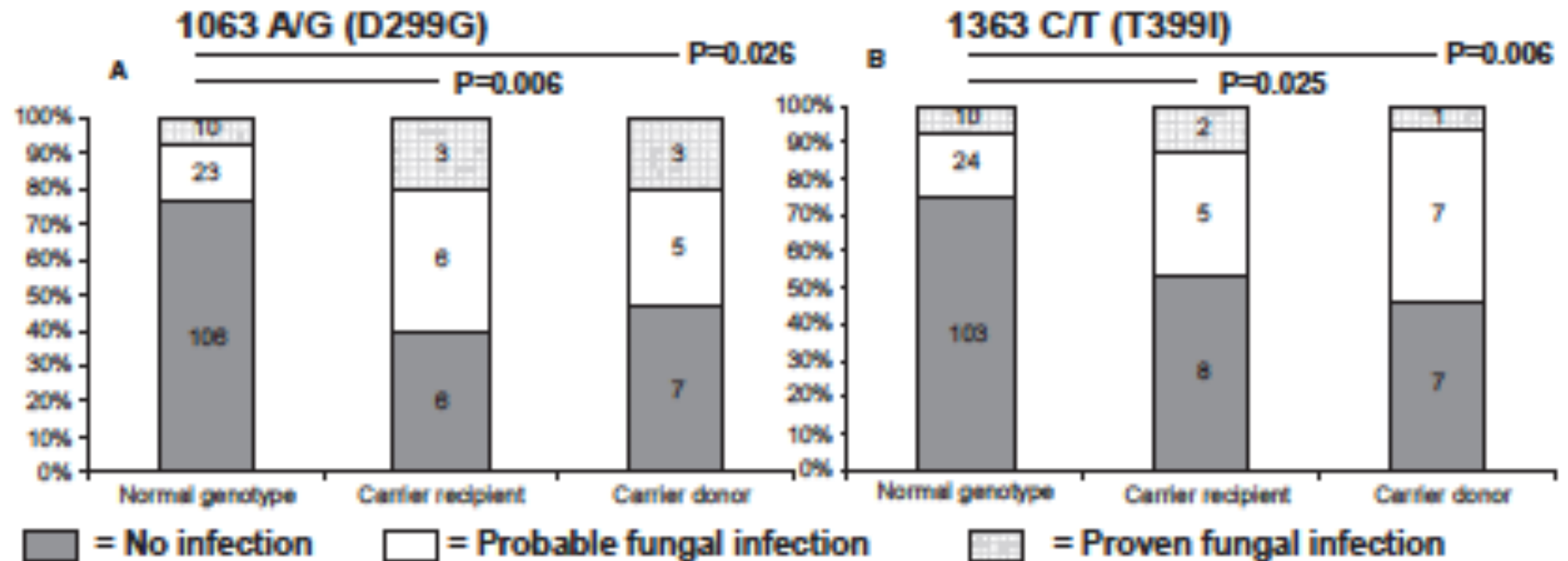
- Technologie en pleine expansion
 - nouvelles techniques d'extraction / **harmonisation**,
 - **perfectionnement** des cibles : région 28S-ITS2

Résultats encourageants à évaluer au sein
d'une stratégie diagnostique et thérapeutique
Critères EORTC/MSG ?

Increased susceptibility for aspergillosis and post-transplant immune deficiency in patients with gene variants of TLR4 after stem cell transplantation

M. Koldehoff, D.W. Beelen, A.H. Elmaagachi

Retrospectif 10 ans - 154 pt HCST pour LA
Polymorphisme gène du TLR4

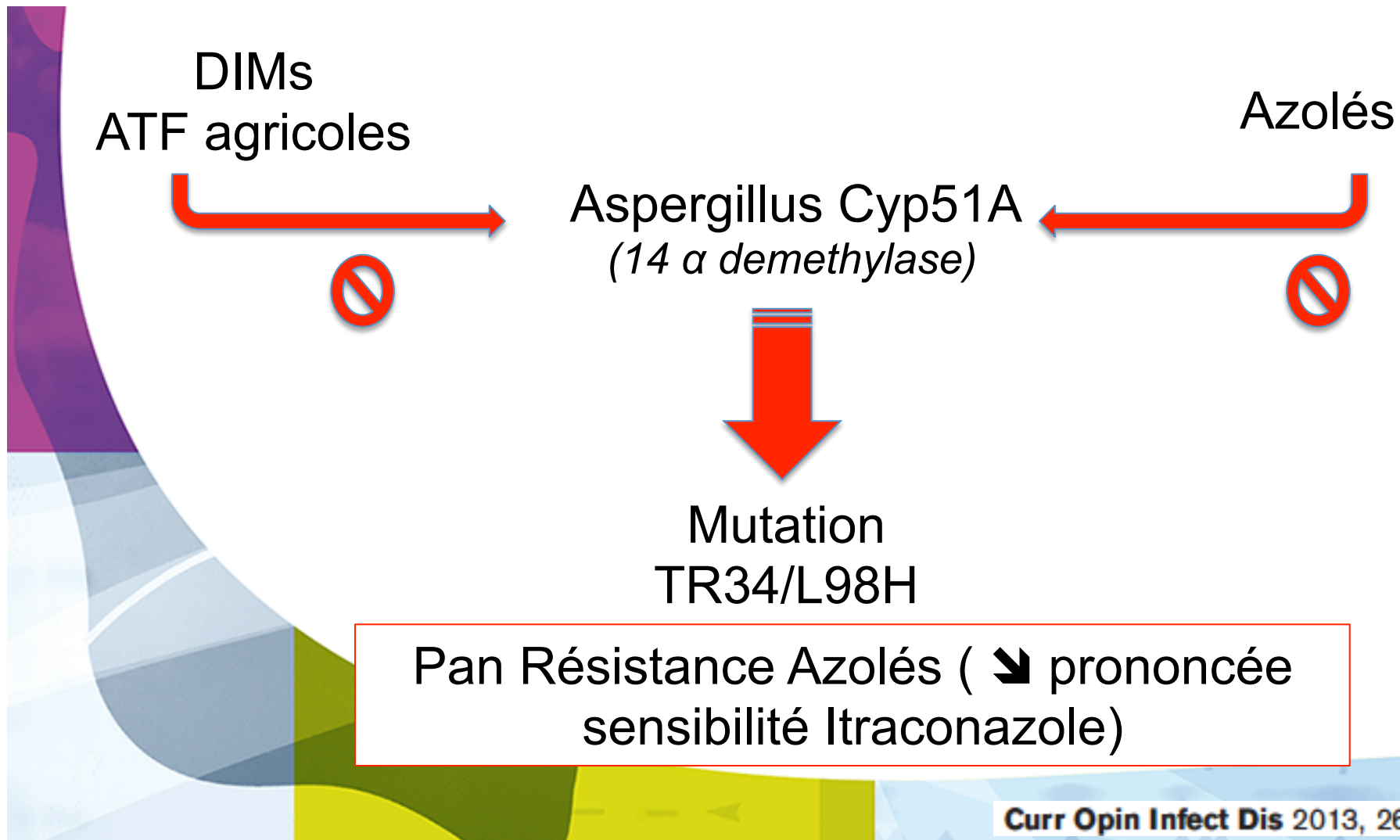


➔ Association significative: TLR4 et AI

➔ Détermination polymorphisme TLR4: un outil ?

Azole resistance in *Aspergillus fumigatus*: a growing public health concern

Edith Vermeulen^a, Katrien Lagrou^{a,b}, and Paul E. Verweij^c



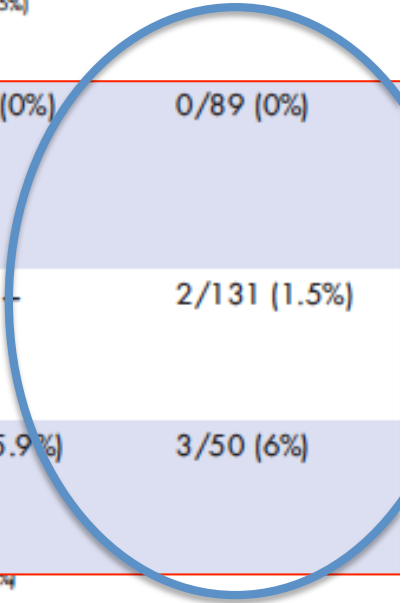
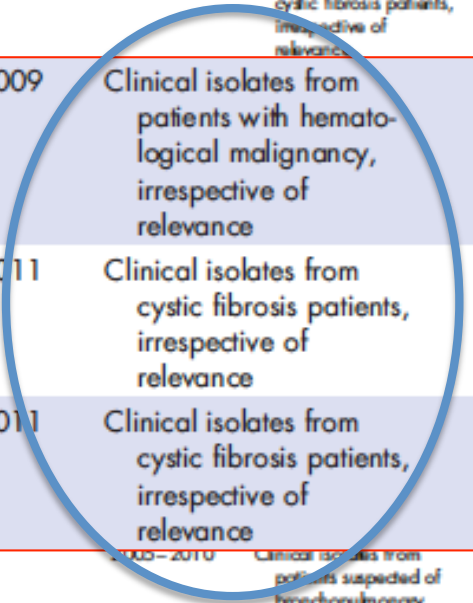


Country, ref.	Study period	Study isolates	Resistance rate	Resistance prevalence	TR _{3d} /L98H rate	TR _{3d} /L98H prevalence
UK, [6]	1997–2007	Clinical isolates, irrespective of relevance; referral center for chronic/allergic aspergillosis	34/519 (6.6%)	20/400 (5%)	2/519 (0.4%)	2/400 (0.5%)
UK, [8]	2008–2009	Clinical isolates sent for susceptibility testing; referral center for chronic/allergic aspergillosis	64/230 (27.8%)	28/157 (17.8%)	0/230 (0%)	0/157 (0%)
The Netherlands, [12]	1994–2007	Clinical isolates, irrespective of relevance	63/2061 (3.1%)	45/1320 (3.4%)	–	39/1320 (3.0%)
The Netherlands, [13]	2007–2009	Clinical isolates, irrespective of relevance	82/1792 (4.6%)	63/1192 (5.3%)	74/1792 (4.1%)	57/1192 (4.8%)
The Netherlands, [14*]	2009–2011	Clinical isolates, irrespective of relevance	–	63/921 (6.8%)	–	47/921* (5.1%)
Spain, [15]	2010–2011	Clinical isolates, irrespective of relevance	1/156 (0.6%)	–	–	–
Spain, [16*]	1999–2011	Clinical isolates from proven or probable invasive aspergillosis or aspergilloma	6/343 (1.8%)	6/148 (4.1%)	0/343 (0%)	0/150 (0%)
Denmark, [10]	2007–2009	Clinical isolates from cystic fibrosis patients, irrespective of relevance	–	6/133 (4.5%)	–	2/133 (1.5%)
France, [17]	2006–2009	Clinical isolates from patients with hematological malignancy, irrespective of relevance	1/118 (0.8%)	1/89 (1.1%)	0/118 (0%)	0/89 (0%)
France, [11]	2010–2011	Clinical isolates from cystic fibrosis patients, irrespective of relevance	–	6/131 (4.6%)	–	2/131 (1.5%)
France, [18**]	2010–2011	Clinical isolates from cystic fibrosis patients, irrespective of relevance	9/85 (10.6%)	4/50 (8.0%)	5/85 (5.9%)	3/50 (6%)
Germany, [19*]	2011–2012	Clinical isolates irrespective of relevance	3.2% (17/527)	–	6/527 (1.1%)	–
Japan, [20*]	1994–2010	Clinical isolates, irrespective of relevance (obtained from Pneumology Dept.)	11.2% (22/196)	–	0/196 (0%)	–
India, [21*]	2005–2010	Clinical isolates from patients suspected of bronchopulmonary aspergillosis	2/103 (1.9%)	2/85 (2.4%)	2/103 (1.9%)	2/85 (2.4%)
Iran, [22*]	2003–2009	Clinical isolates obtained from patients with aspergillus diseases	3.2% (4/124)	–	3/124 (2.4%)	–
USA, [23]	2001–2006	Isolates recovered from transplant recipients with proven or probable invasive aspergillosis	1/181 (0.6%)	–	–	–

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Mutation TR34/L98H chez *Aspergillus fumigatus* dans le monde



c ml

Aspergillosis due to Voriconazole Highly Resistant *Aspergillus fumigatus* and Recovery of Genetically Related Resistant Isolates From Domiciles



Décembre 2009 à janvier 2011

- 1315 isolats (921 pt)
- 21 isolats/ 15 pt Haut niv de R Vorico
- **TR46/Y121F/T289A**
- 8 pt: essai Vorico = échec clinique
- 6/10 prélèvements domiciles = +



Sex/ Age	Month of Isolation/ Site	City	MIC (mg/L)			Underlying Condition	<i>Aspergillus</i> Disease [30]	Previous Azole Exposure ^a	Treatment	Outcome at 12 wk
			ITZ	VCZ	POS					
F/11	Dec 2009/sputum	Utrecht	4	>16	0.25	Relapse ALL, HSCT, GVHD	Probable IA	None	VCZ, CAS	Persistent infection
M/70	Jan 2010/ear	Amsterdam	>16	>16	2	Chronic otitis externa, sinusitis, and paralysis of abducens nerve	IA ^b	None	L-AMB, AND	Persistent infection
F/51	Jan 2010/ abdominal abscess	Nijmegen	2	>16	0.5	Kidney transplant	Proven IA	None	VCZ, POS	Died
F/9	Feb 2010/sputum	Amsterdam	4	>16	0.5	Cystic fibrosis	No IA	None	None	Alive
M/69	Feb 2010/sputum	Amsterdam	>16	>16	2	Lung carcinoma, radiation	No IA	None	None	Alive
M/54	Mar 2010/sputum	Groningen	1	>16	0.25	Multiple myeloma, autologous HSCT, relapse	Probable IA	None	VCZ, L-AMB	Died
F/54	Mar 2010/sputum	Groningen	16	>16	0.5	Cystic fibrosis, bilateral lung transplant	Proven IA	VCZ	L-AMB	Alive
F/65	May 2010/biopsy	Amsterdam	4	>16	1	Chronic otitis after cholesteatoma surgery	Proven IA	None	Surgery, L-AMB	Alive
M/76	May 2010/sputum	Amsterdam	>16	>16	1	Lung fibrosis	None	None	None	Alive
M/70	Jun 2010/sputum	Amsterdam	1	>16	0.25	High energetic trauma, ICU admission	None	None	None	Died
M/59	Jul 2010/brain biopsy	Amsterdam	4	>16	1	β-thalassemia and diabetes mellitus	Proven IA	None	VCZ, L-AMB, CAS	Died
F/21	Sep 2010/sputum	Nijmegen	2	>16	0.5	Cystic fibrosis	ABPA	VCZ	None	Alive
F/49	Oct 2010/sputum	Groningen	>16	>16	2	COPD, unilateral lung transplant	None	None	L-AMB, VCZ	Alive
F/64	Nov 2010/sputum	Leiden	>16	>16	2	COPD	No IA	None	None	Alive
F/50	Jan 2011/sputum	Utrecht	>16	>16	1	NH B-cell lymphoma, allo-SCT	Probable IA	VCZ	VCZ	Died





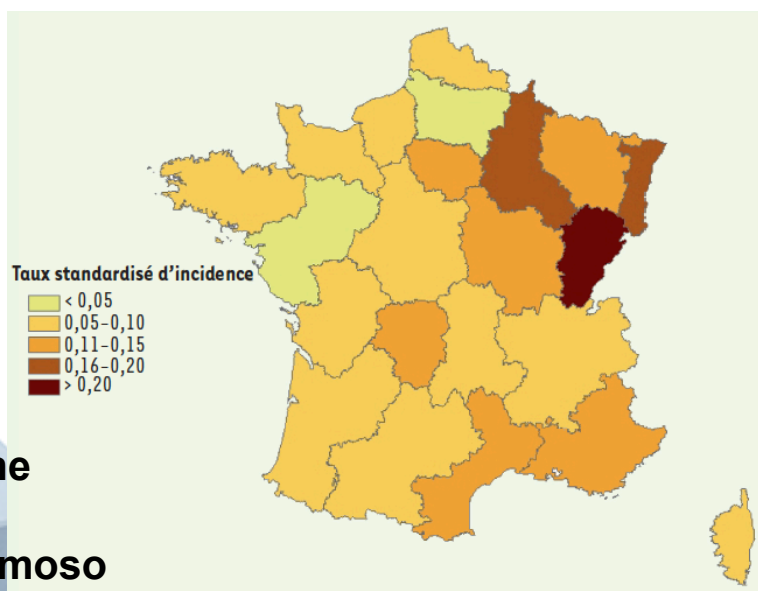
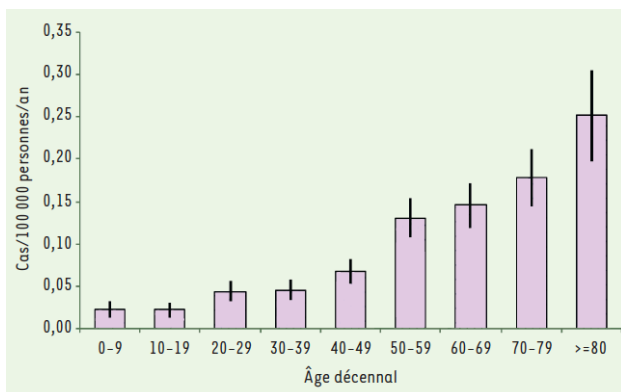
Aspergillus sp.
Résistance aux Azolés: une menace bien réelle

- Multiplication et **propagation** des mécanismes de résistance sur le globe
- Corréliées à l'utilisation d'ATF agricoles

Mucormycoses




médecine/sciences 2013 ; 29 (hors série n° 1) : 25-30



O. Lortolary
D. Bitar, D. Che
R. Herbrecht
D. Garcia-Hermoso
M. Sabou
MP Ledoux
B. Pilmis
F. Lanternier

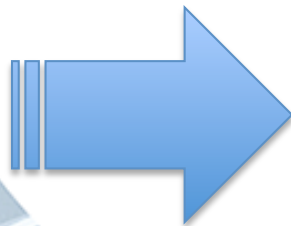
Facteur de risque	Mucormycoses		
	N	%	Évolution annuelle de l'incidence
Hémopathie maligne (HM)	143	34,8 %	10,3 % (p = 0,02)
• avec greffe de moelle	40	9,7 %	ns
• avec neutropénie	53	12,9 %	20,7 % (p = 0,008)
• HM seule	50	12,2 %	ns
VIH/Sida	15	3,6 %	ns
Greffes d'organes solides	9	2,2 %	ns
Cancers des organes solides	19	4,6 %	ns
Maladies inflammatoires	9	2,2 %	ns
Diabète	68	16,5 %	ns
Maladies respiratoires chroniques	13	3,2 %	ns
Insuffisance rénale chronique	8	1,9 %	ns
Autres pathologies*	19	4,6 %	60,8 % (p = 0,004)
Facteur de risque non spécifié**	108	26,3 %	ns
Total	411		7,6 % (p = 0,003)



Quantitative Polymerase Chain Reaction Detection of Circulating DNA in Serum for Early Diagnosis of Mucormycosis in Immunocompromised Patients

Laurence Millon,^{1,2} Fabrice Larosa,³ Quentin Lepiller,^{2,4} Faezeh Legrand,³ Steffi Rocchi,¹ Etienne Daguindau,³
Emeline Scherer,^{1,2} Anne-Pauline Bellanger,^{1,2} Joel Leroy,⁵ and Frederic Grenouillet^{1,2}

Diagnostic difficile



Diagnostic histopathologique difficile
Rendement culture insuffisant ajout PCR/tissu
Problématique de l'obtention de tissu



3 qPCR sériques spécifiques
→ espèces plus fréquentes mucormycose
(*Rhizopus* - *Lichtheimia* species – *Rhizomucor* – *Mucor*)

Etude rétrospective 10 patients mucormycose prouvée
vs 10 témoins sains
vs 10 pt onco-hématologie sans infection
vs 17 AI prouvée/probable (EORTC/MSG)
vs 14 pneumocystoses

entre janvier 2004 et juin 2012 au CHU Besançon




Patient No.	Sex ^a / Age	Underlying Disease	Localization of Infection, Date ^b of First Clinical Symptoms/First Radiological Signs	Date ^b of Fever Onset	Positive Histology (Day 0)/Molecular Diagnosis	Positive Mycological Cultures (Day)/ Identification	Antifungal Therapy (Day of Initiation of L-AmB, POS)	Outcome at Day 90	qPCR Result Cq (Day)	Days From 1st Positive PCR to 1st Clinical Signs ^c	Days From 1st Positive PCR to Positive Histology ^d
1	F/65	Undernutrition, alcoholism	Disseminated D -1/not done	D -5	Skin/not done	Skin, urine (D0)/ <i>L. corymbifera</i>	FLU	Death (D1)	Acory Neg (D -8, D -1)	/	/
2	M/37	HL	Disseminated D -16/not done	D -28	Lung, liver/not done	Lung, liver (D0)/ <i>L. corymbifera</i>	L-AmB (D -1)	Death (D0)	Acory 28 (D -3), Neg (D -10)	+13	-3
3	M/51	ALL	Rhinocerebral D -1/D -1	D -9	Sinus/ <i>Lichtheimia corymbifera</i>	Sinus (D0)/ <i>L. corymbifera</i>	L-AmB (D0) POS (D0)	Death (D3)	Acory 36 (D -8), 32 (D -5), 30 (D -1), 25 (D0), 27 (D2), Neg (D -18, D -15, D -12)	-7	-8
4	F/48	Polytrauma	Cutaneous D -5/not done	D0	Skin/ <i>Lichtheimia</i> sp	Skin (D0)/negative	FLU, CAS	Death (D2)	Acory 27 (D0), Neg (D -2, D -9)	+5	0
5	M/59	NHL, diabetes mellitus	Disseminated D -5/D -22	D -14	Kidney/ <i>Lichtheimia</i> sp	Cerebrospinal fluid (D -10)/ <i>Lichtheimia ramosa</i>	FLU, CAS, L-AmB (D0)	Alive	Acory 32 (D -23) 38 (D -8), Neg (D -1, D5)	-18	-23
6	F/41	PNH, SAA	Disseminated D -30/D -4	D -17	Sinus/ <i>Rhizopus</i> sp	Sinus (D0)/ <i>R. oryzae</i>	CAS, POS (D -11) L-AmB (D0) deferasirox (D0)	Death (D15)	Muc1 36 (D -10), 35 (D -1), 38 (D2), 36 (D6), 38 (D9), 34 (D13)	+20	-10
7	F/13	AML	Pulmonary ^p D -62/D -61	D -65	Lung/ <i>Rhizopus oryzae</i>	Not done	VOR, L-AmB (D -60)	Alive	Muc1 35 (D -68), 40 (D -62), 36 (D -55), 38 (D -21), 36 (D10), Neg (D -75, D29)	-6	-68
8	M/57	MDS, allo-HSCT	Rhino-cerebral D -18/D -11	D0	Sinus/not done	Sinus (D0)/ <i>R. pusillus</i>	ITR (D -96), L-AmB (D0) POS (D0)	Alive	Rmuc 39 (D -18), 39 (D -1), Neg (D -11, D -8, D9, D13)	0	-18
9	F/55	Renal transplant	Disseminated D -24/D -11	D -30	Lung, heart/ <i>Rhizomucor pusillus</i>	Lung, heart (D0)/ <i>R. pusillus</i>	CAS	Death (D0)	Rmuc 29 (D -4), 22 (D -3), Neg (D -24, D -19)	+20	-4
10	M/60	AML	Disseminated D -50/D -47	D -50	Liver, spleen/ <i>Rhizomucor</i> sp	Liver, spleen (D0)/negative	VOR, L-AmB (D -44) POS (D -36)	Alive	Rmuc 41 (D -49), 28 (D -46), Neg (D -52, D -42, D -39, D -36, D -18, D -13, D -1)	+1	-49



PCR Mucormycose

- Prometteuse
- A évaluer
- Ne pas sous estimer co-infections....



Fourth European Conference on Infections in Leukaemia (ECIL-4): Guidelines for Diagnosis and Treatment of Human Respiratory Syncytial Virus, Parainfluenza Virus, Metapneumovirus, Rhinovirus, and Coronavirus

Hans H. Hirsch,^{1,2} Rodrigo Martino,³ Katherine N. Ward,⁴ Michael Boeckh,⁵ Hermann Einsele,⁶ and Per Ljungman^{7,8}

Recommandations diagnostiques IRVC

- **Infection symptomatique VAS/VAI** receveurs/candidats greffe SCH
→ **la recherche de virus communautaire doit être réalisée (AII)**
- **Orienté / Site** (écouvillon/VAS, LBA/VAI, si imposs aspi trachéale) (BII)
- **1^{ère} ligne de PCR virale:** influenza A et B, VRS et HPIV (AII)
- autres PCR: en fonction du risque d'exposition, épidémio locale ou premiers résultats négatifs (BIII)
- **IRB:** discuter **LBA** et tests diagnostiques incluant biopsie pulmonaire (BII)

Recommandations Thérapeutiques greffes CSH et L.A.

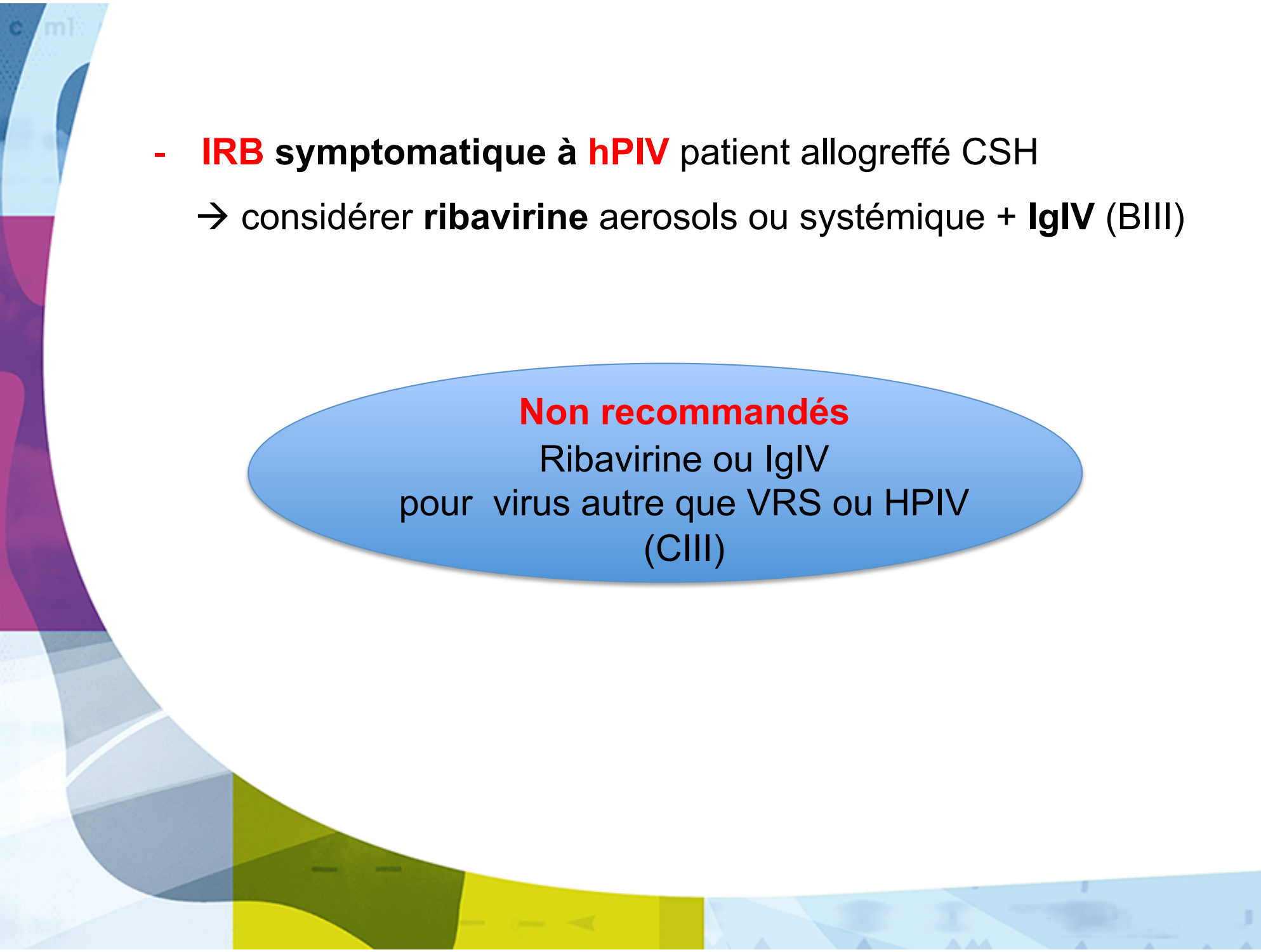
- Infection **symptomatique VAS** à **VRS** conditionnement ou allogreffé de CSH
si FDR de progression vers une atteinte des VAI ou de décès
→ **Ribavirine** aerosol/systémique
+ IgIV (BII)
- Allogreffé CSH **IRB** à **VRS**
→ **Ribavirine** aerosol
ou systémique
+ IgIV
ou anticorps spécifiques anti-VRS (BIII)

Progression to LRTID

- Lymphopenia $<0.2 \times 10^9/L$
- Older age
- Mismatched/unrelated donor
- Allogeneic HSCT <1 mo
- Neutropenia $<500/\mu L$
- No therapy with aerosolized ribavirin + IVIG

Mortality

- Preengraftment
- Lymphopenia $<0.2 \times 10^9/L$
- Allogeneic HSCT <1 mo
- Severe immunodeficiency
- Older age (>65 y)

- 
- **IRB** symptomatique à **hPIV** patient allogreffé CSH
→ considérer **ribavirine** aerosols ou systémique + **IgIV** (BIII)

Non recommandés
Ribavirine ou IgIV
pour virus autre que VRS ou HPIV
(CIII)



*MERCI de
votre attention*

