



Le NGS déjà un outil en maladie infectieuse complexe !

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NGS en maladies infectieuses... déjà un outil

- Epidémiologie
- Infections difficiles à diagnostiquer
- Pathogène non évoqué / non connu / non cultivable

Aujourd'hui : pour diagnostic difficile

Mr B, 66 ans, pas d'antécédent médical notable

- J0: Syndrome pseudo-grippal

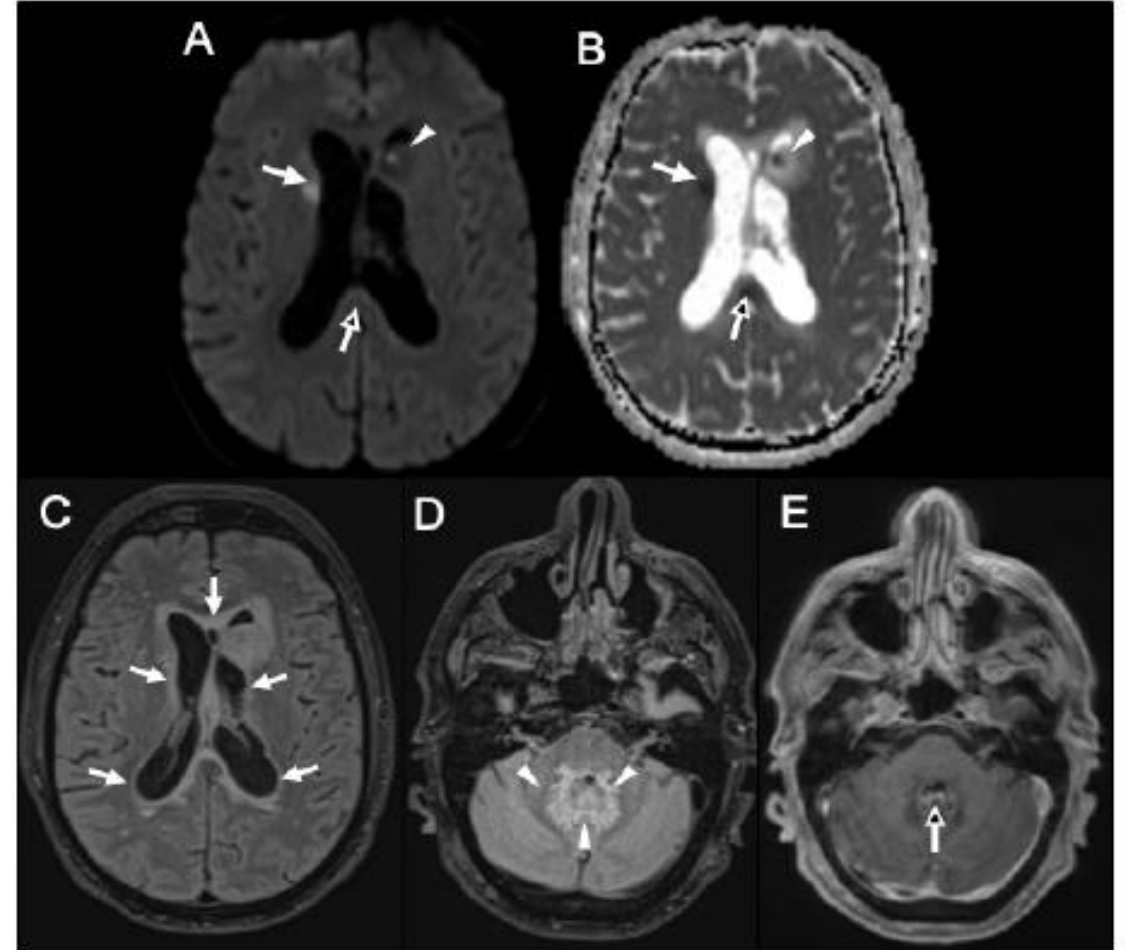
Traitement par Augmentin pendant 10j

- J10 : Fièvre et confusion => SAU
 - LCR : 750 elt/mm³, (76%PNN, hyperproteino, hypoglyco)
 - PCR multiplex neg, Culture neg, 16S neg
 - HIV -
 - Auto-Ac -

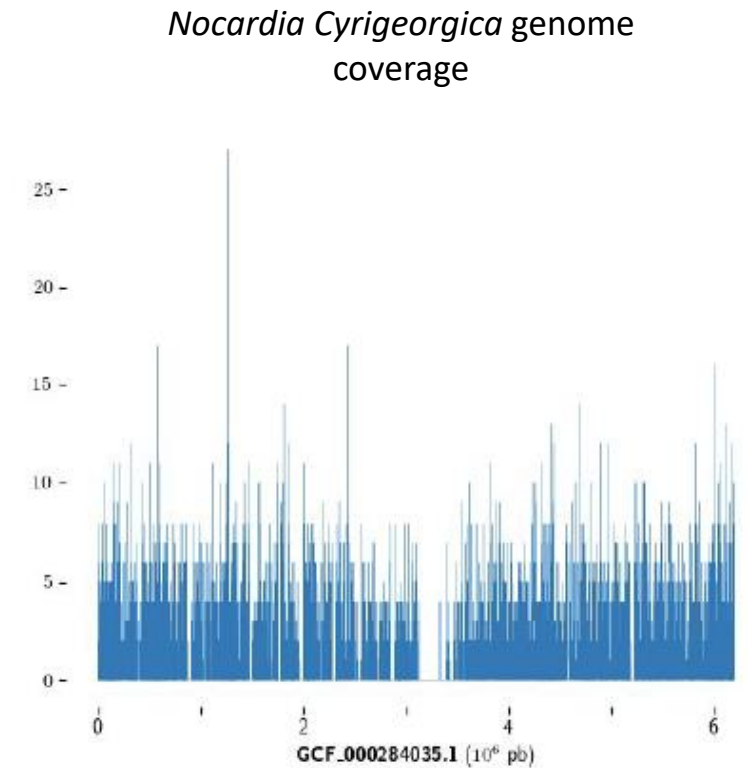
IRM : abcès de 10mm
noyau caudé gauche

J20 : Nouveau LCR
1130 elt/mm³ (82%PNN)
Culture, PCR, 16S négatifs

=> Envoi NGS

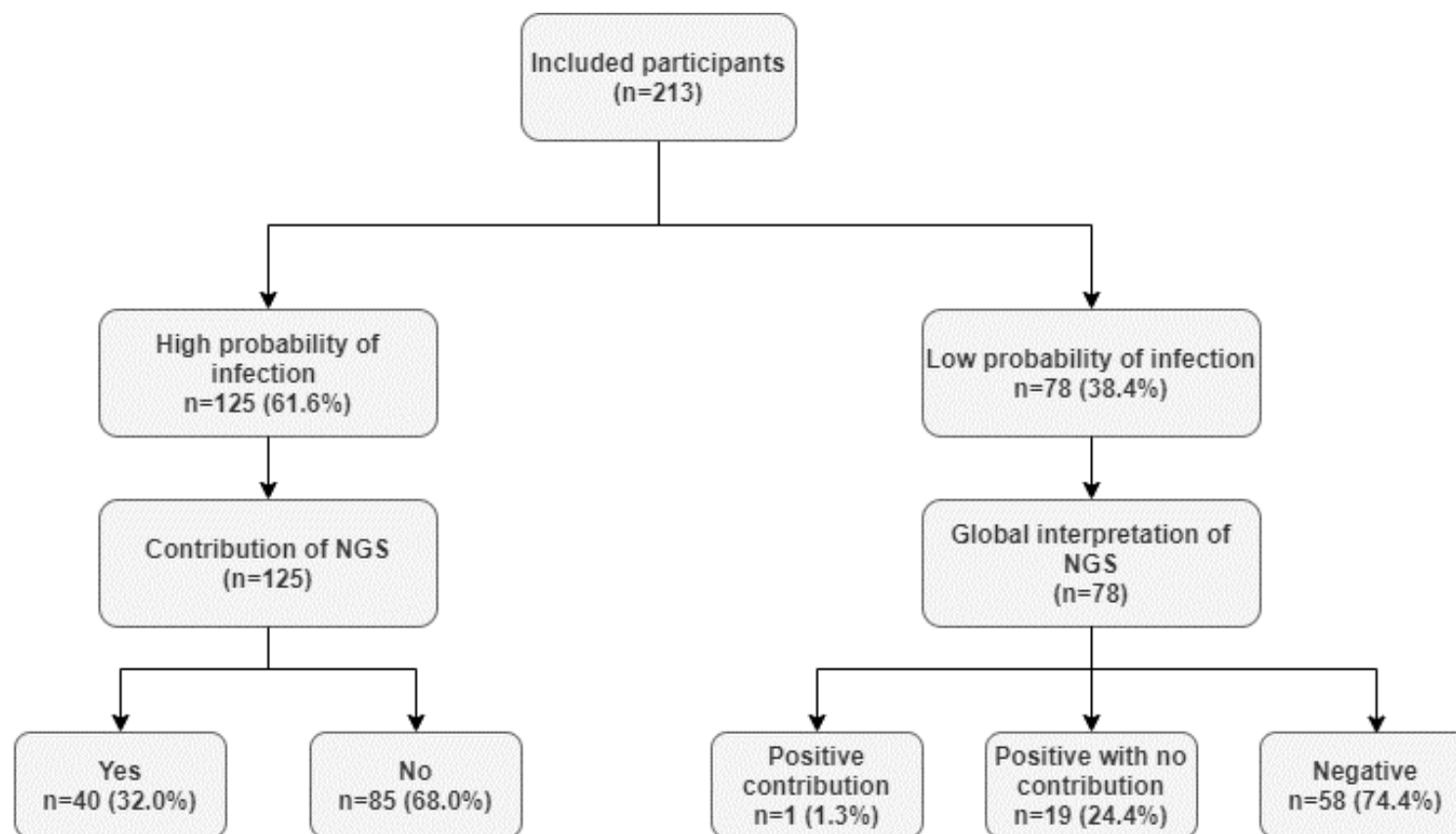


- J30 : MetaMIC : *Nocardia cyriageorgica* (confirmation par CNR *Nocardia*). Traitement par Imipenem/Bactrim
- Recherche d'un déficit immunitaire acquis
Présence d'anti GM-CSF traité par rituximab
- 2019 : arrêt de toutes les thérapies
Patient considéré en rémission complète

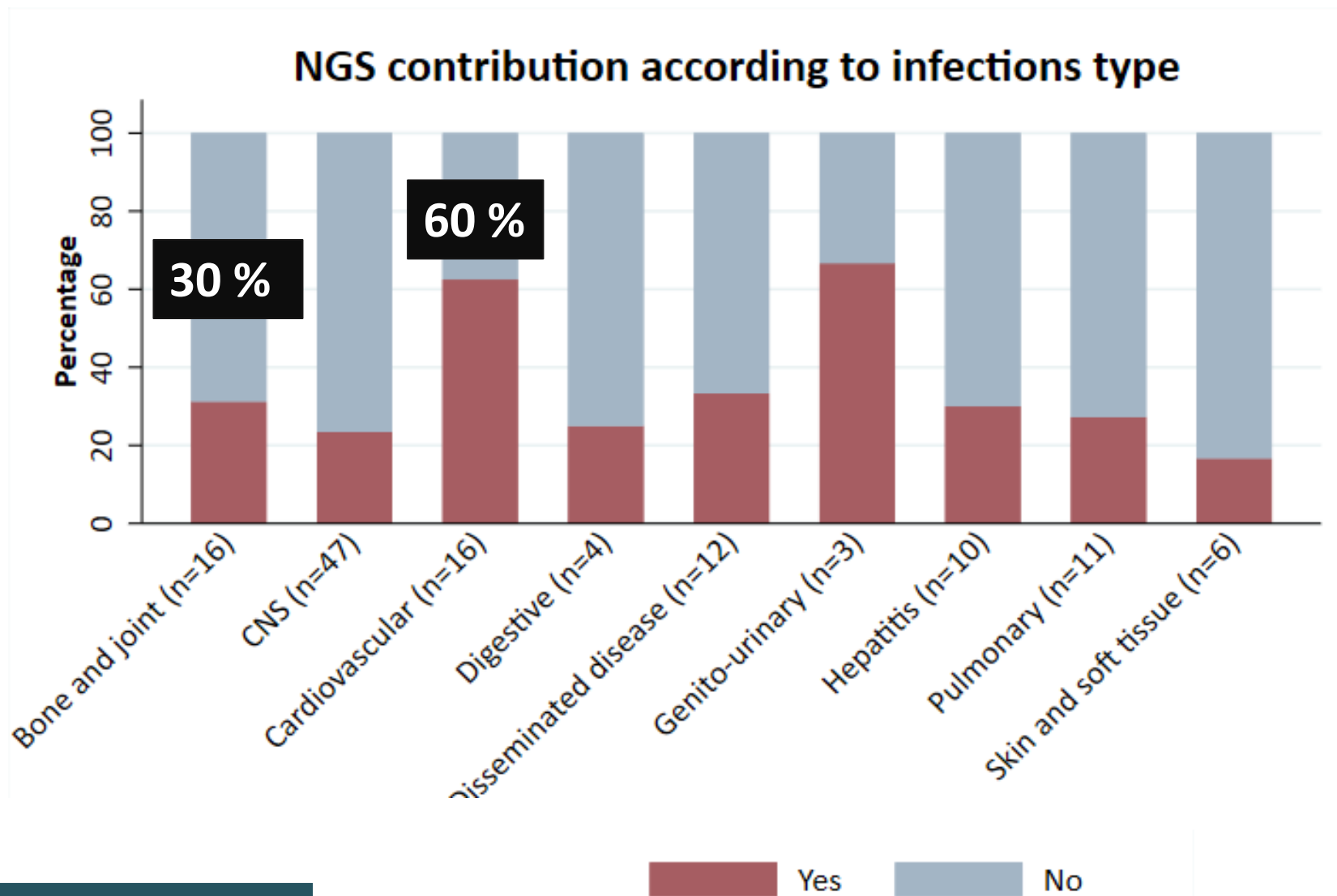


Aujourd'hui : pour diagnostic difficile

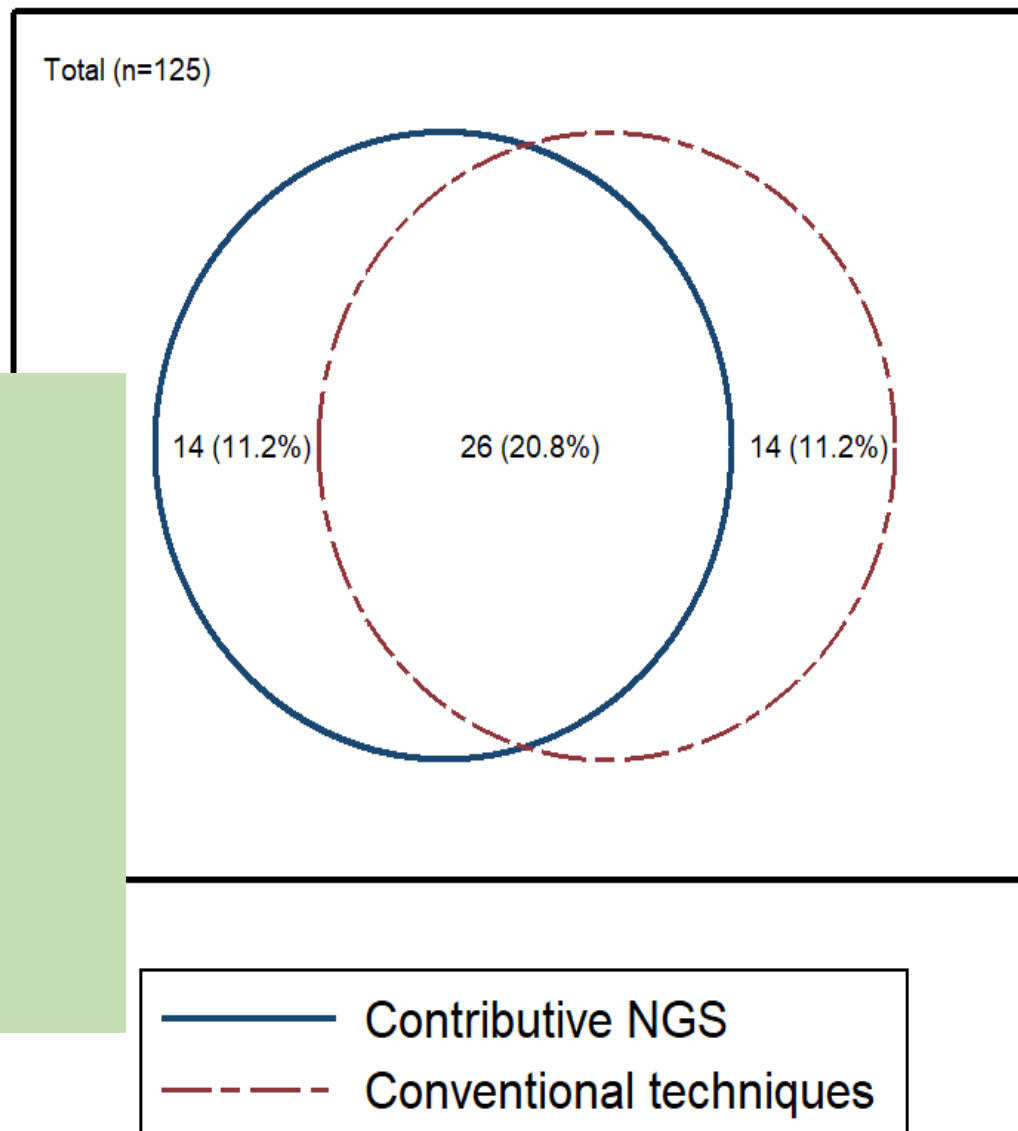
Prospectif
Multicentrique
Tout prélèvement 2018-2020
213 patients



Infections variées



Abscès hépatique à Kp, anaérobies
 Encéphalite à Orthobunyavirus,
 Moraxella, Avulavirus
 Faux positif séro VIH
 Syphilis
 Méningite à erysipelotheix, Bacillus
 Nocardiose pulmonaire
 Ostéite à saccharomyces
 PI CMV
 Abscès rein polymicrobien
 BK



26 individuals are in the intersection of the two sets

7 autres prélèvements
 2 BK
 5 sérologies

Mais déjà des données en première ligne

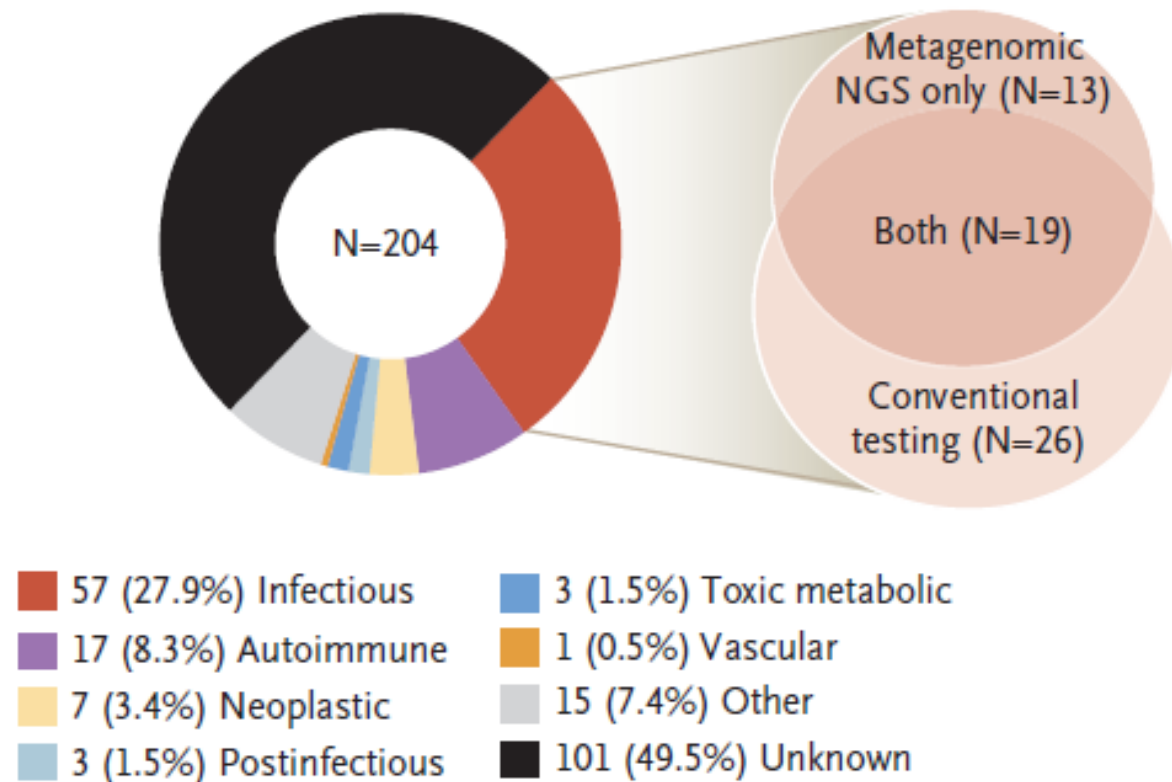
ORIGINAL ARTICLE

Clinical Metagenomic Sequencing
for Diagnosis of Meningitis and Encephalitis

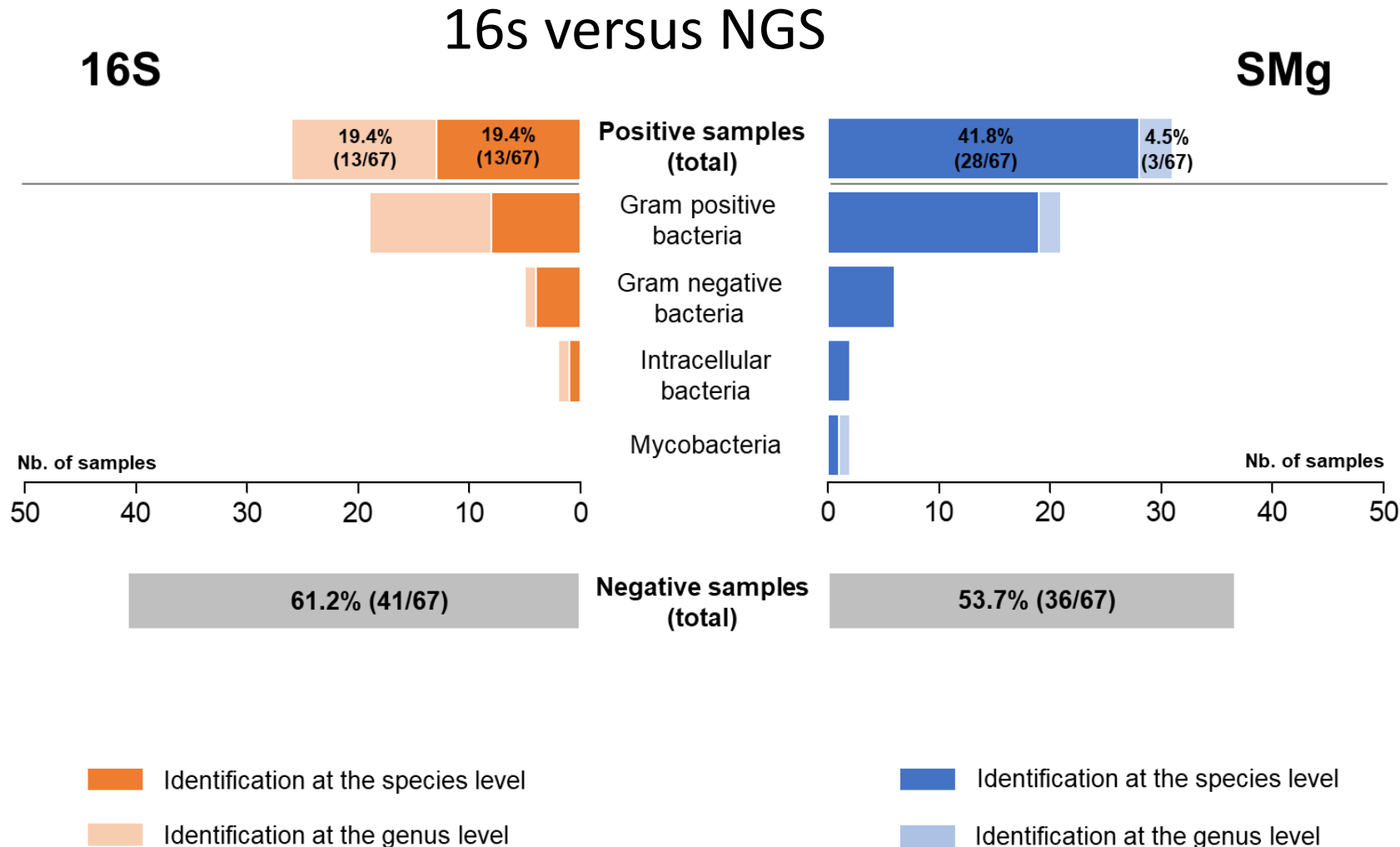
Prospectif
Multicentrique
Méningites/Encéphalites
204 patients

Etiologie retrouvée ½
Infectieux 1/4

A Established Diagnoses in the Study Patients



Dans l'infection ostéoarticulaire



Etude prospective (N=67)

Polymicrobien
BK
+ 10 % de diagnostic
à l'espèce

NGS en maladies infectieuses... et la suite ?

- Epidémiologie
- Infections difficiles à diagnostiquer
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- Résistance aux antimicrobiens

Table 1. Summary of Antimicrobial Susceptibility Data for Various Strain/Antibiotic Combinations

Species	Antibiotic			
	Ceftazidime	Cefepime	Piperacillin-Tazobactam	Meropenem
<i>Escherichia coli</i> (n = 31)				
Resistant ^a , No. (%)	20 (64.5)	18 (58.1)	8 (25.8)	1 (3.2)
MIC range, mg/L	<0.25 to >256	<0.5 to >256	2 to >256	<0.06 to >64
<i>Klebsiella pneumoniae</i> (n = 24)				
Resistant ^a , No. (%)	16 (66.7)	14 (58.3)	10 (41.7)	6 (25.0)
MIC range, mg/L	<0.5 to >256	<0.25 to >256	2 to >256	<0.06 to >64
<i>Pseudomonas aeruginosa</i> (n = 22)				
Resistant ^a , No. (%)	6 (27.3)	10 (45.5)	9 (40.9)	14 (63.6)
MIC range, mg/L	1 to >256	2 to >256	2 to >256	0.12 to >64
<i>Enterobacter cloacae</i> (n = 13)				
Resistant ^a , No. (%)	7 (53.8)	3 (23.1)	7 (53.8)	0 (0.0)
MIC range, mg/L	<0.5 to >256	<0.5 to 32	0.75 to >256	0.25 to 1

Whole-Genome Sequencing Accurately Identifies Resistance to Extended-Spectrum β -Lactams for Major Gram-Negative Bacterial Pathogens

Samuel A. Shelburne,^{1,2,3} Jiwoong Kim,^{4,5} Jose M. Munita,^{3,6,7} Pranoti Sahasrabhojane,¹ Ryan K. Shields,⁸ Ellen G. Press,⁸ Xiqi Li,⁹ Cesar A. Arias,^{3,6,10,11} Brandi Cantarel,⁴ Ying Jiang,¹ Min S. Kim,^{4,5} Samuel L. Aitken,^{3,12} and David E. Greenberg^{3,13,14}

90 souches
Gold standard microdilution

Table 3. Diagnostic Performance of Whole-Genome Sequencing Versus Clinical Microbiology Data Using Broth Microdilution as the Gold Standard

Diagnostic Performance	WGS	Clinical Microbiology	PValue
Sensitivity (95% CI)	0.87 (.81–.92)	0.82 (.76–.88)	.36 ^a
Specificity (95% CI)	0.98 (.96–.999)	0.95 (.92–.98)	.07 ^a
Positive predictive value (95% CI)	0.97 (.94–.999)	0.92 (.88–.97)	.025 ^b
Negative predictive value (95% CI)	0.91 (.88–.95)	0.88 (.84–.92)	.24 ^b

Original article

From genotype to antibiotic susceptibility phenotype in the order
Enterobacterales: a clinical perspective

E. Ruppé ^{1,*†}, A. Cherkaoui ², Y. Charretier ¹, M. Girard ¹, S. Schicklin ³, V. Lazarevic ¹,
J. Schrenzel ^{1,2}

⇒ 1496 isolate/antibiotic combinations (187 isolates × 8 antibiotics)

⇒ 96.4 % were correctly predicted, 1.9 % were major errors (MEs) and
1.7 % were very major errors (VMEs)

⇒ The majority of VMEs were putatively due to the overexpression of
chromosomal genes

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Prediction of Susceptibility to First-Line Tuberculosis Drugs
by DNA Sequencing

The CRyPTIC Consortium and the 100,000 Genomes Project

10 200 souches

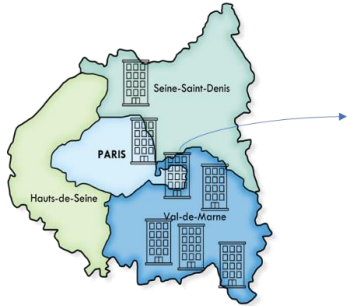
Sensibilité spécificité > 90 %

CONCLUSIONS

Genotypic predictions of the susceptibility of *M. tuberculosis* to first-line drugs were found to be correlated with phenotypic susceptibility to these drugs. (Funded by the Bill and Melinda Gates Foundation and others.)

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- Résistance aux antimicrobiens
- Intégration des données de l'hôte



7 Hospitals in the east of Paris
Intensive Care, Hospitalized and Out Patients

Lead : Henri Mondor Hospital (Créteil, France)
Principal Investigator : Dr C. Rodriguez

Métagénomique J0

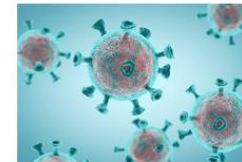
RESEARCH ARTICLE

Viral genomic, metagenomic and human transcriptomic characterization and prediction of the clinical forms of COVID-19

Christophe Rodriguez^{1,2}*, Nicolas de Prost^{3,4}, Slim Fourati^{1,2}, Claudie Lamoureux¹, Guillaume Gricourt^{1,2}, Melissa N'debi^{1,2}, Florence Canoui-Poitaine^{5,6}, Isaac Désveaux¹, Oriane Picard¹, Vanessa Demontant¹, Elisabeth Trawinski¹, Raphaël Lepeule¹, Laure Surgers¹, William Vindrios⁷, Jean-Daniel Lelièvre⁷, Nicolas Mongardon⁸, Olivier Langeron⁸, José L. Cohen^{9,10}, Armand Mekontso-Dessap^{3,4}, Paul-Louis Woerther^{1,11}, Jean-Michel Pawlotsky^{1,2}



Impact on severity COVID-19 ?



SARS-CoV-2



Micro-Organisms

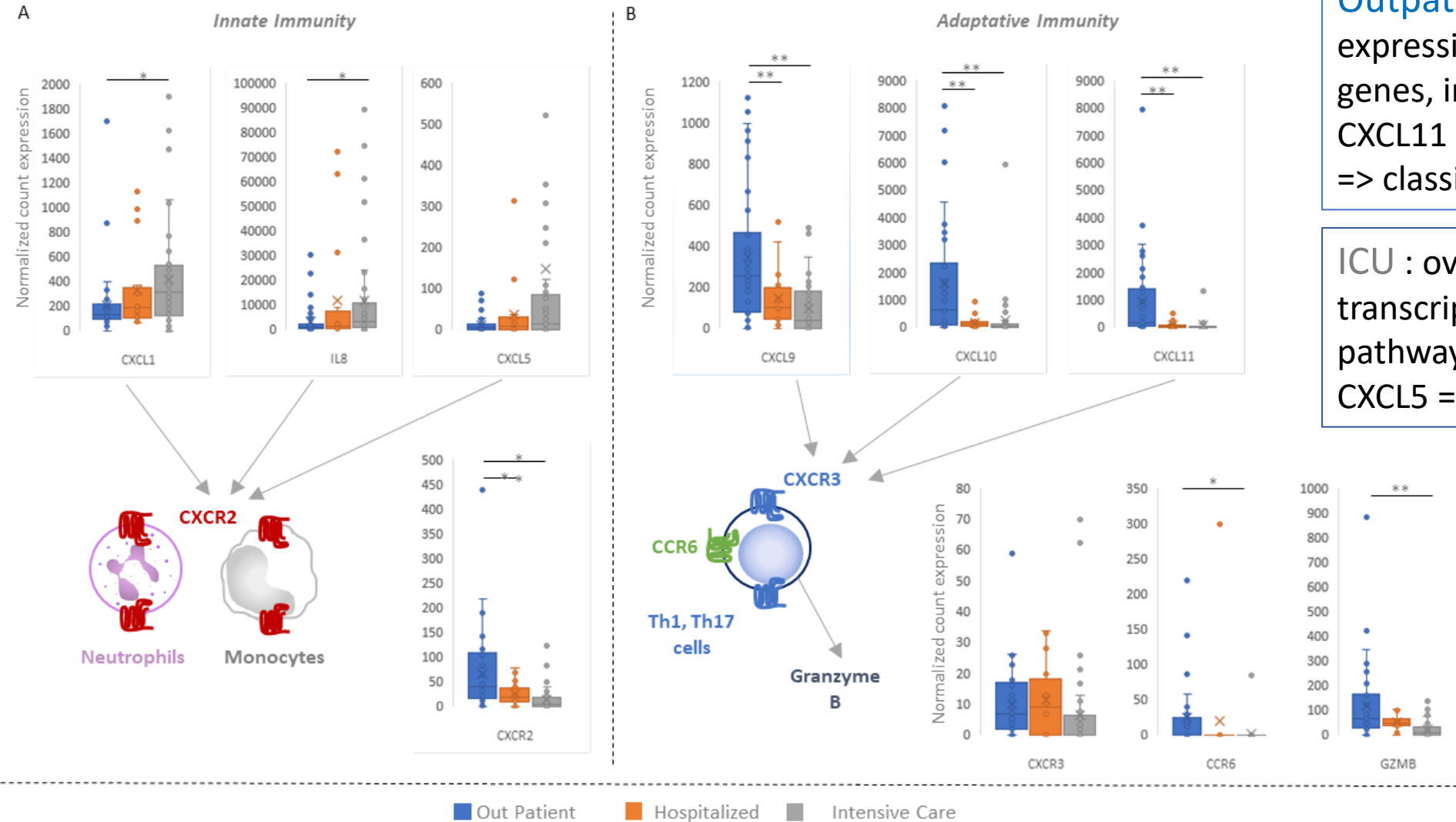


Immunity

COMETS

Patient characteristics	Outpatients (N=42)	Hospitalized patients (N=17)	ICU patients (N=45)
Demographic parameters			
Age, median [range], year (N=104)	50 [19-86]	50 [42-86]	65 [33-90]
Female- n/N (%) (N=104)	30/42 (71.4)	7/17 (41.2)	9/45 (20.0)
Risk factors			
Current smokers- n N (%) (N=88) □	4/34 (11.8)	0/10 (0)	3/44 (6.8)
BMI, median [range] (N = 84) □	25.6 [16.9-34.0]	26.5 [19.8-39.0]	28.0 [20.9-43.3]
Obese patients: BMI ≥30- n N (%) *	7/34 (20.6)	3/10 (30.0)	12/40 (30.0)
Chronic underlying conditions, n N no (%) □			
Chronic obstructive pulmonary disease (N= 88)	2/34 (5.8)	1/10 (10.0)	2/44 (4.5)
Asthma (N=88)	7/34 (20.6)	2/10 (20.0)	1/44 (2.3)
Diabetes (N=88)	1/34 (2.9)	2/10 (20.0)	14/44 (31.8)
Hypertension (N=88)	7/34 (20.6)	6/10 (60.0)	24/44 (54.6)
Cardiac disease (N=88)	2/34 (5.9)	2/10 (20.0)	10/44 (22.7)
Chronic renal disease (N=88)	1/34 (2.9)	0/10 (0)	3/44 (6.8)
Cancer (N=88)	2/34 (5.9)	0/10 (0)	1/44 (2.3)
Immunodeficiency (n = 88)	0/34 (0)	2/10 (20.0)	4/44 (9.1)
Treatments n N (%) □			
Antibiotics (N=89)	11/34 (32.4)	6/11 (54.6)	38/44 (86.4)
Corticosteroids (N=89)	1/34 (2.9)	2/11 (18.2)	1/44 (2.3)
NSAIDs (N=89)	9/34 (26.5)	1/11 (9.1)	5/44 (11.4)
Others immunosuppressive drugs (N=89)	0/34 (0)	2/11 (18.2)	4/44 (9.1)
COVID-19 Disease			
Median interval from symptoms to NSP, days (N=86) □	4	7	6
Neutrophil count, median [range], G/L	/	4.9 [2.4-11.0]	5.6 [2.3-16.4]
Lymphocyte count, median [range], G/L	/	1.1 [0.6-3.2]	0.7 [0.2-1.6]
Ventilation, n N (%)			
Oxygen (N=104)	0/42 (0)	8/12 (66.7)	45/45 (100)
Non-invasive ventilation (N=104)	/	/	21/45 (46.7)
Mechanical ventilation (N=104)	/	/	19/45 (42.3)
SOFA score, n N (%) □			
<6 (n = 43)	/	/	20/43 (46.5)
≥6 (n = 43)	/	/	23/43 (53.5)
Death – n/N (%) (N=104) □	0/42 (0)	1/17 (5.9)	17/45 (37.8)

COMETS



Outpatient : significant increase in the expression of CXCR3 pathway activator genes, including CXCL9, CXCL10, CXCL11 and Granzyme B
=> classical Th1-Th17 T-lymphocyte

ICU : overexpression of cytokine transcripts activating the CXCR2 pathway, including CXCL1, IL-8 and CXCL5 => réponse T inadaptée

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- Résistance aux antimicrobiens
- Intégration des données de l'hôte
 - Risque de forme grave (cf COMET) => intensification traitement ?
 - Profil bactérien / viral => épargne antibiotique
 - Profil évolutif ? => réduction durée traitement

Etudes cliniques pour positionnement en première ligne dans les grands syndromes cliniques +++

Merci pour votre attention