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The urinary microbiote renal failure with infectious lithiasis in children.

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Purpose :
The study of the urinary microbiota makes it possible to establish the epidemiological of urolithiasis infected or coloned sometimes by mutli-resistant and cunning microorganisms necessary in the prevention of infectious or functional complications and control his clinical recurrence.
Methods :
1-Culture and antibiotics sensitivity tests from microorganims stone kidney after fragmentation have been realized .
2-A direct metagenomic ribosome 16 S V3-V4 PCR in EP 2x300 LP (one MiSeq illumine) of the intraoperative pyelic pyuria urine has been analyzed .
Results:
1-The culture of the stone returns positive at bacteria urealytic positive: *Morganella morganii* sensitive to the calforan ; imipenem and resistant to Cefazolin; Augmentin and Ampicillin .
2-A direct metagenomic analysis of intraoperative pyelic pyuria and the bioinformatic according to the presence of an Anaerobic bacterium : *Bacteroides fagilis* and others .
Conclusion : From this genomic experience , we will have to review the threshold microorganisms and especially the place of Anaerobes or emerging agents by a genomic multicenter study of the urinary microbiota of patients with infections urolithiasis to better management of this public health pathology

I-Introduction

Lithiasis infection is an important part of daily medical practice in urology, pediatrics and nephrology today. According to available statistics, it represents between 10 to 15% of urinary lithiasis (Daudon et al .2018).
This proportion is very controversial because, according to experts, it is underestimated. Recent data report its participation, and sometimes its implication, in complex and multifactorial phenomena of the formation of other types of lithiasis; classified so far as metabolic.
Adequate management of this type of lithiasis cannot be achieved without understanding its ethiopathogenesis, based mainly on the specific study of lithiasis and its urinary microbiota.
The study of the urinary microbiota makes it possible to establish the epidemiological profile of the sometimes mutli-resistant and cunning

microorganisms necessary in the prevention of infectious or functional complications and the control of their recurrence (Lange et al .2019) .

II-Methods

1-It Acts M, S 05 year old girl originating in Taref and consanguineous relative A, admitted at the Hospital Mohammed Seghir Nekkache for pyelic lithiasis on final renal insufficiency with urosepsis (fever with 40, shiver, AEG and weakens).Assessment infectious : hyperleucocytoses 12400 elements , hemoglobine: 8.6 and CRP 20 mg/l. Setting under antibiotic treatment (imipenem and Flagyl *and Triflugan*) .Programmed for surgery on the urinary tract thanks to a technique flexible Uretroscopie (URS).

2-Setting Culture and antibiotics sensitivity tests microorganism from stone kidney after fragmentation have been realized according to The Clinical & Laboratory Standards Institute (CLSI).

3-Direct metagenomic ribosome 16 S V3-V4 PCR in EP 2x300 LP (one MiSeq illumine*) of the intraoperative pyelic pyuria urine has been analyzed via **Macrogen** molecular bio-technology platform .The taxa analysis was deduced using the maximum parsimony (MP) method. The Phylogenetic tree was obtained using the Subtree-Pruning-Regrafting (SPR) algorithm (Nei et al. 2000).

4-Bioinformatics software are included four algorithm preprocessing: CleanPrimer, Slice Genome, PCR clean and Sample Size. It included a single metagenomic data simulation algorithm, simulateR for error and correction and the processing to correct errors in Next Generation Sequencing data included : eMER-kmer process coverage .

III-Results

1- The microbiological assumption of responsibility returns with a preoperative ECBU with strong a leucocyturie amicrobienne to the uroculture (direct examination: 1764 Ets /mm and many Red blood corpuscles)

2-The culture of the pyelic pyuria returns positive A bacteria urealytic : *Morganella morganii* sensitive to : calforan ; imipenem and resistant to :Cefazolin. Augmentin and Ampicillin.

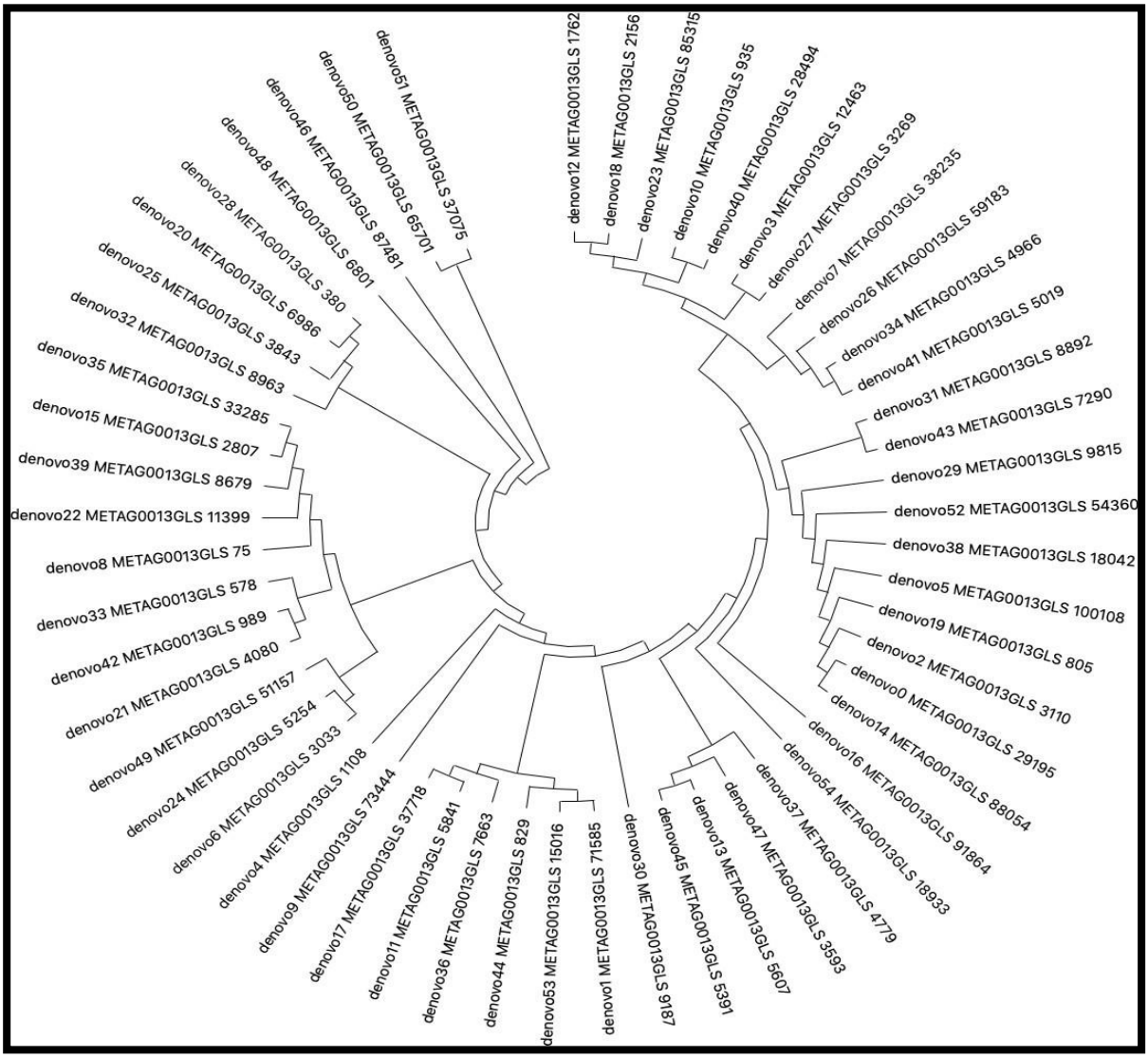
3-After The pejorative evolution of the patient towards the multiple abscess of the left kidney and acute renal insufficiency with 180 creatinin micromoles ,

4-A Contracting and calcification urétérale and fibrin was objectify by the surgeon urologist.

5-A direct **metagenomic analysis** of intraoperative pyelic pyuria and the bioinformatic analysis gave the results after **parental consent** according to: mainly *Entérobactéries*(60.68%,), then *Lactobacillales* (13.28%), *Micrococcacales* (8.68%), *Actinomycetales* (4.12%);*Pseudomonadales*(2.80%), *Bacillales* (2.44%),

Corynebacteriales (1.94%); *Clostridiales* (1.04%) and other bacteria with a percentage which does not exceed the 1% , notification of the presence of an Anaerobic bacterium of the *Bacteroides* and others . (Fig.01)

Figure 01: Phylogenetic tree of bacterial specie of intraoperative pyelic pyuria (Macrogen).



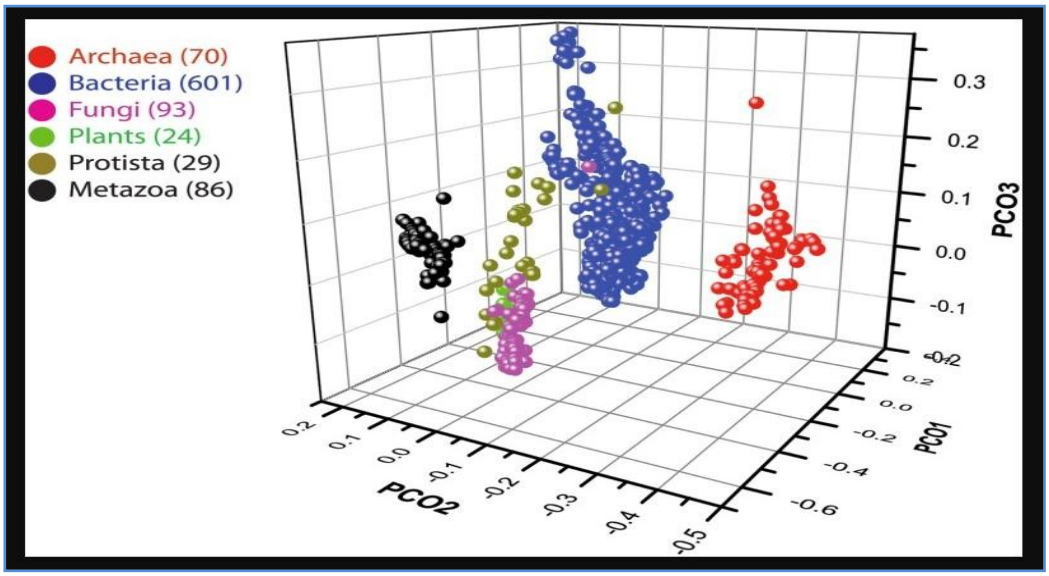
VI-Discussion

1-Genome sequencing of urine samples (Fig.02) in the context of infection, its complementarity with urine culture, automated species identification and the properties of either approach are generally not well studied.

2-To assessthis aspect, we used 16S rDNA sequencing techniques from metagenomics, which offers increased resolution, allowing more specific and expanded taxonomy to the available urobiome data base.

3-The functional classification of sequences, as well as the discovery of new genes and bacterial genomes, isexplored with the aim of having a greater potential for identification and involvement of strains in the infectious process.

Figure 02 : Graphic representation of taxa in metagenomics in urine(Macrogen).



4-Recent articles ; point to the existence of subpopulations (subspecies) in the majority of abundant urinary prokaryotes. They allow a better functional and ecological understanding of the human urinary microbiome associated with urolithiasis (Ruan et al.2019; Janes et al.2017 ; Voroshilova et al.2016 ; Zampini et al.2019) .

5-This dimension is not captured by the sequencing of 16S rDNA. The study convincingly identifies a high number of reads of conventional uropathogen sequences, but also proposes new bacterial species associate dwith the characteristics of infection. It questions the thresholds used to define infection: generally 10^5 Units Form a Colony (CFU) in urine culture.

6-The quantitative nature of the metagenomic approach (NGS) may identify new uropathogens in lower amounts in samples showing signs of infection.

7-It has the ability to identify bacteriathat are difficult to grow, such as a possible pathogen, for example : *Alloscardovia* and *Actinotignumsp.A. schaalii*,which maybe an underestimated cause of urinary tract infections due to its fastidious growth and difficulty in identify ingit by phenotypic methods (Moustafa et al.2018 ; Mansi et al.2016; Hiltet al.2014) .

8-The microorganisms found as *Fusobacterium nucleatum*, *Bacteriodes fragilis*, *Acinetobacter variabilis*, *Pseudomonas glareae* and others, in our exploration by this metagenomic, denote the variability of the urinary microbiome in fatty especially emerging bacteria that may be involved in the promotion urolithiasis infection or renal failure (Curhan et al.2001; Turney et al.2012).

9-In our experience of exploring the pejorative evolution in young girls, it appeared that the most plausible hypothesis was that of Anaerobes involved in this renal failure, in particular *Bacteriodes fragilis* due to its numerous virulence and

chromosomal resistance factors (Extended spectrum β-talactamase) and other phenomena described in parenchymal abscess (Bandoh et al .1991 ; Michon et al .2015 ; Gottschick et al .2017) .

V-Conclusion

From this genomic experience,we will have to review the threshold microorganisms and especially the place of anaerobes or emerging agents by a genomic multicenter study of the urinary microbiota of patients with complicated urolithiasis in order to clarify the areas of current shadow in the etiopathogeny of urolithiasis of any kind and control the desimination of mutliressitant microorganism for better medical care. However, we should alaways correlating all the results to the clinical and epidemiological context of the patients.

VI-Ethics approval and consent to participate: I have had the consent of the parents and the authorization of the attending physician.

EXAMEN DES CARACTERISTIQUES GENETIQUES A DES FI

IDENTIFICATION	IDENTITE DU TITULAIRE DE L'AUTORITE PARENTALE SI MINEUR OU DU TUTEUR
NOM : PRENOM : DATE DE NAISSANCE :	NOM : PRENOM :

CONSENTEMENT

Je soussigné(e), susnommé(e), reconnais avoir été informé(e) par le Docteur sur l'examen des caractéristique génétiques suivant :

Sequençage génétique direct à partir du sang.

Cet examen sera réalisé à partir ☐ du prélèvement qui m'a été effectué
☒ du prélèvement qui a été effectué sur mon enfant mineur

Je donne mon consentement pour ce prélèvement.

Seul le médecin prescripteur ici désigné est habilité à me communiquer les résultats.
Ces derniers ne seront transmis à aucun membre de ma famille. Si cette transmission apparaissait essentielle, elle n'interviendrait qu'avec mon accord.

Fait à : HCA Le : 31/07/2019 Signature :

ATTESTATION

Je soussigné (e) Docteur
atteste avoir reçu ce jour Monsieur
avoir reçu son consentement et l'avoir informe(e) sur :
☒ les caractéristiques de la maladie recherchée,
☒ les résultats susceptibles d'être obtenus,
☒ les modalités de communication des résultats.

Fait à : HCA Le : 31/07/2019 Signature et cachet

Dr. Z. [illegible]
Résidente en Pédiatrie

VII-Consent for publication: was submitted to the ethics committee of the university hospital Mohamed Seghir Nekkache. Algiers

VIII-Availability of data and materials: Thanks to the patients computer file (DPI) of the hospital and Gene life science .

IX- Competing interests: Declaring to have no financial interest

X- Funding: Total financing at my expense

XI-Authors' contributions : Provision of the urologist with the medical file and the intraoperative sample

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XIII-References

[1] Daudon M. Epidemiology of urolithiasis. Paris (France): Elsevier Masson; 2018.

[2] Lange D , Scotland KB. The Role of Bacteria in Urology. 2ème ed. London (UK) :Spinger ; 2019.

[3] Janes VA, Matamoros S, Willemse N, Visser CE, de Wever B et al. Metagenomic sequencing to replace semi-quantitative urine culture for detection of urinary tract infections: a proof of concept. BioRxiv. 2017.

[4] Voroshilova TM. Metagenomic analysis of samples in cystitis. Urologiia. 2016 Feb;(1):29-31.

[5] Zampini A. Definining Dysbiosis in Patients with Urolithiasis. Scientific reports. 2019;9:5425.

[6] Ruan F. Antibiotic resistome profile based on metagenomics in raw surface drinking water source and the influence of environmental factor: A case study in Huaihe River Basin, China Environmental Pollution. 2019;248:438-44.

[7] Mansi M, Goldfarb DS. The role of the microbiome in kidney stone formation. International Journal of Surgery. 2016;36:607e-612

[8] Moustafa A.Microbial metagenome of urinary tract infection .www.nature.com/scientific. 2018.

[9] Hilt, E. E. et al. Urine is not sterile: use of enhanced urine culture techniques to detect resident bacterial flora in the adult female bladder. Journal of clinical microbiology 52, 871–876 (2014)

[10] Curhan, G. C., Willett, W. C., Speizer, F. E. & Stampfer, M. J. Twenty-four–hour urine chemistries and the risk of kidney stones among women and men. Kidney international 59, 2290–2298 (2001)

[11] Turney, B. W., Reynard, J. M., Noble, J. G. & Keoghane, S. R. Trends in urological stone disease. *BJU international* **109**, 1082–1087 (2012)

[12] Bandoh K ,MutoY. Biochemical properties and purification of metallo-B-lactamase from *Bacteroïdes fragiIis*. Antimicrob. Agents Chemother. 1991;35(2):371-372

[13] Michon AL, Dubreuil L, Marchandi H. Anaerobic bacteria: general. 90-05-0035-A] - Doi : 10.1016/S2211-9698(14)61315-7. 2015 Elsevier Masson

[14] Gottschick C.The urinary microbiota of men and women and its changes in women during bacterial vaginosis and antibiotic treatment. Microbiome. 2017 Aug 14;5(1):99. doi: 10.1186/s40168-017-0305-3