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Research article

# A STUDY ON RECTAL CARRIAGE OF MULTIRESISTANT GRAM-NEGATIVE BACILLI IN CHILDREN ATTENDING PEDIATRIC HOSPITAL AMBOHIMIANDRA, MADAGASCAR

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## ABSTRACT

**Introduction:** Gram-negative bacilli are often responsible for rectal colonization in children. These bacilli normally sensitive to third generation cephalosporins (3GC) have acquired disturbing antibiotic resistance in recent years, hence the interest of our study. The aims are to monitor the epidemiological evolution resistance of Enterobacteriaceae with antibiotics in particular β-lactams, estimate the prevalence of enterobacteria responsible for rectal colonization and their resistance to 3GC. **Material and methods**: This is a descriptive prospective study of *Enterobacteriaceae* in children attending Ambohimiandra Hospital for a period of 3 months (from 01 August to 31 October 2015). We have done a rectal swab of all the children whom we have parental consent. The samples were processed at the Laboratory of Medical Biology Faravohitra. The parameters studied are the antecedent of therapy antibiotic, notion of recent hospitalization and the results of microbiological examinations. **Results:** Of the 55 bacteriological samples obtained, 39 strains of *Enterobacteriaceae* were isolated, *Escherichia coli* (n = 12), *Enterobacter cloacae* (n = 13), *Citrobacter spp* (n = 1), *Salmonella spp.* (N = 5) and *Shigella spp* (n = 2), *Proteus mirabilis* (n = 1). Six isolated strains (50%) of *Escherichia coli* showed resistance to ceftriaxone, of which 4 (33.33%) produced ESBL. **Conclusion**: Gram-negative bacilli responsible for colonization of the digestive tract have several resistances to β-lactams, in particular 3GC, by the production of enzymes (Expanded spectrum beta-lactamase or ESBL and cephalosporinase or CASE) which hydrolyze these antibiotics. Measures to combat the spread of these phenomena resistance must be implemented in the Malagasy community to limit them.

**KEYWORDS:** Rectal porous, Gram negative bacillus, ESBL, Multiresistence

## INTRODUCTION

Antimicrobial resistance is a problem of worldwide health systems, resulting in failure, high treatment costs and increased mortality. An increase production of β-lactamase by Gram-negative multiresistant in the organisms, such as extended spectrum β-lactamase (ESBL), or plasmid-mediated carbapenems-producing bacteria, is a particular concern [1]. Let us mention that the main reservoirs of the different Gram-negative species are the environment, intestines of animals and humans, then these organisms can result in infections acquired on the community in humans. Due to the presence of co-resistance with other classes of antibiotics (including fluoroquinolones and aminoglycosides), the antibiotic treatment of infections caused by these organisms is a challenge [2]. The diffusion of these β-Lactamases is facilitated by horizontal transfers via mobile genetic elements (integrons, transposons and plasmids) and bacterial clonal proliferation [3]. Overall, surveillance data for antimicrobial resistance varies from country to country, ranging from the lack of national surveillance programs [1]

In recent years, the emergence of multiresistant bacteria, mainly related to the pressure of antibiotic selection and hygiene, concern to the medical world as in Madagascar [4]. ESBL in Madagascar were initially isolated between 2005 and 2006 of acquired urinary tract infections in the community [5]. They were then isolated in epidemic to two pediatric units in 2006. More recently, ESBL have been isolated from several infections acquired in different surgical and intensive care units Antananarivo [6]. In pediatrics, Andriatahina et al reported a fecal carriage of enteric bacteria producing an enlarged-spectrum β-lactamase (ESBL) before and after hospitalization respectively 21% and 51% of cases with a major

risk factor for the notion of prior hospitalization [7]. In intensive care and surgical services at two hospitals in the capital, Randrianirina and al, found ESBL strains in more than 30% of the bacteria isolated from different bacteriological samples to the patients included [8]. At the community level, Herindrainy and al. Reported a fecal port of Gram-negative ESBL bacillus in 10% of outpatients [9]. Another study on antimicrobial resistance of uropathogenic germs in an extra-hospital environment showed a resistance rate to ceftriaxone in 6% of the *Enterobacteriaceae* [10]. In addition, patients with ESBL infection and other third-generation cephalosporin-resistant bacteria are usually colonized by these bacteria in the intestinal tract. Spontaneous loss of ESBL positive colonizers occurs but is significantly delayed in repeated hospitalizations and antibiotic treatments [11,12].

The objective was to evaluate the resistance profile of enterobacteria isolated from rectal sampling in children attending the Ambohimiandra Hospital

## MATERIALS & METHODS

It is a descriptive observational prospective study, carried out over a period of 3 months (01 August to 31 October 2015), in the laboratory of Medical Biology, Faravohitra of Antananarivo. To better understand the extent of the problem, it is important to expand the databases on the existence of these strains in the various community sectors. In this sense, a preliminary study was carried out in the laboratory of medical biology, Faravohitra of Antananarivo, Madagascar. The objective was to evaluate the resistance profile of enterobacteria isolated from rectal sampling in children attending the Ambohimiandra Hospital. It is a prospective study, carried out over a period of 3 months (01 August to 31 October 2015), in the laboratory of Medical Biology, Faravohitra of Antananarivo. We included all children (1-13years old) hospitalized in the ambohimiandra university hospital with the consent of their parent, children with or without clinical manifestations of a digestive infection. The parameters studied were the history of antibiotic therapy, the history of recent hospitalization, and the results of microbiological examinations. Collected specimens were sent directly to the Laboratory of Training and Research in Medical Biology and immediately plated onto DCL agar (Difco® Laboratories Inc., Detroit, MI, USA) and incubated statically for 24–48 h at 37°C. Every colony type was tested for Gram staining and oxidase activity. Oxidase-negative Gram-negative bacilli were identified with the API 20E system (bioMérieux, Marcy l'Etoile, France). The sensitivity of the isolated Enterobacteriaceae was evaluated using the diffusion technique on solid medium following the recommendations of the Antibiogram Committee of the French Microbiology Society [1]. Ten antibiotics were tested: amoxicillin, ticarcillin, cefalotin, cefoxitin, ceftriaxone, gentamicin, tetracycline, ciprofloxacin, cotrimoxazole, amoxicillin-clavulanic acid. Two types of multiresistant enterobacterium have been particularly sought: ESBL-producing Enterobacteriaceae characterized by their ability to hydrolyze penicillins and cephalosporins of 1st (1CG), 2nd (2CG), 3rd (3CG) and 4th generation but whose action Is inhibited by the presence of an inhibitor of B-lactamase such as clavulanic acid thus making it possible to detect them by the presence of synergy between clavulanic acid and a 3CG; the hyperproductive strains of cephalosporinase whose cephalosporin hydrolysis is not inhibited by clavulanic acid (absence of synergy between clavulanic acid and a 3CG).

## RESULTS

Of the 55 bacteriological samples obtained, 39 strains of *Enterobacteriaceae* were isolated, including *Escherichia coli* (n = 12*), Enterobacter cloacae* (n = 13), *Citrobacter spp* (n = 1), *Salmonella spp*. (N = 5) and *Shigella spp* (n = 2*), Proteus mirabilis* (n = 1).

Six isolated strains (50%) of *Escherichia coli* showed resistance to ceftriaxone, of which 4 (33.33%) produced ESBL. In addition, all strains exhibited resistance associated with fluoroquinolone, cotrimoxazole, and tetracycline while maintaining sensitivity to gentamicin.

Table 1: Carriage rectal of the Enterobacteria according to hospitalization history .

| | Carriage rectal positive (%) | Carriage rectal negative (%) | Total (%) |
|---|---|---|---|
| **Previous Hospitalization** | 18 (90) | 2 (10) | 20 (36.4) |
| **No hospitalization** | 11 (31.4) | 24 (68.5) | 35 (63.6) |
| **p-value = 0,001** | | | |

Table 2: Carriage rectal of the Enterobacteria according to the antecedent administration antibiotic.

| | Positive rectal carriage (%) | Negative rectal carriage (%) | Total (%) |
|---|---|---|---|
| **History of antibiotic therapy** | 9 (64.7) | 5 (35.7) | 14 (25.5) |
| **No antibiotic therapy** | 20 (48.8) | 21 (51.2) | 41 (74.5) |

## DISCUSSION

These results confirm that it is classical to find a predominance of *Escherichia coli* (33,33%) in rectal colonization, this is similar to some study (32.2%)[7].They also confirm the presence of multidrug-resistant strains in the community. The associated resistance to the most accessible antibiotics inexorably limits the choice of antibiotic to be instituted for proper treatment. Moreover, this worrying presence hampers the current probabilistic antibiotic therapy, which is usually inspired by the antimicrobial resistance patterns found in Europe, partly the rate of transport of *Enterobacteriaceae* among young children in the French community (4.6%) [13,14]. Intestinal colonization

by ESBL producing isolates has a high risk of developing ESBL infection [15] and this intestinal transport is a factor in *Escherichia coli* infection producing ESBL[16]. In addition, the prevalence of ESBL transmission in children varies between studies and geographic areas. It ranges from 0.1% in Bolivia and Peru [17] to 31% in Niger [18]. These results suggest that studies of a broader population should be undertaken to statistically support these findings and to assess other risk factors and determinants of the carrying of these multiresistant bacteria in community and hospital settings. These factors include the notion of prior hospitalization and the history of antibiotic therapy. Regarding the relationship between this carriage rectal of multiresistant *Enterobacteriaceae* and the notion of prior hospitalization is significant. According to Andriatahina T. and al hospitalization in the past 30 days (adjusted OR = 7.4 [95% CI: 2.9-18.3]) was one significant risk factors for the acquisition of an ESBL with the previous antibiotic dose (OR adjusted = 4.1 (95% CI: 1.8-9.4)) [19]. This study therefore reinforces the interest of setting up a strategy to combat the spread of these multiresistant bacteria see on the table 1. In the case of Madagascar, the monitoring system for multidrug-resistant bacteria is less well developed, mainly due to a limited diagnostic infrastructure. Cultures are only taken after antibiotic therapy fails and the number of hospital infections is undoubtedly underestimated [20-23], which justifies our study table 2. The main lines of this policy should concern the setting up of a network to monitor the antibiotics sensitivity with the collaboration of all the laboratories in Madagascar; introduction medical training for doctors on the proper use of anti-infective; fight against the illicit sale of medicines which promotes self - medication even with antibiotics [4]. *Escherichia coli* multi resistant and other enteric bacteria producing extended spectrum β-lactamases (ESBLs) have emerged as an important cause of invasive infection. Targeting the primary (intestinal) niche with decolonization can be a valuable approach to reducing the risk of recurrent infections and transmission enteric ESBL pathogens [11].

## CONCLUSION

Finally, nosocomial infections and antimicrobial resistance are becoming increasingly challenging in low-income countries. The Malagasy medical scientific community should propose its own protocols of probabilistic antibiotic therapies because those currently applied are not always in line with the reality of antibiotic resistance profiles in our country. The detection and quarantine of patients with ESBL at admission to hospital is one of the appropriate measures for their eradication. Other studies are needed to show the prevalence of ESBL transport in Malagasy population and to identify its determinants epidemic.

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