Title: High prevalence of fecal carriage of extended-spectrum beta-lactamase-producing Escherichia coli and Klebsiella pneumoniae in a pediatric unit in Madagascar

Authors:

Todisosa Andriatahina (todiandria@yahoo.fr)
Frédérique Randrianirina (frederique@pasteur.mg)
Antoine Talarmin (atarmin@pasteur.mg)
Honoré Raobijaona (raobijaona@yahoo.fr)
Yves Buisson (yves.buisson@auf.org)
Vincent Richard (vrichard@pasteur.mg)

Version: 2 Date: 18 December 2009

Author's response to reviews: see over
Referee 1:

Reviewer's report

Andriatahina et al. describe the prevalence of extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL-PE) in a pediatric unit in Madagascar. Although the authors had mentioned it in the DISCUSSION, the major pitfall of this study is to not using an updated and standardized method to detect and verify ESBL-PE isolates. This would reflect the credibility of the data.

It would be more noteworthy to describe patients in groups; a group that remained to have no ESBL-PE for the entire hospital course; a group that had no carriage on admission and turned to have ESBL-PE carriage; a group that already had ESBL-PE carriage on admission and whether these patients remained to have the same organism or if there were changes in organism carriage. This information will help readers understand better about the evolving pattern of carriage and significance of this study.

*We propose inserting the figure 1 and the correction page 6.

Major Compulsory Revisions

1. The authors did not rationalize whether increasing carriage of ESBL-PE is correlated to infections among these patients, or just asymptomatic carriage. The clinical correlation of acquiring ESBL-PE during hospitalization and what are potential sources of these isolates would be important to mention.

   None of the six patients who have presented a nosocomial infection were ESBL-PE carriers on admission.

2. Because the design was similar to a before-and-after (admission) study for patient group, the authors should note when the medical staff were sampling in order to see the correlation with increasing carriage among patients (if the medical staff were a potential source of ESBL-PE spreading or the medical staff also received the organism during the same study period as patients).

   The medical staff were sampling at the beginning and at the end of the study. No difference was found between the two stages.

3. Since the environmental samples were collected each week, the authors should explain more in the RESULTS whether the positive samples were detected in which week, whether the number of positive samples was increasing during the later weeks, and whether the organisms detected were correlates with data from patients. More details on environmental sampling should make the study more comprehensible.

   We propose inserting figure 2

4. In the RESULTS/subheading risk factors, what the authors said regarding the average number of antibiotics prescribed during hospital was very unclear—it should be rewritten.

   Corrected page 8

5. The discussion should be much more concise and directed to the objectives of this study.

   Discussion was modified
6. Table 2 seems to be less useful because 1) there was no evidence whether the isolates from before and after admission were the same strains and 2) the study period was very short (1 month) to notice the changing resistance pattern. It may be described briefly only in text. Since a large portion of isolates may be acquired during hospitalization, it would be more interesting to instead compare between isolates that were acquired from community versus hospital.

Table 2 corrected

7. Tables 3-4 are very difficult to understand and should be restructured.

Tables corrected and in the results, corrections added.

Minor Essential Revisions
1. In RESULTS, data from medical staff were ?rectal swab cultures? not ?stool cultures?, please correct it.

Corrected page 6

2. There are several misspells throughout the manuscript.

Level of interest An article of limited interest Quality of written English Needs some language corrections before being published Statistical review No, the manuscript does not need to be seen by a statistician.
Declaration of competing interests I declare that I have no competing interests

Referee 2:
Reviewer's report
Major compulsory revisions

This study reported the prevalence of fecal carriage of extended spectrum β-lactamase producing Enterobacteriacea in a pediatric unit at Befelatanana hospital in Antananarivo (Madagascar). This kind of study is very important for the implementation of infection control measures and the management of bacterial resistance to antibiotics. It is an interesting worldwide public health subject.

Comments

Title: As the text is focused on Escherichia coli and Klebsiella pneumoniae I suggested to the authors the following title ?High prevalence of fecal carriage of extended-spectrum β-lactamase-producing Escherichia coli and Klebsiella pneumoniae in a pediatric unit in Madagascar?

Title modified/ page 1

Abstract: The methods used for the study are missed and the authors are advised to put the abstract in BMC Infectious Diseases abstract format.

Correction page 2
Materials and Methods

Design of the study
The authors should precise the duration of the study.

*E.g.* first sentence “Patients <15 years of age hospitalized in the pediatric unit of Befelatanana hospital in Antananarivo were enrolled, after receiving parental consent, in a cohort study from March to April 2008”.

It would be interesting to give the details on demographic characteristics and medical history.

Corrected page 4

Why do the authors record the civil status of the patients (< 15 years old)? The words “Civil status” was erased and replaced by “demographic characteristics”

Laboratory methods

It is important to know which antibiotics the authors used for double-disk test and the list of antibiotics for susceptibility testing. What about the confirmatory test for ESBL-producing strains?

Corrected page 5

ESBL production was also confirmed by Etest (AB Biodisk, Solna, Sweden) with strips containing Caz plus clavulanate versus Caz alone and cefepime plus clavulanate versus cefepime alone.

Statistical analysis

It is important to precise the significance level of p value.

Corrected page 5

Results

Patients

The determination of the mean length of hospital stay is not clear. It seems like a mean for 244 patients.

“Cohort (244 patients) characteristics included a sex ratio of 1.25, a mean age of 38.3 months (95% CI: 38.1-95.5). For all the patients included and for whose discharge happened, the mean length of hospital stay of 5.7 days (95% CI: 5.2 - 6.2).”

Actually, mean length of hospital stay concerned only the patients included in the cohort for whose discharge was happened during the study period.

Table 1: For good comprehension, it is important to give the number of patient by age group and the number of ESBL carriers on admission and discharge.

Table 1 corrected

Of the 254 patients on discharge, it would be interesting to know:

1. The rate of patient which be carrier and became non carrier
2. The number of patient who did not acquire ESBL-producing organism

*e.g.* figure 1

3. The rate of acquisition of ESBL-producing organism On admission, are the authors diagnosed infections with ESBL-producing organism?

Characteristics of ESBL-PE isolates
It would be interesting to know the total number of E. coli and K. pneumoniae strains isolated on admission and discharge for better understanding the frequency of ESBL-PE. As the authors talked about characteristics of ESBL-PE isolates, include strains from medical staff and environment would be good. Could be possible to the authors to give the number of ESBL-PE by species on admission and discharge?

The authors said that all tested strains were susceptible to imipenem and amikacin? But there are no results for imipenem in Table 2.

Results for imipenem were added in Table 2.

In the same Table, the authors give the results of antimicrobial resistance of Klebsiella spp. which is not Klebsiella pneumoniae. How do they explain this? How about the relevance of these results?

It’s a mistake. Corrected


Title modified

As the authors detected ESBL-producing strains by double-disk synergy test, it would be interesting to report in table 2 the activity of combination ?-lactam + inhibitor used.

Table 2 corrected.

Risk factors for ESBL-PE carriage
It is very difficult to comment Tables 3 and 4 as the authors do not give the significance level value of p. In general a p value <0.05 is considered significant. If I applied this value I noticed that on admission prior hospitalization was not the independent risk factor for ESBL-PE carriage; on discharge, the only independent risk factor is Brachial catheter (p= 0.05) which was not significant according to the univariate analysis. So it is difficult to follow the authors as the text is not reported what they have in the tables.

For example the authors wrote that Adjusted OR for prior hospitalization is 7.4 but in table this value is 8.24 (Table 3).

In the table 3 or 4, we presented the result of the first model (in that case, all variables with p-value<0.20 in univariate analysis were included), while in the text we presented the result of the backward stepwise procedure. Only the variable “prior hospitalization” was kept in the model. At each step we eliminated the variable that contributes least to the model.

It is important to have the significance level of p value for the relevance of these results.

We make the choice to present 95% CI but in this corrected manuscript we took in account the first reviewer’s remark and have restructured the tables 3 and 4.
I suggested to include in the Tables (3 &4) non-ESBL-PE carriers and removed the total. We make the choice to present “Total” because it is important to give in the same table descriptive information about all patients. Information concerning Non ESBL-PE carrier could be calculated.

Nosocomial Infections
How the authors could be determined the monthly incidence rate for one month study?
We corrected this information: “incidence rate… during the study period”

It would be interesting to know if isolates were ESBL-producers.
Information given page 9
It would be also important to know if the patients who acquired nosocomial infections were ESBL-PE carriers.
Information given page 9

Discussion
Paragraph 1: changed bacterial multiresistance by multiresistant bacteria
Corrected

Paragraph 2: Table 2 refers to antibiotics resistance not to risk factor.
Table “2” changed by Table “3”.

Paragraph 3: The authors reported for the first time the total number of ESBL-PE. It would be interesting to include this in the results with more details of different species as I already suggested.
Data included in the results

How do the authors solve the problem of production of cephalosporinase by Enterobacter cloacae during double-disk test?
to differentiate between ESBL- and non-ESBL-producing strains of Enterobacter cloacae, a double disc potentiation test was used to show synergy between a cefepime disc and an adjacent co-amoxiclav disc
ESBL production was also confirmed by disc containing cefepime plus clavulanate versus cefepime alone (an MIC ratio > 8 indicated ESBL production)

Paragraph 4: One of the main limitations of the study is the impossibility to know the relatedness between different ESBL-PE strains. Add to this the absence of ESBL confirmatory test.
Information added Page 11

Paragraph 6: there is confusion between ESBL-PE carriers on admission and outpatient.
Corrected

Last paragraph: I suggested to start by ?the detection and isolation of ESBL-PE in patient on admission ?...? instead of ?the detection and isolation of patients with ESBL-PE on admission ?...? We would like to talk about detection of ESBL-PE in patient on admission and quarantine of the patient with ESBL-PE carriage; e.g. correction page 12

References:
The authors should shake and put the name of bacteria species in italic.

*Name of bacteria species corrected*

Level of interest An article whose findings are important to those with closely related research interests Quality of written English Acceptable Statistical review No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests I declare that I have no competing interests