Reviewer's report

Title: Bordetella pertussis in infants hospitalized for acute respiratory symptoms remains a concern

Version: 2 Date: 9 September 2013

Reviewer: Flor M Munoz

Reviewer's report:

The authors have provided an improved manuscript based on the reviewer's comments. Most comments were taken into consideration, but not all.

The new title is more appropriate but reading through the manuscript, the main objective of the study still needs to be clearly described (is it to describe the clinical and laboratory characteristics that would help a clinician consider pertussis in an infant in order to start early treatment?) and important aspects of the methodology need to be better described as well.

The authors continue to speculate that vaccine immunity may be affected by the findings of 2 isolates with different genetic make up in this series, however, this is not supported by their study and it is something that only prospective epidemiologic and vaccine effectiveness studies might be able to ascertain.

Other Essential Revisions:

ABSTRACT
- The sentence used in the background is not a background statement, it is methodology. Please change.
- Methods paragraph: the information in the first parenthesis is unnecessary. Update methods accordingly after making change in "background".
- Results paragraph: Why first sentence on breast feeding? Provide report based on study methodology starting with demographics, etc. Sentence in the middle -"Infants with pertussis more often had no chest sounds on auscultation.." is this correct? Or are you referring to "abnormal chest sounds"? Last sentence in results section is a good addition.

Conclusion - not based on results - no incidence or rate of the disease given among all infants hospitalized for an acute respiratory infection. Are the authors concluding that "characteristics symptoms and laboratory findings of pertussis should be sought in infants with acute respiratory illnesses even during RSV season in order to start early antibiotic treatment"?

MAIN DOCUMENT

Page 3 - First paragraph, third sentence - is pertussis resurgence attributed to incomplete immunization programs or incomplete immunization status of the patient? Also, the citations used in this sentence and this paragraph refer to
European literature mostly, therefore this should be stated - that these variations are described "in Europe".

Second paragraph, first sentence - is "first booster dose at 3 months' correct? A booster would imply completion of a primary series of vaccine - I am not aware of any country that considers a 3 month old ready for boosters...

Same paragraph, although pertussis infection may occur in a patient with RSV or in patients without RSV but at the same time as RSV is circulating, as the authors have shown, the clinical presentation of these two diseases is distinct and these two clinical syndromes "pertussis-like" and "bronchilitis" can be differentiated by a knowledgeable clinician.

Last sentence of the second paragraph is unclear - what does "it" refer to?

Third paragraph - Last sentence, not really true - the clinical and laboratory characteristics of pertussis have been well described, even in neonates, and therefore these are not lacking as suggested by the authors, rather, clinicians' awareness of these characteristics is necessary. Also, one would not consider it "unfortunate" that in clinical practice the diagnosis is usually made without microbiologic confirmation - again, most diagnoses rely on clinical and laboratory features and confirmation by PCR is achieved in places where resources are accessible.

Page 4 - The main purpose of this study needs to be better described. This is a descriptive study, so a description of the clinical and laboratory features of infants hospitalized with laboratory confirmed pertussis is provided and compared to those of infants with laboratory confirmed RSV.

The second and third sentences of this paragraph are describing methods and should be moved to the methods section.

Methods

Patients - first paragraph. Confusing. Mixes methodology with results. The sentence "no B. pertussis clustering was observed..." is a result, not a method. It is not clear WHY was a 1:1 ratio chosen to identify controls to the cases. Usually the ratio in a retrospective study is higher so that at least two or three controls are identified for every case. Please explain why 1:1 was chosen. Also, not clear as to whether patients were hospitalized or seen in the Emergency Department, or both, or Hospitalized after being seen in the ED. Please clarify this and also what the baseline population was. Was it 164 infants that were hospitalized and were the 19 with pertussis among the 164? Can you please add the inclusion criteria for the study - how old did the infants need to be to be included in the study? Where there any other inclusion/exclusion criteria?

Page 5 - if this is a retrospective study, how was a clinical severity score assigned at admission? Are the authors meaning to say that " A clinical severity score was assigned to each child based on symptoms described at the time of admission"?. Again, in this retrospective study, how was the study population selected (inclusion/exclusion criteria) and identified (based on discharge diagnostic codes, based on laboratory reports, both?) and how were parents of subjects asked to signed consent (by letter, in person, with a clinic visit?)
would expect that in a retrospective study some families might not be accessible anymore after some time has passed from their infant's initial hospitalization. Please describe how many potential subjects were identified and how many actually agreed to be in the study.

The next two sections on testing need to indicate how patients are selected for testing (is this the decision of the clinician caring for them in the ED?, is there a protocol? is this for all hospitalized patients? is this routine for all ED patients, even if not hospitalized?)

Page 6 - were the "B.pertussis isolates" or "nasopharyngeal samples" cultured on charcoal agar?

Results
Page 7 - please indicate if "schooling" for the study population refers to out of home care (daycare) given the age of the patients, or if indeed, they are in a school setting.

second paragraph, sentence before last - "Finally, chest examination..." is not clear, please re-phrase. And in the sentence after that, please include the median and range data right next to each of the parameters it describes, not both together.

Discussion
First paragraph - why starting with genetic information and vaccination? Again, the discussion should follow the order of the data results.

In this same paragraph - second sentence, why "clinically" important that the strains found here differ from the ones in the vaccines - is there any difference in clinical presentation or management because of this? If there is, describe, if not, delete "clinically". Also Only 2 isolates were studied, there is no denominator, therefore the sentence "the B.pertussis strains circulating in our Pediatric emergency department seem to differ genetically from those..." is not supported by the findings in this study.

When describing vaccination compliance (which would be better placed later in the discussion), it would be important to include a description of the vaccine recommendations in Italy and how many of the children in the study were eligible vs. not up to date.

Third paragraph - third sentence describing pathogenesis mechanisms is unnecessary.

Page 10 - second paragraph - first sentence - lymphocytosis is not a confirmatory finding of pertussis. Also, the second sentence explaining pathogenesis is unnecessary.

Page 11 - top paragraph, last sentence - consider "..because cultures were negative in 13 infants with PCR confirmed pertussis who were already receiving specific antibiotic...."

Second paragraph - last sentence - is speculation.

page 12 - so, was nasal or nasopharyngeal washing done to establish a
diagnosis?
Entire document: Check spelling and grammar throughout.
Tables 1 and 2 - some columns are shifted, please correct.

Food for thought: If the author’s suggestion (not supported by this study) that the changes in the genetic make up of circulating strains results in diminished response/effectiveness of the available pertussis vaccines is correct, how will maternal immunization with these same vaccines result in better infant protection?

**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'