Author’s response to reviews

Title: Candidiasis caused by Candida kefyr in a neonate.

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Author’s response to reviews: see over
To the Editors

BMC Infectious Diseases
Journal Editorial Office BioMed Central

21st of December 2011

Manuscript ID 1833111555933031 entitled „Systemic candidiasis caused by Candida kefyr in a neonate.“

Dear Editors,

Thank you and the reviewers very much for the constructive and helpful comments and your encouragement to resubmit a revised version of our manuscript “Systemic candidiasis caused by Candida kefyr in a neonate”. Please find below a detailed list of the changes introduced and our response, point by point, to the comments raised by the reviewers. By following all reviewers’ recommendations we could significantly improve the manuscript. We hope that the revised manuscript will meet all points addressed by the reviewers and will now be acceptable for publication in BMC Infectious Diseases.

Sincerely,

Stefan Weichert, MD.
Reviewer 1  
**Reviewer's report**  
**Title:** Systemic candidiasis caused by Candida kefyr in a neonate.  
**Version:** 2  
**Date:** 15 November 2011  
**Reviewer:** Cornelia Lass-Floerl

Reviewer's report:  
Minor Essential Revisions

General comments

**Comment 1.1:** I am not sure whether this case represents a real "systemic candidiasis", as Candida was never isolated from specimens other than urine.

**Comment 1.2:** From the data shown, blood cultures remained sterile? A fact, which should be addressed in the discussion section.

Although our patient had recurrent infections due to *Candida kefyr* and had clinical symptoms of systemic disease the pathogen *Candida kefyr* was only isolated from urine cultures and not from blood cultures or other sites. Two blood cultures in total had been taken and remained sterile. Without question, we have no direct microbiological proof of a systemic infection, such as with positive blood culture results for *Candida kefyr*.

On the other hand amongst clinical signs for systemic disease candiduria may be the only indication for candidaemia and therefore systemic infection. Studies confirmed that blood cultures are 40-75% false negative in patients with candidiasis, as demonstrated in patients with autopsy proven candidiasis [1, 2]. In addition to clinical signs of systemic disease, our patient had renal involvement as well, such as parenchymal changes on ultrasound. As an ascending infection would be expected to result in isolated pelvicalyceal disease and haematogenous spread is the most common route for renal candidiasis [3] we propose that our patient had transient candidaemia, which may have led to renal infection. Nevertheless, it is known that blood cultures are often no longer positive when renal candidiasis becomes manifest [4]. We therefore termed the infection with *Candida kefyr* in our patient “systemic candidiasis”. As a real systemic candidiasis or candidaemia is based on positive blood culture results we propose the following changes to our manuscript’s title: “Candidiasis caused by *Candida kefyr* in a neonate”. As candiduria is regarded as a risk factor for invasive candidiasis [5] clinicians should be aware of this, even though blood cultures might remain negative. We completed our revised manuscript, including the explanations and thoughts mentioned above.


Specific comments

Comment 1.3: How often did you perform in vitro susceptibility testing? With how many strains?

Susceptibility testing against fluconazole, amphotericin B, and caspofungin was performed by ellipsometer test (E-test) and revealed minimal inhibitory concentrations (MIC) of 0.25 µg, 0.047 µg, and 0.25 µg, respectively. All tests were repeated two times with similar results. The inoculum for susceptibility testing was generally performed by pooling of 10-20 individual colonies. No macrolonies were observed in the inhibition zone of the E-test. Further subplating and antibiotic susceptibility testing of individual colonies in order to detect antibiotic susceptibility variants was not performed. After having started with liposomal amphotericin B instillation and systemic fluconazole therapy Candida kefyr was isolated for the last time after 7 days of treatment. We did not repeat susceptibility testing from this isolate, and therefore have no data on possible MIC changes under therapy. Afterwards, all tested urine cultures remained sterile. We completed our revised manuscript with the more detailed E-test methodology and included the last positive urine culture for Candida kefyr under antifungal therapy.
Comment 1.4: How many blood cultures have been taken?

Overall, 2 blood cultures have been taken and yielded no growth. We have included the changes in our revised manuscript.

Reviewer 2

Reviewer's report
Title: Systemic candidiasis caused by Candida kefyr in a neonate.
Version: 2 Date: 27 November 2011
Reviewer: Jorge Garbino

Reviewer's report:

Comments

Comment 2.1: Reference number 5 is not cited in the manuscript.

Numeration of the cited literature has been completed and changed. The former reference number [6] has been changed correctly to reference number [5].

“…This biochemical result was confirmed by sequencing of the internal transcribed spacer (ITS) regions using primer pairs ITS 1 and ITS-4 (ITS1: 5-TCCGTAGGTGAACCTGCGG-3, ITS4: 5-TCCTCCGCTTATTGATATGC-3 and V9D:5-TTAAGTCCCTGCCCTTTTGA-3 and LS266:5-GCATTTCCAAACAAACTCGACTC-3, respectively) [5]. …”


Comment 2.2: Reference 6 in page 5 seems not be related.

Numeration of the cited literature has been completed and changed. The former reference number [6] in page 5 has been replaced by reference number [5] and correctly completed in page 6.
“… So far, good susceptibilities of AMB against most non-*albicans Candida* species were shown, although country specific differences were observed [4, 6, 7]…”


Comment 2.3: Reference 14 not yet published?

Reference 14 is an advance online publication in *Bone Marrow Transplantation*, 30 May 2011; doi:10.1038/bmt.2011.112.


Comment 2.4: Which was the reason to start antifungal treatment with amphotericin B in a patient who had previous renal alterations?

Fluconazole is the antifungal drug with the highest active (concentrations) in the urinary tract without nephrotoxicity.

After having received the first information about fungal growth in the urine culture, the responsible microbiologist had a preliminary suspicion of a non-*albicans Candida* species. The *Candida* species was morphological resembling *Candida krusei* at first, but later turned out to be *Candida kefyr*. We therefore started our patient on liposomal amphotericin B. Considering our patient’s renal alterations we started with the less nephrotoxic liposomal amphotericin B, although the liposomal formulation might reach lesser renal concentrations compared to the non-liposomal formulation [1]. Despite of this, our patient’s kidney function deteriorated over time, which may be either due to the candidiasis, the amphotericin B treatment and/or further damage caused by the grade V reflux. We then changed our treatment to fluconazole, to which the isolated *Candida kefyr* was fully sensitive according to the resistogramm. Thereafter, our patient’s kidney function recovered completely. We completed
our revised manuscript, including the explanations and thoughts mentioned, and optimized the order of the presented data for better understanding of the decision making processes.