Risk factors and epidemiological features of Candida septicemia in neonatal unites at King Hussein Medical Center.

Aghadir Alhadidi MD, Faten Alawaisheh MD*, Abdallah Almherat MD*, Mahdi Frehat MD*, Mohammad Almayta MD**, Yara AkhayatMD**

ABSTRACT

Objective: To review the epidemiology of candidemia in neonatal intensive care unit (NICU), identify the risk factors associated with the development of the infection and suggest a rational approach to starting empirical antifungal treatment in neonate at risk of developing candida infection.

Methods: In this retrospective epidemiologic multicentre pilot study three neonatal intensive care units were included, where all the positive blood cultures for candida in neonates were recorded during 2016 and 2017. Data of Candida species and antifungal drugs susceptibility were collected. The medical records of those neonates were studied and the following information were taken; gestational age, birth weight, gender, age at onset of infection, previous use of antibiotics and steroid drugs with the duration of their use before candida infection, endotracheal intubation, presence of necrotizing enterocolitis, and the use of total parental nutrition.

Results: 18 neonates developed candida infection in the blood during that period with incidence of 0.5% of the total admission. Male to female ratio was 3:1. Most of the neonates were premature <32 weeks (p value 0.015) and very low birth weight<1500 gm (p value 0.015).Non candida albicans was the most common cause(66.6%). Where candida parapsilosis was the most common non albicans (27.7%) and all the candida species were sensitive to most commonly used antifungal drugs. Central line presence (p value 0.01) value, endotracheal tube>1 week (p value 0.01) and prolonged use of antibiotics>1 week (p value 0.002) were the most common associated risk factors. Mortality rate 16.6%

Conclusion: Candida infection is a serious infection in the neonates with increased incidence of non albicans candida species with no emerge of resistant species.

Key words: Candida, Candidemia, Neonate.

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Introduction

Candida infection is a serious infection in neonates with high rate of mortality and is increasing at an alarming rate [(1)]. It is the third most common cause of late onset sepsis in NICU patients [(2)]. Systemic Candida infection is associated with increased short and long term morbidity in extremely low birth weight (ELBW) infants [(2-4)]. It is mainly occurs in neonates <1000gm and <28 weeks, in the babies with gastrointestinal anomalies and in patients who need central line for prolonged period [(1)]. There is huge difference in the incidence of invasive candida infection in neonates reported by different studies 15-cases per 10000 neonatal admission [(5)] and a recent large study done in united states of America found the incidence to be 10.7 to 11.8 cases/100000 births between 2012 and 2015 [(6)] , and 3-23% [(7-9)]in infants less than

From Department of:
* Pediatric, King Hussein Medical Center.
** Microbiologist , princesses Iman for laboratory research.
Correspondence should be addressed to Dr: Aghadir Alhadidi. Email: aghadiralhadidi@yahoo.com
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1000 g, and this depends mainly on whether the hospital gives support to extreme premature infants 22-25 weeks or uses fluconazole as a prophylaxis in the babies less than 1500-1000 gm or less than 32-28 weeks [10,11]. But in general there is an increase of the incidence of candida infection with the increase in the antifungal drugs resistant with limitation of the drugs that can be used [12]. Although the incidence of candida infection is less than bacterial infection but it associated with high mortality rate11.9- 28% in infants less than 1000 g(5,8) and increase in the hospital stay mainly in neonates weight more than 1000 gm.[5] Prophylactic fluconazole significantly reduces the incidence of colonization and systemic infection by Candida species in both ELBW and very low birth weight (VLBW) neonates and decreases the rates of progression from initial colonization to massive colonization and to systemic infection. All VLBW neonates may benefit from fluconazole prophylaxis. [13] More than 90% of invasive infections are caused by Candida albicans, Candida glabrata, Candida parapsilosis, Candida tropicalis and Candida krusei [14] There is change in the most common candida species that cause candidemia in neonate in the recent years and different institutions report different species and this affect the survival. [15] Most common species that cause infection are candida albicans and candida parapsilosisin neonates where no antifungal prophylaxis is used [8]. Candida albicans and parapsilosis are normal flora of human mucosal oral cavity, gastrointestinal tract and vagina. Oral colonization of candida in neonates is associated with the development of invasive candidiasis mainly with candida albicansin NICU [16]. Candida albicans is the most virulent one and associated with higher mortality rate than others candida non-albicans [17] but candida parapsilosis is associated with late recurrent candidaemia [18] .In our research we want to find the incidence of candida infection over a period of two years in King Hussein medical center and find out the spectrum of candida species and its susceptibility which will guide us in choosing the empirical treatment when suspected candida infection and quantify the burden of candida infection in our institution. Recognizing the risk factors for candida infection is another goal of the study in order to more easily recognize patients at risk of candida infection so antifungal infection can be started early and this will improve the outcome. And support which neonates would benefit from prophylactic antifungal.

Methods

In this retrospective epidemiologic multi centre pilot study three neonatal intensive care units were included (neonatal intensive care unit in King Hussein medical hospital, neonatal intensive care unit and surgical unit in Queen Rania hospital for children and neonatal cardiac care unit in Queen Alia hospital for cardiology all of these hospitals are found in King Hussein medical center) A review of the microbiology records in Prince Iman center for laboratory research was done and all positive blood cultures for candida in neonates were recorded during 2016 and 2017.Data about Candida species and antifungal susceptibility were collected. The medical records of those neonates were studied and the following information were taken gestational age, birth weight, gender, age at onset of infection, types of antibiotics and steroid drugs with the duration of their use before candida infection, endotracheal intubation, presence of necrotizing enterocolitis, and the use of total parental nutrition. All blood samples were bedside collected using aseptic technique “the skin was disinfected with 70% isopropyl alcohol, followed by 2% iodine tincture”, after Blood culture bottles were injected with the drawn blood they were sent to microbiology department to be incubated in one of three different detection systems (Versa Trek, Bactec and Bact alert) according to the bottle manufacturer up to five days. After the bottle was flagged as positive during this period it was examined by light microscope using gram stain technique. If yeast was observed it was subcultured on blood chocolate and Sabouraud agar then incubated for 24-48 hours at 37C. The yeast isolates obtained from cultures underwent identification using “vitek 2 YST ID card” with capability to identify 50 different targets of yeasts. And susceptibility testing using “Vitek 2 AST-YS07” by Vitek 2 compact system according

to CLSI recommendations. Yeast were tested for susceptibility against 6 different antibiotics "amphotericin B, micafungin, caspofungin, fluconazole, flucytosine and voriconazole" We used Mean and standard deviation to analyzed Continuous variables such as weight, gestational age, age in days and others clinical features were analysed using frequencies and percentages such as gender. P values are two-tailed (Social Sciences (SPSS) software 10 for Mac OS X; SPSS Inc.,Chicago, IL, USA). We consider p value to be statistically significant to be <0.05 Ethical committee approval was taken from the institute. A written consent from all the parents of the neonates was taken at admission.

Results

The total number of neonatal admissions to the three unit in 2016 and 2017 were (1745) and (1868) neonates respectively**.** The mean days of hospitalization was 6 days [(18)] Neonates developed candidemia during the study period with overall incidence of 0.5%.Male: female ratio was 3:1. Most of the patients who developed candidemia were premature less than 32 week of gestational age(66%) and less than 28 weeks 50%with median gestational age was 30±6 weeks and very low birth weight <1.5 kg (72%) with median birth weight 1250±960 gm. Full term infants who developed candidemia have congenital anomalies who needed central line for prolonged period like congenital heart disease post operation(3 neonates) and posterior urethral valve (one neonate). In this group of patients they developed the candida infection earlier than the premature one(median age of candidemia was 6 days, 15 days respectively) Presence of central line was the most significant risk factor to develop candidemia (94%) with median time of its presence is 12 days followed by the presence of endotracheal tube (77%) and prolonged use of antibiotics (55%) (Table I :clinical data and risk factors in neonates with candidemia).most common antibiotics were imipenem and vancomycin and the most common associate bacterial infection was klebseilla.Postnatal steroid use (p value 0.4)and necrotizing enterocolitis (p value 0.6) were not found as a significant factors in the development of candidemia. In Four of babies the microbiology laboratory reported the result of blood culture as candida sp without determine the subspecies. Non albicans candida is the most common cause 66.6%. Candida albican(33.3%) and candida parapsilosis (27.7%) were the most common cause of candidemia.(figure (1): the distribution of candida species in the 18 neonates.) We found that all the candida species were sensitive to most commonly used antifungal drugs (amphotericin B, micafungin, caspofungin, fluconazole, flucytosine and voriconazole), but we found a delay in starting antifungal empirical treatment which has an effect on the outcome and the mortality.(Table II: relationship of starting antifungal drugs with clearance rate and mortality.) Mortality rate was 16.6% and there was no difference in the mortality according to the candida species. But the most important risk factor was the gestational age<28 week (P value <0.05)

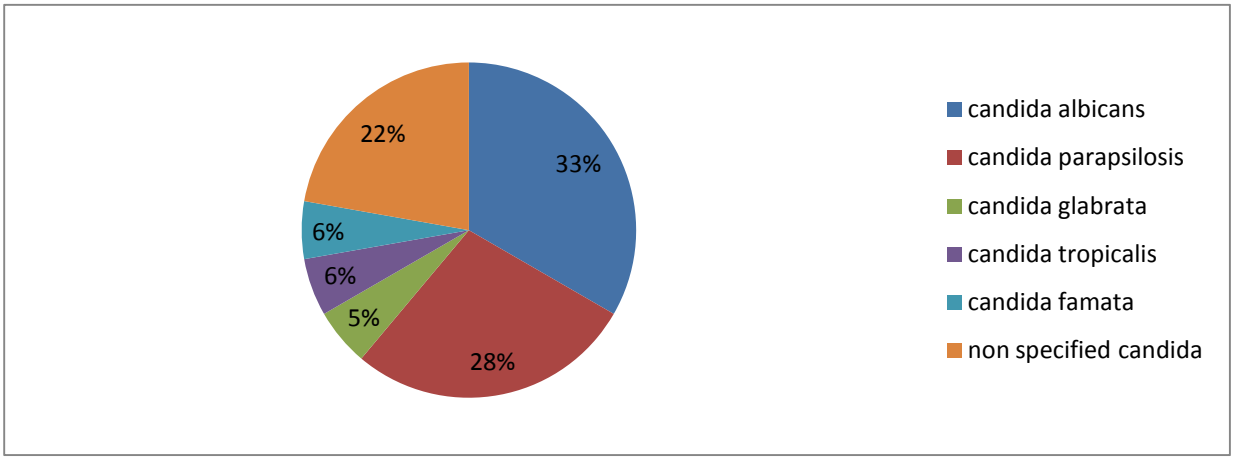
Table I: Clinical data and risk factors in neonates with candidemia

Risk factors	**Number of patients with candidiasis(no 18 pts)**	**No candidiasis (no :3595 pts)**	**P value**
Male/female	12/6 (66.7%:33.3%)	1865/1730 (51.8%:48.2%)	
Gestational age <32 week	12	719	0.015
Gestational age <28 weeks	9	175	0.001
Birth weight <1.5 kg	13	193	0.015
Prolonged use of antibiotics> 1 week	10	84	0.002
Central line >1 week	17	54	0.01
Steroid use postnatally	1	39	0.4
Mechanical ventilation> 1 week	14	94	0.01
Necrotizing enterocolitis	1	86	0.6
Use of TPN (total parental nutrition)	15	193	0.02
Age of developing candidemia>14 days	14	-	

Table II: Relationship of starting antifungal drugs with clearance rate and mortality.

Time of starting antifungal drugs from the blood culture	Number of patients	Negative blood culture after one week of starting treatment	Mortality rate
Within the first 24 hrs	11/18(61%)	10/11 (91%)	0%
After 48 hrs	7/18 (39%)	4/7(57%)	43%

Fig 1: The distribution of candida species in the 18 neonates.



Discussion

The incidence of candidemia in our study was 0.5 %which is considered a low incidence comparing to the other studies reported from Asia 1.1-15.7% [19-22] and Europe (1.1%) [23] and America (6.7%) [8] and (2.3%) Africa [24]although we don't use fluconazole prophylaxis in our unites [11]. This could be explained by the fact we don't resuscitate extreme premature babies less than 25 week as this group of premature babies have a high rate of candidema. In our study we found a high incidence of non-albicans candida (64%). With review of studies done in different countries around the world we notice a geographical variation between different region as most of the studies conducted in Asia showed an increase in the candida non-albicans with proportion range from 39-92% [19-22, 25-32]. Candida albicans still the most common species in Europe, North and South America [6, 33-48] and few studies from Africa [49] .Most of the studies show increase in the incidence of non-albicans candida over the recent years. A study was done in Jordan published in 2008 showed that the percentage of candida albicans 50% but in our study 35% (50)which could be explained by wide spread use of prophylactic antifungal drugs in the extremely low birth weight infants [36, 49] and most common non albicans candida is candida parapsilosis. [21, 37, 36, 49,51] as we found in our study. Others found candida krusei as the most common [22,52], and candida glabrata in other [33].This change in the epidemiology of candida infection will affect the survival rate as candida albicans is the most virulent species of candida as it is presented in two form unicellular yeast form and filamentous hyphal form and can switch between the two form and this increases it's resistance. [53] but in the same time the emerge of resistance in the non albicans candida still an important issue to be studied [52,54]Fluconazole resistance was seen in 30% cases, mostly among non- albicans Candida which warrants its judicious use as a prophylactic agent in hospitals. [55] Another study showed that the resistance and dose-dependent susceptibility rates against fluconazole were 4.2% and 2.1%, respectively. No resistance to amphotericin B and echinocandin was identified. [56] Among the non albican C tropicalis and parapsilosis is sensitive to azole agents but c glabrata and c krusie are more resistant to antifungal agents.[11,52] our finding shows no resistance to antifungal drugs of all candida species [35,36]. fluconazole and amphotericin are the first line of empirical treatment of candidemia in our unit. [51] .This finding may

explain that there is no effect of changing in the epidemiology of candida infection on the survival shown by our result very low birth weight infants (VLBW) and premature babies <32 weeks were the most common group to developed candidemia as those infants have a longer duration of hospitalization [29,36,58] and most of them exposed to endotracheal intubation [29,58], central line placement and the delay of administration of milk with the use of the total parental nutrition [2,21,36,58] . All of these modalities of treatment increase the risk of the formation of the biofilm. the use of amino acids potentiate the proliferation and differentiation of the candida species [55] but we didn't find an association between the use of total parental nutrition and the non albicansspp The study of the risk factors associated with the development of candidemiais very important in improving the outcome with early suspension. A history of third-generation cephalosporin or carbapenem exposure in the 7 days before the blood culture, then the physician should consider administration of empirical antifungal therapy [21,59].Center incidence of candidiasis correlates with average broad-spectrum antibiotics use per infant and average use of broad-spectrum antibiotics with negative cultures per infant [9].Our policy of empirical antibiotics we started with ampicillin or penicillin and aminoglycoside for early onset sepsis and vancomycin with imipenem for late onset sepsis and reserve third generation cephalosporin for meningitis. Stopping the antibiotics after 48 hours of its start if no growth in the blood culture,may also has effect on our low incidence of candidemia. Gram negative sepsis is associated with increased incidence of candidemia,in our study klebsiella sepsis was associated with the development of candida infection. But we couldn't identify if this was due to infection itself or prolonged use of antibiotics due to small number of the cases.Others connect clostridium difficile to the development of candidemia [60] The presence of central line catheter is a major risk factor for developing nosocomial candida infection [22.58,61] as there is a high tendency of the candida to form antibiotic resistant biofilm(a highly organized biological community imbedded in an extracellular matrix) mainly to amphotericin b and fluconazole. [62-64] candida albicans has more tendency to perform biofilm than other species [64] and this explain the high incidence of candidemia in extremely premature infants who need central line for prolonged period.The pervious use of steroid has been shown to be a significant risk factor in the development of candidemia in neonates by many studies [58] but we didn't find the same result in our study and this could be explained by the restrict use of postnatal steroid policy in our center.

Mortality rate in a large study in USA involved 730 neonate from 192 Neonatal intensive care units who have candida separated from the blood, urine, CSF was 19%. [58] other studies showed a higher rate of mortality(26%-35.7) [21, 35, 65] In our study the mortality rate was 16.6% and we found a strong relation with the time of starting antifungal treatment from the development of early symptoms and sign or waiting until 48 hours after blood culture extraction shows a candida growth. In our study we didn't find statistically significant difference in the mortality between patients infected with C parapsilosis and those infected with C albicans same finding was shown by other studies [56] others showed a higher mortality with candida albicans [66]as C parapsilosis is mainly acquired from the hands of health care worker [58] this highlight the importance of hand wash in decreasing its incidence. There is no difference between candida species in the persistent positive blood culture one week after starting treatment [67].

Conclusion: Candida infection is a serious infection in the neonates with increased incidence of non albicans candida species with no emerge of resistant species. Prematurity, low birth weight<1.5kg, prolonged use of antibiotics, mechanical ventilation and prolonged use of central line are significant risk factors.

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