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Neonatal invasive candidiasis in low- and middle-income countries: Data from the NeoOBS study

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Abstract

Neonatal invasive candidiasis (NIC) has significant morbidity and mortality. Reports have shown a different profile of those neonates affected with NIC and of fluconazole-resistant *Candida* spp. isolates in low- and middle-income countries (LMICs) compared to high-income countries (HICs). We describe the epidemiology, *Candida* spp. distribution, treatment, and outcomes of neonates with NIC from LMICs enrolled in a global, prospective, longitudinal, observational cohort study (NeoOBS) of hospitalized infants <60 days postnatal age with sepsis (August 2018–February 2021). A total of 127 neonates from 14 hospitals in 8 countries with *Candida* spp. isolated from blood culture were included. Median gestational age of affected neonates was 30 weeks (IQR: 28–34), and median birth weight was 1270 gr (interquartile range [IQR]: 990–1692). Only a minority had high-risk criteria, such as being born <28 weeks, 19% (24/127), or birth weight <1000 gr, 27% (34/127). The most common *Candida* species were *C. albicans* ($n = 45$, 35%), *C. parapsilosis* ($n = 38$, 30%), and *Candida auris* ($n = 18$, 14%). The majority of *C. albicans* isolates were fluconazole susceptible, whereas 59% of *C. parapsilosis* isolates were fluconazole-resistant. Amphotericin B was the most common antifungal used [74% (78/105)], followed by fluconazole [22% (23/105)]. Death by day 28 post-enrollment was 22% (28/127). To our knowledge, this is the largest multi-country cohort of NIC in LMICs. Most of the neonates would not have been considered at high risk for NIC in HICs. A substantial proportion of isolates was resistant to first choice fluconazole. Understanding the burden of NIC in LMIC is essential to guide future research and treatment guidelines.

Lay Summary

Our study describes neonates from low- and middle-income countries with neonatal invasive candidiasis (NIC). Most of them were outside the groups considered at high risk for NIC described in high-income countries. *Candida* spp. epidemiology was also different. The mortality was high (22%). Further research in these settings is required.

Keywords: neonatal candidemia, low- and middle-income countries, *Candida parapsilosis*, candidiasis, *Candida auris*

Introduction

The World Health Organization (WHO) estimates that 2.4 million children died globally in the first month of life in 2019, with infection being the third commonest cause of death following prematurity- and intrapartum-related complications.[1] The contribution of infection to deaths in the neonatal period is often underappreciated and varies according to geographic location, neonatal characteristics, and whether or not neonates are born in a medical facility.[2–4]

Neonatal invasive fungal infections are mostly caused by *Candida* spp. Reported rates of neonatal invasive candidiasis (NIC) vary significantly globally[5] and are associated with a high crude mortality rate, ranging from 12% to 37% in high-income countries (HICs) and from 8.9% to 75% in low- and middle-income countries (LMICs).[6] In HICs, NIC is most commonly reported in neonates <1000 gr birth weight or <28 weeks gestational age, but recent reports from LMIC neonatal units show the occurrence of NIC outside these specific groups.[3,7,8] Although antifungal-resistant *Candida* spp. infections remain uncommon in HICs,[9,10] LMICs are reporting an increasing proportion of fluconazole-resistant isolates, including C. *parapsilosis*,[11,12] C. *krusei*, and C. *auris*.[13,14]

The aim of this NeoOBS invasive candidiasis sub-study was to describe the epidemiology, antifungal resistance patterns, antifungal treatment, and clinical outcomes of neonates with *Candida* spp. bloodstream infections in LMICs. Data were collected as part of the larger NeoOBS study (https://clinicaltrials.gov/ct2/show/NCT03721302).

Materials and methods

NeoOBS study population

NeoOBS, a global, prospective, longitudinal, observational cohort study of hospitalized infants <60 days postnatal age with sepsis, was conducted at 19 hospitals in 11 countries, between August 2018 and February 2020. Hospitals were a mix of tertiary and district hospitals in Bangladesh ($n = 1$), Brazil ($n = 2$), China ($n = 3$), Greece ($n = 1$), India ($n = 3$), Italy ($n = 1$), Kenya ($n = 1$), South Africa ($n = 3$), Thailand ($n = 2$), Uganda ($n = 1$), and Vietnam ($n = 1$).[15,16]

Infants could be enrolled in the study in two different ways (Fig. 1). The primary cohort of infants was enrolled with clinical sepsis meeting the diagnostic criteria of at least one clinical sign of sepsis plus one clinical or laboratory sign, with a blood culture taken prior to initiating new antimicrobial treatment (referred to as cohort 1). Up to 200 infants from each hospital were enrolled through this route. Infants were excluded if the clinical signs were subsequently deemed to be more likely related to a non-sepsis diagnosis, as determined by the treating clinician (Supplementary Table 1).

In addition, a secondary cohort of infants (referred to as cohort 2) was enrolled based on the isolation of a carbapenem-resistant organism or *Candida* spp. from blood culture or with confirmed bacterial meningitis (Supplementary Table 1). Cohort 2 was designed to better understand specific infections and capture infants with these infections who may not have been enrolled in or eligible for cohort 1. There was no minimum or maximum enrollment number for cohort 2 across hospitals. Infants already enrolled in cohort 1 were not eligible to be enrolled in cohort 2 as all microbiology findings were already captured as part of cohort 1 follow-up.

Exclusion criteria for both cohorts were significant non-infectious-related comorbidity expected to cause death within 72 h, enrollment in an interventional study or previous enrollment in this study. Hospitals were given pragmatic flexibility for enrollment time frames given variability in case numbers and staffing capacity. Full inclusion and exclusion criteria for both cohorts are described in Supplementary Table 1.

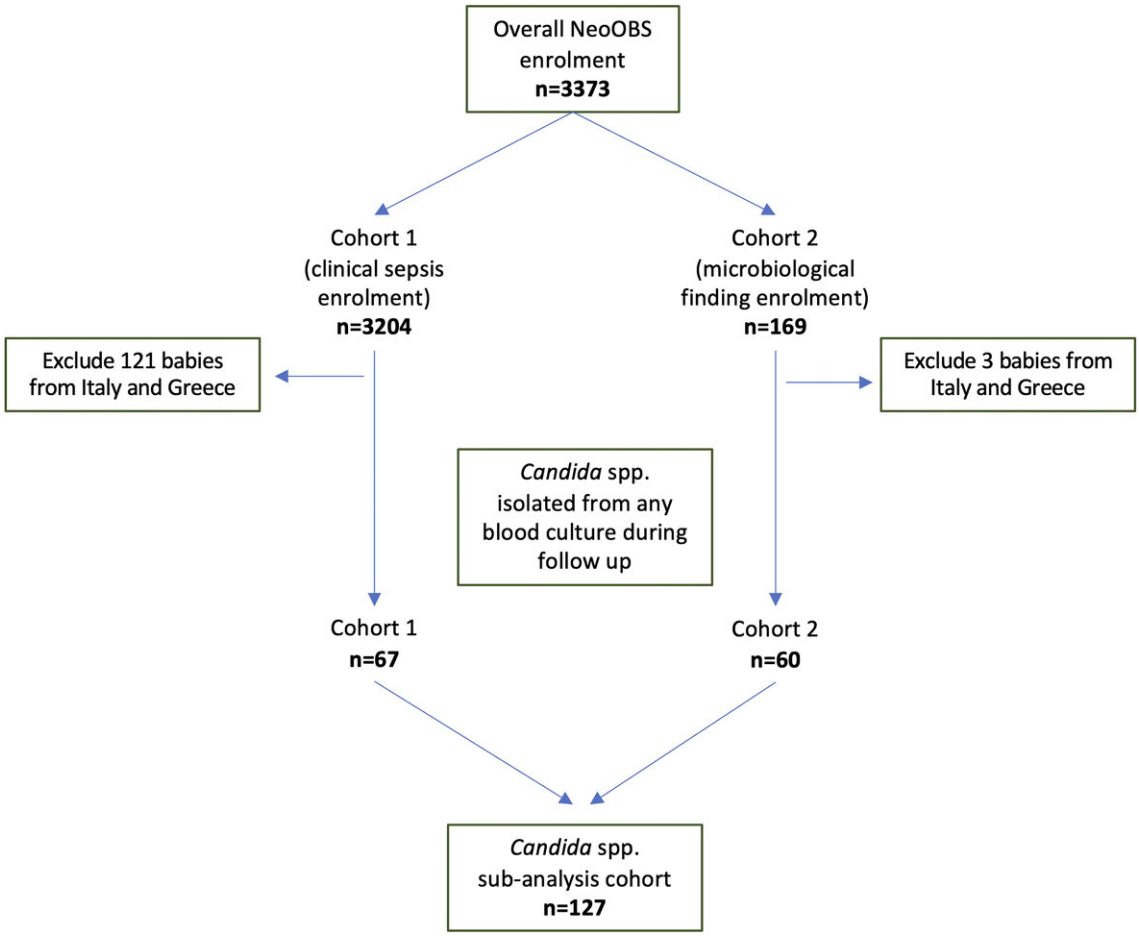

Figure 1. *Candida* spp. sub-study population derived from the overall NeoOBS study. See Supplementary Figure 1 for detailed schematic of the study population indicating the two enrollment cohorts. Note: Overall NeoOBS enrollment: cohort 1 was 3204 babies from 19 hospitals in 11 countries; cohort 2 was 169 babies from 14 hospitals in 10 countries. *Candida* sub-analysis cohort: cohort 1 includes 67 babies from 12 hospitals in 7 countries; cohort 2 was 60 babies from 12 hospitals in 7 countries.

Data collection

Infants meeting the eligibility criteria were enrolled in both cohorts. Infants in cohort 1 were followed prospectively daily for the duration of hospitalization up to day 28 from the day of enrollment.

For infants in cohort 2, daily clinical and antimicrobial treatment data and any laboratory or microbiological investigations were retrospectively collected using medical notes and other available data from the day the culture was taken up to the day of enrollment, and then, prospectively collected from the day of enrollment (Supplementary Fig. 1) to 28 days from when the eligible blood culture was taken. For babies in both cohorts, clinical signs, supportive measures, and antimicrobial treatment were collected daily from the day of enrollment; blood culture, routine laboratory investigations, and other microbiology results were collected as and when conducted. At enrollment, demographics, labor and delivery details, and risk factors were collected.

All treatments and investigations were at the discretion of clinicians at the local hospital and were not determined by the study processes. At discharge or in-hospital death, information on mortality (if applicable), antimicrobial treatment, and both infection- and non-infection-related diagnoses were collected. Infants who were discharged prior to day 28 were telephoned on day 28, to assess vital status and any medical interventions since discharge. The primary outcome of the study was death by day 28, from the day the enrollment blood culture was taken. Primary and secondary causes (if applicable) of death were captured both for infants who died in hospital and those who died post-discharge before day 28.

Microbiological examinations were conducted as per local hospital procedures; however, babies must have had a blood culture taken prior to new antimicrobials being started to be eligible for enrollment in cohort 1. Blood culture results were collected as reported by local hospitals.

Study data were collected by paper case report form and entered and managed using REDCap electronic data capture tools hosted at St. George's, University of London. REDCap is a secure, web-based software platform[17,18] used for the collection and management of research data.

Ethics approval from local and national bodies was received by each hospital prior to commencing recruitment. Informed consent was obtained for all patients prior to enrollment.

Candidemia study population

All infants enrolled in the NeoOBS study via cohort 1 or cohort 2 who had *Candida* spp. isolated from a blood culture at any point during their follow-up up to day 28 (regardless of enrollment diagnosis) were included in this analysis. Analyses

Table 1. Comparison of summary characteristics of neonates with candidemia by survival status.

	Overall ($n = 127$)	Survived ($n = 99$)	Died ($n = 28$)
Sex; Female (%)	59 (47)	44 (44)	15 (54)
Birth weight (gr) (median [IQR])	1270.0[990.0, 1692.5]	1300.00[1022.5, 1724.5]	955.00[772.0, 1655.0]
Gestational age (weeks) (median [IQR])	30 [28, 34]	30 [28, 34]	29 [27, 33]
Age at *Candida* spp. culture (days) (median [IQR])	16 [10.5, 22.0]	16.0 [12.0, 22.0]	14.5 [8.0, 22.5]
Birth status Hospitalized since birth (%)	114 (90)	90 (91)	24 (86)
Organism ($n = 128$) (%)			
Candida albicans	45 (35)	35 (35)	10 (36)
Candida parapsilosis	38 (30)	31 (31)	7 (25)
Candida auris	18 (14)	13 (13)	5 (18)
Other *Candida* spp.[a]	27 (21)	21 (21)	6 (21)
Country (%)			
India	40 (32)	30 (30)	10 (36)
South Africa	55 (43)	44 (44)	11 (39)
Vietnam	13 (10)	10 (10)	3 (11)
Other[b]	19 (15)	15 (15)	4 (14)
Hospital (%)			
Hospital 1	28 (22)	22 (22)	6 (21)
Hospital 2	25 (20)	20 (20)	5 (18)
Hospital 3	21 (17)	17 (17)	4 (14)
Hospital 4	13 (10)	10 (10)	3 (11)
Hospital 5	11 (9)	8 (8)	3 (11)
Other[c]	29 (23)	22 (22)	7 (25)

[a]Other *Candida* spp. include *C. famata* ($n = 1$), *C. glabrata* ($n = 6$), *C. metapsilosis* ($n = 1$), *C. pelliculosa* ($n = 4$), *C. rugosa* ($n = 1$), undefined *Candida* spp. ($n = 10$), and *C. tropicalis* ($n = 4$).
[b]Other countries are comprised of five countries, each contributing <8 participants (range: 1–7 per country).
[c]Other sites are comprised of nine hospitals, each contributing <7 participants (range: 1–6 per site).

were restricted to the first *Candida* spp. isolated from each patient. Regardless of when during follow-up the *Candida* spp. was taken, all infants were censored at 28 days from when the enrollment blood culture for the overall NeoOBS study was taken (see Supplementary Fig. 1) This means some infants may have contributed fewer than 28 days of follow-up from when first *Candida* spp. culture was taken to this candidemia sub-analysis.

Due to differing times in follow-up when *Candida* spp. cultures were taken for these patients, for this sub-analysis, all patients were aligned with day 0 defined as the day the positive *Candida* spp. blood culture was taken (Supplementary Fig. 1). All data collection tools were the same for infants in both cohorts. Analyses were restricted to infants from LMICs only, thus excluding infants from Greece ($n = 3$) and Italy ($n = 0$).

Statistical analysis

Categorical variables were described as relative frequency, and continuous variables were described as median and interquartile range (IQR). Demographic and clinical characteristics between enrollment cohorts were compared using the χ^2 test. Kaplan–Meier curves and Cox proportional hazards model were used to investigate mortality. All data management and analyses were conducted in RStudio v1.4.1717 (R version 4.0.3).

Results

Study population and baseline characteristics

After excluding infants from Greece and Italy, 3083 neonates were enrolled from 17 hospitals in cohort 1, and 166 neonates were enrolled from 14 hospitals in cohort 2. Results from the overall NeoOBS study are described elsewhere.[19] Overall, 127/3249 (4%) infants met the inclusion criteria for the candidemia sub-analysis (67 were from cohort 1 and 60 from cohort 2) (Fig. 1). Infants with candidemia were from 14 hospitals in eight LMICs; however, 85% (108/127) of the infants with candidemia were reported from seven hospitals in three countries (South Africa, India, and Vietnam). Forty-six percent (58/127) of infants had *Candida* spp. isolated during follow-up after enrollment in the overall NeoOBS study, while the remaining 54% (69/127) had *Candida* spp. isolated from their baseline enrollment blood culture.

At the time when the blood culture that grew *Candida* spp. was taken, the median postnatal age was 16 days (IQR: 10.5–21), the median gestational age at birth was 30 weeks (IQR: 28–34), and the median birth weight was 1270 gr (IQR: 990–1692). Fifty-four percent (68/127) of the infants were male. Only 19% (24/127) of the infants were born before 28 weeks of gestation, and 27% (34/127) had birth weights <1000 gr. Infants with candidemia were hospitalized for a median of 14 days (IQR: 6.5–20) prior to when the *Candida* spp. blood culture was taken. Eighty percent (102/127) of infants received at least one broad-spectrum antibiotic in the week prior to that blood culture. The majority of cases, 90% (114/127), were born either at the enrolling hospital or in a referral hospital and remained hospitalized from birth. Baseline characteristics of both enrollment cohorts were similar (Supplementary Table 2). Epidemiological characteristics by survival are summarized in Table 1.

Microbiology findings

The most common *Candida* species in this study were *C. albicans* ($n = 45$, 35%), *C. parapsilosis* ($n = 38$, 30%), and *C. auris* ($n = 18$, 14%). Other species isolated were *C. glabrata*

Figure 2. Reported susceptibility profiles to amphotericin B, fluconazole, and micafungin for the most common *Candida* species.

($n = 6$), *C. pelliculosa* ($n = 4$), *C. tropicalis* ($n = 4$), *C. famata* ($n = 1$), *C. metapsilosis* ($n = 1$), and *C. rugosa* ($n = 1$). There were 10 (8%) unspecified *Candida* spp. (Table 1). Species distribution varied by hospital ($P < .0001$) and country ($P < .0001$) (Supplementary Table 3). One patient had two *Candida* spp. isolates (*C. parapsilosis* and *C. glabrata*) from the same blood culture (a total of 128 *Candida* spp. isolates). Sixty-one percent (11/18) of *C. auris* isolates were found in India, and the remaining 39% (7/18) were from South Africa.

Susceptibility testing was not reported for 13% (16/128) of isolates, and for 17 isolates (13%), only fluconazole susceptibility was reported. Susceptibility results for fluconazole, amphotericin B, and an echinocandin [micafungin] were reported in 80% (103/128), 78% (87/128), and 36% (46/128) of all *Candida* spp. isolates, respectively (Fig. 2). Of these, overall, 41/103 (40%) were fluconazole-resistant, 16/87 (18%) were amphotericin B-resistant, and 0/46 (0%) were micafungin-resistant.

Of the *C. albicans* isolates with susceptibility results reported, 91% (38/42) were susceptible to fluconazole, and 100% (30/30) were susceptible to amphotericin B; however, 15/45 (33%) did not have amphotericin B susceptibility reported.

Reported resistance to fluconazole in *C. parapsilosis* was high (19/32 resistant, 59%); however, the majority were susceptible to amphotericin B (27/29 susceptible, 93%). Almost all the fluconazole-resistant *C. parapsilosis* isolates (17/19, 90%) were reported from South Africa.

There was an expected high reported resistance to fluconazole (15/17, 88%) and amphotericin B (11/13, 85%) in *C. auris* isolates. Most *C. auris* isolates did not have micafungin susceptibility testing done (16/18, 89%). Resistance of *C. auris* isolates to voriconazole was reported in 31% (5/16). Figure 2 illustrates the susceptibility profiles of *Candida* spp. isolates to the three commonly used antifungal agents.

Antifungal drug use

Overall, 14% (18/127) of infants received antifungals for either prophylaxis ($n = 8$) or empirical treatment ($n = 11$) in the week preceding the positive blood culture with *Candida* spp. being taken. Of those, all the infants who received prophylaxis received fluconazole. Of those receiving empirical treatment prior to the blood culture, amphotericin B was received by 73% (8/11) of infants.

Overall, neonatal antifungal prophylaxis was uncommon in the infants that developed candidemia (8/127, 6%). None (0/27) of those born before 28 weeks of gestation, and only 3/34 (9%) of neonates with a birth weight <1000 gr received prophylaxis.

Ninety percent of infants (114/127) received antifungal treatment after taking the blood culture that grew *Candida* spp., including eight infants who continued antifungal treatment that had been started before the culture (fluconazole: $n = 3$ and amphotericin B: $n = 5$). Of the 106, who started any new antifungals after the blood culture was taken, only

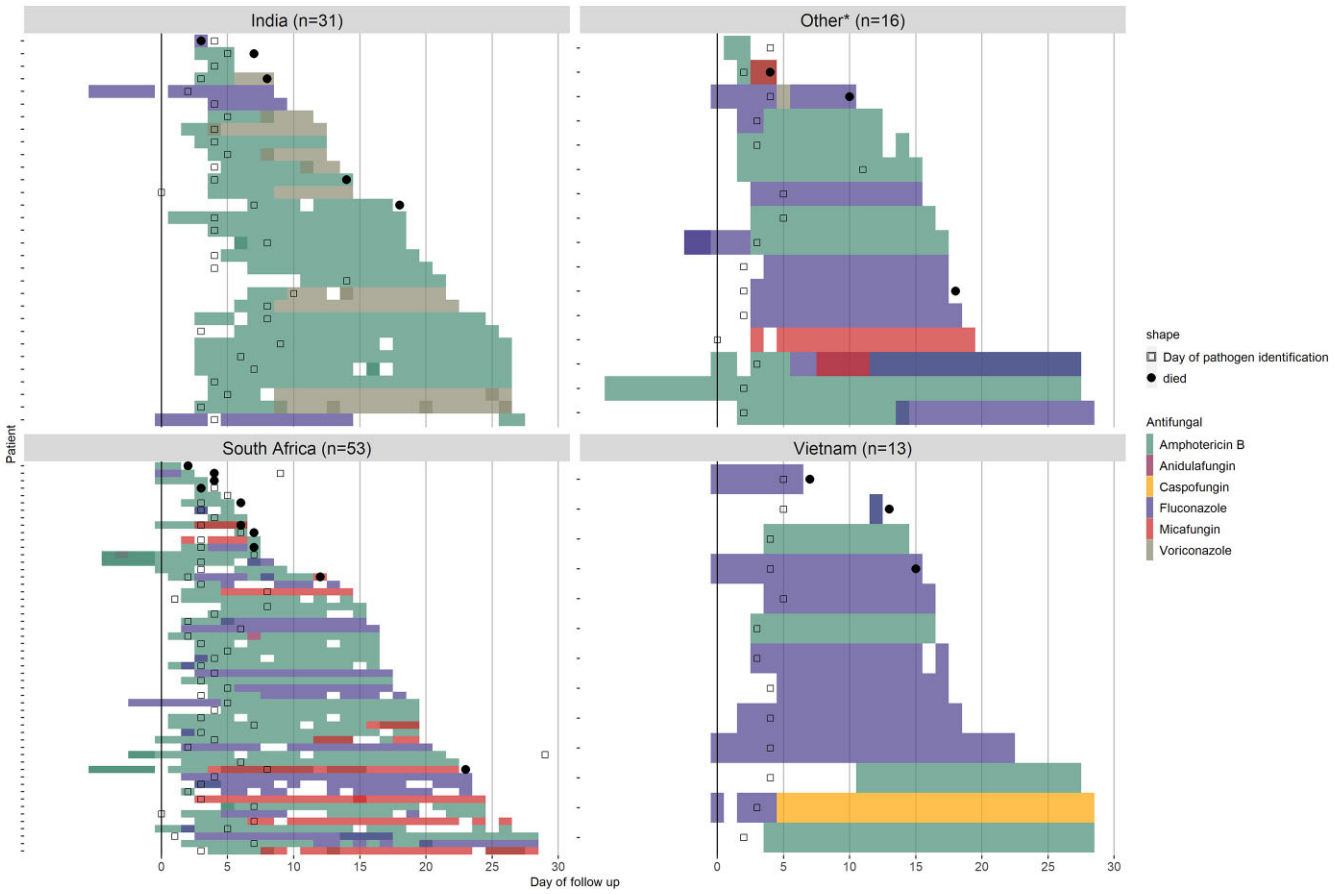

Figure 3. Patient-based antifungal treatment choice by country indicating the day the blood culture was taken (day 0), the day the fungal organism was identified (open squares), and mortality (solid dots). White space indicates calendar days that antifungal treatment was not given.
*Other countries are comprised of five countries, each contributing < 8 participants (range: 1–7 per country).

one had received fluconazole prophylaxis. After taking the blood culture, the median time to start a new antifungal treatment in these 105 infants was 3 days (IQR: 2–4). The first antifungal treatment of choice was amphotericin B in 74% (78/105) of the cases and fluconazole in 22% (23/105) of the cases.

Out of 88 infants, 85 (97%) received appropriate antifungal treatment based on the reported *in vitro* susceptibility profile of the *Candida* species with available susceptibility testing results. In these infants, amphotericin B ($n = 45$) was the most commonly prescribed antifungal, followed by fluconazole ($n = 27$), voriconazole ($n = 10$), and micafungin ($n = 3$). In infants with known susceptibility profile of the *Candida* species, the median time to appropriate antifungal treatment was 3 days from when the blood culture was taken (IQR: 2–5 days, range: 8 days prior to blood culture to 12 days after blood culture).

Antifungal treatment varied by country (Fig. 3) and causative *Candida* species (Fig. 4). Amphotericin B was used in all countries. In infants who received antifungal treatment after blood culture, a higher proportion of infants received amphotericin B in South Africa (48/53, 91%) and India (28/31, 90%) compared to Vietnam (5/13, 38%) (Fig. 3). Fluconazole was less commonly prescribed in India (4/31, 13%) and South Africa (23/53, 43%) compared to Vietnam (9/13, 69%). Voriconazole was used only in India ($n = 10$), micafungin was used predominantly in South Africa ($n = 10$), and caspofungin was used only in Vietnam ($n = 1$). Infants may have received more than one antifungal during their treatment.

Clinical outcome

Death by day 28 post-enrollment was 22% (28/127). Sixty-four percent (81/127) of infants were still in the hospital on day 28 post-enrollment. The median length of follow-up from the day the *Candida* spp. culture was taken was 21 days (IQR: 11.5–27). Among infants who died, the median length of follow-up was 7 days (IQR: 4–12.25). Unadjusted mortality by species is illustrated in Fig. 5. In aunivariable Cox proportional hazards analysis, mortality was strongly associated with birthweight <1000 gr (HR: 3.83; 95% CI: 1.84–7.97) and gestational age <28 weeks (HR: 2.32; 95% CI: 1.08–4.99). There was no significant difference in mortality by species, by hospital, by country, or by study cohort (Table 2).

Discussion

To the best of our knowledge, this is the largest multi-country cohort of neonates with *Candida* spp. bloodstream infections in the LMIC setting. Most of the neonates, included in the NeoOBS invasive candidiasis sub-study, were outside the high-risk groups for NIC as described in HIC, with 81% born after 28 weeks gestation and 73% with a birth weight >1000 gr. Although *C. albicans* was the most frequent species isolated, the species distribution varied significantly between hospitals and

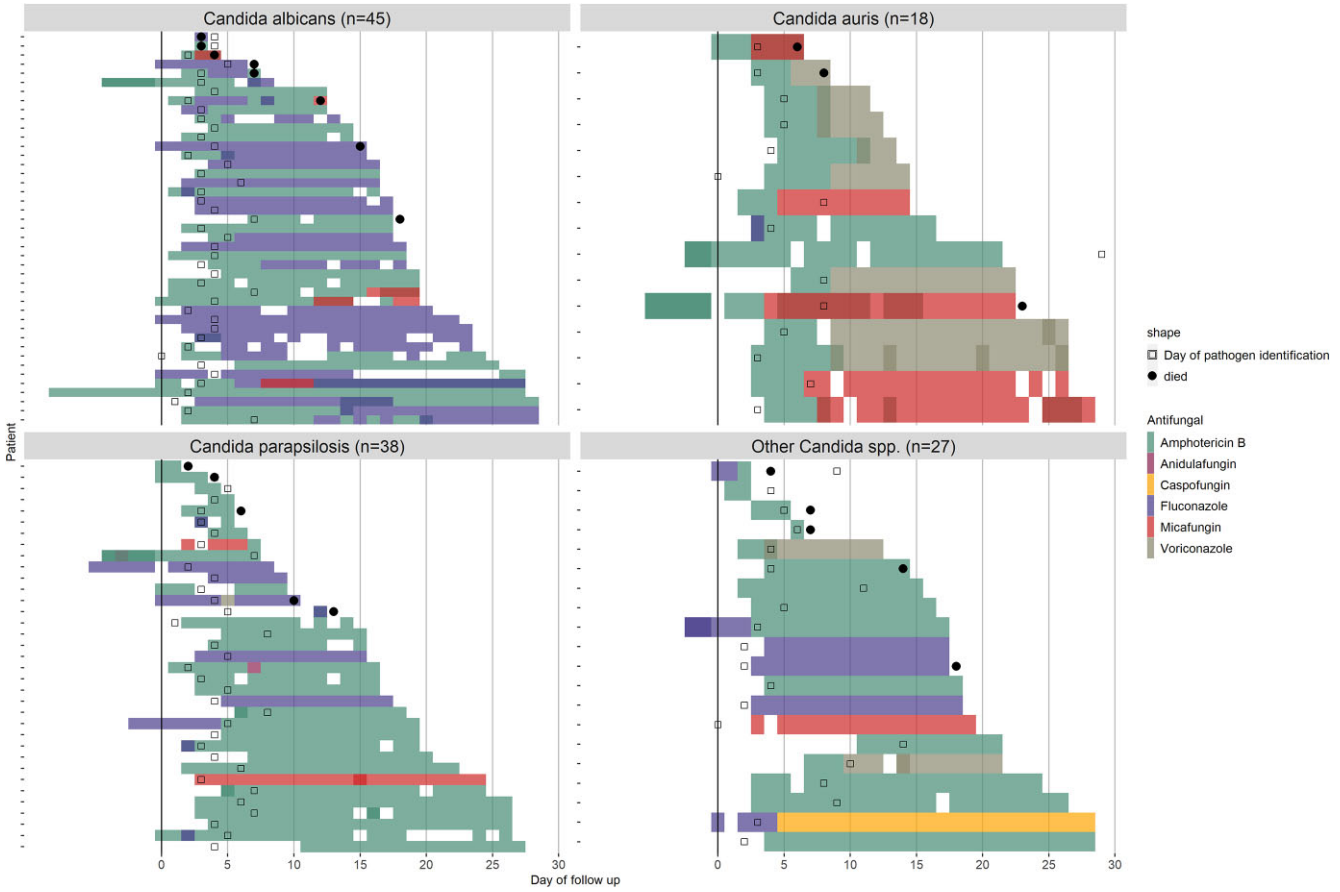

Figure 4. Patient-based antifungal treatment choice by causative *Candida* spp. Day 0 is the day the blood culture was taken that grew *Candida* spp. Day that the *Candida* spp. was identified is indicated with open squares and mortality with solid dots.

countries. Across the *Candida* spp. with known *in vitro* susceptibility profile, 40% were fluconazole-resistant, 18% were amphotericin B-resistant, while no resistance was reported for micafungin. (Albeit many isolates were not tested.) *Candida albicans* was reported to be highly susceptible to fluconazole and amphotericin B. In contrast, a significant proportion of *C. parapsilosis* was reported to be fluconazole-resistant, driven by high resistance rates observed in South Africa.[12] *Candida auris* was the third most common species overall, but its presence varied greatly between countries. Amphotericin B was the most common empiric antifungal used (74%), followed by fluconazole (22%) with echinocandins rarely used. Antifungal prophylaxis was infrequently used in infants who developed candidemia, even for those neonates considered at high-risk. Overall, the mortality was high (22%), and it was significantly associated with low birth weight (<1000 gr) and extreme prematurity (<28 weeks).

In the cohort of neonates with NIC described here, 37% weighed >1500 gr and 38% were ≥32 weeks of gestational age. These results are similar to other studies in LMIC,[11,20] and contrast dramatically with the data from HICs, where extreme prematurity and ELBW neonates are the main high-risk groups for NIC.[21] In 2018, e.g., the DeNIS study reported unusually high rates of NIC in a cohort of neonates in India born outside the hospital; more than a quarter of neonates with a positive blood culture (90/339, 26.5%) had *Candida* spp. isolated. Remarkably, 61.5% of those neonates weighed >1500 gr, and 73.3% were born at or after 32 weeks gestation.[3]

Candida albicans has been reported as the most common causative species in NIC.[10,22] Increasingly, a shift in epidemiology of NIC globally has been described, with a higher rate of non-*albicans Candida* isolates in LMICs compared to HICs.[6] The rise in non-*albicans Candida* spp. in NIC is associated with reduced susceptibility to fluconazole. This has been described in India, where MDR strains of *C. krusei* and *C. auris* have been reported[13]; and in South Africa,[23] where surveillance has shown an increase in the number of fluconazole-resistant *C. parapsilosis*.[12] For example, Govender et al. reported a significant shift toward *C. parapsilosis* in neonates, with 53% of all *C. parapsilosis* isolates being fluconazole-resistant and 44% and 70% cross-resistant to voriconazole and posaconazole, respectively.[12] Other South African series report similar results.[7,24,25] In the cohort presented here, *C. albicans* and *C. parapsilosis* accounted for 35% and 30% of the isolates, respectively. Whereas *C. albicans* remained mostly susceptible to fluconazole (91% cases), 59% of the *C. parapsilosis* isolates were fluconazole resistant, mostly from South Africa.

Candida auris was the third most commonly reported pathogen (14% of all the cases) in this cohort, with significant variability between countries (0%–27.5%). *Candida auris* is a rapidly emerging, multi-drug resistant, nosocomial pathogen, with high reported resistance to fluconazole and amphotericin

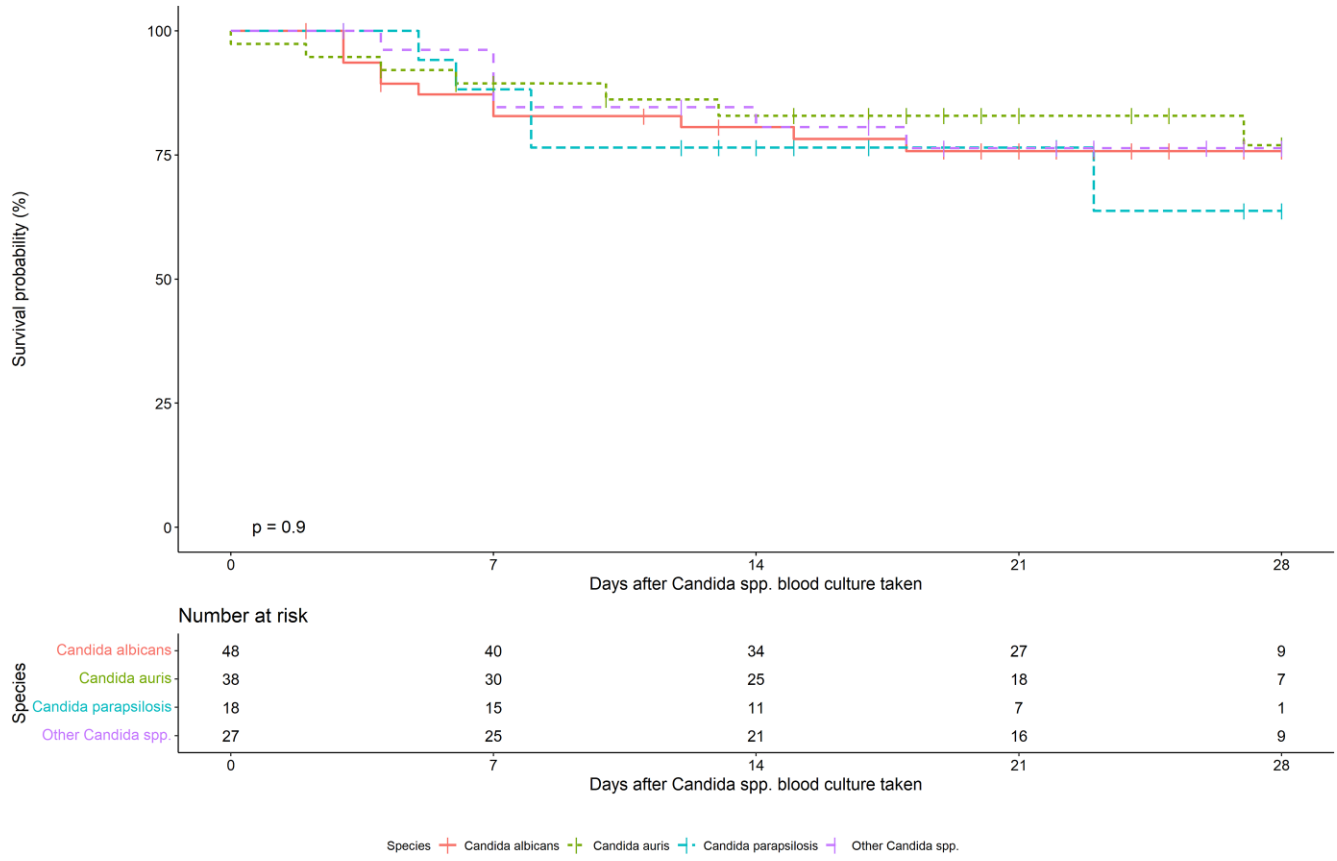

Figure 5. Kaplan–Meier curve for mortality from day of culture for each *Candida* species.

B.[25–27] There have been scant reports focused on invasive *C. auris* infections in neonates[28–30]; most of them are from India.[28] Chakabrati et al. published a multi-center prospective study from 2011 to 2012; where amongst 273 neonates from three hospitals with NIC, the proportion of *C. auris* isolates was 2.2%.[13] More recent data,[11,14,28] together with our observations, show that *C. auris* has quickly become one of the most commonly encountered species causing NIC in LMICs.

Reported neonatal mortality attributable to NIC in LMICs varies from 20% to as high as 50%.[7,31] In the cohort described here, mortality was strongly associated with low birth weight (<1000 gr) and gestational age (<28 weeks); however, we did not find a clear association with causative species or susceptibility profiles.

Antifungal prophylaxis with fluconazole targeted to neonates <1000 gr birth weight and/or <28 weeks gestation, as well as those infants with birth weight of 1000–1500 gr with additional risk factors, is a recommended strategy in neonatal units in HICs to prevent NIC.[32–36] Based on our data, NIC in LMICs affects mostly neonates with a birth weight >1500 gr, putting in doubt the relevance of HIC neonatal fungal prophylaxis guidelines for LMICs. In addition, a high prevalence of fluconazole resistance poses an important barrier to the use of fluconazole prophylaxis in these countries. Future studies determining the clinical and health economic benefit of neonatal antifungal prophylaxis in LMIC settings are needed. Fluconazole is not the only available drug to be considered; prophylaxis with nystatin, a low cost oral antifungal, which has also been proven to have an impact on neonatal mortality and is included in the essential medicines list (EML)[37–39] can also be considered.

Compared with the burden of bacterial sepsis, where *Klebsiella pneumoniae*, *Acinetobacter* spp., and *Escherichia coli* are the most commonly reported pathogens in LMICs,[40,41] the burden of fungal sepsis, particularly caused by *Candida* spp. has been poorly described. For this reason, it is not possible to provide an accurate estimated burden of NIC in LMICs.[6,11] Reported incidence of invasive candidiasis in pediatric intensive care units is significantly higher in LMICs (42.7 cases per 1000 admissions) compared to HICs (0.043–0.47 cases per 1000 admissions).[6] In general, *Candida* spp. are likely to be an underreported pathogen and mostly linked to healthcare-associated infections.

This study has some key limitations. First, not all neonates with *Candida* spp. bloodstream infections presenting at these hospitals were enrolled in the study, introducing the risk of selection bias. The aims of the NeoOBS study were to describe presentation, management, and outcomes of infants with sepsis not describe incidence of these infections, and thus we were unable to quantify incidence of NIC in this cohort due to this enrollment bias. It is also possible that the seven hospitals contributing majority of the infants to this candidemia cohort had higher repeat blood culture rates than other hospitals in the NeoOBS study. Cohort 2 enrollment may have missed some infants who died prior to a positive culture result and who were unable to be consented, potentially contributing survivor bias and lower mortality for certain *Candida* species. Differences in baseline characteristics and mortality between the two enrollment cohorts were explored (Supplementary Table 1), and no significant differences were found in key risk factors, *Candida* species, or mortality, which supported combining patients from these two cohorts into one

Table 2. Univariable hazard ratios from Cox proportional hazards analysis.

Mortality		*N* (%)	Univariable hazard ratio (95% CI, *P*)
Birthweight (1000 gr)	**≥1000 gr**	**95 (72.5)**	-
	<1000 gr	**36 (27.5)**	**3.83 (1.84–7.97, *P* < .001)**
Gestational age	**≥28 weeks**	**104 (79.4)**	-
	<28 weeks	**27 (20.6)**	**2.32 (1.08–4.99, *P* = .032)**
Organism	*Candida albicans*	48 (36.6)	-
	Candida parapsilosis	38 (29.0)	0.84 (0.32–2.16, *P* = .711)
	Candida auris	18 (13.7)	1.28 (0.44–3.70, *P* = .645)
	Other *Candida* spp.	27 (20.6)	0.92 (0.34–2.48, *P* = .868)
Country	Country 1	40 (30.5)	-
	Country 2	55 (42.0)	0.82 (0.35–1.94, *P* = .658)
	Country 3	13 (9.9)	0.77 (0.21–2.78, *P* = .685)
	Other*	23 (17.6)	0.80 (0.27–2.34, *P* = .681)
Hospital	Hospital 1	28 (21.4)	-
	Hospital 2	25 (19.1)	0.98 (0.30–3.22, *P* = .977)
	Hospital 3	21 (16.0)	1.10 (0.31–3.91, *P* = .883)
	Hospital 4	13 (9.9)	0.92 (0.23–3.70, *P* = .911)
	Hospital 5	11 (8.4)	1.16 (0.29–4.63, *P* = .837)
	Other*	33 (25.2)	1.11 (0.38–3.20, *P* = .851)
Enrollment cohort	Cohort 1	69 (52.7)	-
	Cohort 2	62 (47.3)	0.60 (0.29–1.27, *P* = .186)

Note: Significant covariates are bolded.
*Other should correspond to Other sites are comprised of nine hospitals, each contributing < 7 participants (range: 1–6 per site).

analysis. Additionally, we used hospital-reported identification and phenotypic susceptibility testing results for this analysis, which may be less accurate than MIC values and/or molecular identification techniques; moreover, the use of interpretation guidelines of antimicrobial resistance (e.g., CLSI, EUCAST, and BSAC) may vary by hospital. Finally, there were a number of isolates that did not have any susceptibility testing done or were only tested for fluconazole. Therefore, we are unable to fully evaluate resistance and appropriateness of choice of the antifungal treatment.

In conclusion, this study demonstrates that NIC is associated with significant mortality in the LMIC setting. The optimal method of prevention and treatment of this life-threatening infection requires further targeted studies. These studies should consider the epidemiological differences of NIC in LMICs compared to HICs, with an increased incidence of NIC in neonates outside the 'high-risk' group (<28 weeks and/or <1000 gr) and, although with significant variability between settings, higher rates of fluconazole resistances in non-*albicans Candida* species. Insights into the fungal epidemiology and susceptibility profiles are of utmost relevance in order to develop management guidelines for NIC in LMICs. Diagnostics for *Candida* species, including susceptibility testing, need to be made available and improved. Although no micafungin resistance was observed (within the few isolates tested), the role of empiric therapy with micafungin in LMICs for NIC needs to be a research priority. Micafungin has been included in the WHO-EML for children in 2021,[37] but there are still limitations for its use in neonates, such as the lack of a defined optimal dose for those cases with meningoencephalitis.[42,43] Finally, studies in LMICs are required to define which neonates might benefit from antifungal prophylaxis. As our study shows the current recommendations used in HIC targeting 'high-risk' neonates do not entirely apply to neonates in LMICs.

Supplementary material

Supplementary material is available at *Medical Mycology* online.

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Declaration of interest

There are no conflicts of interest to declare.

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