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Microorganisms Causing Bloodstream Infections in Pediatric Burn Patients: Distribution and Resistance Pattern

Pediyatrik Yanık Hastalarında Kan Dolaşımı Enfeksiyonlarına Neden Olan Mikroorganizmaların Dağılımı ve Direnç Paterni

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Abstract

Objective: The risk of infection increases, particularly in children, due to the deterioration of skin integrity, which serves as a protective barrier as a result of burns, the necessity for intensive care, invasive procedures, and catheter implementations. Burn-related infections have unfavorable effects on morbidity and mortality and continue to challenge the healthcare system. In this study, we investigated the distribution and antimicrobial drug resistance of causative pathogens of bloodstream infection in children with burns.

Material and Methods: This cross-sectional study was conducted at the tertiary child care hospital between May 2015 and September 2022. Pediatric cases in which microorganisms were isolated from cultures (including blood and/or catheter) and bloodstream infection was detected are included.

Results: Gram-positive bacteria were the most common causative agents of bloodstream infections in patients with burns (n= 38, 43.7%), followed by *Candida* species (n= 26, 29.9%) and gram-negative bacteria (n= 23, 26.4%). Gram-positive agents predominantly (n= 22; 64.7%) on the first seventh day of the burn, *Candida* species (%56.3, n= 9) in the second and third weeks, and gram-negative agents (%37, n= 10) after the third week were observed in bloodstream infection.

Conclusion: Recognizing the time distribution of the factors detected in pediatric burn cases is critical in determining the empirical regimen

Öz

Giriş: Yanma sonucu koruyucu bariyer olan cilt bütünlüğünün bozulması, yoğun bakım ihtiyacı olması, girişimsel uygulamalar ve kateter uygulamalarıyla özellikle çocukluk çağında enfeksiyon görülme riski artar. Yanık ilişkili enfeksiyonların morbidite ve mortalite üzerinde olumsuz etkileri olup özellikle pediyatrik vakalarda sağlık sisteminde zorluk teşkil etmeye devam etmektedir. Bu çalışmada, yanığa maruz kalan çocuklarda kan dolaşım enfeksiyonunda saptanan etkenlerin dağılımları ve antimikrobiyal direnç paternlerinin araştırılması planlanmıştır.

Gereç ve Yöntemler: Bu kesitsel çalışmaya, Mayıs 2015-Eylül 2022 tarihleri arasında, üçüncü basamak çocuk hastanesi yanık ünitesinde izlenen ve kan ve/veya kateter kültürlerinden en az birinde mikroorganizma izole edilip, kan dolaşım enfeksiyonu saptanan pediyatrik vakalar dahil edilmiştir.

Bulgular: Kan dolaşım enfeksiyonunda en sık saptanan ajanlar gram-pozitif mikroorganizmalar (n= 38, 43.7%) olup, bunu sırasıyla *Candida* türleri (n= 26, 29.9%) ve gram-negatif etkenler (n= 23, 26.4%) izlemiştir. Yanığın ilk yedinci gününde baskın olarak gram-pozitif ajanlar (n= 22; 64.7%), 2-3. haftalarda *Candida* türleri (%56.3, n= 9), üçüncü haftadan sonra ise gram-negatif ajanların (%37, n= 10) ürediği görülmüştür.

Sonuç: Pediyatrik yanık vakalarında saptanan etkenlerin zamana göre dağılım olasılıklarının iyi bilinmesi ampirik başlanacak olan rejimi belirlemede önemlidir. Ayrıca etken ajanı tedavi etmek için uygun rejim, klinik

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to be initiated. Furthermore, the appropriate treatment regimen for the causative agent should be based on clinical strain susceptibility data, and antimicrobial colonization rates and resistance patterns of relevant centers should be properly identified.

Keywords: Child, burn, microorganism, bloodstream infection

suşların duyarlılık verilerine dayanmalıdır ve bu duyarlılık verilerinde ilgili merkezlerin antimikrobiyal kolonizasyon oranları ve direnç paternleri iyi bilinmelidir.

Anahtar Kelimeler: Çocuk, yanık, mikroorganizma, kan dolaşım enfeksiyonu

Introduction

Burns are one of the most common accidents in childhood and especially affect the younger age group (1). Children under the age of two make up almost a quarter of burn cases (2).

The risk of infection increases, especially in childhood, with the deterioration of skin integrity, which is the protective barrier, the need for intensive care, interventional applications and catheter applications (3). The risk of morbidity and mortality directly proportional to infection also increases, and pediatric burn cases continue to pose a particular challenge for healthcare providers (4).

One of the most important factors that makes the treatment process difficult is the presence of multi-drug-resistant microorganisms. Although studies are limited in the pediatric age group, more resistant and deadly invasive pathogens such as methicillin-resistant *Staphylococcus aureus* (MRSA), *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Acinetobacter baumannii-calcoaceticus* complex have also been reported to cause bacteremia (5). Among multidrug-resistant microorganisms reported at increasing rates, *Acinetobacter baumannii* and *Pseudomonas aeruginosa* rapidly become multidrug resistant due to their intrinsic resistance mechanisms, which creates a very worrying situation in the health system (6,7). Recent studies have shown that infections with these multidrug-resistant microorganisms prolong hospital stay, mechanical ventilation support and antibiotic use, and consequently increase the risk of sepsis, multi-organ failure, and mortality (8).

Despite current and tremendous advances in critical care and supportive treatments in burn patient approach and wound care, infection remains the most common cause of death in pediatric burn patients (9). In this study, it was planned to examine the rates of bloodstream infection and its agents, the rates of multi-drug resistant agents and their distribution over a seven-year period in children exposed to burns.

Materials and Methods

Pediatric patients with a burn diagnosis who were hospitalized in the tertiary pediatric hospital burn unit between May 1, 2015 and September 1, 2022 were included in this cross-sectional study. The burn center of our hospital is a two-bed intensive care unit, with a total of 12 beds, and the annual number of inpatients is around 4500. According to the institution's antimicrobial protocol, prophylactic antibiotics are not recommended and antifungal prophylaxis is not routinely given.

Patient information was reviewed retrospectively from files and computer records. Age, sex, length of stay, burn severity, burn type, burn percentage, interventions, infection, and treatments of the patients included into the study were recorded. Types of burns were classified as flame burn, scalding burn, electrical burn. Microorganisms were isolated in at least one of the blood and/or catheter cultures of the patients, and the agents considered as bloodstream infections and their antimicrobial susceptibility were recorded. Except for bloodstream infections, patients with growth in surgical area, burn area, urine and tracheal aspirate samples were not included in the study. Age, sex, percentage of burns, and length of hospital stay of the patients with mortality were noted.

Definition

Patients with at least two coagulase-negative staphylococci (CNS) isolations in their blood cultures during the 24-hour sampling period, and patients with clinical accompaniment in case of opportunistic pathogen detection and without any other clinical cause were also included in the study. For multiple isolations of the same causative microorganism, the first isolation time was accepted. Different causative growths in the same patient were evaluated as different episodes.

Carbapenem resistance for *Pseudomonas aeruginosa* and *Acinetobacter baumannii*, broad spectrum β-lactamase resistance for *Klebsiella* and *Enterobacter* species, resistance to methicillin and vancomycin antibiotics for *Staphylococcus* spp. were evaluated.

Ethics committee approval was received by the local ethics committee (Decision no: 2022/16-02).

Microbiological Analysis

Blood culture samples were incubated in the BacT/ALERT 9240 automated system (bioMérieux, Marcy l'Etoile, France) for seven days or until positive (11). A blood sample from blood cultures that gave a positive signal was inoculated on chocolate, eosin methylene blue, and blood agar plates and incubated for 48 hours at 37.8°C, 5% CO^2 in accordance with the guidelines (12). Microorganisms, antibiotic susceptibility testing, minimal inhibitory concentration (MIC) levels, presence of broad-spectrum β-lactamase (ESBL) and carbapenem resistance were identified by the VITEK-2

compact system (bioMérieux), including manufacturer's instructions and Clinical and Laboratory Standards Institute (CSLI)/European Committee on Antimicrobial Susceptibility (EUCAST) criteria (12,13). Identification and antibiotic susceptibility testing of gram-positive bacteria were performed according to the manufacturer's instructions using the gram-positive identification card AST-P592 and the automated VITEK-2 system, an additional e-test (bioMérieux, Durham, NC, USA) and an appropriate disk diffusion test (14). Vancomycin-resistant *Enterococcus* strains (VRE) and MRSA were also identified using the automated VITEK-2 system. This system has also been used for identification of gram-negative bacteria with AST-N325, AST-N326 and AST-N327 gram-negative identification cards and antibiotic susceptibility tests (15). Yeast identification was performed using API 20C AUX (bioMérieux), fungal antifungal resistance identification was evaluated by e-test/antifungal MIC levels (16).

Statistical Analysis

Statistical analyzes of the study were performed with the SPSS 22.0 package program. Categorical variables were presented as frequency and percentage and numerical variables were presented using tables as (mean ± standard deviation) or (median) range (minimum, maximum). Chi-square analysis method was preferred to determine the relationship between categorical variables. A p value of <0.05 was considered statistically significant in the entire study.

Results

A total of 62 patients were included in the study covering a seven-year period. Of the patients included in the study, 43 (69.4%) were males and 19 (30.6%) were females. Median age was 20 months (the youngest patient one month-oldest 13 years). Mean hospital stay was 30 days (minimum four days-maximum 131 days). Of the patients, 80.7% (n= 50) were scalded and 14.5% (n= 9) were exposed to flame burns. Patients in the second-degree burn group were in majority (n= 57, 91.9%), the rate of patients in the third-degree burn group was 8.1% (n= 5), and the group with first-degree burns was not found in the study. Among burn percentages, the majority of patients had 16-30% burns (n= 22, 35.5%), and patients with 50% or more burns were 17.7% (n= 11). Demographic and characteristic data are summarized in Table 1.

Table 1. Demographic data of patients followed in the burn unit

Demographic data	Total number: 62 (%)
Age, months (median, min-max)	20 (min 1-max 156)
Gender, n (%)	
Male	43 (69.4)
Female	19 (30.6)
Duration of hospitalization (median, min-max)	30 (min 3-max 131)
Type of burn injury, n (%)	
Scald burn	50 (80.7)
Flame burn	9 (14.5)
Unknown reason	3 (4.8)
Total body surface area burn, n (%)	
0-15%	15 (24.2)
16-30%	22 (35.5)
31-50%	11 (17.7)
51-69%	7 (11.3)
≥70%	4 (6.5)
Unknown	3 (4.8)
Degree of burn	
Second-degree burns	57 (91.9)
Third-degree burns	5 (8.1)
Mortality, n (%)	5 (8.1)

n: Number, min: Minimum, max: Maksimum.

During the study period, 486 blood and catheter culture samples were sent. One hundred and forty-five agents were isolated from cultures and 87 bloodstream infections were observed during the study.

Distribution of Causative Agents of Bloodstream Infection

The most common agents detected in bloodstream infection were gram-positive microorganisms (n= 38, 43.7%), followed by *Candida* species (n= 26, 29.9%) and gram-negative agents (n= 23, 26.4%), respectively.

Median time for the detection of gram-positive microorganisms in the culture after exposure to burns was seven days (minimum 0 days-maximum 75 days), median time for the detection of fungi was 17 days (minimum two days-maximum 49 days), and for the detection of gram-negative microorganisms, median time taken was 20 days (minimum 0 days-maximum 80 days). Gram-positive agents were predominantly detected in the first seven days after exposure to the burn (n= 22; 64.7%). On the other hand, *Candida* species became more dominant by the 15th day, and it was observed that these factors constituted 56.3% (n= 9) at 2-3 weeks. Gram-negative agents, on the other hand, were most prevalent after the third week with a rate of 37% (n= 10) (detailed in Figure 1).

Evaluation of Factors According to the Presence of Resistance

Resistance was not detected in 44 (50.6%) of 87 agents detected in total, and resistance results could not be reached for eight agents. Among the factors; four out of five *Escherichia coli* (80%) and one in two *Klebsiella pneumoniae* (50%) were ESBL positive. Carbapenem resistance was detected in one (33.3%) of the three *Acinetobacter* species and two (50%) of the

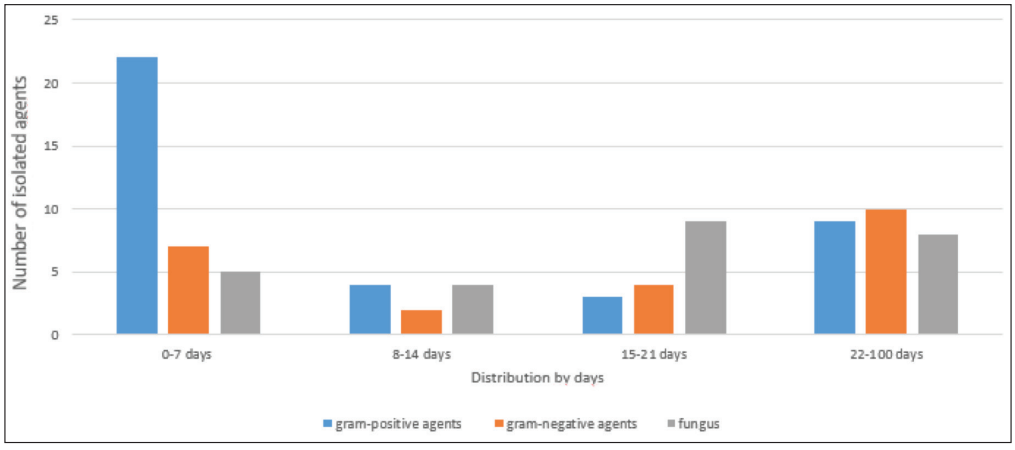

Figure 1. Distribution of bloodstream infection causative agents by weeks.

four *Pseudomonas aeruginosa* agents. Methicillin resistance was observed in one (50%) of the two *Staphylococcus aureus* agents and in two (13.3%) of 15 coagulase-negative staphylococci. One patient was found to be positive for VRE (1/17, 5.9%).

It was observed that seven (27%) of 26 fungal agents were resistant to all azoles and/or polyene group antifungals. When these seven factors were classified among themselves, two *Candida parapsilosis*, one *Candida krusei* and one *Candida albicans* were resistant to all azole antifungals, including voriconazole and posaconazole. The remaining three resistant agents were *Candida tropicalis* and were resistant to both amphotericin B and all azole groups. Among the total agents, 6.5% (n= 4) opportunistic pathogen was found to be the causative agent, among them *Ochrobactrum intermedium* ESBL was positive. The distribution of the factors according to the weeks and the resistance rates are detailed in Table 2.

Thirty-day mortality rate in the study was 8.1% (n= 5). In one of the patients who developed mortality, *Candida parapsilosis* resistant to all azole group antifungals grew, one carbapenem-resistant *Pseudomonas aeruginosa* and one ESBL positive *Escherichia coli* grew. One of the two patients with other mortality had growths of *Enterococcus faecium* (vancomycin sensitive) and CNS. While the percentage of burns was 70% and above in two of the patients with mortality, the percentage of burns was 51-69% in one of the remaining three patients, and burn percentage was 31-50% in the remaining two patients.

Discussion

Burn-related infections have adverse effects on morbidity and mortality and continue to pose a challenge in the healthcare system, especially in pediatric cases. Studies in the literature are mostly adult-based, and pediatric cases have recently been included in the studies. In this study, patients hospitalized in the burn ward in a period of seven years with a growth in their blood and/or catheter culture were evaluated, and their periodic distribution and resistance patterns were discussed.

In the study, 87 agents were found in burn-related bloodstream infections, and gram-positive microorganisms constituted the highest rate of 47.3% (n= 38) followed by *Candida* species (n= 26, 29.9%) and gram-negative agents (n= 23, 26.4%), respectively. While gram-negative factors are the most common factors found in the literature, different from our study (7,17), gram-positive factors were similarly found at a higher rate in the study conducted by Devrim et al. between 2008 and 2015 (3). These results once again emphasize that the epidemiological data, surveillance and resistance patterns of burn centers occupy an important place in the initiation of treatment for the agent. *Staphylococcus aureus* was the most frequently detected agent in studies, and methicillin resistance and vancomycin resistance caused poor prognosis and morbidity in burn-related infections, unlike in our study, enterococci and subsequent coagulase-negative staphylococci were the leading agents, and according to the literature, the relative rates of methicillin and vancomycin resistance were found to be was found to be lower (18). As emphasized in the literature, while the possibility of developing resistant strains increases with the widespread use of vancomycin, these results in our study were attributed to the avoidance of unnecessary antibiotic use by making clinical and patient-based decisions on empirical antibiotic use (19).

Compared to the rates in the literature, it was observed that *Candida* agents were detected at higher rates in our study (3,17). As is known, *Candida* infections are detected as causative agents in clinically critical patients and in long-term follow-ups (20). In our study, it was seen that four of the patients with *Candida* had flame burns and it included all patients with burns over 50% of the total body surface. And this result was directly associated with large burn wounds, type of burn and length of stay, similar to studies (21,22). Among *Candida* agents, *Candida albicans* was the most common, followed

Table 2. Distribution and resistance rates of determined microorganisms according to weeks

Gram-positive bacteria, n (%)	**n (%)**	**0-7 days, n (%)**	**8-14 days, n (%)**	**15-22 days, n (%)**	**>22 days, n (%)**	**Number of resistant microorganisms, n (%)**
Enterococcal species	17	10 (58.8)	3 (17.6)	2 (16.7)	2 (16.7)	1 (5.9)[a]
Staphylococcus aureus	2	2 (100)	0	0	0	1 (50)[b]
Coagulase-negative *staphylococcus*	15	6 (40)	1 (6.7)	1 (6.7)	7 (46.7)	2 (13.3)[b]
Streptococcus pneumoniae	1	1 (100)	0	0	0	0
Viridans group streptococci	3	3 (100)	0	0	0	0
Total	38	22 (57.9)	4 (10.5)	3 (7.9)	9 (23.7)	4 (10.5)
Gram-negative bacteria, n (%)	**n (%)**	**0-7 days, n (%)**	**8-14 days, n (%)**	**15-22 days, n (%)**	**>22 days, n (%)**	**Number of resistant microorganisms, n (%)**
Acinetobacter species	3	0	0	1 (33.3)	2 (66.7)	1 (33.3)[c]
Escherichia coli	5	2 (40)	0	1 (20)	2 (40)	4 (80)[d]
Enterobacter species	4	1 (25)	1 (25)	1 (25)	1 (25)	0
Klebsiella pneumoniae	2	0	0	0	2 (100)	1 (50)[d]
Pseudomonas aeruginosa	4	1 (25)	0	0	3 (75)	2 (50)[c]
Salmonella newington	1	1 (100)	0	0	0	0
Opportunistic pathogens, n (%)	**n (%)**	**0-7 days, n (%)**	**8-14 days, n (%)**	**15-22 days, n (%)**	**>22 days, n (%)**	**Number of resistant microorganisms, n (%)**
Chryseobacterium indologenes	1	0	1 (100)	0	0	0
Ochrobactrum intermedium	1	1 (100)	0	0	0	1 (100)[d]
Sphingomonas paucimobilis	1	1 (100)	0	0	0	0
Stenotrophomonas maltophilia	1	0	0	1(100)	0	0
Total	23	7 (30.4)	2 (8.7)	4 (17.4)	10 (43.5)	9 (39.1)
***Candida* species, n (%)**	**n (%)**	**0-7 days, n (%)**	**8-14 days, n (%)**	**15-22 days, n (%)**	**>22 days, n (%)**	**Number of resistant microorganisms, n (%)**
Candida albicans	12	3 (25)	1 (8.4)	4 (33.3)	4 (33.3)	1 (8.3)[e]
Candida glabrata	1	1 (100)	0	0	0	0
Candida kefyr	1	0	0	1 (100)	0	0
Candida krusei	1	0	1 (100)	0	0	1 (100)[e]
Candida parapsilosis	5	0	1 (20)	1 (20)	3 (60)	2 (40)[e]
Candida tropicalis	6	1 (16.7)	1 (16.7)	3 (50)	1 (16.7)	3 (50)[e,f]
Total	26	5 (19.2)	4 (15.4)	9 (34.6)	8 (30.8)	7 (27)

a: Vancomycin resistance, b: Methicillin resistance, c: Carbapenem resistance, d: Extended spectrum β-lactamase, e: Azole group antifungal resistance, f: Amphotericin-B resistance.

by *Candida tropicalis* and *Candida parapsilosis*. These three factors constituted high rates of 94.6% in the literature, as well as 88.5% of *Candida* agents in our study (23,24). While azole resistance was observed in only one of the *Candida albicans* strains, resistance rates were higher in non-*albicans* strains, similar to the literature (19). It has been stated in studies for a long time that non-*albicans Candida* species are increasing gradually, and it was observed that *Candida albicans* strains were still dominant in our study, and this situation was associated with avoidance of off-label antifungal prophylaxis or treatment (22,25).

Gram-negative agents also occupy an important place in burn patients, especially in terms of resistance development and prognosis of infection (7). While it is stated in the literature that gram-negative agents are the most common microorganisms in burn-associated bloodstream infections, in our study, gram-negatives were ranked third as burn-associated bloodstream infections, contrary to the literature (17,26). In our study, the most common gram-negative agent was *Escherichia coli*, followed by *Pseudomonas aeruginosa* and other *Enterobacter* species. As a resistance pattern, gram-negative agents were found to have a higher probability of resistant strains than gram-positive agents, and this result is similar to the literature (27). Another point that differs from the literature in the study is that opportunistic pathogens that are not commonly detected in burn-associated bloodstream infections were observed. In the study of Lin et al., it has been stated that *Stenotrophomonas maltophilia* is seen more frequently than *Pseudomonas aeruginosa*, *Staphylococcus aureus*, with rates such as 13% (n= 12). It has been stated that *Chryseobacterium indologenes* is seen more frequently than *Pseudomonas aeruginosa* and *Staphylococcus aureus* with a rate of 5.1% (n= 5), but it is mostly polymicrobial, and each of them has been found to be a single agent in one patient in our country. The high rates in the study of Lin et al. were associated with the injury of patients in the same accident, and it should not be forgotten that although rare, opportunistic pathogens can be seen in burn patients due to

their immunosuppression, the presence of a catheter, and the need for long-term hospitalization (28).

Colonization rates of bacteria and fungi vary in relation to the post-burn time. In our study, gram-positive pathogens were predominantly detected in the first week, *Candida* infections after the second week and in the long-term period, and infections with gram-negative agents were observed after the third week. Similar to our study, in the literature, while gram-positive agents such as *Staphylococcus aureus* were prominent at a rate of more than 80% in the first weeks, gram-negative agents such as *Pseudomonas aeruginosa* became prominent as of the third week (29). Knowing the day of exposure in the treatment selection is necessary in determining the agent to be chosen empirically.

Our study was a retrospective study and had inherent limitations when compared to randomized clinical trials. Due to the retrospective study, there were few patients whose antibiogram results and resistance results could not be reached. On the other hand, it is among the rare studies in the literature in terms of dealing with bloodstream infectious agents and resistance patterns in pediatric burn cases covering a period of seven years.

Conclusion

In conclusion, it should not be forgotten that the factors detected in pediatric burn cases may differ in terms of timing and resistance pattern. Knowing the distribution probabilities of the factors according to time is important in determining the regime to be started empirically. In addition, the appropriate regimen to treat the causative agent should be based on susceptibility data of clinical strains, and antimicrobial colonization rates and resistance patterns of relevant centers should be well known in these susceptibility data.

Ethics Committe Approval: This study approval was obtained from SBU İzmir Dr. Behçet Uz Pediatric Diseases and Surgery Training and Research Hospital Clinical Research Ethics Committee (Decision no: 240, Date: 29.09.2022).

Informed Consent: Patient consent was obtained.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept- EC; Design- EC, NB, İD; Supervision- EC, NB, İD; Data Collection and/or Processing- EC, ŞŞ, MYÇ, MG, PK, MNA, ÖO, KP, ADP, AO; Analysis and/or Interpretation- EC, AAK, GGÖ, FYA; Literature Search- EC; Writing- EC; Critical Review- NB, İD.

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