Isolation of Pathogenic Bacteria and Their Antibiotic Sensitivity Profiles in Hospitalized Febrile Neutropenic Children with Acute Lymphoblastic Leukaemia

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Abstract

Background: Acute lymphoblastic leukemia (ALL) is the commonest malignancies in childhood. Common obstacle in the treatment of ALL is febrile neutropenia and its complications. **Objectives:** To identify bacteria causing infection, their isolation rate and antibacterial sensitivity pattern in hospitalized febrile neutropenic children with ALL in different cycle of chemotherapy. **Methodology:** This observational study conducted in 2014 - 2015 in the department of paediatric haematology and oncology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka Bangladesh. Sixty febrile neutropenic episodes from 52 diagnosed cases of ALL aged 0 to 18 years were included. Complete blood count, blood culture, urine microscopy and culture, serum alanine aminotransferase, serum creatinine were done in every patient. X-ray chest, stool microscopy and culture, pus, wound, throat and aural swab for culture & sensitivity were done in selective patient. **Results:** Bacterial infection was confirmed by culture in 15 (25%) episodes from 60 febrile neutropenic episodes. Fifteen (25%) organisms were isolated from the study subjects from sample of blood (60%), pus (13.3%), aural swab (13.3%), wound swab (6.7%) and throat swab (6.7%) respectively. All isolates were gram negative. The organism isolated were *Klebsiella* spp. 5 (33.31%), *E. coli* 4 (26.7%), *Acinetobacter* 3 (20%), *Pseudomonas* 2 (13.3%) and only one (6.7%) *Enterobacter* species. All the isolates of the *Klebsiella* spp., *E. coli* and *Acinetobacter* spp. were resistant to amoxicillin. All isolated *E. coli* were resistant to cotrimoxazole, ceftazidime, ceftriaxone, cefotaxime and ciprofloxacin, *Acinetobacter* spp. Isolated were 100% sensitive to imipenem, colistin sulphate & piperacillin-tazobactam and resistant to cotrimoxazole and cephadrine. All *Pseudomonas* spp. showed 100% sensitivity to imipenem, amikacin, ciprofloxacin & colistin and resistances to ceftazidime. **Conclusion:** The species of *Klebsiella* were the predominant causative bacterial agent followed by *Escherichia coli*, *Acinetobacter* spp, *pseudomonas* spp. and *Enterobacter* spp. They showed resistance to commonly prescribed antibiotics ceftazidime, gentamicin, ceftriaxone & ciprofloxacin and sensitive to imipenem, colistin-sulphate & piperacillin-tazobactam.

Keywords: Neutropenia, Pathogenic Bacteria, Culture and sensitivity.

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Introduction

Acute leukemia is one of the most common childhood malignancies. Childhood acute lymphoblastic leukaemia (ALL) is also common in our country. Cancer treatment is associated with infections, in spite of the progress made in the development of antibiotics, is still a major concern for healthcare professionals. On the other hand, assessment of the type of infection presented after chemotherapy plays a major role in achieving a better treatment outcome¹. A correct prediction of the type of infection at the immediate onset of fever before culture results is available. On the contrary, late decision-making and wrong predictions can not only lead to the spread of infection, but can also result in failure of chemotherapy, delayed completion of remission, and poor treatment outcome. Febrile neutropenia is the most frequent manifestation of a potentially lethal complication of current intensive chemotherapy regimens. And it is also oncology related medical emergency^{2,3}. The pathogens isolated from culture in neutropenic fever have changed over time and may be location dependent.

Culture isolates from febrile neutropenic patients were once predominantly gram negative-organisms⁴. There were major changes in the type and range of pathogens causing infection in neutropenic patients during the last decades⁵. Clinicians should be aware of the predominant pathogens and an antibiogram that depicts the in vitro susceptibility patterns of the most prevalent pathogens in their own institution to select an efficient initial empirical therapy⁶. So, with this view, aim of our study is to assess the frequency of bacterial infection and their antibacterial sensitivity profiles in hospitalized febrile neutropenic children with acute lymphoblastic leukemia in different cycle of chemotherapy.

Methodology

This was a cross sectional observational study conducted from November 2014 to November 2015 in the department of paediatric haematology and oncology, Bangabandhu Sheikh Mujib Medical University (BSMMU). Sixty febrile neutropenic episodes from 52 diagnosed cases of ALL with febrile neutropenic children aged 0 to 18 years were included during one-year study period after fulfilling the criteria of case definition. Informed consent from respective parents and legal guardian obtained prior to inclusion

of any child. History was taken for any symptom of sepsis. Then proper physical examination carried out. Complete blood count (CBC), blood culture and sensitivity, urine routine microscopic examination and culture & sensitivity, serum glutamic pyruvic transaminase, serum creatinine were done in every patient. X-ray chest, stool routine microscopic examination and culture & sensitivity, pus, wound, throat and aural swab for culture & sensitivity were done in selected patient according to sites of infection in the respective department of BSMMU.

Study procedure

Their full initial presentations, initial total count, history of recent chemotherapy with their intensity, history of any associated co morbidity like diarrhea, shock, DIC, severe bleeding manifestation & full physical examination were recorded. Complete blood count (CBC) was done by Sysmax® automated analyzer, serum glutamic pyruvic transaminase, and serum creatinine were done in every patient in the department of Paediatric Haematology and Oncology. X-ray chest was done in the radiology department, BSMMU and on emergency cases bed side x-ray was done in selective patients. All aseptic precautions were maintained carefully before and after specimen collection. Blood culture and sensitivity (Automated BACTEC 9240 machine), urine routine microscopic examination & culture and sensitivity were done in every patients and stool routine microscopic examination & culture and sensitivity, pus, wound, throat & aural swab for culture and sensitivity were done in selected patient according to sites of infection in the department of microbiology, BSMMU. Venous blood was collected for complete blood count, metabolic workup and blood culture.

Statistical analysis of data

After collection, data editing, compilation and analysis was done manually. Statistical analysis was done both manually and window-based software devised with Statistical Packages for the Social Sciences (SPSS).

Ethical implication

Prior to the commencement of this study, the thesis protocol was approved by the Institutional Review Board, BSMMU, Dhaka. The thesis protocol was explained to parents or legal guardians about my study

before taking the consent. Then a written consent was taken from each patient's legal guardian. Every precaution was taken so that study will not cause any harm or delay the treatment of cases. They had liberty to exclude their child from the study at any time.

Results

Total 60 febrile neutropenic episodes from 52 acute lymphoblastic leukaemic children were studied, 35 males and 17 females, with a mean age 5.5 years. (Fig 1, Table 1) Primary site of infection could not be suspected clinically in 19 (31.6%) episodes. Bacterial infection was confirmed by culture in 15 (25%) episodes from 60 febrile neutropenic episodes.

Table 1: Distribution of febrile neutropenic episodes with ALL patients according to age (n = 60).

Age Groups (years)	n (%)
≤2	10 (16.7)
2.1 - 5.0	27 (45.0)
5.1 - 10.0	17 (28.3)
>10	6 (10.0)
Total	60 (100.0)
Mean ± SD	5.47 ± 3.57
Range (Min-Max)	0.5 – 16.0

Figure 1: Sex distribution of the patients.

Seven patients were admitted in more than one episode and two of them were culture positive in more than one episode. Fifteen (25%) organisms were isolated from the study subjects of which blood 9 (60%), pus 2 (13.3%), aural swab 1 (13.3%), wound swab 1 (6.7%) and throat swab 1 (6.7%). All isolates were gram negative. (Fig. 2)

Figure 2: Result of culture positivity of different specimen of febrile neutropenic episodes.

The most common organism was *Klebsiella* spp 5 (33.31%), followed by *E. coli* 4 (26.7%), *Acinetobacter* 3 (20%), *Pseudomonas* 2 (13.3%) and only one (6.7%) was *Enterobacter* species. Maximum isolation of bacteria 13 (32.5%) of 40 episodes was in induction phase of chemotherapy. (Fig. 3)

Figure 3: Isolation of Bacterial species from Culture

All the isolated of the *Klebsiella* spp., *E. coli* and *Acinetobacter* spp. were resistant to amoxicillin and Cephadrine. Eighty (80%) of *Klebsiella* spp. were resistant to cotrimoxazole, ceftazidime, ceftriaxone, cefotaxime and cefuroxime. All isolated the *E. coli* were resistant to cotrimoxazole, ceftazidime, ceftriaxone, cefotaxime and ciprofloxacin followed by 75% were resistant to amikacin. All isolated the *Acinetobacter* spp. 100% sensitive to imipenem, colistin sulphate & piperacillin-tazobactam and resistant to cotrimoxazole and cephadrine. All *Enterobacter* spp. showed 100% sensitive of imipenem & amikacin and resistant to other antibiotics used in the study. All *Pseudomonas* spp. showed 100% to imipenem, amikacin, ciprofloxacin & colistin and resistant to ceftazidime. (Table 2)

Table 2: Resistance pattern of antibiotics against commonly isolated pathogens (gram negative) of febrile neutropenic episodes with ALL.

Antibiotics	Escherichia Coli	Acinetobacter spp	Klebsiella spp	Pseudomonas spp	Enterobacter spp
Amoxycilin	4 (100.0)	3 (100.0)	5 (100.0)	-	1 (100.0)
Cotrimoxazole	4 (100.0)	3 (100.0)	4 (80.0)	-	1 (100.0)
Nalidixic	4 (100.0)	2 (66.7)	3 (60.0)	-	-
Gentamicin	-	1 (33.3)	2 (66.7)	1 (50.0)	1 (100.0)
Ceftazidime	4 (100.0)	2 (66.7)	4 (80.0)	2 (100.0)	1 (100.0)
Amikacin	3 (75.0)	1 (33.3)	2 (40.0)	0	0
Imipenem	2 (50.0)	0	1 (20.0)	0	0
Cephadrine	2 (100.0)	3 (100.0)	4 (100.0)	-	1 (100.0)
Ciprofloxacin	4 (100.0)	1 (33.3)	1 (20.0)	0	1 (100.0)
Ceftriaxone	4 (100.0)	2 (66.7)	4 (80.0)	-	1 (100.0)
Cefotaxime	4 (100.0)	2 (66.7)	4 (80.0)	-	1 (100.0)
Cefuroxime	4 (100.0)	2 (66.7)	4 (80.0)	-	1 (100.0)
Aztreonam	4 (100.0)	2 (66.7)	4 (80.0)	1 (50.0)	1 (100.0)
Netilmicin	2 (50.0)	1 (33.3)	1 (20.0)	0	1 (100.0)
Cefepime	-	-	-	1 (50.0)	-
Tazobactam	2 (50.0)	0	-	1 (50.0)	-
Piperacillin	-	-	-	-	-
Ticarillin	-	2 (66.7)	-	0	-
Colistin	-	0	-	0	-

Discussion

Infections in the immunocompromised host as a result of cancer chemotherapy is an important problem in the present day-to-day treatment care, as they are associated with an increased incidence of neutropenic infectious complication, which in turn influences the outcome of the chemotherapeutic response, and thereby morbidity and mortality, in these patients⁷.

One study discussed that blood was the most common site of infection and 85.4% of them had gram-negative bacteria and also stated that source of 56% of isolated organisms was blood and gram-negative bacteria accounted for 78% of isolated organisms from febrile neutropenic patients in hematological malignancies⁸.

The most common organisms were *Escherichia coli* (43%) and *Klebsiella pneumonia* (17.4%) and revealed among gram negative-organism *E.coli* (30.2%) was followed by *Klebsiella Pneumoniae* (20.9%)⁹. In this study, all (100%) the isolated organisms were gram negative and 60% of these were from blood but there were no growth in stool and urine. The most common isolated organisms were *Klebsiella* spp. (33.3%) followed by *Esch. Coli* (26.7%) and *Acinetobacter* spp. (20.0%). In this study, all the isolated of the *Klebsiella* spp., *E. coli* and *Acinetobacter* spp. were resistant to amoxicillin and Cephadrine. Eighty (80%) of *Klebsiella* spp. were resistant to cotrimoxazole, ceftazidime, ceftriaxone, cefotaxime and cefuroxime. All isolated the *E. coli* were resistant to cotrimoxazole, ceftazidime, ceftriaxone, cefotaxime and ciprofloxacin followed by 75% were resistant to amikacin. All isolated the *Acinetobacter* spp. showed 100% sensitive to imipenem, colistin sulphate & piperacillin-tazobactam and resistant to cotrimoxazole and cephradine. All *Enterobacter* spp. showed 100% sensitive of imipenem & amikacin and resistant to other antibiotics used in the study. All *Pseudomonas* spp. showed 100% sensitive to imipenem, amikacin, ciprofloxacin & colistin and resistant to ceftazidime. Mahallawy et al. (2005) has reported very high resistance rates for gram negative pathogens, reaching 60% for ceftazidime, 50% for amikacin and imipenem and 40% for piperacillin-tazobactam, cefepime and ciprofloxacin¹⁰. Another popular study Aslan et al. (2012) has revealed the antimicrobial sensitivity patterns of gram-negative organisms to imipenem (89.2%). Kwon et al. (2013) has also stated

that the resistance rate of gram-negative bacteria was amikacin 10.6%, imipenem 4.4%. In this study very high resistance rates were reported to gram negative bacteria¹¹. The most probable explanation for these findings is the increased use of antibiotics in the hospital, as evidenced by a profound resistance to both amikacin and ceftazidime, which have been used extensively as first-line empirical therapy.

Conclusion

Febrile neutropenic patients with acute lymphoblastic leukaemia underwent chemotherapy were in a major risk for infection. The present study, 25% of febrile neutropenic episodes were bacterial culture positive in different phases of chemotherapy and all were gram-negative with *klebsiella* spp. as major isolate followed by *Escherichia coli*, *Acinetobacter* spp, *pseudomonas* spp. and *enterobacter* spp. Sixty (60%) of these bacteria were isolated from blood and mostly 13 (32.5%) during induction phase of chemotherapy. These bacteria were highly resistance to ceftazidime, gentamicin, ceftriaxone, ciprofloxacin, amikacin & cefotaxime and sensitive to imipenem, colistin-sulphate & piperacillin-tazobactam.

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