

## Causative Pathogens of Febrile Neutropaenia in Children Treated for Acute Lymphoblastic Leukaemia

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### Abstract

**Introduction:** Treatment of acute lymphoblastic leukaemia (ALL) using intensive chemotherapy has resulted in high cure rates but also substantial morbidity. Infective complications represent a significant proportion of treatment-related toxicity. The objective of this study was to describe the microbiological aetiology and clinical outcome of episodes of chemotherapy-induced febrile neutropaenia in a cohort of children treated for ALL at our institution. **Materials and Methods:** Patients with ALL were treated with either the HKSGALL93 or the Malaysia-Singapore (Ma-Spore) 2003 chemotherapy protocols. The records of 197 patients who completed the intensive phase of treatment, defined as the period of treatment from induction, central nervous system (CNS)-directed therapy to reinduction from June 2000 to January 2010 were retrospectively reviewed. **Results:** There were a total of 587 episodes of febrile neutropaenia in 197 patients, translating to an overall rate of 2.98 episodes per patient. A causative pathogen was isolated in 22.7% of episodes. An equal proportion of Gram-positive bacteria (36.4%) and Gram-negative bacteria (36.4%) were most frequently isolated followed by viral pathogens (17.4%), fungal pathogens (8.4%) and other bacteria (1.2%). Fungal organisms accounted for a higher proportion of clinically severe episodes of febrile neutropaenia requiring admission to the high-dependency or intensive care unit (23.1%). The overall mortality rate from all episodes was 1.5%. **Conclusion:** Febrile neutropaenia continues to be of concern in ALL patients undergoing intensive chemotherapy. The majority of episodes will not have an identifiable causative organism. Gram-positive bacteria and Gram-negative bacteria were the most common causative pathogens identified. With appropriate antimicrobial therapy and supportive management, the overall risk of mortality from febrile neutropaenia is extremely low.

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**Key words:** Infective complications, Bacterial infections, Mortality, Toxicity

### Introduction

Acute lymphoblastic leukaemia (ALL) is the most common form of childhood cancer in Singapore, accounting for 44% of all causes of childhood cancer annually.<sup>1</sup> Treatment of childhood ALL consists of a period of intensive chemotherapy followed by maintenance chemotherapy for up to 2 years, with the objective of producing lasting remission of the disease. In Singapore, chemotherapy protocols that have been used in the treatment of childhood ALL include the Malaysia-Singapore (Ma-Spore)-ALL

2003 and the HKSGALL 93 protocols. These have resulted in high cure rates, with 5-year overall survival rates of more than 80%.<sup>2,3</sup>

The initial intensive phase of treatment typically consists of an induction phase followed by central nervous system (CNS)-directed therapy and reinduction chemotherapy. Myelosuppression and generalised immunosuppression are expected consequences of this intensive phase of therapy, leading to episodes of infection that can result in treatment delays, dropouts and mortality.<sup>4</sup> The objective

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Table 1. Episodes of Febrile Neutropaenia According to Treatment Risk Groups and Phase of Treatment

Risk Group	No. of Patients	No. of Episodes of Febrile Neutropaenia in Each Phase of Treatment			Total	Mean No. of Episodes Per Patient
		Induction	Protocol M	Reinduction		
Standard risk	70	93	21	95	209	2.99
Intermediate risk	109	141	29	139	309	2.83
High risk	18	23	14	32	69	3.83
Total	197	257	64	266	587	
Mean		1.30	0.32	1.35		

of the study was to describe the microbiological aetiology and clinical outcome of episodes of chemotherapy-induced febrile neutropaenia in children being treated for ALL at our institution.

## Materials and Methods

This is a retrospective cohort study of 197 patients who were treated for ALL at KK Women's and Children's Hospital from June 2000 to January 2010. Patients included in the study were those who developed febrile neutropaenia during the intensive phase of treatment, defined as the period of treatment from induction, CNS-directed therapy to the end of reinduction therapy.

The treatment protocol used in our institution from June 2000 to December 2006 was the HKSGALL 93 protocol.<sup>2</sup> From January 2007, the Ma-Spore ALL 2003 protocol<sup>3</sup> was used instead. Both protocols are based on the Berlin Frankfurt Munster (BFM) backbone and only differ in terms of treatment strategy, with the Ma-Spore ALL 2003 being a minimal residual disease (MRD)-directed treatment protocol. Treatment intensity differed according to risk stratification. Patients who were treated with either protocol were analysed together and not differentiated.

Neutropaenia was defined as an absolute neutrophil count of less than  $1 \times 10^9/L$ . Fever was defined as a temperature more than or equal to  $38^\circ C$  on 2 or more occasions or a temperature of  $38.5^\circ C$  or higher on a single occasion. All patients admitted for febrile neutropaenia were extensively investigated for a source with blood, urine, stool cultures and other tests as clinically indicated, according to the protocols in place in our institution. The individual case records of the patients were analysed to gather information on the clinical presentation and other supportive investigations, including chest X-rays and other radiological findings, to determine if positive isolates were causative organisms and not merely commensals.

Severe episodes of neutropaenic fever were defined as cases requiring admission into a high dependency unit

(HDU) or intensive care unit (ICU) due to any clinical presentation requiring a higher level of care.

Data was analysed using SPSS Version 12. This study was approved by the Singhealth Central Institutional Review Board.

## Results

There were a total of 587 episodes of febrile neutropaenia in 197 patients, translating to an overall rate of 2.98 episodes per patient during the intensive phase of chemotherapy. Patients on high-risk protocols who received the most intensive treatment had the highest mean number of episodes per patient (3.83). The treatment phases associated with the higher mean number of episodes per patient were the induction (1.30) and reinduction (1.35) phases of therapy. The frequency of episodes of febrile neutropaenia according to treatment phase and risk group is given in Table 1.

## Causative Pathogens

A causative pathogen was isolated from either blood, urine, stool cultures or nasopharyngeal aspirates in 133 out of 587 (22.7%) episodes of febrile neutropaenia. Multiple organisms were isolated in some cases. Organisms identified in order of frequency were Gram-positive bacteria (36.4%) and Gram-negative bacteria (36.4%), followed by viral pathogens (17.4%), fungal pathogens (8.4%), and other bacteria (1.2%). The majority of positive bacterial and fungal isolates were from peripheral or central line blood cultures. The most common bacteria isolated were *Staphylococcus aureus* (27/124, 21.8%), *Escherichia coli* (20/124, 16.1%), *Pseudomonas aeruginosa* (13/124, 10.5%), *Bacillus* sp (8/124, 6.5%) and *Klebsiella pneumoniae* (8/124, 6.5%). A breakdown of the specific organisms identified is given in Table 2 and Figure 1.

We further analysed the microbiological profile of severe episodes of febrile neutropaenia requiring admission into a high dependency unit (HD) or intensive care unit (ICU). There were 20 of such episodes, accounting for 3.4% of

Table 2. Types of Causative Pathogens Isolated

Type	Organism	No.	%
Gram-positive bacteria	<i>Staphylococcus aureus</i>	27	36.4
	<i>Bacillus</i> species	8	
	Coagulase-negative <i>Staphylococcus</i>	6	
	Other <i>Staphylococcus</i> species	6	
	<i>Enterococcus faecalis</i>	3	
	<i>Streptococcus salivarius</i>	2	
	Other <i>Streptococcus</i> species	2	
	<i>Micrococcus</i> species	2	
	<i>Dermabacter hominis</i>	1	
	<i>Streptococcus agalactiae</i>	1	
	<i>Streptococcus mitis</i>	1	
	<i>Streptococcus pyogenes</i>	1	
	<i>Streptococcus viridans</i>	1	
Gram-negative bacteria	<i>Escherichia coli</i>	20	36.4
	<i>Pseudomonas aeruginosa</i>	13	
	<i>Klebsiella pneumoniae</i>	8	
	<i>Proteus mirabilis</i>	4	
	<i>Acinetobacter baumannii</i>	3	
	<i>Salmonella</i> species	3	
	Unidentified Gram-negative bacilli	2	
	<i>Aeromonas hydrophilia</i>	1	
	<i>Chryseobacterium indologenes</i>	1	
	<i>Enterobacter cloacae</i>	1	
	<i>Enterobacter hormaechei</i>	1	
	<i>Fusobacterium nucleatum</i> *	1	
	<i>Morganella morganii</i>	1	
	<i>Neisseria</i> species	1	
	<i>Proteus vulgaris</i>	1	
Other bacteria	<i>Mycoplasma pneumoniae</i>	2	1.2
Viral	Respiratory syncytial virus	8	17.4
	Influenza A	6	
	Parainfluenza 3	4	
	Rotavirus	4	
	Varicella zoster	3	
	Parainfluenza 1	2	
	Epstein-Barr virus	1	
	Influenza B	1	
Fungal	<i>Candida albicans</i>	4	8.4
	<i>Aspergillus fumigatus</i>	2	
	<i>Candida tropicalis</i>	2	
	Other <i>Candida</i> species	3	
	<i>Candida parapsilosis</i>	1	
	<i>Pneumocystis jirovecii</i>	1	
	<i>Ulocladium</i> species	1	

\*Anaerobic bacteria

Table 3. Types of Causative Pathogens Isolated from Severe Cases of Febrile Neutropaenia Requiring HD/ICU Admissions

Type	Organism	No.	%
Gram-negative bacteria	<i>Escherichia coli</i>	1	38.5
	<i>Klebsiella pneumoniae</i>	1	
	<i>Enterobacter hormaechei</i>	1	
	<i>Aeromonas hydrophilia</i>	1	
	<i>Pseudomonas aeruginosa</i>	1	
Gram-positive bacteria	<i>Staphylococcus aureus</i>	1	30.8
	<i>Streptococcus viridans</i>	1	
	<i>Bacillus</i> species	2	
Fungal	<i>Candida albicans</i>	1	23.1
	<i>Candida tropicalis</i>	1	
	<i>Aspergillus fumigatus</i>	1	
Viral	Influenza A	1	7.7

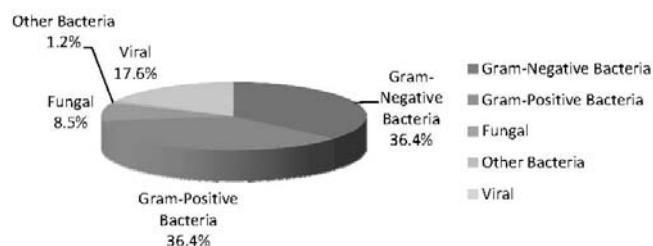


Fig. 1. Chart showing the types of organisms that cause febrile neutropenia.

all cases of febrile neutropaenia. Causative pathogens were isolated in 9/20 (45%) of these episodes (Table 3). Multiple organisms were again isolated in some cases. Organisms isolated in order of frequency were Gram-negative bacteria (38.5%), Gram-positive bacteria (30.8%), fungal pathogens (23.1%) and viral pathogens (7.7%).

## Outcome

The majority of cases of febrile neutropaenia resolved with appropriate antimicrobial therapy and supportive management. There were 3 deaths from infection, accounting for 1.5% of all patients and 0.5% of all episodes of febrile neutropaenia. Cases requiring HD/ICU admission had a significantly higher mortality rate of 15%, reflecting the clinical severity of these cases. Two of these deaths occurred in patients with underlying trisomy 21. One occurred during the initial induction phase of treatment when he presented with febrile neutropaenia in acute respiratory distress to the ICU. Despite extensive workup for a source of infection, no organisms were isolated.

The other patient with trisomy 21 succumbed to systemic *Candida albicans* infection during the reinduction phase of chemotherapy, with the organism isolated from urine, stool and bronchial washings. The third death occurred in a child without any comorbidities during the reinduction phase of chemotherapy. Blood and endotracheal cultures were positive for *Pseudomonas aeruginosa*. Novel influenza A (H1N1) virus was also isolated from bronchial washings. The child eventually died of acute respiratory distress syndrome despite aggressive use of appropriate antibiotics and oseltamivir (Tamiflu).

## Discussion

In our study, a causative pathogen was isolated in only 22.7% of all episodes of febrile neutropaenia. This concurs with prior studies which found that the aetiology for the majority of episodes of febrile neutropaenia will remain unknown despite comprehensive diagnostic workup.<sup>4,5</sup> For microbiologically documented episodes, the pathogen profile in our cohort was also consistent with findings in similar studies conducted both within Southeast Asia<sup>6,7</sup> and from other regions of the world,<sup>5,8</sup> with bacterial organisms being the predominant cause of infections.

Gram-positive and Gram-negative bacteria were the most common causative organisms. This finding suggests that empirical antimicrobial therapy for episodes of febrile neutropaenia should include agents that have activity against both Gram-positive and Gram-negative bacteria. The empirical antimicrobial therapy in use at our institution from 2000 to 2010 was a combination of intravenous ceftriaxone and gentamicin. However, subsequent review of antibiotic susceptibility patterns from 2006 to 2011 showed a trend in increasing antibiotic resistance to cephalosporins, especially amongst Gram-negative bacteria. Empirical antimicrobial therapy for treatment of neutropaenic fever in our institution was hence amended to piperacillin/tazobactam from 2012, which provided better coverage against Gram-positive, Gram-negative and anaerobic bacteria.

Severe infections requiring HD or ICU care are of particular concern, as these are the cases with a higher risk of morbidity and mortality. The rate of identification of a causative organism was higher in these cases, with just under half of all episodes associated with positive microbial isolates. Although bacterial organisms were again the predominant pathogens identified, fungal organisms accounted for a much larger proportion of these severe cases requiring HD or ICU care compared to clinically milder cases (23.1% vs 8.4%). Our findings suggest that empirical antifungal agents in addition to antibiotics should be commenced at presentation for clinically severe cases of febrile neutropaenia, which is the current practice at our institution.

The highest number of episodes of febrile neutropaenia occurred during the induction and reinduction phases of chemotherapy, and also in patients on high-risk protocols who received the most intensive treatment which would result in greater myelosuppression. This is not a surprising finding, as the degree of neutropaenia has been found to correlate with the frequency and severity of febrile neutropaenia.<sup>9</sup> Despite the multiple episodes of febrile neutropaenia seen in our cohort, the mortality rate was low at 1.5%, as compared to previously reported infection-related mortality rates of 0.6% to 19.2% in paediatric febrile neutropaenia studies.<sup>6,7,8</sup>

Two of the 3 deaths recorded within the duration of the study occurred in patients with underlying trisomy 21. This concurs with existing literature which showed that ALL patients with Down syndrome had increased rates of infective complications with higher treatment-related morbidity and mortality.<sup>10</sup> This particularly vulnerable group of patients would need to be monitored carefully during periods of neutropaenia for development of sepsis, and treated aggressively with antibiotics and antifungal agents at presentation during episodes of febrile neutropaenia even before any organisms are isolated.

## Conclusion

Febrile neutropaenia continues to be of concern in ALL patients undergoing intensive chemotherapy. The majority of episodes will not have an identifiable causative pathogen. Gram-negative and Gram-positive bacteria were the most common causative pathogens identified. With appropriate antimicrobial therapy guided by antibiotic sensitivity patterns of each individual institution and supportive management, the overall risk of mortality from febrile neutropaenia is extremely low.

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