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Invasive fungal infections in children with acute lymphoblastic leukaemia: results from four Australian centres, 2003 – 2013

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Abbreviations	
IFI	Invasive fungal infections
ALL	Acute lymphoblastic leukaemia
TERIFIC	The Epidemiology and Risk Factors for Invasive Fungal Infections in Immunocompromised Children study
HSCT	Haematopoietic stem cell transplant
EORTC	European Organization for Research and Treatment of Cancer/Invasive Fungal Infections Cooperative Group
PCR	Polymerase chain reaction
MSG	National Institute of Allergy and Infectious Diseases Mycoses Study Group
COG	Children's Oncology Group
CCG	Children's Cancer Group
SR	Standard Risk
HR	High Risk
GM	Galactomannan
BAL	Bronchoalveolar Lavage

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ABSTRACT

Background: Invasive fungal infections (IFI) are an important complication of acute lymphoblastic leukaemia (ALL) treatment. Our study describes the prevalence and outcomes of IFI in children with ALL.

Methods: IFI episodes in children with primary or relapsed ALL, identified for The Epidemiology and Risk Factors for Invasive Fungal Infections in Immunocompromised Children (TERIFIC) study, were analysed. IFI were classified according to EORTC criteria with a 'modified possible' category included.

Results: A total of 123 IFI episodes in 119 patients with ALL were included. A proven, probable, possible and modified possible IFI was diagnosed in 56 (45.5%), 22 (17.9%), 39 (31.7%), 6 (4.9%) episodes, respectively. The prevalence was 9.7% (95% CI 8-11.4%) overall and 23.5% (95% CI 14.5-32.5%) for relapsed/refractory ALL. For non-relapsed ALL the IFI prevalence was significantly higher for children with high-risk compared to standard-risk ALL (14.5% vs 7.3%, p=0.009), and IFI were more common during induction, consolidation and delayed intensification phases. Mould infections occurred more frequently than non-mould infections. Thirteen children (10.9%) died within 6 months of IFI diagnosis with 5 deaths (4.2%) attributable to an IFI.

Conclusions: IFI is more common in children with high-risk ALL and in relapsed disease. Overall survival was encouraging, with IFI contributing to very few deaths.

INTRODUCTION

Acute lymphoblastic leukaemia (ALL) is the most common cancer in children and adolescents.[1] Over the last five decades, refinements in risk-directed therapy and use of allogeneic haematopoietic stem cell transplant (HSCT) have led to dramatic improvements in treatment outcomes, with five year survival as high as 90% for certain risk groups.[2] However, despite improvements in supportive care, treatment-related deaths occur in one of every 25 children treated for ALL.[3] An important cause of morbidity and mortality in children with ALL is infection, of which invasive fungal infection (IFI) remains one of the most challenging to prevent, diagnose and treat.[4]

Predictors for IFI in children with cancer include neutropenia, HSCT, graft versus host disease and prolonged corticosteroid exposure.[5,6] The degree of IFI risk has also been ascribed to underlying cancer diagnosis, with overall risk often considered low for ALL.[5] However, children with ALL are a heterogeneous group and this low risk categorisation may not be applicable to patients with underlying high-risk leukaemia and during intensive chemotherapy phases.[7] To date, few paediatric studies have explored the changing pattern of IFI risk during ALL treatment phases and between risk categories with many combining data for all types of acute leukaemia and HSCT.[8-13]

The prevalence of IFI in children with ALL is reported to range between 4 and 35%, depending on era, chemotherapy protocol, risk category and prophylaxis regimen.[8-13] In an early Australian study, the IFI prevalence in children with high-, standard- and low-risk ALL of was 35%, 30% and 6%, respectively.[13] However this included patients treated on an intensive chemotherapy protocol that is now no longer used and may, in part, explain the high IFI rates observed.[14] An evaluation in the US of infection-related complications in children with ALL identified an increased incidence of IFI during the more intensive induction and re-induction treatment phases.[7] Similarly, a recent European study

of children with ALL found the majority of proven and probable IFI occurred during the induction phase, with an overall attributable mortality of 13%.[8] However, the proportion of patients with relapsed ALL or post HSCT were not reported in several of these studies and may have impacted the overall rates.

The Epidemiology and Risk Factors for Invasive Fungal Infections in Immunocompromised Children (TERIFIC) study is a retrospective, multi-centre, cohort study of IFI in immunocompromised children in Australia. The epidemiological data from this large cohort has been previously described.[15] The aim of this study is to describe the prevalence and outcomes of IFI in children with ALL.

METHODS

Episodes of IFI in immunocompromised children at four Australian tertiary paediatric centres in Brisbane (Queensland Children's Hospital), Melbourne (Royal Children's Hospital), Perth (Perth Children's Hospital) and Sydney (Sydney Children's Hospital) were retrospectively identified (Perth enrolled between 2003 to 2014, Brisbane and Melbourne enrolled between 2004 to 2014, and Sydney between 2012 to 2013).

Participants. Detailed methodology, including participant identification and inclusion and exclusion criteria have been described.[15] Briefly, children with IFI were identified from hospital pharmacy, microbiology and oncology databases and hospital diagnostic coding. Data collected included patient demographics, IFI details, clinical management and treatment outcome over a 6-month period from date of IFI diagnosis. Data were entered into a centralised and secure web-based application. Ethics approval was granted at each individual site.

Invasive fungal infections were classified as proven, probable or possible as per the European Organization for Research and Treatment of Cancer/Invasive Fungal Infections Cooperative Group (EORTC) criteria.[16] A positive *Aspergillus* polymerase chain reaction (PCR) was also included as mycological evidence within the diagnostic criteria for probable IFI. Although not included in the current EORTC criteria, the EORTC/MSG (National Institute of Allergy and Infectious Diseases Mycoses Study Group) have proposed future inclusion within the diagnostic criteria for probable invasive aspergillosis.[17] Furthermore, given the real-world difficulties of applying the EORTC criteria to children, a further category of 'modified possible' was also included in this analysis. 'Modified possible' was defined as (i) presence of host factors and clinical criteria suggestive of IFI not listed by EORTC criteria (i.e. lesions suggestive of hepatosplenic candidiasis with negative blood cultures) or ii) host and mycology criteria but no EORTC clinical criteria. Adapted. After primary data collection, children with a diagnosis of ALL were extracted from the database. IFI episodes occurring in children with ALL post HSCT were excluded from the analysis.

Across all sites, prophylaxis was physician dependant and not routinely prescribed to patients with ALL. Data on the total number of patients with ALL receiving prophylaxis was unavailable. Regarding diagnostic workup for IFI, patients were typically investigated after three to five days of persistent fever. Investigations available at all sites included radiological (x-ray, CT and MRI) and microbiological (fungal culture, galactomannan and Aspergillus PCR). Investigations performed were physician dependent and individualised to the patient and presenting symptoms.

Denominator data. The total number of patients diagnosed with ALL at each centre during the study period was collected from Perth (n=224), Brisbane (n=375) and Melbourne (n=617). Denominator

data were unavailable for Sydney. Compared to our previous publication, denominator data from Perth were also included for the year 2003.[15]

To allow for risk group comparison, data were collected on the number of children treated with specific ALL protocols. The treatment protocols were classified by two paediatric haematologists (RSK, AM) into standard risk (Children's Oncology Group (COG) trials AALL0331, AALL0932 and the Children's Cancer Group (CCG) 1991 trial) and high risk (COG trials AALL0631, AALL1122, AALL0232, AALL1131, AALL0031, AALL0434, AALL08P1, P9407 and the CCG1961 trial). The standard-risk protocols incorporated all children that were classified as standard, average or low risk, whereas the high-risk protocols incorporated all children that were classified as high or very high risk according to COG criteria. For children treated on alternative protocols, the ALL risk status documented in the medical record was used.

Data were obtained from individual centres regarding the number of patients treated on high-risk protocols (Perth, 88; Brisbane, 153), standard-risk protocols (Perth, 113; Brisbane, 200), and relapse protocols (Perth, 23; Melbourne 63). Data on number of patients treated on high- and standard-risk protocols were unavailable for Melbourne.

Statistical methods. Descriptive statistics were presented using median and interquartile range (IQR) for continuous data and frequency and percentage for categorical data. Chi-square test or Fisher's exact test were used to assess the relationship between two categorical variables.

Crude prevalence of IFI in ALL was calculated by dividing the number of patients with IFI by the total number of new patients diagnosed with ALL during the study period and was reported with 95% confidence interval (CI). Due to availability of specific denominator data, IFI prevalence for relapsed disease was calculated for Melbourne and Perth and for standard- and high-risk disease for Brisbane and Perth. Overall survival was calculated from the earliest date that the IFI was suspected or confirmed to date of death from all causes or date of last follow-up with patients censored at six months. Mean overall survival was calculated using Kaplan-Meier estimates and reported with standard error (SE) and Kaplan-Meier curves. Median time from diagnosis of ALL to the first documented diagnosis or treatment for IFI was calculated and reported with IQR. All tests were 2-tailed and a p-value of <0.05 was considered statistically significant. Analyses were performed using R statistical software.[18]

RESULTS

There were 348 episodes of IFI in 331 children. Of these, 149 episodes occurred in 142 children with ALL. Twenty-six IFI episodes occurred in children who had undergone HSCT for ALL a median of 48.5 days (IQR 9.5-274 days) prior to IFI diagnosis. After excluding these, there were 123 episodes in 119 patients included in this analysis (table 1). Ninety-four IFI episodes occurred in 94 patients with primary, non-relapsed disease and 29 IFI episodes occurred in 25 patients with relapsed or refractory disease (4 patients had two IFI's).

Of the children with B-cell ALL, the most common standard risk study protocol was COG AALL0331 (NCT00103285) (51.4% of all episodes with standard risk B-cell ALL) and the most common high-risk study protocol was COG AALL0232[19] (50.0% of all episodes with high risk B-cell ALL). For children with T-cell ALL, the most common protocol was AALL0434[20] (85.7%). For children with relapsed

disease the most common treatment protocol was UKALLR3[21] (n=16, 55.2%). Prevalence of IFI according to each treatment protocol is available in Supplementary Appendix 1.

Overall, a proven, probable and possible IFI was diagnosed in 56 (45.5%), 22 (17.9%) and 39 (31.7%) episodes, respectively. A modified-possible IFI was diagnosed in 6 episodes (clinical details in Supplementary Appendix 2). In the 94 patients with non-relapsed ALL a proven, probable, possible and modified-possible IFI was diagnosed in 42 (44.7%), 18 (19.1%), 29 (30.9%) and 5 (5.3%), respectively. For these 94 patients, 28 (29.8%) occurred during induction, 26 (27.7%) during consolidation and 25 (26.6%) during delayed intensification (Figure 1). The median time from diagnosis of ALL to IFI diagnosis was 3.3 months (IQR 1-7.5 months).

The median age was 6.5 years (IQR 3-11 years), 8 years (IQR 6-14 years) and 9 years (6.75-9.75 years) in the proven/probable group, possible and modified-possible group, respectively. For the non-relapsed group, there was no difference between the proven/probable, possible and modified-possible groups with respect to disease risk status, with 34 patients (57%) in the proven/probable group, 21 (72%) in the possible group and 4 (80%) in the modified-possible group being high-risk (p=0.29). Similarly, no difference was found between the three groups with respect to treatment phase (Intensive vs. Maintenance, p=0.27). A total of 50 (86%) proven/probable IFIs (with a known treatment phase), 28 (97%) possible IFIs and 3 (100%) modified-possible IFIs were identified in intensive chemotherapy phases.

Prevalence

The IFI prevalence was calculated for episodes occurring in Melbourne, Brisbane and Perth. During the study period, there were 1216 patients with ALL treated at these three sites (Table 2). Overall the prevalence was 9.7% (95% CI 8-11.4) for proven/probable/possible/modified-possible IFI and 6.2% (95% CI 4.8-7.5) for proven/probable IFI. There was no significant difference in the overall IFI prevalence between centres (p>0.05, data not shown).

The IFI prevalence in standard-risk ALL was 7.3% (95% CI 4.5-10.2) for proven/probable/possible/modified-possible IFI and 5.8% (95% CI 3.2-8.3) for proven/probable IFI. The IFI prevalence in high-risk ALL was 14.5% (95% CI 10.1-19.0) for proven/probable/possible/modified-possible IFI and 8.7% (95% CI 5.1-12.3) for proven/probable IFI (mould n=17; yeast n=4). IFI prevalence was significantly higher for children with high-risk ALL compared to those with standard-risk ALL (14.5% vs 7.3%; p=0.009). Considering the individual centres separately, the IFI prevalence in high-risk ALL was 12.5% (95% CI 9.9-21.4) in Perth and 15.7% (95% CI 9.9-21.4) in Brisbane (p=0.62). The IFI prevalence for standard risk ALL was 3.5% (95% CI 0.1-6.9) for Perth and 9.5% (5.4-13.6) in Brisbane (p=0.08).

The prevalence in relapsed/refractory ALL was 23.5% (95% CI 14.5-32.5) for proven/probable/possible/modified-possible IFI and 12.9% (95% CI 5.8-20.1) for proven/probable IFI (mould n=7; yeast n=4). For non-relapsed disease, the prevalence was 6.2% (95% CI 4.4-7.9) for proven/probable/possible/modified-possible IFI and 3.8% (95% CI 2.4-5.1) for proven/probable IFI (mould n=18; yeast n=10). The IFI prevalence was significantly higher in patients with relapsed/refractory ALL than in non-relapsed ALL (23.5% vs 6.2%; p<0.001) for

proven/probable/possible/modified-possible IFI and for proven/probable IFI (12.9% vs 3.8%; p=0.001).

Microbiology

There were 82 fungal pathogens (54 mould and 28 non-mould) identified by culture, molecular techniques or histopathology in the 78 episodes with a proven or probable IFI (Table 3) (3 episodes had 2 mould pathogens and 1 episode had 2 non-mould pathogens). *Aspergillus* species was the most frequently identified mould, followed by *Lomentospora* (formerly *Scedosporium) prolificans* and *Exserohilum* species. Among the non-mould infections, *Candida* species accounted for the majority of infections, of which *Candida albicans* and *Candida parapsilosis* were most frequently detected. There was no significant relationship between disease status (relapse versus non-relapse) and type of fungus (p=0.58). Mould infection accounted for 38/60 (63.3%) episodes in patients with non-relapsed disease compared to 13/18 (72.2%) episodes in relapsed/refractory disease.

Among the proven/probable mould IFI, the most frequent primary site of infection was lung (n=31), followed by skin and soft tissue (n=10), sinonasal (n=3), central nervous system (n=3), blood (n=2) and bone (n=2). For non-mould infections, the most frequent site of infection was blood (n=27).

Impact of prophylaxis

Overall, there were 34 (27.6%) IFI episodes in which anti-fungal prophylaxis had been administered for at least seven consecutive days in the month prior to diagnosis of IFI. The median duration of prophylaxis prior to IFI diagnosis was 32 days (IQR 18-92 days, range 8-215 days).

A total of 89 (72.4%) IFI episodes occurred in patients on no prophylaxis, of which 31 (34.8%) were mould and 28 (31.4%) were non-mould (no pathogen identified in 30). Twenty-six (21.1%) IFI episodes occurred in patients on fluconazole prophylaxis, of which 17 (65.4%) were mould and two (7.7%) were non-mould (*Pichia kudriavzevii* and *Rhodotorula* species) (no pathogen identified in 7). *Aspergillus* species was cultured in 10 of the 17 (58.8%) mould infections, with a further four patients having indirect evidence of *Aspergillus* species with a positive PCR. Finally, eight (6.5%) IFI episodes occurred in patients on mould-active prophylaxis (intermittent liposomal amphotericin n=3, itraconazole n=5). Of the episodes occurring on mould-active prophylaxis, two had pulmonary aspergillosis (*Aspergillus niger* and *Aspergillus flavus*) diagnosed by core biopsy, one had *P. kudriavzevii* (formerly *Candida krusei)* cultured from the blood and two had probable pulmonary aspergillosis with positive galactomannan on bronchoalveolar lavage specimen (no pathogen identified in 3). Therapeutic drug levels for itraconazole prophylaxis were only performed in two patients and both were subtherapeutic.

Treatment and outcome

The median duration of antifungal treatment was 131 days (IQR 37.5-236.5) overall and 157 days (IQR 24.7-264.0) for proven IFI, 108 (IQR 58.5-197.0) for probable, 108 (IQR 41.5-188.0) for possible and 275 (IQR 185.8-317.8) for modified possible IFI.

Thirteen (10.9%) children died within 6 months of IFI diagnosis, 6 of whom had relapsed disease and 2 had refractory ALL (Figure 2). Five deaths (4.2%) were attributed to IFI. Causative organisms in these five patients included *Scedosporium prolificans* (n=2), *Aspergillus flavus* (n=1), *Aspergillus fumigatus* complex (n=1), *Trichosporon asahii* and *Candida parapsilosis* (both infections in one patient). The mortality according to pathogen for patients with proven *L. prolificans* and *Aspergillus* spp was 50.0% (n=2/4) and 17.6% (n=3/17), respectively.

DISCUSSION

In this large, multicentre study of IFI in children with ALL, the overall IFI prevalence was 9.7%. The prevalence was significantly higher in children with relapsed or refractory ALL compared to non-relapsed disease (23.5% versus 6.2%). It was also higher for children treated on high-risk chemotherapy protocols compared to standard-risk (14.5% versus 7.3%) and most infections occurred during the dose-intensive chemotherapy phases. Across all groups, mould infections occurred more frequently than non-mould infections.

Together with our previously published paediatric study describing the epidemiology of IFI in immunocompromised Australian children, this is one of the few attempts at extending EORTC criteria to accommodate the nuances of IFI diagnosis in children.[15] This particularly relates to the difficulties in obtaining computed tomography or magnetic resonance imaging in those requiring a general anaesthetic,[22] as well as the absence of typical nodular and ground glass radiological changes in the younger population.[23,24] Furthermore, the inclusion of a 'modified-possible' group reflects the realities of clinical practice where patients are treated for IFI without fulfilling criteria originally intended for research purposes.[16] While highly specific criteria are important for clinical trials assessing new diagnostic markers or drugs, they may be less relevant to understanding the

epidemiology of IFI in children with cancer, the overall clinical impact of these infections and to informing prospective, real-time surveillance and prophylaxis strategies.

To date, few paediatric studies have explored the contribution of chemotherapy intensity and ALL risk status on the prevalence of IFI. This may, in part, explain the broad range of prevalence previously described.[8-13], In keep with our results, a recent publication of children with ALL treated on the UKALL2001 protocol found the IFI rate was significantly higher in children treated with a more intensive chemotherapy regimen (11% versus 4%).[25] Similarly, the study also showed the majority of IFI occurred in the dose-intense treatment phases of induction, consolidation and delayed intensification that are typically associated with prolonged periods of neutropenia and high-dose steroids.

Limited conclusions can be drawn from this study regarding the impact of antifungal prophylaxis on development of IFI as the total number of patients treated for ALL that received antifungal prophylaxis is unknown. The high prevalence of IFI in children with relapsed/refractory ALL in our study is in keeping with guidelines recommending routine antifungal prophylaxis in this group.[26] In contrast, few paediatric guidelines recommend prophylaxis for children with non-relapsed ALL on high-risk treatment protocols. Our study suggests that that this group may also benefit from antifungal prophylaxis, particularly during the more intensive chemotherapy phases. While choice of antifungal agent is frequently limited by drug-chemotherapy interactions, mould-active cover should be considered due to the proportionally higher rates of mould infections in this group. Given the complexities of antifungal use in this population, particularly around adverse effects, drug interactions, optimal dosing and therapeutic drug monitoring, further research in this area is urgently required.[26,27]

Overall survival in this ALL cohort is encouraging, with a 6-month IFI attributable mortality rate of 4.2%. At an individual pathogen level, 6-month mortality associated with proven invasive aspergillosis (17.6%) was also better than previously reported, with mortality as high as 50%.[28,29] Unfortunately our study did not capture the challenges associated with treating these infections, including interruptions to chemotherapy, requirements for dose adjustments and drug interactions. As treatment durations extended beyond 130 days for half of these episodes, these are likely to be significant.

Whilst our study is one of the larger reports of IFI in children undergoing treatment for ALL, it is limited by its retrospective nature. Prevalence assessments for ALL risk groups were grouped according to site data availability, although when considered separately there was no significant difference between sites. There was, however a trend toward a higher IFI prevalence at Brisbane, which is unlikely to be explained by differences in approaches to prophylaxis or diagnostic workup as these were similar between sites. Furthermore, our overall prevalence of 9.7% was slightly different to the 10.6% reported in our previous publication, due to our inclusion of 2003 data from the Perth site.[15] However, data produced from the TERIFIC study cohort is unique as it is the first to define a separate diagnostic group (modified possible IFI) that accommodates IFI diagnostic challenges in children as well as the realities of treating clinical findings suspicious of IFI in immunosuppressed children.[15] Moving forward, such criteria may be useful in informing epidemiological studies as well as surveillance criteria in this population.

This study provides further insights into the burden of IFI in children with ALL and highlights the importance of considering standard- and high-risk subgroups, as well as dose-intense treatment

phases, separately. Our study may also inform prophylaxis strategies for children with relapsed ALL and those on high-risk chemotherapy protocols. With the rapid evolution of ALL treatment a collaborative approach is required to ensure we remain abreast of changing patterns of IFI in this population and consideration should be given to inclusion of additional criteria, such as 'modified possible,' into definitions used to monitor prevalence rates.

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FIGURE LEGENDS

Figure 1. IFI diagnoses according to ALL treatment phase, n=94. *IFI, invasive fungal infection; ALL, Acute Lymphoblastic Leukaemia; IM, Interim Maintenance; DI, Delayed Intensification; IM2, Interim Maintenance 2.*

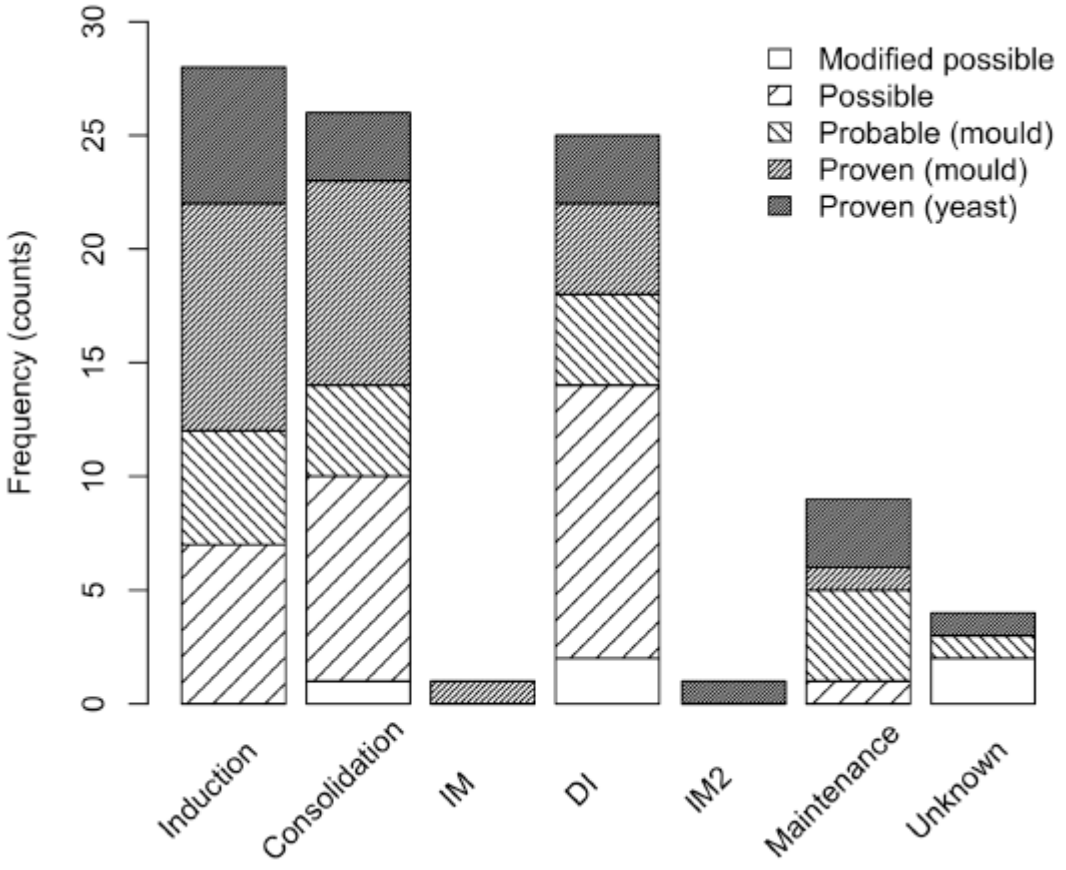

Figure 2. Overall Survival for patients with Acute Lymphoblastic Leukaemia

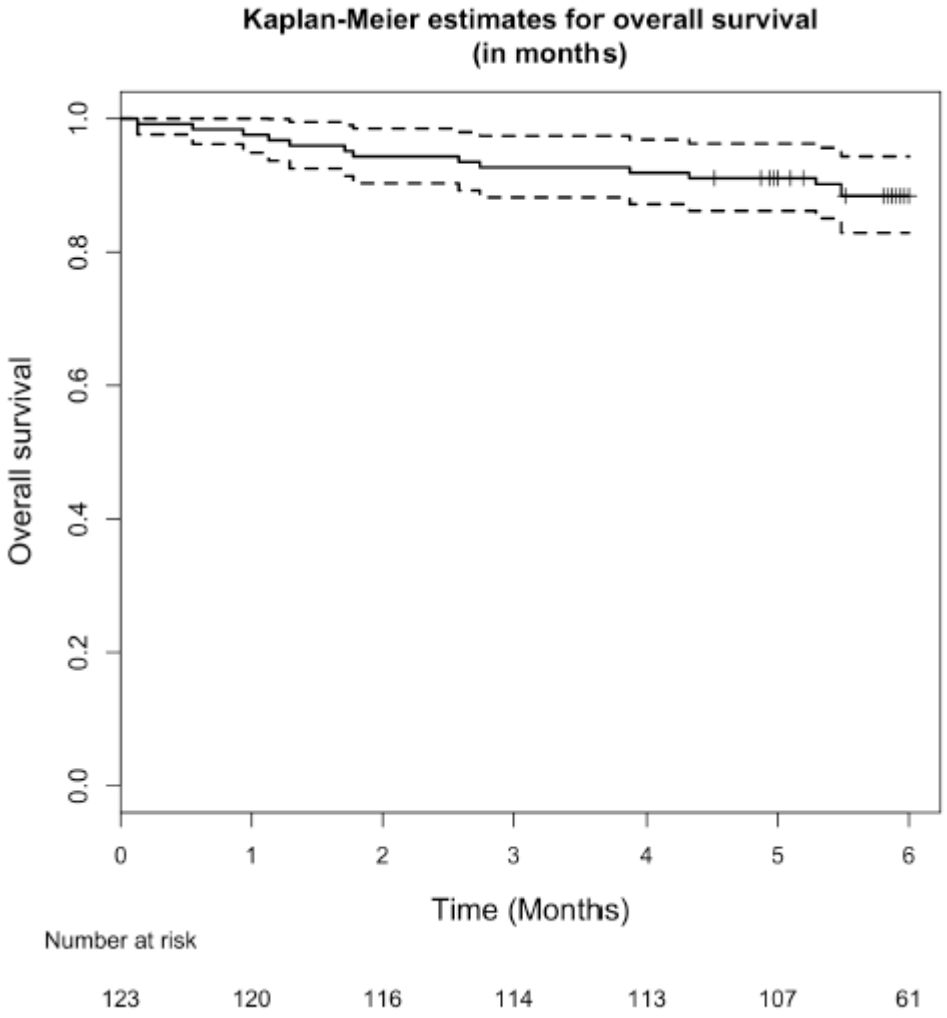

TABLE 1 Patient Characteristics per IFI Episode

Total number of IFI episodes	**123**
Median age, years (IQR)	7 (4-12)
Male sex, n (%)	63 (51.2)
Diagnosis, n (%) - Primary B cell ALL → Standard risk → High risk - Primary T cell ALL - Relapsed/refractory ALL → B cell → T cell	 87 (70.7) 35 52 7 (5.7) 29 (23.6) 24 5
Treatment phase in non-relapsed disease, n (%) Induction Consolidation Interim maintenance Delayed intensification / Reinduction Maintenance Unknown	94 28 (29.8) 26 (27.7) 2 (2.1) 25 (26.6) 9 (9.6) 4 (4.2)
CVAD in situ, n (%)	113 (91.9)
Fungal prophylaxis, n (%) - Yeast-active: Fluconazole - Mould-active - Itraconazole - Liposomal amphotericin	34 (27.6) 26 (76.5) 8 (23.5) 5 3
Neutropenia (<0.5 μmol/L) within 30 days, n (%) Median duration of neutropenia prior to IFI diagnosis, days (IQR)	116 (94.3) 16 (11-27)

IFI, Invasive Fungal Infection; IQR, Inter-Quartile Range; ALL, Acute Lymphoblastic Leukaemia; CVAD, Central Venous Access Device

TABLE 2 IFI episodes according to study site and ALL risk status

		Melbourne	Brisbane	Perth	Sydney	Total*
Patients diagnosed with ALL during study period	**TOTAL**	**617**	**375**	**224**	**NA**	**1216**
	SR	545	200	113	NA	1099
	HR		153	88	NA	
	Relapsed/refractory	62	NA	23	NA	85
	Infant	10	NA**	NA**	NA	10
	Unknown	-	22	-	NA	22
Proven	**TOTAL**	**14**	**29**	**11**	**2**	54
	SR	4	14	3	0	21
	HR	3	8	5	1	16
	Relapsed/refractory	5	7	2	0	14
	Infant	2	0	1	1	3
Probable	**TOTAL**	**12**	**7**	**2**	**1**	21
	SR	3	1	0	1	4
	HR	6	5	1	0	12
	Relapsed/refractory	3	0	1	0	4
	Infant	0	1	0	0	1
Possible	**TOTAL**	**21**	**11**	**5**	**2**	37
	SR	3	3	1	1	7
	HR	9	7	4	1	20
	Relapsed/refractory	9	1	0	0	10
	Infant	0	0	0	0	0
Modified Possible	**TOTAL**	**1**	**5**	**0**	**0**	6
	SR	0	2	0	0	2
	HR	1	2	0	0	3
	Relapsed/refractory	0	1	0	0	1

	Infant	0	0	0	0	0

ALL, Acute Lymphoblastic Leukaemia; NA, Not Available; SR, Standard Risk; HR, High Risk

**data from Sydney not included in Total; **included in the high risk group*

TABLE 3 Details of proven and probable IFI

	Relapsed disease		**Non relapsed disease**	
	Proven	**Probable**	**Proven**	**Probable**
Mould				
-*Aspergillus fumigatus complex*	3	1	3	4
-*Aspergillus flavus*	2	-	1	1
-*Aspergillus niger*	-	-	1	-
-*Aspergillus* spp	1	3^	2	11^^
-*Exserohilum* spp	-	-	4	-
-Zycomycetes			2	-
- *Lomentospora* prolificans	1*	-	3	-
-*Fusarium* spp	-	-	1	-
-*Acremonium* spp	-	-	1	-
-*Penicillium* spp	-	-	1	-
-*Paecilomyles lilacinus*	-	-	1	-

-Hyphomycete	-	-	-	1
-Hyphae on tissue biopsy	3	NA	2	NA
Non-mould				
-*Candida albicans*	3	-	7	-
-*Candida parapsilosis*	1	-	6	-
-*Pichia kudriavzevii*	1	-	1	-
-*Candida guillermondii*	-	-	2	-
-*Candida tropicalis*	-	-	2	-
-*Candida* spp.	-	-	1	-
-*Candida rugosa*	-	-	1	-
-*Cryptocococcus neoformans*	-	-	1	-
-*Trichosporon asahii*	-	-	1**	-
- *Rhodotorula* spp	-	-	1	-

*1 episode had 2 pathogens (*A. fumigatus* and *L. prolificans*)

**1 episode had 2 pathogens (*T. asahii* and *C. rugosa*)

^3 identified on Polymerase Chain Reaction (PCR); ^^10 identified by PCR and 1 by galactomannan on Bronchoalveolar Lavage (BAL) specimen