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Early and treatment-related deaths in childhood acute myeloid leukaemia in the Nordic countries: 1984–2003

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Summary

Despite major improvements in the cure rate of childhood acute myeloid leukaemia (AML), 5–15% of patients still die from treatment-related complications. In a historical prospective cohort study, we analysed the frequency, clinical features and risk factors for early deaths (ED) and treatment-related deaths (TRD) in 525 children included in the Nordic Society of Paediatric Haematology and Oncology (NOPHO)-AML-84, -88 and -93 trials. Seventy patients (13%) died before starting treatment or from treatment-related complications. The death rate rose from 11% in NOPHO-AML-84 to 29% in -88, but then fell to 8% in -93. Sixteen patients (3%) died within the first 2 weeks, mainly from bleeding or leucostasis. Hyperleucocytosis, age <2 or ≥10 years were risk factors. After day 15, 10% of patients died from treatment-related complications with infection as the main cause of death. Risk factors were age <2 or ≥10 years and treatment according to the NOPHO-AML-88 protocol. The number of EDs and TRDs in AML is high. Therefore optimal antifungal prophylaxis is essential, and studies on the benefit of antibacterial prophylaxis and individual risk factors for ED and TRD are needed.

Keywords: acute myeloid leukaemia, children, early deaths, treatment-related mortality, infection.

The last two decades have seen a considerable improvement in the prognosis for children with acute myeloid leukaemia (AML) owing to the introduction of more intensive chemotherapy, better risk-group stratification and improved supportive care. The overall survival rate in childhood AML has reached 60–65% (Kaspers & Creutzig, 2005). Most deaths are due to progressive disease, but 5–15% die from toxic effects of treatment (Riley *et al*, 1999; Creutzig *et al*, 2004; Rubnitz *et al*, 2004; Slats *et al*, 2005). Further improvement in AML survival rates will probably require the introduction of individualized therapy, where targeted and more leukaemia-specific drugs are used in an effort to arrest progressive disease, in combination with better supportive care to prevent early deaths (ED) and treatment-related deaths (TRD).

The aim of the present study was to identify the frequency, clinical features and possible risk factors for EDs and TRDs in childhood AML in the Nordic countries.

Patients and methods

Eligibility and methods

Since July 1984, all children diagnosed with AML in the Nordic countries (Denmark, Finland, Iceland, Norway and Sweden) have been registered and treated on common protocols conducted by the Nordic Society of Paediatric Haematology and Oncology (NOPHO)-AML Study Group. The study is population-based for patients younger than 15 years of age, whereas 15- to 18-year-old patients are enrolled according to local practice. Patients diagnosed between 1 July 1984 and 31 December 2003 were identified in the database and included in the present historical prospective cohort study. Excluded were patients treated according to other protocols than NOPHO-AML-84/88/93 or pre-treated with cytostatic drugs for more than 14 d, patients with myeloid leukaemia of Down syndrome, Fanconi anaemia, Kostmann syndrome, extrame-

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dullary myeloid tumour without significant bone marrow involvement, preceding myelodysplastic syndrome or therapy-related AML. A total of 525 patients fulfilled the inclusion criteria. Among these, 489 were younger than 15 years at the time of diagnosis and 36 were 15–18 years old. The patients were diagnosed in 21 hospitals in Denmark ($n = 114$), Finland ($n = 97$), Iceland ($n = 6$), Norway ($n = 106$) and Sweden ($n = 202$). Allogeneic transplants were performed in eight centres.

The Nordic AML database contains information about the time and cause of death, and 10 children who died before starting treatment, and 78 who died from treatment-related complications were identified. Information about patients registered as dying from treatment-related complications in second or third complete remission is not reported. Additional data concerning the remaining 70 patients were collected by reviewing each patient's file using a specific registration form. The files of 29 patients registered as non-responders were reviewed to verify the correctness of the data. Leukaemia-related data, such as white blood cell count (WBC), platelet count and presence of central nervous system (CNS) leukaemia at diagnosis, French-American-British (FAB)-type and cytogenetics were collected from the database. Performance status at diagnosis was not available.

NOPHO-AML-84/88/93 treatment

The treatment regimens of the NOPHO-AML trials are shown in Fig 1 and the treatment elements including cumulative drug doses are summarized in Tables I and II. Details concerning patient characteristics and clinical outcome have been reported elsewhere (Lie *et al*, 2005).

The NOPHO-AML-84 protocol was of moderate intensity with an induction of three courses of cytarabine, 6-thioguanine and doxorubicin followed by four consolidation courses with high-dose cytarabine. To reduce the frequency of non-response and relapse in the NOPHO-AML-88 study, etoposide and mitoxantrone were added in selected courses during induction and consolidation, and the interval between induction courses was reduced. The event-free survival rose from 29% to 41%, but the NOPHO-AML-88 protocol suffered from an unacceptably high frequency of toxic deaths during induction and in early consolidation. The NOPHO-AML-93 used the same induction blocks as the NOPHO-AML-88, but the dose intensity was modified: children with a good response after the first course were given their second course only following haematological recovery. Consolidation was identical with NOPHO-AML-88. The event-free survival at 5 years was 48% in the NOPHO-AML-93 protocol.

Intrathecal methotrexate was given on the first day of all courses.

In the NOPHO-AML-84/88/93 studies, allogeneic haematopoietic stem cell transplantation (HSCT) in first complete remission (CR1) was recommended to all patients with a human leucocyte antigen (HLA)-matched family donor. For patients without a donor, autologous HSCT was optional in the NOPHO-AML-84, -88 and the first part of -93, and consequently, autologous HSCT was performed on a non-randomized basis decided by the responsible physician. HSCT conditioning regimens and supportive care followed local guidelines.

Recommendations for supportive care measures in the NOPHO-AML-84/88/93 protocols included trimethoprim/sulfamethoxazole prophylaxis for *Pneumocystis jiroveci*. Systemic antifungal prophylaxis and granulocyte-colony-stimulating factor (G-CSF) were not recommended. Cytoreduction prior to chemotherapy was performed at the discretion of the attending physician. All-*trans*-retinoic acid (ATRA) was recommended for patients with acute promyelocytic leukaemia (APL) as from 1999.

Definitions

The *diagnosis of AML* was established by morphological analysis of bone marrow (BM) aspirates according to the FAB and WHO classifications.

Central nervous system (CNS) involvement was defined as more than five leucocytes per microlitre in the cerebrospinal fluid (CSF) in combination with detectable leukaemic cells in the cytospin and/or presence of neurological symptoms, such as cranial nerve palsy.

Complete remission (CR) was defined according to the CALGB criteria (Cheson *et al*, 1990) with minor deviations: ≤5% leukaemic blasts in the BM with signs of normal haematopoiesis in the BM and with clear signs of regeneration of normal blood cell production in the peripheral blood (PB) (platelet count $>80 \times 10^9$/l without transfusions and neutrophil count $>1{\cdot}0 \times 10^9$/l), and no leukaemic cells in the PB or in the CSF. Response after induction was evaluated close to day 15; a BM blast cell count <5% was considered as good response.

Non-responders (NR) included all patients surviving the first 42 d of treatment without achieving CR. Patients with CR criteria lasting <4 weeks were also classified as NR. NRs at day 42 ($n = 29$) were censored in the analysis of TRD.

Early death (ED) was defined as a fatal event occurring before or within the first 42 d of treatment. ED was subclassified into: (i) $ED_{0–14}$ before or during and after the first therapy course (<15 d of treatment) and (ii) $ED_{15–42}$ in aplasia between days 15–42 from treatment start. This classification reflects the ED rate and its association with initial problems (hyperleucocytosis, leucostasis) or aplasia after induction therapy.

Treatment-related death (TRD) was defined as death unrelated to progressive disease occurring after day 42 of treatment start in patients who achieved CR. In this paper, only TRDs in CR1 are reported.

Hyperleucocytosis was defined as WBC $\geq 100 \times 10^9$/l at diagnosis.

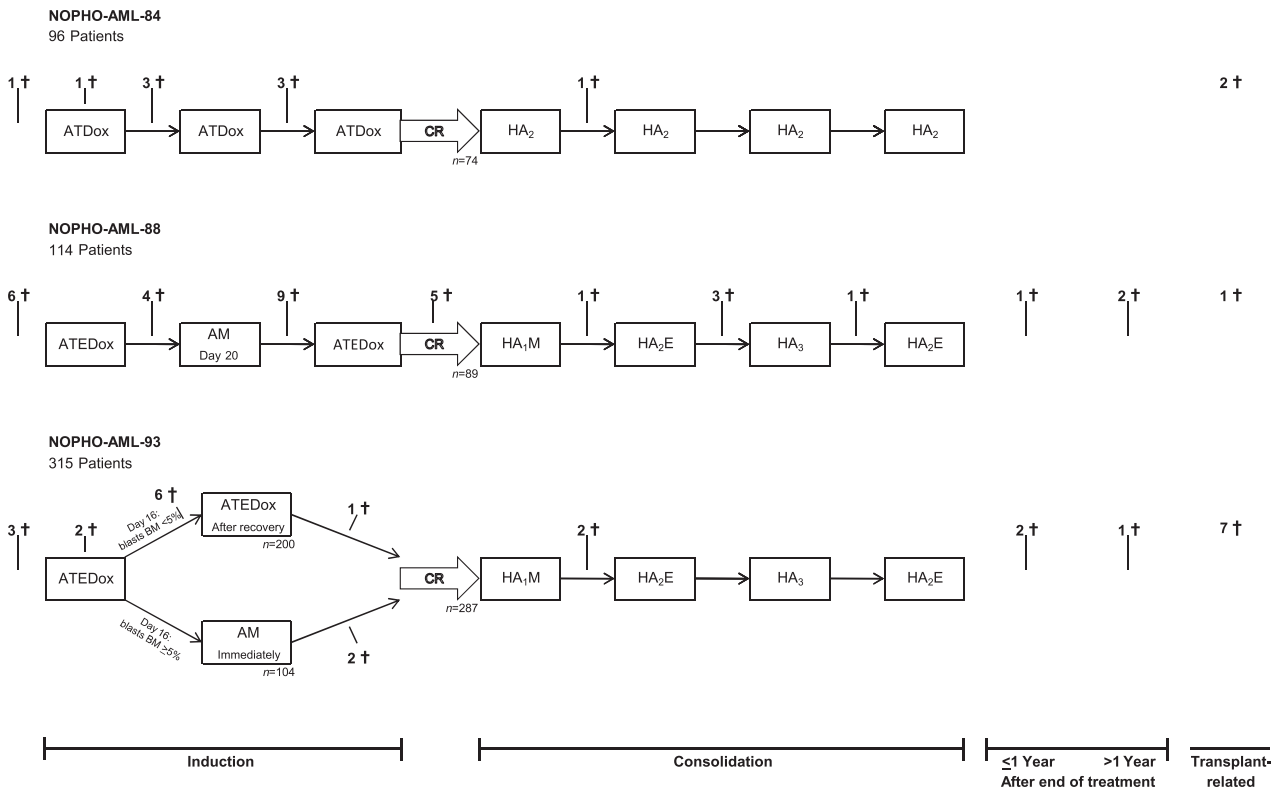

Fig 1. Flow diagram of the NOPHO-AML-84/88/93 protocols with indication † of early deaths and treatment-related deaths (n = 70). The treatment elements are described in Tables I and II. Among patients who suffered ED day 0–14; 10 died before starting treatment, three during the first induction, and three after finishing the first induction course. ED day 15–42 included seven patients dying after the first induction course and eight after the second. Abbreviations: A, cytarabine; T, 6-thioguanine; Dox, doxorubicin; CR, complete remission; *n*, number; HA, high-dose cytarabine; E, etoposide; M, mitoxantrone; BM, bone marrow; ED, early death; TRD, treatment-related death.

Table I. NOPHO-AML-84/88/93 treatment elements.

ATDox	Cytarabine 100 mg/m^2 per d continuous infusion day 1–4 6-Thioguanine 100 mg/m^2 orally every 12 h day 1–4 Doxorubicin 75 mg/m^2 8-h infusion day 5
HA_2	Cytarabine 2 g/m^2 2-h infusion every 12 h day 1–3
ATEDox	Cytarabine 200 mg/m^2 per d continuous infusion day 1–4 6-Thioguanine 100 mg/m^2 orally every 12 h day 1–4 Etoposide 100 mg/m^2 per d continuous infusion day 1–4 Doxorubicin 75 mg/m^2 8-h infusion day 5
AM	Cytarabine 100 mg/m^2 per d continuous infusion day 1–5 Mitoxantrone 10 mg/m^2 30-min infusion day 1–3
HA_1M	Cytarabine 1 g/m^2 2-h infusion every 12 h day 1–3 Mitoxantrone 10 mg/m^2 30-min infusion day 3–5
HA_2E	Cytarabine 2 g/m^2 2-h infusion every 12 h day 1–3 Etoposide 100 mg/m^2 per d 1-h infusion day 2–5
HA_3	Cytarabine 3 g/m^2 2-h infusion every 12 h day 1–3
CNS therapy	Methotrexate age-adjusted 6–12 mg intrathecal on the first day of all courses

For children <2 years of age, drug doses were calculated according to body weight with 1 m^2 equalling 30 kg.

Infections were categorized as clinically, radiologically or microbiologically documented. Viral disease was registered when both relevant disease symptoms and virus were detected. Fungal infections were defined as proven invasive infection (positive histopathological findings or positive culture) (Ascioglu *et al*, 2002).

Cytogenetics: Patients with t(8;21)(q22;q22), t(9;11)(p22;q23), t(15;17)(q22;q12-21), inv16(p13q22) and t(16;16)(p13;

Table II. NOPHO-AML-84/88/93 cumulative doses of cytarabine, etoposide and anthracyclines.

	Cytarabine (g/m^2)	Etoposide (mg/m^2)	Doxorubicin/Mitoxantrone Cumulative dose of anthracyclines* (mg/m^2)
NOPHO-AML-84	50·4	0	225/0 Cum. dose = 225
NOPHO-AML-88	50·1	1600	150/60 Cum. dose = 450
NOPHO-AML-93	GR: 49·6 PR: 61·3	GR: 1600 PR: 1600	GR: 150/30 PR: 75/60 Cum. dose GR = 300, PR = 375

GR, good responders; PR, poor responders.
*Calculated applying the following conversion factors to daunorubicin-equivalents: doxorubicin 1×, mitoxantrone 5×.

q22) were classified as having a favourable karyotype (Grimwade *et al*, 1998).

Statistics

Subgroups of patients were compared by Chi-square or Fischer's exact tests. Kaplan-Meier plots were used for survival analysis, including estimation of cumulative incidence of EDs and TRDs. The main event in the analysis of possible risk factors was composite endpoints consisting of $ED_{15–42}$ or TRD. Time was defined as time from day 15 after diagnosis to date of death. Patients who experienced a relapse ($n = 203$) or a second malignant neoplasm ($n = 2$) were censored at the time of this event. Patients without events were censored at the time of last follow-up. Cox proportional hazard regression analysis was performed with the composite endpoints as events and time to this as outcome measure. The following covariates were included: age at diagnosis (<2, 2–9, ≥10 years), WBC at diagnosis (<20, 20–99, ≥100 × 10^9/l) and protocol (NOPHO-AML-84 vs. NOPHO-AML-88 or NOPHO-AML-93). For patients who underwent HSCT, a time-dependent covariate was defined as zero before, and one after the date of HSCT, and for all other patients, this time-dependent covariate was defined as zero.

A stepwise, multiregression analysis according to Cox was also performed for composite endpoints consisting of $ED_{15–42}$ or TRD caused by infection. The analysis was adjusted for age at diagnosis and protocol.

The *P*-values reported are two-tailed, and *P*-values <0·05 were considered significant. The statistical analyses were performed using Stata Statistical Software, Release 10 (StataCorp 2007, College Station, TX, USA).

Results

In the Nordic countries, 13% (70/525) of children diagnosed with AML died before starting treatment or from treatment-related complications (Table III). ED and TRD rose from 11%

Table III. Number of early deaths, treatment-related deaths, non-responders and relapses in NOPHO-AML-84/88/93 protocols.

		NOPHO AML-84		NOPHO AML-88		NOPHO AML-93		Total	
Status	Period	No. of patients	%	No. of patients	%	No. of patients	%	No. of patients	%
No. of patients		96		114		315		525	
Early death (day 0–42)	ED day 0–14	3	3	7	6	6	2	16	3
	ED day 15–42	3	3	9	8	3	1	15	3
Treatment-related death (>day 42)	During chemotherapy								
	Induction	2	2	8	7	5	2	15	3
	Consolidation	1	1	6	5	4	1	11	2
	Transplant-related	2	2	1	1	7	2	10	2
	>1 year after end of treatment	0	0	2	2	1	0·3	3	1
Total ED/TRD		11	11	33	29	26	8	70	13
Death without CR (non-responders)	NR >day 42	14	15	1	1	14	4	29	6
Relapse (cumulative incidence)		44	46	39	34	120	38	203	39

ED, early death; TRD, treatment-related death; CR, complete remission; NR, non-responders.

in the NOPHO-AML-84 protocol to 29% in -88 and then declined to 8% in -93. The frequency of EDs and TRDs in the first half of the protocol period compared with the second half of the protocol was 9% vs. 14% in NOPHO-AML-84 ($P = 0{\cdot}5$), 35% vs. 24% in NOPHO-AML-88 ($P = 0{\cdot}2$), and 9% vs. 8% in NOPHO-AML-93 ($P = 0{\cdot}8$).

The analysis was performed in February 2010 with a median follow-up of 11·8 years (range, 2·8–25·1 years) for patients registered as alive.

Early death day 0–14

The main causes of $ED_{0–14}$ and characteristics of these patients are given in Table IV. The death rate was largely unchanged over time with 3%, 6% and 2% in the NOPHO-AML-84, -88 and -93 protocols, respectively ($P = 0{\cdot}4$ and $P = 0{\cdot}05$ when comparing NOPHO-AML-93 with -84 and -88, respectively). Ten (63%) patients died before protocol treatment was begun and three shortly after the first induction course. Among the 16 patients (3%) who died early, 11 (69%) died from intracranial haemorrhage, three (19%) from leucostasis and two (12%) from sepsis. Age was a risk factor for $ED_{0–14}$: 87% ($n = 14$) of the patients were <2 years or ≥10 years at diagnosis ($P = 0{\cdot}02$). Hyperleucocytosis was also a risk factor. It was present in 75% of the $ED_{0–14}$ patients compared with 15% of the AML cases in general ($P < 0{\cdot}001$). FAB M4 or M5 was not a significant risk factor for $ED_{0–14}$ compared with other FAB-types ($P = 0{\cdot}2$). However, 55% of the patients who died from a cerebral haemorrhage had FAB types M4 or M5, vs. 38% in the AML population ($P = 0{\cdot}1$).

Early death day 15–42

The principal cause of death among the 15 patients (3%), who died between days 15–42, was infection (93%) (Table V). Four (29%) already had signs of infection at the time of their last course of chemotherapy, which was the first induction course in one case. One patient died from a gastrointestinal bleed after the second induction course.

Treatment-related death (>day 42)

TRD occurred in connection with *chemotherapy* in 26 (5%) patients. The main cause of death was infection ($n = 22$, 85%) (Table V). Eight (36%) of these patients had signs of infection (fever and/or respiratory symptoms) when the last course of chemotherapy before death was being administered. One patient with FAB M4 died from cerebral bleeding. Two patients died from acute respiratory distress syndrome (ARDS). A 13-year-old girl died unexpectedly 1 d after the first consolidation course. She had been complaining of chest pain, but the autopsy revealed no signs of heart disease or other causes of death.

TRD occurred in ten (7%) of 150 patients *transplanted in CR1*. Eight (80%) of these deaths were caused by a severe infection. One patient died from veno-occlusive disease (VOD) and another from graft-*versus*-host-disease (GVHD) with bronchiolitis obliterans.

Three patients, all treated with chemotherapy only, died from TRD *more than 1 year after end of AML treatment*. A 13-year-old female died from cardiac failure 11 years after end of treatment. She had received a heart transplant 7 years prior to her death due to anthracycline-induced cardiomyopathy. A 17-year-old boy had candidiasis in the liver and spleen during induction. Later, liver cirrhosis with oesophageal varices developed. He died from multiorgan failure 7 years after end of treatment, following a month with symptoms of pulmonary infection. A 7-month-old girl with AML FAB M4, WBC 170×10^9/l and platelets 75×10^9/l presented at diagnosis with cerebral bleeding and died 12 years later as a consequence of severe brain damage.

Risk factors for $ED_{15–42}$ or TRD

Both univariate and multiple Cox regression analyses of the composite endpoints consisting of $ED_{15–42}$ or TRD showed that age at diagnosis was a significant prognostic factor (Table VI). Eighteen (13%) of 141 patients <2 years of age ($P = 0{\cdot}03$) and 23 (14%) of 169 patients ≥10 years ($P = 0{\cdot}01$) died from treatment-related complications as compared with 13 (6%) of 215 children between 2 and 9 years of age. Another risk factor was treatment according to the NOPHO-AML-88 protocol ($P = 0{\cdot}01$). The number of $ED_{15–42}$ or TRDs was 26 (23%) in the NOPHO-AML-88 as compared with 8 (8%) and 20 (6%) in the NOPHO-AML-84 and -93, respectively. WBC 20–99 $\times 10^9$/l at diagnosis seemed to be a risk factor as well, though this endpoint only reached statistical significance in the multiple regression analysis ($P = 0{\cdot}04$). FAB M3 was a risk factor for $ED_{15–42}$ or TRD in the multiple Cox regression analysis (11% vs. 5%, $P = 0{\cdot}05$), but only a few patients had this FAB type. Allogeneic HSCT was also associated with a slight excess risk of death from treatment-related complications (HR, 2·0; 95% confidence interval (CI), 0·8–4·9; $P = 0{\cdot}1$). Sex, platelet count and presence of CNS leukaemia at diagnosis, cytogenetics and size of the treating department were not statistically significant risk factors.

Death in FAB M3

Eight (29%) of 28 patients with FAB type M3 suffered ED or TRD, two (25%) within the first 2 weeks of diagnosis from either a CNS bleed before starting chemotherapy or ATRA (Table IV). The other six all died from infections. FAB M3 was not a significant risk factor compared with other FAB types for $ED_{0–14}$ (7% vs. 3%, $P = 0{\cdot}2$), but it was a significant risk factor for $ED_{15–42}$ or TRD (11% vs. 5%, $P = 0{\cdot}05$).

Table IV. Characteristics of patients who suffered early death day 0–14 in NOPHO-AML-84/88/93 protocols.

Patient no.	Survival after diagnosis (d)	Age at diagnosis	Sex	WBC at diagnosis ($\times 10^9$/l)	Platelets at diagnosis ($\times 10^9$/l)	Cytogenetics	FAB	Treatment	Causes of death
								NOPHO-AML-protocol, surgery, other	
1	0	9 years	F	550	69	Normal	M1	None	Bleeding (CNS)
2	0	11 years	M	290	45	Normal	M4	None	Bleeding (CNS)
3	1	0 months	F	450	20	Other	M4	No chemotherapy. Exchange transfusion	Leucostasis
4	1	15 years	F	41	15	No data	M3	None	Bleeding (CNS)
5	2	11 years	M	338	65	No data	M4	No chemotherapy. Neurosurgery	Bleeding (CNS)
6	2	14 years	M	173	76	t(15;17)(q22;q21)	M3	Started 1st induction	Bleeding (CNS)
7	2	15 years	F	365	12	No data	M1	None (Jehovah's Witness)	Bleeding (CNS)
8	3	0 months	F	400	No data	No data	M5	None	Leucostasis
9	3	7 months	F	442	35	Other *MLL* than t(9;11)	M0	Started 1st induction. Neurosurgery	Bleeding (CNS)
10	3	12 years	M	317	35	Normal	M4	Started 1st induction	Bleeding (CNS)
11	3	14 years	F	245	89	Normal	M5a	Cytoreduction (cytarabine, hydroxycarbamide)	Bleeding (CNS)
12	5	8 months	F	458	158	t(9;11)(p22;q23)	M5a	No chemotherapy. Neurosurgery	Bleeding (CNS)
13	9	8 years	M	247	17	Normal	M1	None (Jehovah's Witness)	Leucostasis
14	11	4 months	F	10	10	No data	RAEB-T	1st induction. Antibiotics for 3 d before death, no antifungal treatment	Sepsis (Pseudomonas aeruginosa)
15	11	11 years	F	7	108	Normal	M4	1st induction. Antibiotics for 7 d, antifungals for 1 d before death	Sepsis (no positive culture), bleeding (lungs)
16	14	3 months	M	30	22	No data	M5	1st induction	Bleeding (CNS)

WBC, white blood count; F, female; M, male.

Table V. Causes of early deaths and treatment-related deaths in NOPHO-AML-84/88/93 protocols.

Status	Period	No. of patients	Bleeding/ Leucostasis	Infection				Clinically/radiologically documented, no positive culture	Cardiac	Other	Unknown
				Bacteria	Fungal	Viral	Mixed				
Early death (day 0–42)	ED day 0–14	16	14	1	0	0	0	1	0	0	0
	ED day 15–42	15	1	3	4	0	1	6	0	0	0
Treatment-related death (>day 42)	During chemotherapy										
	Induction	15	0	5	4	1	1	2	0	2	0
	Consolidation	11	1	3	1	0	1	4	0	0	1
	Transplant-related	10	0	2	2	1	1	2	0	2	0
	>1 year after end of treatment	3	0	0	0	0	0	0	1	1	1
Total ED/TRD		70	16	14	11	2	4	15	1	5	2

ED, early death; TRD, treatment-related death.

Cause of death

Death from infection. Infection was the main reason for ED or TRD ($n = 46$; 66%) (Table V). After day 15, infection was considered the cause of death in 44 (81%) of 54 deaths. Infection was the cause of death in eight (80%) of 10 patients dying from treatment-related causes during HSCT.

In 14 patients, fatal complications caused by infections occurred as a result of bacterial pathogens; a gram-positive isolate in six cases and a gram-negative isolate in eight. *Pseudomonas aeruginosa* ($n = 7$) were predominant among gram-negative pathogens, *Klebsiella* was isolated in a single case. The gram-positive pathogens were Streptococci ($n = 2$), Staphylococci ($n = 2$), *Bacillus cereus* ($n = 1$) and *Gemella* ($n = 1$). Eleven patients died of fungal infection. This was caused by *Aspergillus* in six patients, by *Candida* in four patients and in one case no specific microorganism could be identified. Nine (82%) of 11 cases of fungal infection were only diagnosed by autopsy. Viruses causing TRD were cytomegalovirus ($n = 1$) and respiratory syncytial virus ($n = 1$). In four cases, the infection was polybacterial or polymicrobial. Fifteen patients died of clinically/radiologically documented infections without having a pathogen identified.

Age <2 ($P = 0{\cdot}02$) or ≥10 years ($P = 0{\cdot}01$) at diagnosis were the only statistically significant risk factors for infection-related $ED_{15–42}$ or TRD (Table VII).

Death from cardiac failure. Only one patient receiving a cumulative dose of doxorubicin of 150 mg/m^2 and mitoxantrone 60 mg/m^2 died from cardiac failure. Six patients who died from infection had cardiac insufficiency a few days preceding their death but in all cases, cardiac failure was considered part of septic shock.

Discussion

The three consecutive NOPHO-AML protocols have shown a successive improvement in outcome from a 5-year overall survival of 38% in the NOPHO-AML-84 to 64% in the NOPHO-AML-93 (Lie *et al*, 2005). This analysis shows that 13% of children diagnosed with AML in the Nordic countries died before starting treatment or from treatment-related complications. The rates of ED and TRD rose from 11% in NOPHO-AML-84 to 29% in -88, but then fell to 8% in -93. A total of 88% of the patients with $ED_{0–14}$ died due to haemorrhage or leucostasis. The risk factors for death before day 15 were hyperleucocytosis, age <2 years or ≥10 years. After day 15, infection caused 81% of $ED_{15–42}$ and TRDs. Risk factors were age (<2 years or ≥10 years at diagnosis), WBC 20–99 × 10^9/l, FAB M3 and treatment according to the NOPHO-AML-88 protocol.

The number of EDs and TRDs has been estimated in different AML trials as shown in Table VIII (Riley *et al*, 1999; Creutzig *et al*, 2004, 2006; Rubnitz *et al*, 2004; Dluzniewska *et al*, 2005; Entz-Werle *et al*, 2005; Gibson *et al*, 2005; Kardos

Table VI. Risk factors for early death day 15–42 or treatment-related death in NOPHO-AML-84/88/93 protocols.

	All patients		$ED_{15–42}$ or TRD		HR (95% CI)	Adjusted HR* (95% CI)
Risk factors	*n* = 525	%	*n* = 54	%	Simple regression	Multiple regression
Sex						
Female	272	52	30	56	1·2 (0·7–1·0)	1·2 (0·7–2·0)
Male	253	48	24	44	1·0	1·0
Age at diagnosis (years)						
<2	141	27	18	33	2·3 (1·1–4·6)	2·3 (1·1–4·7)
2–9	215	41	13	24	1·0	1·0
≥10 (10–14)	169 (133)	32	23 (14)	43	2·4 (1·2–4·7)	2·4 (1·2–4·8)
Leucocytes at diagnosis ($\times 10^9$/l)						
<20	285	54	25	46	1·0	1·0
20–99	159	30	22	41	1·7 (0·9–3·0)	1·8 (1·0–3·2)
≥100	81	16	7	13	1·2 (0·5–2·7)	1·0 (0·4–2·3)
Platelets at diagnosis ($\times 10^9$/l)						
<20	108	20	12	22	1·3 (0·6–2·8)	1·4 (0·6–3·0)
20–99	273	52	29	54	1·2 (0·6–2·3)	1·2 (0·6–2·3)
≥100	140	27	13	24	1·0	1·0
No data	4	1	0	–	–	–
CNS leukaemia at diagnosis						
Yes	29	6	4	7	1·6 (0·6–4·4)	1·3 (0·5–3·6)
No	485	92	48	89	1·0	1·0
No data	11	2	2	4	–	–
FAB-type						
M0	19	4	1	2	0·5 (0·1–4·3)	1·0 (0·1–8·3)
M1	81	15	8	15	1·0	1·0
M2	131	25	14	26	1·0 (0·4–2·5)	1·2 (0·5–2·9)
M3	28	5	6	11	2·3 (0·8–6·5)	2·9 (1·0–8·4)
M4	106	20	9	17	0·8 (0·3–2·2)	0·9 (0·4–2·4)
M5	95	18	11	20	1·2 (0·5–2·9)	1·5 (0·6–3·7)
M6	14	3	2	4	1·4 (0·3–6·4)	2·8 (0·6–13·6)
M7	21	4	0	–	–	–
Others/No data	30	6	3	5	1·0 (0·3–3·7)	1·0 (0·3–3·7)
Cytogenetics						
Normal	93	18	14	26	1·4 (0·7–3·1)	1·4 (0·7–3·1)
Cytogenetic favourable†	100	19	12	22	1·0	1·0
Other *MLL* than t(9;11)	32	6	4	8	1·2 (0·4–3·6)	2·4 (0·7–7·9)
Others	144	27	12	22	0·7 (0·3–1·6)	0·7 (0·3–1·5)
No data	156	30	12	22	–	–
Allogeneic HSCT						
Yes	150	28	10	19	2·1 (0·9–4·9)	2·0 (0·8–4·9)
No	374	71	44	81	1·0	1·0
No data	1	1	0	–	–	–
Protocol						
NOPHO-AML-84	96	18	8	15	1·0	1·0
NOPHO-AML-88	114	22	26	48	2·8 (1·3–6·3)	2·8 (1·3–6·1)
NOPHO-AML-93	315	60	20	37	0·7 (0·3–1·6)	0·7 (0·3–1·6)
Size of treating department						
<2 AML patients per year	287	55	33	61	1·0	1·0
≥2 AML patients per year	238	45	21	39	0·8 (0·5–1·4)	0·7 (0·4–1·3)

$ED_{15–42}$, early death day 15–42; TRD, treatment-related death; HR, hazard ratio; 95% CI, 95% confidence interval; HSCT, haematopoietic stem cell transplantation; CNS, central nervous system.

*Adjusted for age (2–9 vs. <2 or ≥10 years), leucocytes at diagnosis (<20 vs. 20–99 or ≥100 $\times$ 10^9/l) and protocol (NOPHO-AML-84 vs. NOPHO-AML-88 or NOPHO-AML-93).

†Favourable cytogenetics: t(8;21)(q22;q22), t(9;11)(p22;q23), t(15;17)(q22;q12-21), inv16(p13q22) and t(16;16)(p13;q22).

Table VII. Risk factors for early death day 15–42 or treatment-related death caused by infection in NOPHO-AML-84/88/93 protocols.

	All patients		$ED_{15–42}$ or TRD caused by infection		HR (95% CI)	Adjusted HR* (95% CI)
Risk factors	*n* = 525	%	*n* = 46	%	Simple regression	Multiple regression
Sex						
Female	272	52	26	57	1·2 (0·7–2·2)	1·2 (0·7–2·2)
Male	253	48	20	43	1·0	1·0
Age (years)						
<2	141	27	16	35	2·9 (1·3–6·6)	2·8 (1·2–6·3)
2–9	215	41	9	19	1·0	1·0
≥10 (10–14)	169	32	21 (21)	46	3·1 (1·4–6·9)	3·1 (1·4–6·7)
Leucocytes at diagnosis ($\times 10^9$/l)						
<20	285	54	23	50	1·0	1·0
20–99	159	30	18	39	1·5 (0·8–2·7)	1·6 (0·9–2·9)
≥100	81	16	5	11	0·9 (0·4–2·4)	0·7 (0·3–2·0)
Protocol						
NOPHO-AML-84	96	18	8	17	1·0	1·0
NOPHO-AML-88	114	22	20	44	2·2 (1·0–5·1)	2·1 (0·9–4·7)
NOPHO-AML-93	315	60	18	39	0·6 (0·3–1·5)	0·6 (0·3–1·4)

$ED_{15–42}$, early death day 15–42; TRD, treatment-related death; HR, hazard ratio; 95% CI, 95% confidence interval.
*Adjusted for age (2–9 vs. <2 or ≥10 years) and protocol (NOPHO-AML-84 vs. NOPHO-AML-88 or NOPHO-AML-93).

Table VIII. Number of early deaths and treatment-related deaths in recent paediatric AML studies from major groups.

Study group	Years of enrolment	No. of patients	Age range (years)	Early death rate (%)	TRD rate (%)	Overall survival rate (5 years) (%)
AIEOP-92 (Kaspers and Creutzig, 2005 and Pession *et al*, 2005)	1992–2001	160	0–14	6	7	60
AML-BFM 98 (Creutzig *et al*, 2004, 2006)	1998–2002	430	0–18	3	4*	62†
AML-PPLLSG 98 (Dluzniewska *et al*, 2005)	1998–2002	104	0–14	8	10	50
AML99 (Tsukimoto *et al*, 2009)	2000–2002	240	0–18	2‡	4	76
CCG 2891 (Smith *et al*, 2005)	1989–1995	750	0–14	4	15	47
DCOG AML-92/94 (Kardos *et al*, 2005)	1992–1998	78	0–14	10	16	50
EORTC-CLG 58921 (Entz-Werle *et al*, 2005)	1993–2000	166	0–14	2	6	62
LAME 91 (Perel *et al*, 2005)	1991–1998	247	0–14	4	6	61
MRC AML 12 (Gibson *et al*, 2005)	1995–2002	455	0–14	4	6	66
NOPHO-AML-93	1993–2003	292	*0–14*	5	2	65
NOPHO-AML-93	1993–2003	315	*0–18*	3	5	64
POG 8821 (Ravindranath *et al*, 2005)	1988–1993	511	0–14	4	8	42
St. Jude-AML91 (Ribeiro *et al*, 2005)	1991–1997	62	0–14	3	Not reported	57

TRD, treatment-related death; AIEOP, Associazione Italiana di Ematologia e Oncologia Pediatrica; AML99, Japanese Leukaemia/Lymphoma Study Group; BFM, Berlin-Frankfurt-Münster Group; PPLLSG, the Polish Paediatric Leukaemia/Lymphoma Study Group; CCG, the Children's Cancer Group; DCOG, Dutch Childhood Oncology Group; EORTC, European Organization of Research and Treatment of Cancer; LAME, Leucémie Aiguë Myéloblastique Enfant; MRC, Medical Research Council; POG, Paediatric Oncology Group.
*Without non-responders who are included in the AML-BFM study on early death and treatment-related death.
†Is from the paper (Creutzig *et al*, 2006) which included 473 patients in the period from 1998 to 2003.
‡Patients who died before initiation therapy were excluded (*n* = 3).

et al, 2005; Kaspers & Creutzig, 2005; Perel *et al*, 2005; Pession *et al*, 2005; Ravindranath *et al*, 2005; Ribeiro *et al*, 2005; Slats *et al*, 2005; Smith *et al*, 2005; Tsukimoto *et al*, 2009). ED occurred in 2–10% and TRD in 2–16%. The percentage was higher with a high rate of HSCT. With a 5-year overall survival of 60% or above, our ED and TRD results are similar to those reported by others (Riley *et al*, 1999; Creutzig *et al*, 2004). Several groups have reported a decrease in toxic death rates over time without concomitant changes in therapy or even with more intensive therapy. However in the 1982–1997 period, the Dutch childhood oncology group observed that chemotherapy-related mortality

rose from 3% to 8% during consolidation, which was perhaps related to the scheduling of intensification blocks (Slats *et al*, 2005). This illustrates the delicate balance between minor changes in treatment and the risk of treatment-related mortality. Within the different NOPHO-AML protocols, no significant reduction in the number of EDs and TRDs occurred over time in contrast to the learning curve demonstrated in the Children's Cancer Group (CCG)-2961 study (Lange *et al*, 2008).

The causes of ED and TRD were similar in the different AML studies despite variations in supportive care practices (Lehrnbecher *et al*, 2009). In contrast to the excess mortality rate due to cardiac causes (standardized mortality rate = 5·0) in the Childhood Cancer Survivor Study (CCSS) (Armstrong *et al*, 2009), the frequency of cardiac mortality in our population was low.

Three patients died 7–12 years after end of treatment from late effects of treatment or complications in relation to initial AML manifestations, which underlines the importance of continued long-term follow-up. In the CCSS, 4% of 272 5-year AML survivors treated without HSCT had died from other causes than relapse 8–26 years after diagnosis (Mulrooney *et al*, 2008).

In the AML-BFM-93 and -98 trials, risk factors for death before day 15 were low performance status at presentation, initial CNS involvement, hyperleucocytosis and FAB M5 (Creutzig *et al*, 2004). After day 15, age above 10 years and FAB M0 were risk factors. Further studies are needed to determine whether pharmacokinetic differences account for the increased death rate among children <2 years and ≥10 years of age (Palle *et al*, 2006, 2009). We had no data on body mass index at diagnosis, but the CCG-2961 showed that overweight and underweight children and adolescents with AML were more likely to experience treatment-related mortality (Lange *et al*, 2005). The high rate of ED and TRD in APL patients in the NOPHO-AML studies may be due to the low incidence of FAB M3 in Northern Europe and therefore limited experience and late introduction of ATRA.

In the AML-BFM-78 and -83 studies, the risk of ED due to leucostasis and/or haemorrhage was significantly increased in children with FAB M5, hyperleucocytosis, and extramedullary organ involvement (Creutzig *et al*, 1987). These findings are in line with the results from our studies. We also found a higher proportion of EDs from leucostasis/haemorrhage in children <2 years than in children between 2 and 9 years of age. We observed a small decline in the incidence of hyperleucocytosis over time, from 18% in the NOPHO-AML-84 and 19% in the NOPHO-AML-88 to 13% in the NOPHO-AML-93 protocol. The low number of EDs in the NOPHO-AML-84 (3%) and -93 (2%) precludes any conclusion as to whether the lower frequency of hyperleucocytosis reduced ED as shown in the study from St. Jude Children's Research Hospital (Inaba *et al*, 2008).

Next to relapses, infection remains the most important cause of morbidity and mortality among children with AML. Infections caused 85% of all EDs and TRDs when deaths before starting treatment were excluded. The infection-related mortality of 9% (46/525) is comparable to that reported in other recent AML studies, which quoted frequencies of 5–11% (Riley *et al*, 1999; Lehrnbecher *et al*, 2004; Brunet *et al*, 2006; Sung *et al*, 2007). However, it is difficult to compare results and draw conclusions about trends in infection-related mortality due to the use of different definitions and underestimation of the true incidence of these infections. Our Nordic trials included a large amount of infection-related deaths where no microorganism could be identified. Like in other studies, a significant number of invasive fungal infections were diagnosed only at autopsy (Groll *et al*, 1996). Risk factors for infection-related deaths in the CCG 2961 trial were underweight, non-white race and age above 16 years (Sung *et al*, 2007). In our study, age <2 or ≥10 years at diagnosis were risk factors for death due to infection. NOPHO-AML-88 used the principle of timed sequential induction, which caused an unacceptable increase in ED and TRD. This is similar to the results from the CCG-2891 trial, where fatal infections were more common during intensive than during standard-timed induction (Sung *et al*, 2009).

To allow comparison between cooperative AML groups and to improve our understanding of infection outcomes, standard infection definitions should be used (Sung *et al*, 2008). To reduce infection-related mortality, it is important to improve both prevention and treatment through prophylaxis, early diagnosis and aggressive management. The current NOPHO-AML 2004 protocol includes prophylactic antifungal therapy with fluconazole but no prophylactic G-CSF or anti-bacterial therapy because there is no solid evidence that these measures reduce mortality in children with AML. However, a recent study from St. Jude Children's Research Hospital reduced the odds of bacterial sepsis by 93% using intravenous vancomycin with oral ciprofloxacin or a cephalosporin (Kurt *et al*, 2008). A relatively small number of patients were studied and larger multi-institutional studies are needed before recommendations on these prophylactic antibiotic regimens can be made. The development of targeted and more leukaemia-specific drugs may reduce the toxic effect on normal cells and thereby reduce infection-related morbidity. Identification of possible immunogenetic risk profiles at the start of therapy could potentially be helpful for risk-adapted and individualized supportive care (Hartel *et al*, 2007). Initial data have demonstrated that interleukin-6 and chitotriosidase polymorphisms may modify infection risk (Lehrnbecher *et al*, 2005). In a study from the CCG, polymorphisms in the gene encoding cytidin deaminase were associated with an increased risk of treatment-related mortality with cytarabine-based therapy for AML (Bhatla *et al*, 2009).

Our study has some limitations. AML is rare in children and the relatively low number of patients therefore hampers statistical analysis of risk factors at subgroup level. Limited

information in the medical records sometimes made it difficult to categorize the cause of death. In some cases, it was difficult to assess whether the patient was in CR, and to determine whether the cause of death was treatment- or disease-related.

The number of EDs and TRDs in childhood AML in the Nordic countries remains substantial. Further studies should be conducted to explore the pathogenesis behind leucostasis/haemorrhage and prospective studies of the effect of cytoreduction should be performed. Optimal antifungal prophylaxis is essential, and studies on the benefit of antibacterial prophylaxis and individual risk factors for ED and TRD are needed.

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Authorship

Lene Molgaard-Hansen participated in the design of the study, provided clinical data, handled the data, performed the statistical analysis and drafted the manuscript. Henrik Hasle participated in the design of the study, participated in the interpretation of the data and the drafting of the manuscript. Merja Möttönen, Heidi Glosli, Guðmundur K Jónmundsson and Jonas Abrahamsson provided clinical data in their capacity as national coordinators of this study and participated in the interpretation of the data. All authors read and approved the final manuscript.

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