Impact of central nervous system involvement in adult patients with Philadelphia-negative acute lymphoblastic leukemia: a GRAALL-2005 study

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Abstract

Whereas the prognosis of adult patients with Philadelphia-negative acute lymphoblastic leukemia (ALL) has greatly improved since the advent of pediatric-inspired regimens, the impact of initial central nervous system (CNS) involvement has not been formerly re-evaluated. We report here the outcome of patients with initial CNS involvement included in the

pediatric-inspired prospective randomized GRAALL-2005 study. Between 2006 and 2014, 784 adult patients (aged 18-59 years) with newly diagnosed Philadelphia-negative ALL were included, of whom 55 (7%) had CNS involvement. In CNS-positive patients, overall survival was shorter (median 1.9 years *vs*. not reached, HR=1.8 [1.3-2.6], *P*<0.001). While there was no statistical difference in cumulative incidence of relapse between CNS^+ and CNS^- patients (HR=1.5 [0.9-2.5], *P*=0.11), non-relapse mortality was significantly higher in those with initial CNS disease (HR=2.1 [1.2-3.5], *P*=0.01). This increase in toxicity was mostly observed in patients randomized to the high-dose cyclophosphamide arm and in those who received allogeneic stem cell transplantation. Exploratory landmark analyses did not show any association between either cranial irradiation or allogeneic stem cell transplantation and outcome. Despite improved outcome in young adult ALL patients with pediatric-inspired protocols, CNS involvement is associated with a worse outcome mainly due to excess toxicity, without improved outcome with allogeneic SCT.

Introduction

Acute lymphoblastic leukemia (ALL) often involves the central nervous system (CNS), which is considered a sanctuary for leukemic cells. It has been recently shown that ALL cells could use neural migratory pathways to invade the CNS and adapt to the CNS microenvironment by modifying metabolic pathways.[1,2] As relapse can develop from occult CNS disease, CNS-directed therapies such as high-dose methotrexate and intrathecal chemotherapy are mandatory in all patients with ALL whether they have CNS involvement or not.[3] At diagnosis, 4-11% of patients with ALL have CNS involvement.[4-8] These patients are more likely to have other extramedullary site involvement, T-cell ALL, and a higher white blood cell (WBC) count at diagnosis.[4,6] In adults with ALL, the prognosis of initial CNS involvement is controversial, but has been associated with higher rates of primary resistant disease and early death in one series, and with decreased overall survival in another.[4,6] The role of allogeneic stem cell transplantation (SCT) was also controversial in these two historical series.[4,6]

Whereas more recent studies have shown that CNS involvement is associated with a worse outcome in children with ALL,[9,10] no study has re-evaluated the impact of CNS involvement in adults in more recent years during which time they are increasingly receiving pediatric-inspired protocols.[11] In comparison to conventional adult protocols, these pediatric-inspired regimens, which include higher doses of corticosteroids, vincristine, asparaginase, and increased CNS-directed therapy with lower doses of other cytotoxic chemotherapy, have greatly improved the outcome of adult patients.[7,8,11-15] Meanwhile, indications for allogeneic SCT have changed, relying more on measurable residual disease (MRD).[16,17] We report here the association of CNS involvement with survival in adults with ALL included in the pediatric-inspired prospective GRAALL-2005 study.

Methods

Patients and settings

The Group for Research on Adult Acute Lymphoblastic Leukemia 2005 trial (GRAALL-2005) was conducted between 2006 and 2014 at 57 French, 8 Belgian, and 8 Swiss centers. This randomized trial evaluated the impact of hyperfractionated cyclophosphamide and rituximab in adult patients aged 18-59 years with ALL without *BCR-ABL*.[14,18] The full GRAALL 2005 protocol is available in *Online Supplementary Figure S1*. Historical definitions from the Children's Oncology Group were used to classify the initial CNS status based on cerebrospinal fluid (CSF) conventional cytospin (CC) (*Online Supplementary Table S1*).[9] Flow cytometry was not routinely used for CSF evaluation at diagnosis.

All patients received a 5-drug induction therapy including native *Escherichia coli* L-Asparaginase (L-Asp). Patients in complete morphological remission (CR) received two consolidation courses including high-dose methotrexate and high-dose cytarabine. MRD was based on patient-specific *Ig/TCR* gene rearrangement monitoring using standardized quantitative real-time polymerase chain reaction (qRT-PCR) with a sensitivity of at least 10^{-4}, centrally performed on bone marrow (BM) samples after the first induction course. High-risk patients aged <55 years, defined in the *Online Supplementary Methods*, were eligible for allogeneic SCT in first CR with a conditioning regimen including cyclophosphamide and total body irradiation (TBI). Patients in persistent CR who did not proceed to allogeneic stem cell transplantation (SCT) received a late intensification followed by one consolidation course and a 2-year maintenance.

CNS-directed prophylaxis included one methotrexate intrathecal injection during steroid pre-phase followed by six triple intrathecal injections. CNS irradiation was recommended for all patients (i.e., 15 grays for patients proceeding to allogeneic SCT and 24 grays for other patients). Besides CNS irradiation, CNS-positive patients received an additional eight triple intrathecal therapy during induction and were eligible for allogeneic SCT in first CR. During induction therapy, they received fewer L-Asp injections (5 instead of 8) to reduce the risk of CNS thrombosis.

Informed consent was obtained from all patients at entry into this trial, which was conducted in accordance with

the Declaration of Helsinki and approved by the Institutional Ethics Committee Ile-de-France VI, France. This trial was registered at *www.clinicaltrials.gov* (NCT00327678).

Statistical analysis

Categorical variables were presented as numbers with proportions and compared using the χ^2 test or the Fisher's exact test, as appropriate. Continuous variables were presented as medians with interquartile range (IQR) and compared using the Mann and Whitney and Kruskal-Wallis tests, as appropriate. Univariable and multivariable Cox regression models were used to analyze the association between patients' characteristics and overall survival (OS). The Kaplan-Meier method was used to estimate median OS and survival curves were compared using the log-rank test. Cumulative incidence of relapse (CIR; with non-relapse mortality [NRM] as a competing event) and NRM (death without prior relapse with relapse as a competing risk) were summarized using cumulative incidence estimates and compared using Gray's test. Cox proportional hazards regression models were used to estimate cause-specific Hazard Ratio (HR). Landmark analyses were performed to study the impact of cranial irradiation and allogeneic SCT.[19] All tests were two-sided. $P<0.05$ was considered statistically significant. Statistical analyses were performed with R (R Foundation for Statistical Computing, Vienna, Austria; http://www.r-project.org).

Results

Incidence of initial central nervous system involvement and clinical presentation

Between 2006 and 2014, 784 adult patients with newly diagnosed Ph-negative ALL were included of whom 55 (7%) had initial CNS involvement. The first lumbar puncture was performed during pre-phase steroid therapy in most patients (672/784 patients, 86%). CNS-positive patients were more likely to have T-cell ALL (P=0.004), leukocytes ≥30x 10^9/L (P=0.016), and higher hemoglobin levels (P=0.02) (Table 1). Most CNS-positive patients (n=47, 85%) were classified as CNS-3 (>5 white blood cells/μl and a positive CC and/or clinical signs) whereas 7 patients (13%) were classified as CNS-2 (<5 white blood cells/μl and a positive CC) (Table 2). Initial presentation was heterogeneous since 27 patients (49%) had clinical symptoms (9/27, 33% with concurrent positive CC) and 24 patients (44%) had only CSF findings. The most prominent clinical sign was trigeminal anesthesia that was reported in 41% of patients with clinical symptoms. Imaging (mostly magnetic resonance imaging), performed in 26 patients (47%), was only positive in 5: 4 with clinical symptoms and one with CSF findings.

Association between central nervous system involvement and outcome

A patient flowchart is shown in Figure 1. First CR rate, in-

Table 1. Characteristics of patients with or without central nervous system involvement.

Characteristic	All, N=784	CNS-negative, N=729	CNS-positive, N=55	*P*
Age, median (IQR), years	36 (25-48)	37 (25-48)	30 (24-44)	0.15
Female, N (%)	312 (40)	294 (40)	18 (33)	0.27
BMI, median (IQR), kg/m^2	23.6 (21.1-27.2)	23.7 (21.1-27.3)	23.5 (21.1-26.2)	0.75
Phenotype, N (%)				0.004
B-cell	523 (67)	496 (68)	27 (49)	
T-cell	261 (33)	233 (32)	28 (51)	
WBC at diagnosis, median (IQR), x10^9/L	12 (4-42)	11 (4-41)	23 (9-66)	0.15
Hb at diagnosis, median (IQR), g/dL	10.2 (8.2-12.3)	10.1 (8.2-12.2)	11.1 (8.8-13)	0.02
PLT at diagnosis, median (IQR), x10^9/L	72 (33-154)	72 (32-154)	78 (36-137)	0.16
Poor early PB blast clearance, N (%)	187 (24)	172 (24)	15 (27)	0.50
Not evaluable	3	3		
Poor early BM blast clearance, N (%)	306 (39)	285 (39)	21 (38)	0.58
Not evaluable	40	36	4	
CR, N (%)	722 (92)	672 (92)	50 (91)	0.79
Induction death, N (%)	44 (6)	41 (6)	3 (6)	>0.99
MRD1 negativity, N (%)	126 (16)	119 (16)	7 (13)	0.26
Not evaluable	445	416	29	
Allogeneic SCT in first CR, N (%)	278 (35)	249 (34)	30 (55)	0.002

BM: bone marrow; BMI: body mass index; CNS: central nervous system; CR: complete remission; Hb: hemoglobin; IQR: interquartile range; MRD1: measurable residual disease after induction; N: number; PB: peripheral blood; PLT: platelet count; SCT: stem cell transplantation; WBC: white blood cell count.

duction death, and achievement of negative MRD after induction (*Ig/TCR* <10^{-4}) were similar between CNS-negative and CNS-positive patients (92% *vs*. 91%, *P*=0.79; 6% *vs*. 6%, *P*=1; 38% *vs*. 27% in patients in whom MRD was evaluated, *P*=0.26, respectively) (Table 1). OS was, however, shorter in CNS-positive patients (median 1.9 years *vs*. not reached, HR=1.8, [1.3-2.6], *P*<0.001) (Figure 2A). OS was similar when patients who received allogeneic SCT in first CR were censored at the time of transplant (*P*=0.01) (*Online Supplementary Figure S2A*). CNS involvement at diagnosis was not associated with a statistically different CIR (HR=1.5 [0.9-2.5], *P*=0.11), but it was significantly associated with higher NRM (HR=2.1 [1.2-3.5], *P*=0.01) (Figure 2B, C). Causes of NRM were similar between CNS-positive and CNS-negative patients, including infection (69% *vs*. 52%), transplant-related (13% *vs*. 15%), and thrombosis (6% *vs*. 5%) while two were undetermined in CNS-positive patients (13%). Other causes of NRM in CNS-negative patients included bleeding (7%), second cancer (4%), and other causes (4%), while 15 (13%) were undetermined. At three years, 34% (20-47%) of CNS-positive patients *versus* 26% (23-29%) of CNS-negative patients relapsed. CNS-positive patients had a non-significant higher risk of CNS relapse (6% [0-13%] *vs*. 2% [1-3%] at 3 years, *P*=0.095) whereas combined and isolated BM relapses were similar to CNS-negative patients (4% [0-10%] *vs*. 2% [1-3%], *P*=0.41 and 21% [10-33%] *vs*. 20% [17-23%] at 3 years, *P*=0.79, respectively) (*Online Supplementary Figure S3*). However, after censoring patients at allogeneic SCT, the CIR was higher in CNS-positive patients (HR=2.2 [1.2 -3.9], *P*=0.01) whereas there was no difference in NRM between the two groups (HR=1.4 [0.7-3.1], *P*=0.4) (*Online Supplementary Figure S2B, C*).

Because the GRAALL-2005 study randomized patients to standard (Standard-C arm) and hyperfractionated (hyper-C arm) cyclophosphamide, we evaluated the impact of the randomization arm in patients with initial CNS involvement. The characteristics of CNS-positive patients were similar according to the randomization arm (*Online Supplementary Table S2*). The proportion of patients receiving cranial irradiation (57% *vs*. 40% for the standard and hyper-C arms, respectively, *P*=0.28) and allogeneic SCT (53% *vs*. 56%, *P*=1) was also similar. CNS-positive patients randomized to the hyper-C arm had shorter OS (median 1.1 years *vs*. not reached, *P*<0.001) whereas CNS-negative patients had similar outcomes (*Online Supplementary Figure S4A*). CIR was similar according to the randomization arm but there was a significant increase in NRM in CNS-positive patients randomized to the hyper-C arm (*Online Supplementary Figure S4B, C*).

Analysis of factors associated with overall survival

In a univariable Cox regression model, age (HR=1.3 [1.2-1.4], *P*<0.001), body-mass index (HR=1.4 [1.2-1.8], *P*<0.001), leukocytes ≥ $30x10^9$/L (HR=1.4 [1.1-1.7], *P*=0.1), CNS involvement (HR=1.3 [1.8-2.6], *P*=0.001), and poor early BM blast clearance (HR=1.3 [1.1-1.7], *P*=0.02) were associated with lower OS (Table 3). The significant association between CNS involvement at diagnosis and lower OS was also observed in a multivariable Cox regression model (HR=2.1 [1.4-3], *P*<0.001). Age (HR=1.3 [1.2-1.4], *P*<0.001) and leukocytes ≥ $30x10^9$/L (HR=1.4 [1-1.8], *P*=0.03) were the only two other factors associated with OS in this multivariable model (Table 3). After adjustment for randomization arm in a Cox regression model, initial CNS involvement was still associated with worse OS (HR=1.8 [1.3-2.6], *P*=0.002). In CNS-positive patients, factors associated with OS were female gender (HR=3.5 [1.5-8.3], *P*=0.005) and poor early BM blast clearance (HR=4.4 [1.8-11], *P*<0.001) after multivariable analysis. Neither age nor ALL subtype were associated with OS after adjustment (HR=1 [0.7-1.4], *P*=0.8 and HR=1.1 [0.5-2.5], *P*=0.9, respectively) (*Online Supplementary Figure S5*).

Table 2. Initial clinical and cerebrospinal fluid presentation of patients with central nervous system involvement.

	CNS-positive, N=55
CNS status, N (%)	
CNS-2	7 (13)
CNS-3	47 (85)
NA	1
Presentation, N (%)	
CSF only	24 (44)
Clinical signs only	18 (33)
CSF + clinical signs	9 (16)
NA	4
Clinical signs, N (%)	27 (49)
Trigeminal anesthesia	11 (20)
Facial paralysis	4 (7)
Paresthesia: extremities	4 (7)
Visual signs	3 (5)
Meningeal syndrome	2 (4)
Motor deficit	2 (4)
Confusion	2 (4)
Radiological signs, N (%)	
Present	5 (9)
Absent	21 (38)
NA	29

CNS: central nervous system; CSF: cerebrospinal fluid; NA: not available.

Post-remission treatment

Twenty-seven (49%) CNS-positive patients received cranial irradiation as part of consolidation therapy, after a median of 160 days, including 18 patients before allogeneic SCT. As recommended by the study protocol, patients with CNS involvement were more likely to receive allogeneic SCT (55% *vs*. 34%, *P*=0.002), after a median interval of 170

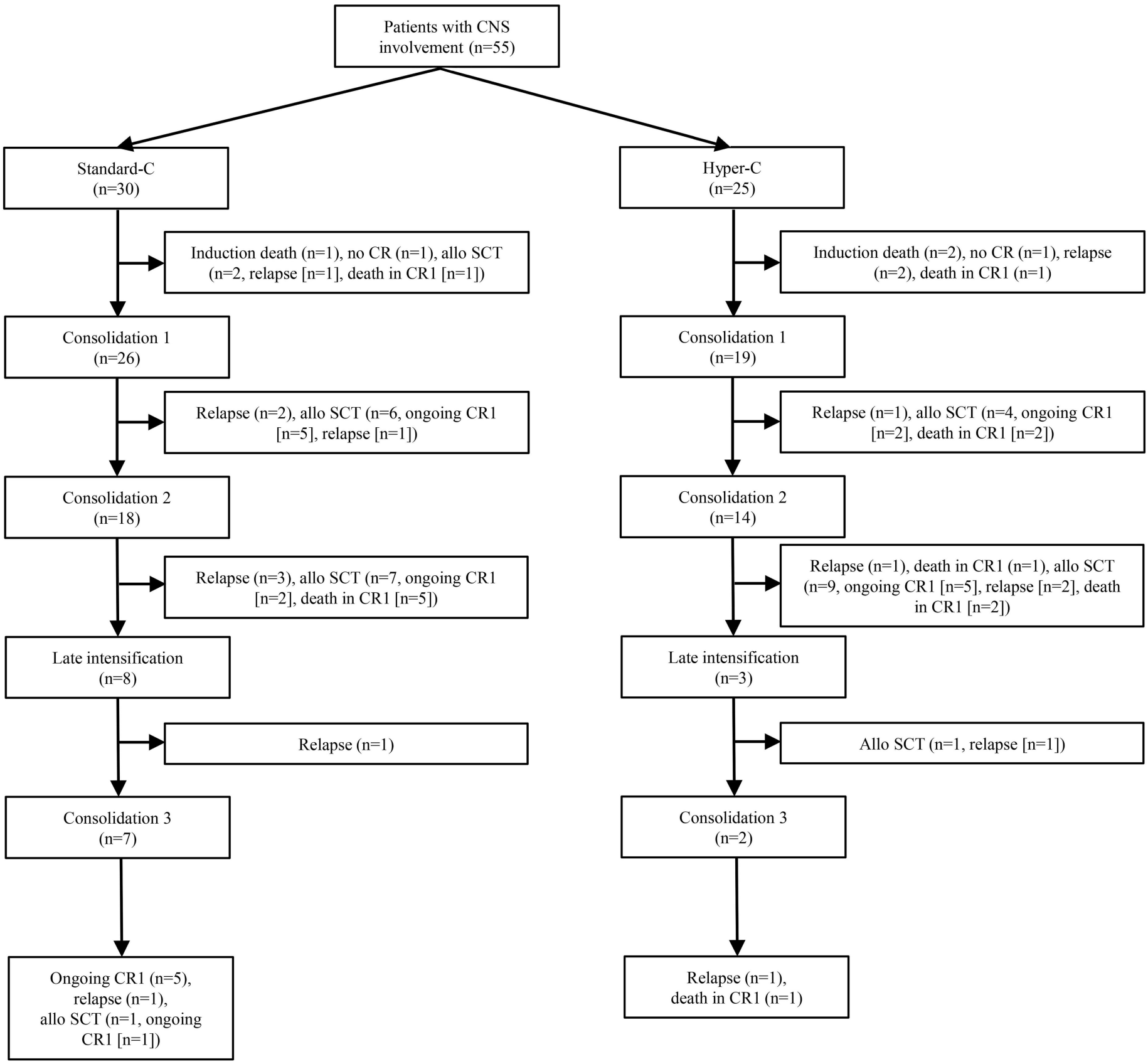

Figure 1. Flowchart of patients with central nervous system involvement at diagnosis. C: cyclophosphamide; CNS: central nervous system; CR1: first complete remission; SCT: stem cell transplantation.

days (*vs*. 142 days for patients without initial CNS involvement proceeding to allogeneic SCT, *P*=0.003). Twenty-five CNS-positive patients did not undergo allogeneic SCT, including 11 who did not have a suitable donor, seven who relapsed before allogeneic SCT could be carried out, six who died from toxicity before allogeneic SCT, and one who was deemed unfit for allogeneic SCT. We therefore performed two landmark analyses for these patients: one at day 160 to study cranial irradiation and one at day 170 to study allogeneic SCT and their association with outcome. After excluding 14 CNS-positive patients who experienced an event (relapse, death) before the day 160 landmark point, there was no difference in OS (HR=0.7 [0.3-1.6], *P*=0.4) between patients who received cranial irradiation and those who did not (*Figure 3A*). Of nine CNS-positive patients who received cranial radiation therapy without allogeneic SCT, only one death resulting from toxicity was observed. There were 11 relapses, four in those who received cranial radiation (2 BM and 2 CNS relapses) and seven in those who did not (5 BM and 2 combined relapses). In addition, after excluding 16 CNS-positive patients who had an event before the day 170 landmark

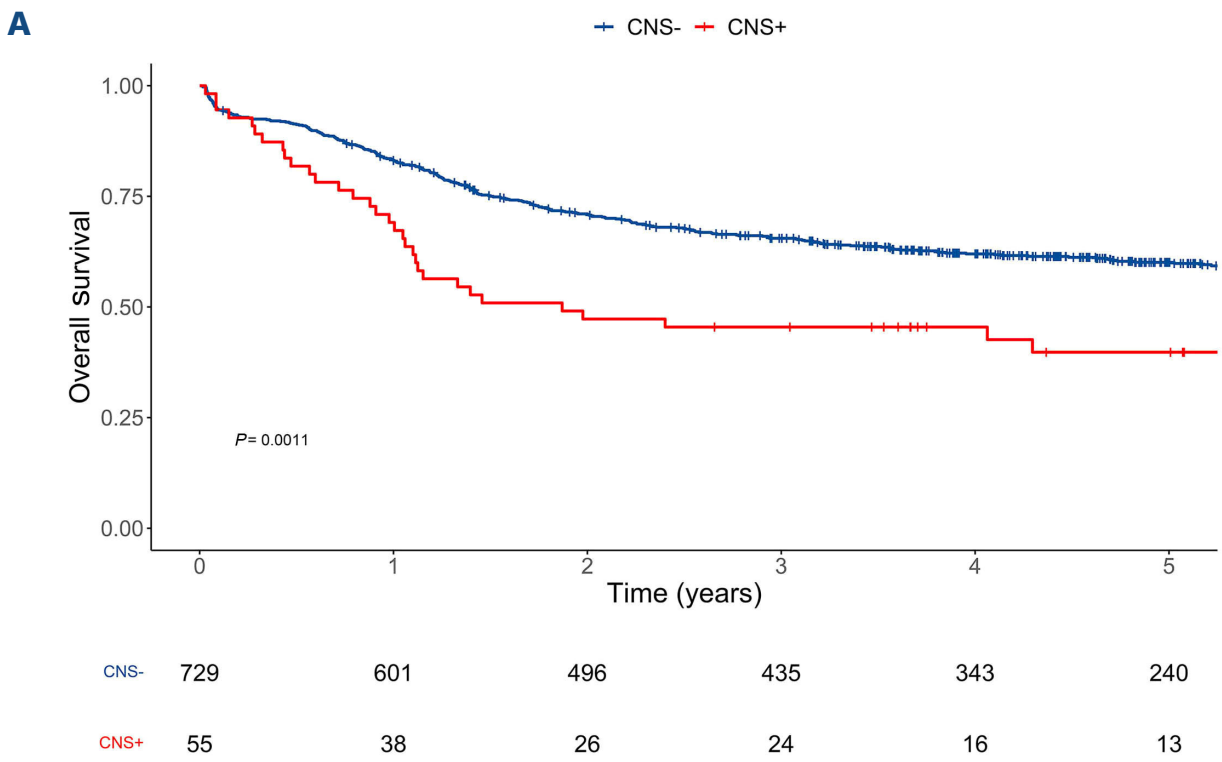

Figure 2. Outcomes according to central nervous system status at diagnosis. (A) Kaplan-Meier curves for overall survival, (B) cumulative incidence of relapse, and (C) cumulative incidence of non-relapse mortality according to (CNS) involvement at diagnosis.

Table 3. Univariable and multivariable analysis of factors associated with non-relapse mortality, relapse, and overall survival by Cox regression.

Factors	Non-relapse mortality				Relapse				Overall survival			
	Univariable analysis		Multivariable analysis		Univariable analysis		Multivariable analysis		Univariable analysis		Multivariable analysis	
	HR	*P*	HR	*P*	HR	*P*	HR	*P*	HR	*P*	HR	*P*
Age/10	1.6 (1.4-1.9)	<0.001	1.5 (1.3-1.8)	<0.001	1.1 (1-1.2)	0.1	1.1 (1-1.2)	0.1	1.3 (1.2-1.4)	< 0.001	1.3 (1.2-1.4)	< 0.001
Female	1.4 (1-1.9)	0.06	1.3 (0.9-1.9)	0.2	1 (0.7-1.3)	0.7	0.9 (0.7-1.2)	0.6	1.1 (0.9-1.4)	0.4	-	-
BMI/10	1.8 (1.3-2.4)	<0.001	1.4 (1-2)	0.1	1.1 (0.9-1.5)	0.4	1.1 (0.8-1.6)	0.4	1.4 (1.2-1.8)	< 0.001	1.2 (1-1.5)	0.1
T-cell phenotype	0.7 (0.5-0.99)	0.04	0.9 (0.6-1.4)	0.6	1 (0.7-1.3)	0.8	0.8 (0.6-1.1)	0.2	0.8 (0.7-1.1)	0.2	-	-
WBC count at diagnosis x10^9/L	0.9 (0.6-1.4)	0.7	0.9 (0.5-1.4)	0.6	1.7 (1.3-2.2)	<0.001	1.7 (1.3-2.4)	<0.001	1.4 (1.1-1.7)	0.01	1.3 (1-1.7)	0.07
CNS involvement	2.1 (1.2-3.5)	0.01	2.8 (1.6-4.7)	<0.001	1.5 (0.9-2.5)	0.1	1.6 (0.9-2.7)	0.08	1.8 (1.3-2.6)	< 0.001	2.1 (1.4-3)	< 0.001
Poor early PB blast clearance	1.1 (0.7-1.6)	0.7	1.2 (0.7-2)	0.5	1.4 (1.1-1.9)	0.02	1.1 (0.8-1.6)	0.6	1.3 (1-1.6)	0.05	1.1 (0.8-1.5)	0.5
Poor early BM blast clearance	1.2 (0.8-1.7)	0.3	1.2 (0.8-1.8)	0.3	1.5 (1.1-1.9)	0.01	1.4 (1-1.9)	0.03	1.3(1.1-1.7)	0.02	1.3 (1-1.7)	0.04

BM: bone marrow; BMI: body mass index; CNS: central nervous system; HR: Hazard Ratio; PB: peripheral blood; WBC: white blood cell count.

point, allogeneic SCT was not associated with OS (HR=0.8 [0.3-2.3], *P*=0.7) (*Figure 3B*). In a sensitivity analysis, at day 220, when 90% of patients had received allogeneic SCT, similar results were observed (*data not shown*).

Discussion

In adults with Ph-negative ALL, the impact of CNS disease at presentation is controversial with no recent large-scale series, especially for patients receiving pediatric-inspired approaches.[4,6] Treatment strategy usually includes CNS-directed therapy such as intrathecal chemotherapy, high-dose systemic methotrexate, or cranial radiation therapy and/or allogeneic SCT.[4,6] For such patients included in the prospective randomized GRAALL-2005 trial, we observed an adverse outcome which seems to be mostly driven by an increased risk of toxicity, whether due to the hyper-fractionated cyclophosphamide arm or to allogeneic SCT, rather than an increased risk of relapse.

As in previous reports, we confirm that CNS involvement is a rare event in adults at diagnosis (7% of patients).[4-8] In our study, most patients (85%) were classified as CNS-3, which contrasts with previous reports in children in which there was a more balanced distribution or, in some studies, more patients with CNS-2 disease.[9,10,20-22] We cannot exclude the possibility that some patients with CNS-2 disease could have been under-reported in our study while

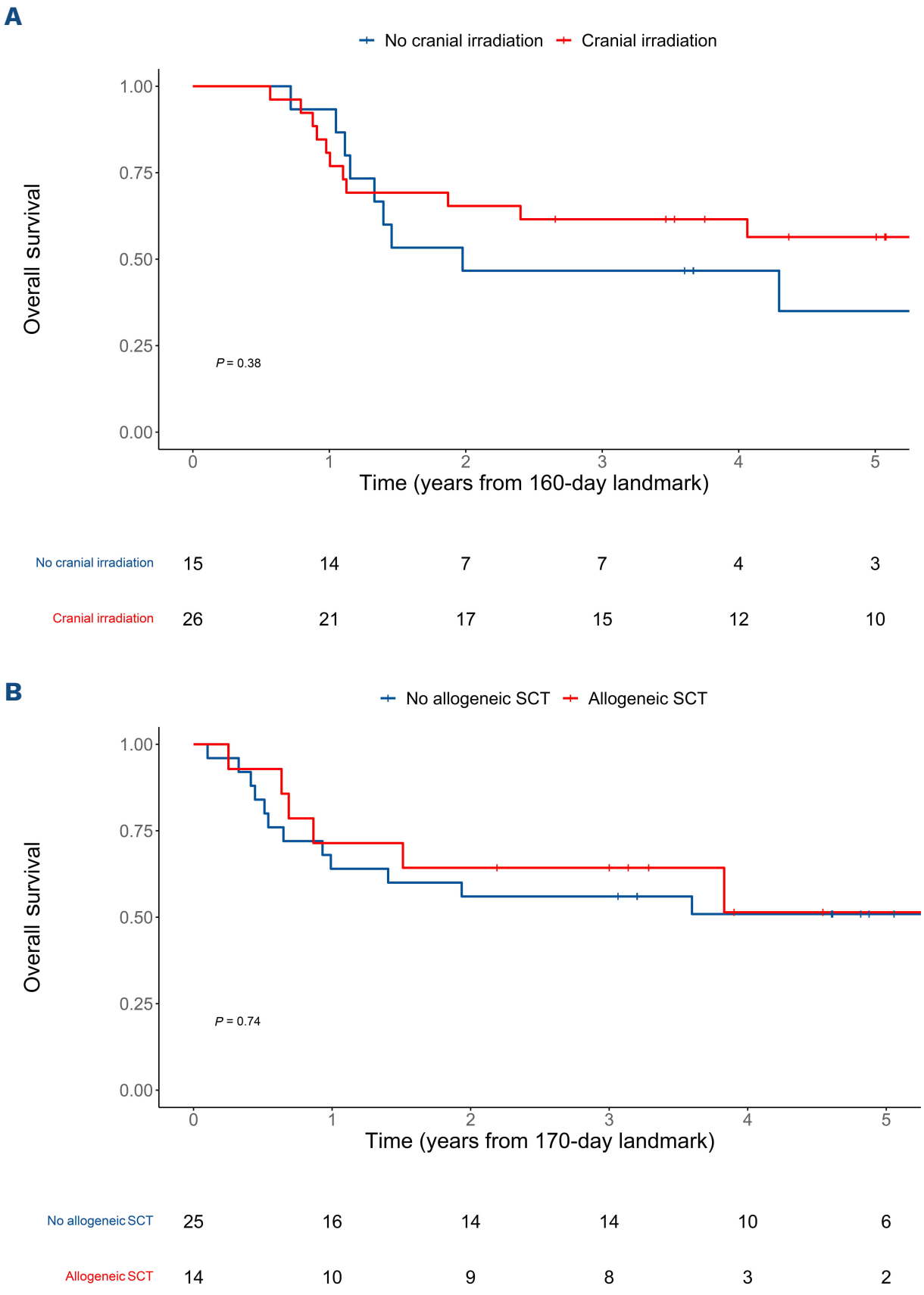

Figure 3. Outcomes of patients with central nervous system disease at diagnosis. (A) Overall survival of patients who received cranial radiation therapy by a 160-day landmark analysis and (B) overall survival of patients who received allogeneic stem cell transplantation (SCT) by a 170-day landmark analysis.

the poor outcome of children with CNS-2 is a question of debate.[9,10,20] CSF flow cytometry, which can increase the likelihood of identifying patients from 6-13% to 18-25%,[23-26] was not routinely available at a multicentric level at the time of the GRAALL-2005 study since it requires either special conditioning or rapid processing.[23,24] CNS involvement has been associated with an increased risk of relapse in some[23-25,27] but not all studies.[26] Garcia *et al.* reported that additional intrathecal therapy (twice weekly until the CSF and symptoms had cleared, then once weekly for 4 weeks) did not mitigate the worse outcome, despite the rapid clearance of blast cells in the CSF.[26]

In our cohort, the CR rate of CNS-positive patients was over 90%, similar to historical series, meaning that patients with initial CNS disease at diagnosis did not have a more resistant disease.[4] Moreover, CNS involvement neither modified induction death rates nor MRD status after induction, despite patients receiving fewer L-asp injections (5 *vs.* 8 in patients without CNS disease) to reduce the rate of CNS thrombosis due to increased intrathecal treatment. This contrasts with both the MRC UKALL XII/ECOG E2993 trial, in which patients with CNS involvement had both higher mortality rates in remission and an increased risk of relapse,[4] and with a previous study of our group (GET-LALA) conducted before the pediatric-inspired treatment era, where the induction death rate was twice as high (10%) in these patients.[6]

Patients included in the MRC UKALL XII/ECOG E2993 trial received intrathecal therapy and cranial radiation as CNS-directed therapy and were likely to proceed to allogeneic SCT.[4] Their systemic CNS-directed therapy was, however, less intense, with high-dose methotrexate administered later during the treatment course.[4] In contrast to this study, our previous GET-LALA trial had showed that patients with initial CNS involvement receiving autologous or allogeneic SCT had better outcomes than patients receiving chemotherapy alone. However, the global results of this older protocol were far inferior to modern regimens, with few patients receiving systemic chemotherapy with good CNS penetration or cranial radiation therapy.[6] Moreover, the rate of CNS relapse remained high after allogeneic SCT, including TBI or TBI-free conditioning regimens, in patients with previous CNS involvement.[28-31] Although the number of patients is limited, this questions whether allogeneic SCT is the most efficient way to reduce relapse in patients with CNS disease at diagnosis, as it might for patients with other high-risk features such as MRD.[16]

The inferior outcome in our patients with initial CNS disease was more likely due to toxicity, since higher mortality in remission was observed in patients who received hyperfractionated cyclophosphamide and/or proceeded to allogeneic SCT. Due to the small number of patients with initial CNS disease, these conclusions are, however, speculative. There is no clear explanation for increased toxicity of hyperfractionated cyclophosphamide in CNS patients as it was not observed in the entire cohort of randomized patients.[14] Higher NRM was no longer observed after censoring patients who proceeded to allogeneic SCT, suggesting that many patients suffered from transplant-related toxicity. On the other hand, the cumulative incidence of relapse was higher in patients who did not proceed to allogeneic SCT. This largely explains why there was no difference in OS between patients receiving SCT or not in the landmark analysis. It might be argued that, while some relapses may be prevented with allogeneic SCT, there was no benefit in OS due to increased toxicity.

Despite our small number of patients with CNS involvement, we found no association between cranial irradiation during consolidation and outcome in the landmark analysis. As in other studies, cranial irradiation was recommended for patients with CNS involvement in many clinical trials but was seldom performed (27/55 patients in the GRAALL-2005 study).[4] In children with initial CNS involvement, the omission of cranial irradiation was associated with a slight increase in risk of CNS relapse in some studies but did not affect overall survival when both intrathecal and systemic CNS-directed chemotherapy were intensified.[10,22,32-35] As cranial irradiation does not completely mitigate the risk of CNS relapse but may induce neurocognitive impairment, many co-operative groups have now abandoned cranial irradiation in all children, including those with initial CNS involvement.[22,33-42] In one study analyzing the outcome of 467 adult patients with ALL who did not receive cranial irradiation during first-line treatment, the risk of CNS relapse was not increased in the 18 patients with initial CNS involvement.[5] To our knowledge, no other study has evaluated the omission of cranial irradiation in adult patients with overt CNS disease. We could not evaluate the association between cranial irradiation and outcomes in CNS-negative patients as this procedure was not exhaustively recorded in CNS-negative patients.

In conclusion, despite improved outcome in adults with ALL included in pediatric-inspired protocols, CNS involvement is still associated with reduced survival, mainly due to excessive toxicity. The historical use of allogeneic SCT did not improve outcome. Because of the rarity of CNS involvement, it is unlikely that different treatment approaches will be tested in these patients in prospective controlled trials. However, it will be important to evaluate the outcome of these patients in the GRAALL-2014 protocol that no longer retains CNS involvement as an indication for allogeneic SCT. Whereas the treatment strategy has remained unchanged for CNS-positive patients, including cranial radiation, our approach to prophylaxis has been modified in CNS-negative patients, with higher doses of systemic methotrexate during consolidation for patients under the age of 45 and an additional seven triple intrathecal injections during therapy,

while cranial radiation is only recommended for patients undergoing allogeneic SCT.

Disclosures

CO acted as a consultant to Incyte and Novartis. MHB was a member of the advisory board of Erytech© and acted as a consultant to Jazz Pharmaceuticals. YC acted as a consultant to MSD, Novartis, Incyte, BMS, Pfizer, Abbvie, Roche, Jazz Pharmaceuticals, Gilead, Amgen, Astra Zeneca, and Servier, and received travel support from MSD, Roche, Gilead, Amgen, Incyte, Abbvie, Janssen, Astra Zeneca, and Jazz Pharmaceuticals.

Contributions

PC, FH, NI, HD, NB and MHB are responsible for study conception and design. CO, NB and MHB are responsible for data analysis and interpretation. All authors are responsible for the data collection and assembly and writing the paper, and approved the final version for publication.

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Data-sharing statement

The datasets analyzed during the current study are available from the corresponding author upon reasonable request.

References

1. Yao H, Price TT, Cantelli G, et al. Leukaemia hijacks a neural mechanism to invade the central nervous system. Nature. 2018;560(7716):55-60.
2. Savino AM, Fernandes SI, Olivares O, et al. Metabolic adaptation of acute lymphoblastic leukemia to the central nervous system microenvironment depends on stearoyl-CoA desaturase. Nat Cancer. 2020;1(10):998-1009.
3. Omura G, Moffitt S, Vogler W, Salter M. Combination chemotherapy of adult acute lymphoblastic leukemia with randomized central nervous system prophylaxis. Blood. 1980;55(2):199-204.
4. Lazarus HM, Richards SM, Chopra R, et al. Central nervous system involvement in adult acute lymphoblastic leukemia at diagnosis: results from the international ALL trial MRC UKALL XII/ECOG E2993. Blood. 2006;108(2):465-472.
5. Sancho J-M, Ribera J-M, Oriol A, et al. Central nervous system recurrence in adult patients with acute lymphoblastic leukemia: frequency and prognosis in 467 patients without cranial irradiation for prophylaxis. Cancer. 2006;106(12):2540-2546.
6. Reman O, Pigneux A, Huguet F, et al. Central nervous system involvement in adult acute lymphoblastic leukemia at diagnosis and/or at first relapse: results from the GET-LALA group. Leuk Res. 2008;32(11):1741-1750.
7. DeAngelo DJ, Stevenson KE, Dahlberg SE, et al. Long-term outcome of a pediatric-inspired regimen used for adults aged 18-50 years with newly diagnosed acute lymphoblastic leukemia. Leukemia. 2015;29(3):526-534.
8. Stock W, Luger SM, Advani AS, et al. A pediatric regimen for older adolescents and young adults with acute lymphoblastic leukemia: results of CALGB 10403. Blood. 2019;133(14):1548-1559.
9. Winick N, Devidas M, Chen S, et al. Impact of Initial CSF findings on outcome among patients with National Cancer Institute standard- and high-risk B-cell acute lymphoblastic leukemia: a report from the Children's Oncology Group. J Clin Oncol. 2017;35(22):2527-2534.
10. Sirvent N, Suciu S, De Moerloose B, et al. CNS-3 status remains an independent adverse prognosis factor in children with acute lymphoblastic leukemia (ALL) treated without cranial irradiation: results of EORTC Children Leukemia Group study 58951. Arch Pediatr. 2021;28(5):411-416.
11. Boissel N, Auclerc M-F, Lhéritier V, et al. Should adolescents with acute lymphoblastic leukemia be treated as old children or young adults? Comparison of the French FRALLE-93 and LALA-94 trials. J Clin Oncol. 2003;21(5):774-780.
12. Huguet F, Leguay T, Raffoux E, et al. Pediatric-inspired therapy in adults with Philadelphia chromosome-negative acute lymphoblastic leukemia: the GRAALL-2003 study. J Clin Oncol. 2009;27(6):911-918.
13. Ram R, Wolach O, Vidal L, Gafter-Gvili A, Shpilberg O, Raanani P. Adolescents and young adults with acute lymphoblastic leukemia have a better outcome when treated with pediatric-inspired regimens: systematic review and meta-analysis. Am J Hematol. 2012;87(5):472-478.
14. Huguet F, Chevret S, Leguay T, et al. Intensified therapy of acute lymphoblastic leukemia in adults: report of the randomized GRAALL-2005 clinical trial. J Clin Oncol. 2018;36(24):2514-2523.
15. Advani AS, Larsen E, Laumann K, et al. Comparison of CALGB 10403 (Alliance) and COG AALL0232 toxicity results in young adults with acute lymphoblastic leukemia. Blood Adv. 2021;5(2):504-512.
16. Dhédin N, Huynh A, Maury S, et al. Role of allogeneic stem cell transplantation in adult patients with Ph-negative acute lymphoblastic leukemia. Blood. 2015;125(16):2486-2496.
17. Ribera J-M, Morgades M, Ciudad J, et al. Chemotherapy or allogeneic transplantation in high-risk Philadelphia chromosome-negative adult lymphoblastic leukemia. Blood. 2021;137(14):1879-1894.
18. Maury S, Chevret S, Thomas X, et al. Rituximab in B-lineage adult acute lymphoblastic leukemia. N Engl J Med. 2016;375(11):1044-1053.
19. Delgado J, Pereira A, Villamor N, Lopez-Guillermo A, Rozman C. Survival analysis in hematologic malignancies: recommendations for clinicians. Haematologica. 2014;99(9):1410-1420.
20. Burger B, Zimmermann M, Mann G, et al. Diagnostic cerebrospinal fluid examination in children with acute lymphoblastic leukemia: significance of low leukocyte counts with blasts or traumatic lumbar puncture. J Clin Oncol. 2003;21(2):184-188.
21. Levinsen M, Taskinen M, Abrahamsson J, et al. Clinical features and early treatment response of central nervous system involvement in childhood acute lymphoblastic leukemia: CNS

involvement in childhood ALL. Pediatr Blood Cancer. 2014;61(8):1416-1421.
22. Tang J, Yu J, Cai J, et al. Prognostic factors for CNS control in children with acute lymphoblastic leukemia treated without cranial irradiation. Blood. 2021;138(4):331-343.
23. Del Principe MI, Buzzatti E, Piciocchi A, et al. Clinical significance of occult central nervous system disease in adult acute lymphoblastic leukemia. A multicenter report from the Campus ALL Network. Haematologica. 2019;106(1):39-45.
24. Thastrup M, Marquart HV, Levinsen M, et al. Flow cytometric detection of leukemic blasts in cerebrospinal fluid predicts risk of relapse in childhood acute lymphoblastic leukemia: a Nordic Society of Pediatric Hematology and Oncology study. Leukemia. 2020;34(2):336-346.
25. Córdova-Serrano RD, Almanza-Huante E, Fernández-Sánchez E, Hernández-Alcántara A, Espinosa-Bautista K. Central nervous system (CNS) involvement has an adverse impact on survival in newly diagnosed adult acute lymphoblastic leukemia (ALL) assessed by flow cytometry. Leuk Lymphoma. 2021;62(13):3264-3270.
26. Garcia KA, Cherian S, Stevenson PA, et al. Cerebrospinal fluid flow cytometry and risk of central nervous system relapse after hyperCVAD in adults with acute lymphoblastic leukemia. Cancer. 2022;128(7):1411-1417.
27. Ranta S, Nilsson F, Harila-Saari A, et al. Detection of central nervous system involvement in childhood acute lymphoblastic leukemia by cytomorphology and flow cytometry of the cerebrospinal fluid: diagnosis of CNS leukemia in children by flow cytometry. Pediatr Blood Cancer. 2015;62(6):951-956.
28. Chantepie SP, Mohty M, Tabrizi R, et al. Treatment of adult ALL with central nervous system involvement at diagnosis using autologous and allogeneic transplantation: a study from the Société Française de Greffe de Moelle et de Thérapie Cellulaire. Bone Marrow Transplant. 2013;48(5):684-690.
29. Aldoss I, Al Malki MM, Stiller T, et al. Implications and management of central nervous system involvement before allogeneic hematopoietic cell transplantation in acute lymphoblastic leukemia. Biol Blood Marrow Transplant. 2016;22(3):575-578.
30. Shigematsu A, Kako S, Mitsuhashi K, et al. Allogeneic stem cell transplantation for adult patients with acute lymphoblastic leukemia who had central nervous system involvement: a study from the Adult ALL Working Group of the Japan Society for Hematopoietic Cell Transplantation. Int J Hematol. 2017;105(6):805-811.
31. Esfandbod M, Enshaei M, Monzavi SM, Kabootari M, Behfar M, Hamidieh AA. Radiation-free myeloablative allogeneic hematopoietic stem cell transplantation for adult acute lymphoblastic leukemia: a comparison of outcomes between patients with and without central nervous system involvement. Leuk Res. 2021;111:106703.
32. Wilejto M, Giuseppe GD, Hitzler J, Gupta S, Abla O. Treatment of young children with CNS-positive acute lymphoblastic leukemia without cranial radiotherapy: treating children with CNS ALL without CRT. Pediatr Blood Cancer. 2015;62(11):1881-1885.
33. Vora A, Andreano A, Pui C-H, et al. Infuence of cranial radiotherapy on outcome in children with acute lymphoblastic leukemia treated with contemporary therapy. J Clin Oncol. 2016;34(9):919-926.
34. Taskinen M, Oskarsson T, Levinsen M, et al. The effect of central nervous system involvement and irradiation in childhood acute lymphoblastic leukemia: lessons from the NOPHO ALL-92 and ALL-2000 protocols. Pediatr Blood Cancer. 2017;64(2):242-249.
35. Pui C-H, Bowman WP, Ribeiro RC, et al. Treating childhood acute lymphoblastic leukemia without cranial irradiation. N Engl J Med. 2009;360(26):2730-2741.
36. Jeha S, Pei D, Choi J, et al. Improved CNS control of childhood acute lymphoblastic leukemia without cranial irradiation: St Jude Total Therapy Study 16. J Clin Oncol. 2019;37(35):3377-3391.
37. Spiegler BJ, Kennedy K, Maze R, et al. Comparison of long-term neurocognitive outcomes in young children with acute lymphoblastic leukemia treated with cranial radiation or high-dose or very high-dose intravenous methotrexate. J Clin Oncol. 2006;24(24):3858-3864.
38. Zhou C, Zhuang Y, Lin X, Michelson AD, Zhang A. Changes in neurocognitive function and central nervous system structure in childhood acute lymphoblastic leukaemia survivors after treatment: a meta-analysis. Br J Haematol. 2020;188(6):945-961.
39. Zając-Spychała O, Pawlak M, Karmelita-Katulska K, et al. Anti-leukemic treatment-induced neurotoxicity in long-term survivors of childhood acute lymphoblastic leukemia: impact of reduced central nervous system radiotherapy and intermediate-to high-dose methotrexate. Leuk Lymphoma. 2018;59(10):2342-2351.
40. Marwaha RK, Kulkarni KP, Bansal D, Trehan A. Central nervous system involvement at presentation in childhood acute lymphoblastic leukemia: management experience and lessons. Leuk Lymphoma. 2010;51(2):261-268.
41. Waber DP, Turek J, Catania L, et al. Neuropsychological outcome from a randomized trial of triple intrathecal chemotherapy compared with 18 Gy cranial radiation as CNS treatment in acute lymphoblastic leukemia: findings from Dana-Farber Cancer Institute ALL Consortium Protocol 95-01. J Clin Oncol. 2007;25(31):4914-4921.
42. Jacola LM, Krull KR, Pui C-H, et al. Longitudinal assessment of neurocognitive outcomes in survivors of childhood acute lymphoblastic leukemia treated on a contemporary chemotherapy protocol. J Clin Oncol. 2016;34(11):1239-1247.