American Society of Hematology
2021 L Street NW, Suite 900,
Washington, DC 20036
Phone: 202-776-0544 | Fax 202-776-0545
editorial@hematology.org

Age‐adapted chemotherapy and MRD‐oriented transplant for Ph-negative acute lymphoblastic leukemia - GRAALL-2014 trial

Tracking no: BLD-2025-029611R3

Nicolas Boissel (Hopital Saint-Louis, AP-HP, France) sylvie chevret (saint louis hospital, France) Françoise Rigal-Huguet (Institut de Cancérologie de Toulouse, France) Thibaut Leguay (Hopital du Haut-Lévèque, France) Mathilde Hunault (CHU, France) Carlos Graux (CHU UCL Namur - Godinne, Belgium) Yves Chalandon (University Hospital of Geneva, Switzerland) Eric Delabesse (University hospital of Toulouse, university of Toulouse 3, Center of Research on Cancer of Toulouse, France) Yosr Hicheri (Institut Paoli Calmettes, France) Patrice Chevallier (Centre Hospitalo-Universitaire, France) Marie Balsat (Hôpital Lyon sud, France) Cedric Pastoret (Rennes University Hospital, France) Martine Escoffre-Barbe (CHU Rennes, France) Florence Pasquier (Gustave Roussy, France) Magalie Joris (CHU Amiens, France) Anne Thiebaut (CHU de Grenoble, France) Anne Huynh (IUCT Oncopole, France) Nathalie Dhedin (SAINT LOUIS HOSPITAL, France) Emilie Lemasle (centre Henri Becquerel, France) Caroline Bonmati (Service d'Hématologie, Centre Hospitalier Universitaire de Nancy, France) Sebastien Maury (Hopital Henri Mondor, France) Gaelle Guillerm (CHRU Brest, France) Ana Berceanu (Besançon University Hospital, France) Urs Schanz (University Hospital of Zurich, Switzerland) Thomas Cluzeau (Côte d'Azur university, CHU of Nice, France) Pascal Turlure (chu limoges, France) Philippe Rousselot (Centre Hospitalier de Versailles, University Versailles Paris-Saclay, France) Bernard De Prijck (CHU de Liège, Belgium) Nathalie Grardel (CHRU de lille, France) Marie-Christine Béné (University Hospital Nantes, France) Marina Lafage-Pochitaloff (Aix-Marseille University, France) Wendy Cuccuini (Saint-Louis Hospital, France) Norbert Ifrah (CHU Angers, France) Véronique Lheritier (Centre Hospitalier Lyon Sud, France) Vahid Asnafi (Laboratory of Onco-Hematology, Assistance Publique-Hopitaux De Paris (AP-HP), Hopital Necker Enfants-Malades, France) Emmanuelle Clappier (Saint-Louis Hospital, AP-HP, France) Hervé Dombret (Paris Saint-Louis Leukemia Institute, Université Paris Cité, Hôpital Saint-Louis, Assistance Publique - Hôpitaux de Paris (AP-HP), France)

Abstract:
The Group for Research in Adult Acute Lymphoblastic Leukemia (GRAALL)-2014 trial (ClinicalTrials.gov: NCT02617004, NCT02619630) evaluated an intensive, age-adapted protocol for adults aged 18-59 years with Philadelphia chromosome-negative acute lymphoblastic leukemia (ALL). The trial was motivated by findings from the previous GRAALL-2005 study, which reported excessive toxicity from pediatric-inspired therapy in older patients and no added benefit from allogeneic hematopoietic stem cell transplantation (alloHSCT) in those with an early favorable response to treatment. Thus, the GRAALL-2014 protocol aimed to reduce treatment-related toxicity in patients aged ≥45 years and to limit alloHSCT to patients with poor measurable residual disease (MRD) responses. A total of 743 patients was included, and outcomes were compared to those of the GRAALL-2005 trial. The GRAALL-2014 demonstrated reduced early mortality and higher complete remission rates in patients aged ≥45 years. MRD-guided transplant decisions reduced alloHSCT indications by approximately 50%. While older patients experienced a higher cumulative incidence of relapse, no significant difference in disease-free survival (DFS) was observed compared to historical cohorts across age subgroups. The overall 4-year DFS was 57.1% (95% CI, 53.4-61.1). Notably, 4-year overall survival improved significantly, from 65.5% (95%CI, 61.7-69.8) to 71.7% (95%CI, 67.7-76.0) in younger patients (p=0.031) and from 49.6% (95%CI, 43.5-56.5) to 59.5% (95%CI, 53.5-66.3) in older patients (p=0.011). These findings highlight the value of individualized treatment strategies, balancing efficacy and safety. Future studies should investigate the integration of immunotherapy to further reduce treatment intensity and improve outcomes.

Conflict of interest: COI declared - see note

COI notes: NB received honoraria from Amgen, Novartis, Pfizer, Servier, Gilead. HD received honoraria from Amgen and Servier. FH received honoraria from Amgen, Novartis, Pfizer, Servier, Gilead. AH : received honoraria from Novartis, Sanofi, Immedica, Pierre Fabre, and travel support from Medac, Sanofi, Novartis and Pierre Fabre. PC received honoraria from Takeda, Servier, Abbvie, Priothera, and travel support from Abbvie. TL received honoraria from Servier, Amgen

Preprint server: No;

Author contributions and disclosures: V.L. provided administrative support; N.B., F.H., T.L., M.H., C.G., Y.C., Y.H., P.C., M.B., M.E.-B., F.P., J.-P.M., A.T., A.H., N.D., E.L., C.B., S.M., G.G., A.B., U.S., T.C., P.T., P.R., B.D.P., N.I., and H.D. provided study materials or patients; N.B., V.L., E.C., V.A., and H.D. performed collection and assembly of data; E.D., C.P., N.G., M.-C.B., M.L-P., V.A., E.C. R.K., E.C., and W.C. performed central review of molecular and cytogenetic data; S.C., N.B., and H.D. performed data analysis and interpretation; N.B., S.C., V.L., and H.D. wrote the manuscript; and all authors performed conception and design and provided final approval of the manuscript.

Non-author contributions and disclosures: No;

Agreement to Share Publication-Related Data and Data Sharing Statement: Not applicable

Clinical trial registration information (if any): The GRAALL-2014 trial included 2 sub-studies for Ph-negative B-lineage ALL (B-ALL) (ClinicalTrials.gov Identifier: NCT02617004) and T-lineage ALL (T-ALL) (NCT02619630) reported in this manuscript.

Age-adapted chemotherapy and MRD-oriented transplant for Ph-negative acute lymphoblastic leukemia - GRAALL-2014 trial

Nicolas Boissel[1,2], Sylvie Chevret[2,3], Françoise Huguet[4], Thibaut Leguay[5], Mathilde Hunault[6], Carlos Graux[7], Yves Chalandon[8], Eric Delabesse[9], Yosr Hicheri[10], Patrice Chevallier[11], Marie Balsat[12], Cédric Pastoret[13], Martine Escoffre-Barbe[14], Florence Pasquier[15], Magalie Joris[16], Anne Thiebaut[17], Anne Huynh[4], Nathalie Dhédin[1,2], Emilie Lemasle[18], Caroline Bonmati[19], Sébastien Maury[20], Gaëlle Guillerm[21], Ana Berceanu[22], Markus Manz[23], Thomas Cluzeau[24], Pascal Turlure[25], Philippe Rousselot[26], Bernard de Prijck[27], Nathalie Grardel[28], Marie C. Béné[11], Marina Lafage-Pochitaloff[29,30], Wendy Cuccuini[30,31], Norbert Ifrah[6], Véronique Lhéritier[32], Vahid Asnafi[33], Emmanuelle Clappier[2,31], Hervé Dombret[2,34.]

Affiliations:

1. Adolescent and Young Adult Hematology Department, St-Louis Hospital, APHP, Paris, France.
2. UMR 1342, Saint-Louis Research Institute, Université Paris Cité, Inserm, Paris, France.
3. Team of Biostatistics, Saint-Louis Hospital, APHP, Paris, France.
4. Department of Hematology, Institut Universitaire du Cancer-Oncopole, CHU de Toulouse, Toulouse, France.
5. Hematology Department, Bordeaux University Hospital, Pessac, France.
6. Department of Hematology, CHU Angers, Angers, France.
7. Université Catholique de Louvain, CHU UCL Namur, Yvoir, Belgium.
8. Department of Hematology, Hôpitaux Universitaires de Genève, Geneva, Switzerland.
9. Laboratory of Hematology, University Hospital of Toulouse, Toulouse, France.
10. Department of Hematology, Institut Paoli-Calmettes, Marseille, France.
11. Hematology Department, Nantes University Hospital, Nantes, France.
12. Hematology Department, Centre Hospitalier Lyon Sud, Lyon, France.
13. Hematology Laboratory, University Hospital of Rennes, Rennes, France.
14. Hôpital de Pontchaillou, Centre Hospitalier Universitaire de Rennes, Rennes, France.
15. Département clinique d'hématologie, Gustave Roussy, INSERM UMR1170, Villejuif, France.
16. Hematology Department, Amiens University Hospital, Amiens, France.
17. Department of Hematology, CHU Grenoble Alpes, Grenoble, France.
18. Hematology Department, Henri-Becquerel Cancer Center, Rouen, France.
19. Hematology Department, CHU Nançy, Vandoeuvre-les-Nancy, France.
20. Service d'Hématologie clinique, Hôpitaux universitaires Henri-Mondor, AP-HP, Créteil, France.
21. Department of Hematology, Brest University Hospital, Brest, France.
22. Clinical Hematology, Besançon University Hospital, Besançon, France.
23. Department of Medical Oncology and Hematology, University Hospital Zurich, Zurich, Switzerland.
24. Department of Hematology, Université Côte d'Azur, Centre Hospitalier Universitaire, Nice, France.
25. Department of Clinical Hematology, Limoges University Hospital, Limoges, France.
26. Hematology Department, Centre Hospitalier de Versailles, Le Chesnay Rocquencourt, France.
27. Laboratory of Hematology, Centre Hospitalier Universitaire Sart Tilman, Liège, Belgium.
28. Laboratory of Hematology, Centre Hospitalier Universitaire Lille, Lille, France.
29. Hematological cytogenetics laboratory , Timone Hospital, Aix Marseille University, Marseille, France.
30. Groupe Francophone de Cytogénétique Hématologique , Paris, France
31. Laboratory of Hematology, Saint-Louis Hospital, APHP, Paris, France.
32. Group for Research on Adult Acute Lymphoblastic Leukemia (GRAALL), Centre Hospitalier Lyon Sud, Lyon, France.
33. Laboratory of Onco-Hematology, Necker University Hospital, Paris, France.
34. Adult Hematology Department, Saint-Louis Hospital, APHP, Paris, France.

Correspondance:

Nicolas Boissel, Saint-Louis Hospital, APHP
1 avenue Claude Vellefaux, 75010 PARIS, France
nicolas.boissel@aphp.fr

Key points:

- In patients over 45 years old, age-adapted protocol improved complete remission rate and decreased non-relapse mortality
- MRD-based transplant decisions reduced transplant indications and ensured the procedure was limited to patients most likely to benefit

Running title

MRD-oriented transplant for adult Ph- ALL

Data Sharing Statement:

Dataset can be transmitted upon request to the corresponding author.

Word count: 3499

References: 23

Figures: 4 Tables, 4 figures

Abstract: 237 words

Abstract

The Group for Research in Adult Acute Lymphoblastic Leukemia (GRAALL)-2014 trial (ClinicalTrials.gov: NCT02617004, NCT02619630) evaluated an intensive, age-adapted protocol for adults aged 18-59 years with Philadelphia chromosome-negative acute lymphoblastic leukemia (ALL). The trial was motivated by findings from the previous GRAALL-2005 study, which reported excessive toxicity from pediatric-inspired therapy in older patients and no added benefit from allogeneic hematopoietic stem cell transplantation (alloHSCT) in those with an early favorable response to treatment. Thus, the GRAALL-2014 protocol aimed to reduce treatment-related toxicity in patients aged ≥45 years and to limit alloHSCT to patients with poor measurable residual disease (MRD) responses. A total of 743 patients was included, and outcomes were compared to those of the GRAALL-2005 trial. The GRAALL-2014 demonstrated reduced early mortality and higher complete remission rates in patients aged ≥45 years. MRD-guided transplant decisions reduced alloHSCT indications by approximately 50%. While older patients experienced a higher cumulative incidence of relapse, no significant difference in disease-free survival (DFS) was observed compared to historical cohorts across age subgroups. The overall 4-year DFS was 57.1% (95% CI, 53.4–61.1). Notably, 4-year overall survival improved significantly, from 65.5% (95%CI, 61.7-69.8) to 71.7% (95%CI, 67.7-76.0) in younger patients (p=0.031) and from 49.6% (95%CI, 43.5-56.5) to 59.5% (95%CI, 53.5-66.3) in older patients (p=0.011). These findings highlight the value of individualized treatment strategies, balancing efficacy and safety. Future studies should investigate the integration of immunotherapy to further reduce treatment intensity and improve outcomes.

Introduction

Before the advent of immunotherapy, significant advances in the treatment outcomes of adults with Philadelphia chromosome-negative (Ph-negative) acute lymphoblastic leukemia (ALL) were achieved with more intensive chemotherapy regimens.[1–3] Our research group and others have demonstrated substantial survival gains in patients up to 60 years old using pediatric-inspired treatment strategies.[1,2,4,5] However, these therapeutic modifications have raised concerns, particularly about increased toxicity with age, which questions the applicability of these protocols in older patients.[2,6] Furthermore, they have prompted a reevaluation of the indications for allogeneic hematopoietic stem cell transplantation (alloHSCT) in first remission.[7,8]

In the earlier GRAALL-2003 and GRAALL-2005 protocols, our group demonstrated the benefits of intensive strategies but also highlighted challenges in treating older adults.[1,2] Notably, we observed an increase in non-relapse mortality with age, particularly beyond 45 years, mainly due to the early use of asparaginase and a higher incidence of infectious episodes during post-remission phases. The GRAALL-2014 protocol adapted the pediatric-inspired regimen to patient age, aiming to reduce exposure to corticosteroids, asparaginase, and anthracyclines during induction and late intensification to lower induction-related and non-relapse mortality.

Although alloHSCT has historically been central to consolidate the treatment of younger adults with ALL,[9] reduced relapse rates with more intensive protocols have questioned its necessity in first complete remission (CR). In the GRAALL-2005 protocol, where we maintained broad transplant indications based on baseline disease characteristics and early response to therapy, patients who benefited from alloHSCT were mostly those with high

measurable residual disease (MRD) levels at the end of induction or consolidation.[10] While transplant indications remain heterogeneous in adults,[7] the GRAALL-2014 protocol specifically restricted alloHSCT indications in first CR to patients with a poor early MRD response.

In this study, we report the outcomes of the GRAALL-2014 trial, aimed at patients aged 18 to 59 years with Ph-negative ALL. Patient characteristics and outcomes were compared with those from the historical GRAALL-2005 study, both overall and within the age groups of 18-44 and 45-59 years.

Patient and methods

Trial oversight

The GRAALL-2014 trial was conducted within the 79 French, Belgian and Swiss centers of the GRAALL network (Supplementary materials). The trial was designed and reviewed by the GRAALL scientific committee and approved by the French Ethical Committee Review Board (Comité de Protection de la Personne) Ile-de-France VI. In accordance with the Declaration of Helsinki, informed consent was obtained before study entry. The trial included three sub-studies for Ph-negative B-lineage ALL (B-ALL) (ClinicalTrials.gov Identifier: NCT02617004), T-lineage ALL (T-ALL) (NCT02619630), and Ph-positive B-ALL (NCT02611492). The results of the Ph-positive sub-study have been recently published.[11]

Study population

Patients 18 to 59 years of age with newly diagnosed Ph-negative B-ALL or T-ALL were eligible in the absence of other evolving malignancy, pregnancy, HIV infection, active viral hepatitis, or other condition that contraindicated intensive chemotherapy. Patients were required to have undergone sampling for a centralized characterization of ALL.

Diagnosis and response assessment

The initial evaluation was conducted at local sites, including cytomorphology, immunophenotyping, cytogenetics (chromosome banding and FISH analyses including *BCR::ABL1* and *KMT2A* screening), and *BCR::ABL1* molecular screening, following standard protocols.[12] Molecular analyses of B-ALL and T-ALL were centralized at Saint-Louis and Necker University Hospitals in Paris, respectively. Response assessment was based on cytomorphology at Day 35. MRD was evaluated at four timepoints (TP): after induction (D35, TP1), after first (TP2) and second (TP3) consolidations, and either before maintenance therapy or day 100 post-alloHSCT (TP4), using IG/TR qPCR as per EuroMRD guidelines in six accredited centers.

Response criteria

Prednisone response was assessed on day 8 (D8) following the initiation of prednisone pre-phase treatment and defined by peripheral blood blast count of less than $1x10^9$/L. Complete remission (CR) was achieved when the patient exhibits less than 5% blasts in the bone marrow, no evidence of disease in other sites and full recovery of peripheral blood counts (platelets > $100x10^9$/L, absolute neutrophil count [ANC] >$1x10^9$/L). Relapse was defined by the recurrence of leukemia cells in the bone marrow (greater than 5% blasts) after achieving complete remission, or by the appearance of leukemia cells in any extra-medullary site.

Risk-adapted therapy

Table 1 presents the treatment regimens outlined in the age-adapted GRAALL-2014 protocol, highlighting key modifications from the preceding GRAALL-2005 trial. The protocol initiated with a 7-day prednisone pre-phase, followed by a five-drug induction regimen. In cases of

induction failure, a salvage course was administered. For patients with persistent toxicity that precluded further planned chemotherapy courses, up to two standby blocks were permitted. The consolidation regimen incorporated high-dose methotrexate and high-dose cytarabine. AlloHSCT was indicated in patients with very high-risk (VHR) criteria that included late CR (CR after salvage), MRD at TP1 (MRD1) ≥ 10^{-3} and/or MRD at TP2 (MRD2) ≥ 10^{-4} (Supplementary Tables 1 and 2). Recommended donors were compatible sibling, matched, or 9/10 mismatched unrelated donors. The recommended conditioning regimen consisted of total body irradiation (TBI) at 12 Gy and cyclophosphamide at 60 mg/kg/day for two days for patients under 45, and TBI at 8 Gy with fludarabine at 30 mg/m^2/day for four days for those aged 45 to 59 years old. The recommended sources of stem cells were bone marrow for TBI 12 Gy and cyclophosphamide and peripheral blood for TBI 8 Gy and fludarabine conditioning regimen. Transplantation was scheduled after the second consolidation course. Patients ineligible for transplantation proceeded to late intensification, a third consolidation course, and a two-year maintenance phase. Initial CNS involvement warranted additional intrathecal therapy during the pre-phase, induction, and first consolidation, plus CNS irradiation prior to alloHSCT (boost) or during the maintenance phase. Additionally, two nested phase 2 studies temporarily included patients having high-risk (HR) criteria for treatment with blinatumomab (for B-ALL, N=94, from July 2018) or nelarabine (for T-ALL, N=88, from April 2017 to December 2020) during consolidation phases (Figure 1B-D). High-risk (HR) criteria were defined as TP1 MRD ≥ 10^{-4} (including late complete remission), and/or the presence of *KMT2A* rearrangement (*KMT2A*-r) or *IKZF1* intragenic deletion (*IKZF1*del) in B-ALL, and an unfavorable 4-gene (*NOTCH1*, *FBXW7*, *RAS*, and *PTEN*) classifier in T-ALL as previously reported (Supplementary Table 2). The results of these two nested studies will be reported elsewhere. Patients without high-risk features were classified as standard-risk (SR).

Outcomes

The main endpoint was disease-free survival (DFS) at 4 years, defined as the time from CR to either relapse or death, whichever occurred first. Secondary outcomes were cumulative incidence of relapse (CIR), non-relapse mortality (NRM), and overall survival (OS), all measured from study enrollment.

Statistical methods

Sample size justification. The primary objective was to demonstrate, through a non-controlled study, that the 4-year DFS rate in the SR population was non-inferior to the historical reference of 60%, allowing for a 15% inferiority margin, with a requirement of 110 patients, a one-sided type I error rate of 0.05, and a statistical power of 0.90.

According to the protocol, primary analyses were performed on the whole population of included patients, i.e., whatever the treatments received and whatever the objective of superiority or non-inferiority demonstration of the sub-studies. Data cutoff was February, 2, 2024. We used individual patient data from the GRAALL-2005 protocol as the external control group.

Statistical analysis. We report median and interquartile ranges (IQR) or percentages for summary statistics, unless specified otherwise. Differences across protocols in binary variables including the rates of CR, early death (death from any cause occurring during induction phase), or salvage among groups, were assessed using the exact Fisher test. For continuous variables, differences were tested using nonparametric Wilcoxon rank sum test. Time-to-event data, including OS and DFS, were calculated from study enrollment or CR, respectively, up to event occurrence or last follow-up, compared by a two-sided log-rank

test. Non-relapse mortality and relapse were analyzed as competing risk events, with cumulative incidence estimated using nonparametric estimators and compared using Gray test. Cox proportional hazards models were fitted to quantify the size of protocol effect on each endpoint, after checking the proportional hazards assumption by examination of Schoenfeld residuals and Grambsch and Therneau tests.[13,14] Gail's interaction statistic was used to test the heterogeneity in the protocol effect on the outcomes across age groups (<, ≥45). In addition, two post-hoc sensitivity analyses were performed. To assess the potential impact of patients who received blinatumomab or nelarabine in the GRAALL-2014 nested sub-studies, a first sensitivity analysis compared outcomes between GRAALL-2005 and the subgroup of patients enrolled in GRAALL-2014 prior to the activation of these sub-studies (April 2017). The second post-hoc sensitivity analysis explored outcomes according to lineage, specifically comparing B-ALL and T-ALL. Finally, Simon and Makuch survival estimates were used to evaluate disease-free and overall survival according to alloHSCT as a time-dependent variable.[15] This method accounts for the fact that patients may move from the non-transplanted to the transplanted group over time, thus allowing an appropriate estimation of survival probabilities in the context of time-dependent treatment allocation. Analyses were performed using the R statistical software version 4.1.1.

Results

Patient characteristics

Between December 2015 and December 2020, 768 patients were included in the two Ph-negative B-ALL and T-ALL sub-studies. Among these 768 patients, 23 had non-eligibility criteria (12 with Ph-positive ALL, 5 with not confirmed ALL, 2 with wrong lineage ALL, 2 with Burkitt leukemia, 1 with no baseline centralized assessment, 1 with no health insurance

coverage) and 2 patients withdrew consent. These 25 patients were excluded from the primary modified intention-to-treat (ITT) analysis (Figure 1A).

The modified ITT population thus included 743 patients with Ph-negative B-ALL (n=489) or T-ALL (n=254), with consistent demographic and clinical characteristics close to those observed in the GRAALL-2005 trial (Table 2). Notably, the median age was 35 years (interquartile range, 24-48), with 32.2% of the cohort aged 45 years and older. Over the 10-year interval between the studies, the incidence of T-ALL (34.2%), CNS involvement (CNS-3 status, 8.0%), and the median white blood cell count at diagnosis ($10.6x10^9$/L) remained stable. Additionally, the proportions of B-ALL/T-ALL patients with a baseline white blood cell count (WBC) ≥$30/100x10^9$/L were 21.5%/19.3%, respectively, indicating no significant change from the previous trial. Although the GRAALL-2014 trial noted an increase in the rate of patients benefiting from cytogenetic characterization in B-ALL and molecular characterization in T-ALL compared to the GRAALL-2005 trial, the prevalence of key genetic aberrations such as *KMT2A*-r, *IKZF1*del, or an unfavorable T-ALL classifier showed no significant variation (Table 2).

Response to induction and salvage

In the GRAALL-2014 trial, 76.2% (565/742) of patients demonstrated a favorable D8 prednisone response (peripheral blasts below $1x10^9$/L after one week), consistent with the previous trial (Table 3). At the end of induction course, 89.2% (663/743) achieved a CR, with a higher CR rate in patients <45y (90.9%) compared to those ≥45y (85.8%). The GRAALL-2014 recorded a significant reduction in early mortality among patients aged ≥45y compared to the previous GRAALL-2005 study (3.3% *vs.* 11.0%, p=0.0013), with all early deaths occurring during induction and none during salvage. Therefore, patients ≥45y were more frequently

proposed to receive a salvage course to achieve CR (9.2% *vs.* 4.5%, p=0.047). Complete remission was achieved in 61.2% (30/49) of patients requiring salvage, with no variation by age or among trials. Overall, CR after one or two courses was reached in 93.3% (693/743) of patients, with a notably higher rate in those ≥45y when compared to prior trial (91.6% *vs.* 85.7%, p=0.045), attributed to reduced early death combined with a higher chance to receive salvage course.

Eligibility to alloHSCT

Among the 693 patients who achieved CR, 213 (30.7%) were classified as VHR and therefore candidates for transplantation. Of these VHR patients, 30 (14.1%) achieved a late CR following salvage course, 140 (65.7%) had a TP1-MRD $\geq 10^{-3}$, and 43 (20.2%) had a TP2-MRD $\geq 10^{-4}$ without prior criteria (late CR or TP1-MRD $\geq 10^{-3}$). Of note, all these assessments were performed prior to patients being exposed to blinatumomab or nelarabine (Supplementary Figure 1). The rate of VHR patients was significantly lower in patients <45y (27.4%) compared to patients ≥45y (37.9%). This population was also significantly enriched in B-ALL with high WBC (p=0.002), *KMT2A*-r (p=0.04) and *IKZF1*del (p<0.001) (Supplementary Table 3). In comparison to the GRAALL-2005 trial and as expected, the fraction of patients eligible for alloHSCT was drastically reduced by more than half (Table 3), specifically by a 2.5 factor in patients <45y and 1.9 in patients ≥45y (p<0.001). A total of 159 patients was eventually transplanted in first CR (22.9%). Among the 213 VHR patients in CR, the rate of transplantation was 66.7% overall (142/213 *vs* 256/476, 54.8% in the GRAALL-2005, p=0.002), 67.7% in younger patients (88/130 *vs* 191/331, 57.7% in GRAALL-2005, p=0.057), and 65.1% in older patients (54/83 *vs* 65/136, 47.8% in GRAALL-2005, p=0.017). Additionally, 17 patients in continuous CR1 who did not meet VHR criteria were transplanted based on

investigator decision. The majority of transplants were performed after a TBI-based conditioning regimen (141/159, 88.7%). The donor was a sibling in 51/159 (32.1%) cases, a matched unrelated donor in 57/159 (35.8%) cases, a mismatched unrelated donor 8/10 or 9/10 in 21/159 (13.2%) cases, a haplo-identical donor in 28 (17.6%) cases, a cord blood in 1 case, and 1 patient with missing data. The choice of donors differed significantly between the two protocols, with especially more haploidentical donors (p<0.001), which contributed to the increased rate of eligible patients actually transplanted in the GRAALL-2014 trial (Supplementary Table 4).

Outcomes

Among the 693 patients who achieved CR, 253 experienced a relapse, 246 patients died, including 38 who were in continuous CR (Table 4). The reduction in dose-intensity and the decreased use of alloHSCT in the GRAALL-2014 trial led to a significant reduction of NRM (sub-distribution hazard ratio [SHR] 0.45; 95%CI, 0.31-0.66; p<0.0001) (Table 4, Figure 2). This reduction was particularly pronounced in older patients (SHR 0.38; 95%CI, 0.22-0.65; p=0.00054) but also observed in younger adults (SHR 0.49, 95%CI, 0.29-0.84, p=0.0093), though with no evidence of treatment-by-age group heterogeneity (p=0.48) (Table 4, Figure 3). Conversely, an increased CIR was observed (SHR 1.25; 95%CI, 1.04-1.50; p=0.016), with no evidence of any significant heterogeneity detected between age groups (p=0.28), though somewhat more pronounced in older patients (SHR 1.45; 95%CI, 1.05-2.01; 40.9% *vs* 29.9%, p=0.026), than in younger adults (SHR 1.16; 95%CI, 0.94-1.45; 36.1% *vs* 29.5%; p=0.17) (Table 4, Figure 3). Overall, there was no improvement in DFS between the two trials (HR 0.99; 95%CI, 0.84-1.16; p=0.89), with 4-year DFS of 57.1% (95%CI, 53.4-61.1) overall and no heterogeneity across age groups (p=0.81). However, OS was improved (HR 0.78; 95%CI, 0.66-

0.92; p=0.0034), with no age-by-protocol interaction (p=0.71), as illustrated by the effect in older (HR 0.74; 95%CI, 0.57-0.96; p=0.011) and in younger patients (HR 0.79; 95%CI, 0.63-0.98; p=0.031). This discrepancy between DFS and OS may be at least partially explained by the recent diversification of salvage therapies, particularly the use of immunotherapy in B-ALL. Supporting this hypothesis, OS improvement was more pronounced in B-ALL (HR 0.73; 95%CI, 0.59-0.89), than in T-ALL (HR 0.89; 95%CI, 0.67-1.19), though such a heterogeneity in effect was not significant (p=0.26) (Supplementary Figure 1). Furthermore, post-relapse overall survival has improved in B-ALL (HR 0.65; 95%CI, 0.50-0.85), while stable in T-ALL (HR 0.86; 95%CI, 0.60-1.23), though such heterogeneity was not significant (p=0.21) (Supplementary Figure 2).

As a subset of patients received blinatumomab or nelarabine starting from consolidation 2 onwards (Figure 1B-C), we conducted sensitivity analyses restricted to the 156 patients enrolled before activation of these nested studies in April 2017 (Supplementary Table 5). These analyses confirmed that the GRAALL-2014 strategy had a consistent impact on NRM and CIR prior to the introduction of these novel agents. Importantly, the limited exposure to blinatumomab or nelarabine among later patients had no significant impact on outcomes within the GRAALL-2014 trial (Supplementary Table 6).

Finally, among the 259 SR patients achieving CR, 4-year DFS was estimated at 63.7% (95%CI, 57.9-70.1). Noninferiority was concluded, as the lower limit of the one-sided 95% CI was above 45% (Supplementary Figure 3).

Benefit of alloHSCT

Given that the allocation to allo-HSCT was not randomized, and to assess the accuracy of the new MRD-based eligibility criteria for alloHSCT, we performed a Simon and Makuch

estimation of DFS and post-CR OS according to this time occurrence in the eligible population, consisting of VHR patients (Figure 4). In the GRAALL-2014, there was evidence of improved outcomes following alloHSCT in terms of post-CR OS (HR 0.48; 95%CI, 0.31-0.74; p=0.008) and DFS (HR 0.58; 95%CI, 0.37-0.91; p=0.019). In the prior GRAALL-2005, we had previously shown no benefit of alloHSCT based on historical eligibility criteria.[10]

Discussion

In this study, we report the outcomes of adult Ph-negative ALL patients aged 18-59 years treated with an age-adapted intensive protocol (GRAALL-2014), where the decision to propose alloHSCT was based on early MRD responses only. By applying such MRD-based transplant eligibility, we successfully reduced the rate of patients eventually transplanted by about 50%. Compared to historical trial results (GRAALL-2005), a reduction of treatment dose-intensity for patients 45 years and older resulted into a significant reduction in early death rates, an improved CR rate, and a substantial reduction in NRM. Despite an increased risk of relapse in these older adults, this overall deescalating strategy did not significantly impact DFS, whatever the age subgroup.

Before the era of immunotherapy, major improvement of the therapy for adult ALL has resulted from the intensification of chemotherapy backbones and the wide implementation of MRD monitoring for risk stratification.[16] For many adult cooperative groups, the use of more intensive induction schedules have led to subsequent adjustments prompted by the excess of toxicities, mostly driven by asparaginase specific safety profile. In the GMALL/07/2003 trial, PEG-asparaginase dosing was age-adapted with reduced doses given in patients 55 years and older.[17] In the UKALL14 protocol, two infusions of pegylated asparaginase were shown to be too toxic for patients older than 40 years. The protocol was

amended to omit D4 infusion in patients over 40 and to decrease the total amount of anthracycline.[18] In the present study, de-escalation of asparaginase but also anthracycline and steroid dosing during induction in patients aged 45 years or more led to a more than 3-fold reduction in early death and an overall CR improvement. The complete remission (CR) rate after the first induction course remained unchanged; however, a higher proportion of patients with resistance to the initial induction were able to receive salvage therapy and achieve CR without toxic deaths.

Eligibility criteria for alloHSCT in adults remain controversial, while most pediatric groups rely on MRD-based stratification to identify the small subset of patients for whom alloHSCT is offered as first remission consolidation. In the context of pediatric-inspired regimens, the question of who benefits the most from transplant has been extensively debated. Many cooperative groups or institutions use both MRD- and baseline biological features to select eligible patients, including high WBC, specific immunophenotypic features (pro-B, early T, mature T), and certain oncogenetic characteristics (*KMT2A*-r, Ph-positive, Ph-like, etc.).[7,16] In the GRAALL-2005 trial, we have used a broad definition of criteria based on disease characteristics as well as early response to treatment (prednisone response, BM blast clearance) to indicate alloHSCT. Among these criteria, patients with slow BM blast clearance or lack of CR after a single induction course appeared to specifically benefit from transplant. The study also indicated that MRD levels after induction and first consolidation were the best predictors of alloHSCT benefit.[10] In the present GRAALL-2014 study, we demonstrate using a time-dependent analysis that poor MRD responders benefited from alloHSCT in first CR, while this MRD-based strategy allowed the overall transplant rate to be reduced by half. Whether patients with high-risk genetics ALL who achieve a favorable MRD response to induction and consolidation would also benefit from alloHSCT remains to be clarified.

Interestingly, the GMALL recently reported on the randomization of alloHSCT in high-risk patients (pro-B, *KMT2A*-r, early-/mature T, WBC > 30 G/L in B-ALL, or failure to achieve CR after induction I) who reached molecular CR following induction. In the overall population, no difference in DFS or OS was observed. Curiously, the non-transplant arm showed a significant advantage in HR T-ALL, while the opposite was seen in HR B-ALL though this did not reach statistical significance for OS.[19]

Despite similar DFS in both trials, OS significantly improved across all age subgroups in the GRAALL-2014, suggesting that advancements in second-line strategies contributed to this outcome. This improvement may be attributed to enhanced salvage therapies and a higher likelihood of transplantation in second complete remission for patients who were not initially transplanted. Notably, the selective improvement in post-relapse survival among B-ALL patients strongly indicates a pivotal role for newly approved immunotherapies.[20–23]

The use of blinatumomab as consolidation therapy has dramatically improved the prognosis of frontline Ph-negative B-ALL, benefiting both patients with unfavorable and favorable MRD responses to chemotherapy.[24–26] While blinatumomab decreases the risk of relapse, its favorable safety profile also creates an opportunity to reduce chemotherapy intensity and further improve NRM. With the broad use of blinatumomab as a frontline consolidation therapy, the indications for alloHSCT will be further challenged. One key question is whether high-risk patients, defined genetically or by poor response to induction chemotherapy, who achieve complete MRD response after blinatumomab still benefit from transplant. The GRAALL-2024 trial is currently investigating this question by randomizing alloHSCT in this specific patient population.

Contribution: V.L. provided administrative support; N.B., F.H., T.L., M.H., C.G., Y.C., Y.H., P.C., M.B., M.E.-B., F.P., J.-P.M., A.T., A.H., N.D., E.L., C.B., S.M., G.G., A.B., U.S., T.C., P.T., P.R., B.D.P., N.I., and H.D. provided study materials or patients; N.B., V.L., E.C., V.A., and H.D. performed collection and assembly of data; E.D., C.P., N.G., M.-C.B., M.L-P., V.A., E.C. R.K., E.C., and W.C. performed central review of molecular and cytogenetic data; S.C., N.B., and H.D. performed data analysis and interpretation; N.B., S.C., V.L., and H.D. wrote the manuscript; and all authors performed conception and design and provided final approval of the manuscript.

COI disclosure: NB received honoraria from Amgen, Novartis, Pfizer, Servier, Gilead. HD received honoraria from Amgen and Servier. FH received honoraria from Amgen, Novartis, Pfizer, Servier, Gilead. AH: received honoraria from Novartis, Sanofi, Immedica, Pierre Fabre, and travel support from Medac, Sanofi, Novartis and Pierre Fabre. PC received honoraria from Takeda, Servier, Abbvie, Priothera, and travel support from Abbvie. TL received honoraria from Servier, Amgen

The study was funded by a grant from Programme Hospitalier de Recherche Clinique Cancer 2012 – PHRC Cancer 2012 (Ministère de la Santé)

References

1. 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 Off J Am Soc Clin Oncol*. 2009;27(6):911-918. doi:10.1200/JCO.2008.18.6916
2. 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 Off J Am Soc Clin Oncol*. 2018;36(24):2514-2523. doi:10.1200/JCO.2017.76.8192
3. Boissel N. New developments in ALL in AYA. *Hematol Am Soc Hematol Educ Program*. 2022;2022(1):190-196. doi:10.1182/hematology.2022000336
4. Toft N, Birgens H, Abrahamsson J, et al. Results of NOPHO ALL2008 treatment for patients aged 1-45 years with acute lymphoblastic leukemia. *Leukemia*. 2018;32(3):606-615. doi:10.1038/leu.2017.265
5. 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. doi:10.1182/blood-2018-10-881961
6. Huguet F, Leguay T, Thomas X, et al. The Upper Age Limit for a Pediatric-Inspired Therapy in Younger Adults with Ph-Negative Acute Lymphoblastic Leukemia (ALL)? Analysis of the Graall-2005 Study. *Blood*. 2016;128(22):762-762.
7. Giebel S, Marks DI, Boissel N, et al. Hematopoietic stem cell transplantation for adults with Philadelphia chromosome-negative acute lymphoblastic leukemia in first remission: a position statement of the European Working Group for Adult Acute Lymphoblastic Leukemia (EWALL) and the Acute Leukemia Working Party of the European Society for Blood and Marrow Transplantation (EBMT). *Bone Marrow Transplant*. 2019;54(6):798-809. doi:10.1038/s41409-018-0373-4

8. Arslan S, Pullarkat V, Aldoss I. Indications for Allogeneic HCT in Adults with Acute Lymphoblastic Leukemia in First Complete Remission. *Curr Treat Options Oncol*. 2021;22(7):63. doi:10.1007/s11864-021-00860-1
9. Goldstone AH, Richards SM, Lazarus HM, et al. In adults with standard-risk acute lymphoblastic leukemia, the greatest benefit is achieved from a matched sibling allogeneic transplantation in first complete remission, and an autologous transplantation is less effective than conventional consolidation/maintenance chemotherapy in all patients: final results of the International ALL Trial (MRC UKALL XII/ECOG E2993). *Blood*. 2008;111(4):1827-1833. doi:10.1182/blood-2007-10-116582
10. Dhedin N, Huynh A, Maury S, et al. Allogeneic Hematopoietic Stem Cell Transplantation (HSCT) In Adults With Philadelphia Chromosome (Ph)-Negative Acute Lymphoblastic Leukemia (ALL): Results From The Group For Research On Adult ALL (GRAALL). *Blood*. 2013;122(21):552-552.
11. Chalandon Y, Rousselot P, Chevret S, et al. Nilotinib with or without cytarabine for Philadelphia positive acute lymphoblastic leukemia. *Blood J*. Published online 7 March 2024:blood.2023023502. doi:10.1182/blood.2023023502
12. Lafage-Pochitaloff M, Baranger L, Hunault M, et al. Value of Cytogenetic Abnormalities in Adult Patients with Philadelphia Chromosome (Ph)-Negative Acute Lymphoblastic Leukemia (ALL) Treated in the Pediatric-Inspired Trials from the Group for Research on Adult ALL (GRAALL). *Blood*. 2014;124(21):492-492.
13. Schoenfeld D. Partial residuals for the proportional hazards regression model. *Biometrika*. 1982;69(1):239-241. doi:10.1093/biomet/69.1.239
14. Grambsch PM, Therneau TM. Proportional hazards tests and diagnostics based on weighted residuals. *Biometrika*. 1994;81(3):515-526. doi:10.1093/biomet/81.3.515
15. Simon R, Makuch RW. A non-parametric graphical representation of the relationship between survival and the occurrence of an event: application to responder versus non-responder bias. *Stat Med*. 1984;3(1):35-44. doi:10.1002/sim.4780030106
16. Gökbuget N, Boissel N, Chiaretti S, et al. Management of ALL in adults: 2024 ELN recommendations from a European expert panel. *Blood*. 2024;143(19):1903-1930. doi:10.1182/blood.2023023568
17. Lanvers-Kaminsky C, Niemann A, Eveslage M, et al. Asparaginase activities during intensified treatment with pegylated E. coli asparaginase in adults with newly-diagnosed acute lymphoblastic leukemia. *Leuk Lymphoma*. 2020;61(1):138-145. doi:10.1080/10428194.2019.1658099
18. Patel B, Kirkwood AA, Dey A, et al. Pegylated-asparaginase during induction therapy for adult acute lymphoblastic leukaemia: toxicity data from the UKALL14 trial. *Leukemia*. 2017;31(1):58-64. doi:10.1038/leu.2016.219
19. Goekbuget N, Schneller F, Nachtkamp K Sr, et al. Chemotherapy or Stem Cell Transplantation in Adult High Risk Ph/BCR::ABL1-Negative ALL Patients with Early MRD Negativity: Results of the Randomized GMALL Trial 08/2013. *Blood*. 2024;144(Supplement 1):961. doi:10.1182/blood-2024-199060
20. Kantarjian HM, DeAngelo DJ, Stelljes M, et al. Inotuzumab Ozogamicin versus Standard Therapy for Acute Lymphoblastic Leukemia. *N Engl J Med*. 2016;375(8):740-753. doi:10.1056/NEJMoa1509277
21. Kantarjian H, Stein A, Gökbuget N, et al. Blinatumomab versus Chemotherapy for Advanced Acute Lymphoblastic Leukemia. *N Engl J Med*. 2017;376(9):836-847. doi:10.1056/NEJMoa1609783

22. Maude SL, Laetsch TW, Buechner J, et al. Tisagenlecleucel in Children and Young Adults with B-Cell Lymphoblastic Leukemia. *N Engl J Med*. 2018;378(5):439-448. doi:10.1056/NEJMoa1709866
23. Shah BD, Ghobadi A, Oluwole OO, et al. KTE-X19 for relapsed or refractory adult B-cell acute lymphoblastic leukaemia: phase 2 results of the single-arm, open-label, multicentre ZUMA-3 study. *Lancet Lond Engl*. 2021;398(10299):491-502. doi:10.1016/S0140-6736(21)01222-8
24. Gökbuget N, Dombret H, Bonifacio M, et al. Blinatumomab for minimal residual disease in adults with B-cell precursor acute lymphoblastic leukemia. *Blood*. 2018;131(14):1522-1531. doi:10.1182/blood-2017-08-798322
25. Litzow MR, Sun Z, Mattison RJ, et al. Blinatumomab for MRD-Negative Acute Lymphoblastic Leukemia in Adults. *N Engl J Med*. 2024;391(4):320-333. doi:10.1056/NEJMoa2312948
26. Bassan R, Chiaretti S, Della Starza I, et al. Up-front Blinatumomab Improves MRD Clearance and Outcome in Adult Ph- B-lineage ALL: the GIMEMA LAL2317 Phase 2 Study. *Blood*. Published online 26 February 2025:blood.2024027500. doi:10.1182/blood.2024027500

Figure legends

Figure 1. Flow chart and schematic representation of the GRAALL-2014 sub-studies
(A) Flow chart showing the definition of the modified intention-to-treat (ITT) analysis population according to lineage (B-ALL *versus* T-ALL)
(B) Flow chart showing activation dates and number of patients included in each substudy within the modified ITT analysis population
(B) Schematic overview of the blinatumomab substudy (GRAALL-2014/B-QUEST)
(C) Schematic overview of the nelarabine substudy (GRAALL-2014/T-ATRIALL)

Figure 2. Outcome of patients enrolled in the GRAALL-2014 trial

Figure 3. Comparison of outcomes between the GRAALL-2005 and GRAALL-2014 trials by age subgroup (<45y, ≥45y). (A,E) overall survival, (B,F) disease-free survival, (C,G) cumulative incidence of relapse, (D,H) cumulative incidence of non-relapse mortality

Figure 4. Simon and Makuch plot for transplant analysis. (A) disease-free survival, (B) post-complete remission survival. Note that the Simon and Makuch method evaluates the group of individuals who remain at risk at each event time, that is, whether or not they received alloHSCT at each time. More specifically, it allows the calculation of event-free probabilities while allowing individuals to move from one group (no allograft) to the other group (allograft) over time. The calculations of probabilities are then the same as the Kaplan-Meier method, in that the conditional survival estimates are a function of the ratio of the number of events to the number at risk at each event time. Number of patients at risk under the x-axis are thus computed over time, but averaged at specific time points; for instance, 108 patients were allografted within the first 6 months (thus exposed to die, OS curve), including 1 whose disease relapsed, explaining the 107 exposed patients for DFS.

Table 1. Overview of GRAALL-2005 and 2014 protocols

Treatment Phase	Drug	Age < 45 Years	Age ≥ 45 Years	GRAALL-2005
Prephase[1]				Same as GRAALL-2014
	Prednisone	60 mg/m²/d (PO) D-7 to D-1		
	IT (MTX)	between D-7 and D-4		
Induction[1,2]				Same as GRAALL-2014
	Prednisone	60 mg/m²/d (PO) D1 to D14	60 mg/m²/d (PO) D1 to D7	with no age limit
	Vincristine	2 mg/d (IVD) D1, D8, D15, D22		(<45y schedule for all)
	Daunorubicin	50 mg/m²/d (IV 30 min) D1-D3 30 mg/m²/d (IV 30 min) D15, D16	30 mg/m²/d (IV 30 min) D1-D3 30 mg/m²/d (IV 30 min) D15, D16	
	Cyclophosphamide	750mg/m²/d (IV 3h) D1, D15		
	L-Asparaginase[2]	6,000 IU/m²/d (IV 1h) D8, D10, D12, D20, D22, D24, D26, D28	6,000 IU/m²/d (IV 1h) D8, D10, D12, D20, D22, D24	
	G-CSF	5 µg/kg/d (SC or IV) D18 until ANC >1G/L		
	IT (MTX + Ara-C + Depomedrol)	D1, D8		
Salvage[3]				Same as GRAALL-2014
	Idarubicin	12 mg/m²/d (IV 1h) D1-D3		
	Aracytine	2000 mg/m²/12h (IV 3h) D1-D4		
	G-CSF	5 µg/kg/d (SC or IV) D8 until ANC >1G/L		
Consolidation[1]				Same as GRAALL-2014
Standby block[4]	**VP-16**	150 mg/m²/d (IV 1h) D1		with no age limit
	Cytarabine	30 mg/m²/12h (SC) D1,D2		(<45y schedule for all)
Block S1	**Cytarabine**	2,000 mg/m²/12h (IV 2h) D1, D2		Asparaginase 10,000 IU/m² at D16
	Dexamethasone	10 mg/12h (IV) D1, D2		
	G-CSF	5 µg/kg/d (SC or IV) D8 to D12		
Block S2	**Methotrexate**	5,000 mg/m²/d (CIV 24h) D15	3,000 mg/m²/d (CIV 24h) D15	
	Vincristine	2 mg/d (IVD) D15		
	6-mercaptopurin	60 mg/m²/d (PO) D15 to D21		
	G-CSF	5 µg/kg/d (SC or IV) D22 to D26		
	IT (MTX + Ara-C + Depomedrol)	D16		
Block S3	**Cyclophosphamide**	500 mg/m²/d (IV 3h) D29, D30		
	VP-16	75 mg/m²/d (IV 1h) D29, D30		
	Methotrexate	25 mg/m²/d (IV) D29		
	G-CSF	5 µg/kg/d (SC or IV) D31 until ANC >1G/L		
	IT (MTX + Ara-C + Depomedrol)	D29		
Consolidation 2	**Bock S4, S5 and S6**	same as block S1, S2, and S3 respectively		Same as consolidation 1
Late intensification[2,3,5]				Same as GRAALL-2014
CR after induction	**Prednisone**	60 mg/m²/d (PO) D1 to D14	60 mg/m²/d (PO) D1 to D7	with no age limit
	Vincristine	2 mg/d (IVD) D1, D8, D15, D22		(<45y schedule for all)
	Daunorubicin	30 mg/m²/d (IV 30 min) D1, D2, D3 30 mg/m²/d (IV 30 min) D15, D16	30 mg/m²/d (IV 30 min) D1, D2, D3	
	Cyclophosphamide	750mg/m²/d (IV 3h) D1, D15		
	L-Asparaginase[2]	6,000 IU/m²/d (IV 1h) D8, D10, D12, D20, D22, D24, D26, D28	6,000 IU/m²/d (IV 1h) D8, D10, D12, D20, D22, D24	
	G-CSF	5 µg/kg/d (SC or IV) D18 until ANC >1G/L		
	IT (MTX + Ara-C + Depomedrol)	D1, D8		
CR after salvage[3]	**Idarubicin, Aracytine, G-CSF, IT**	Same as salvage (see above)		
Consolidation 3	**Bock S7, S8 and S9**	same as block S1, S2, and S3 respectively		Same as consolidation 1
CNS irradiation[1,6]	**24 Gy in 12 fractions**	for CNS-positive patients		whatever CNS infiltration[6]
Maintenance Therapy[1] **(2 years)**				Same as GRAALL-2014
	Vincristine	2 mg/d (IVD) D1, Month 1 to 12		with no age limit
	Prednisone	40 mg/m²/d (PO), D1 to D7		(<45y schedule for all)
	Methotrexate	60 mg/m²/d (PO), for 2 years		
	MTX	25 mg/m²/wk (PO), for 2 years		
	IT (MTX + Ara-C + Depomedrol)	D1 of months 1, 3, and 5		

Abbreviations: Ara-C, Cytarabine; CNS, central nervous system; G-CSF, granulocyte colony stimulating factor; IT, intrathecal therapy; MTX, methotrexate

1. For CNS-positive patients, additional triple IT were given : 1 during prephase, 4 during induction (D4, D11, D15, and D18), and 2 during consolidation 1 (D8, D24). No IT were given after CNS irradiation.
2. Substitution with erwinase 25.000 IU/m2 was planned in case of clinical allergy, asparaginase activity < 100 IU/mL at D14 or D26, or detection of anti-asparaginase antibody during consolidation.
3. Salvage regimen was given in case of induction failure. In this case, the same course was given as late intensification.
4. The standby block could optionally be given to patients who could not receive consolidation due to post-induction toxicity. This block could be repeated once (2 blocks in total).
5. Patients who received alloHSCT were transplanted before late intensification
6. 12 Gy were given as cranial boost before alloHSCT. In the GRAALL-2005, 18 Gy was given as CNS prophylaxis.

Table 2. Patient characteristics according to trial.

	GRAALL-2005 N=787	GRAALL-2014 N=743	p
Age (years)			
Median (IQ)	36 (25;48)	35 (24;48)	0.26
<45 years, N (%)	542 (68.9)	504 (67.8)	0.70
≥45 years, N (%)	245 (31.1)	239 (32.2)	
Sex			
male, N (%)	472 (60.0)	463 (62.3)	0.37
female, N (%)	315 (40.0)	280 (37.7)	
Phenotype			
B-ALL, N (%)	525 (66.7)	489 (65.8)	0.75
T-ALL, N (%)	262 (33.3)	254 (34.2)	
CNS involvement			
CNS-3, N (%)	55 (7.0)	59 (8.0)	0.50
WBC (10^9/L)			
Median (IQ)	11.8 (3.9;41.9)	10.6 (3.7;38.9)	0.34
≥30 in B-ALL, N (%)	118/525 (22.5)	105/489 (21.5)	0.70
≥100 in T-ALL, N (%)	52/262 (19.8)	55/254 (21.7)	0.91
Oncogenetics			
B-ALL			
KMT2A-r	64 (12.2)	56 (11.5)	0.77
LH/NT	37 (7.0)	38 (7.8)	0.72
High hyperdiploidy	31 (5.9)	33 (6.7)	0.61
TCF3::*PBX1*	22 (4.2)	21 (4.3)	1.00
ETV6::*RUNX1*	2 (0.4)	9 (1.8)	0.03
iAMP21	4 (0.8)	6 (1.2)	0.53
B-other	286 (54.5)	321 (65.6)	<0.001
Not available	79 (15.0)	5 (1.0)	<0.001
*IKZF1*del	62/259 (23.9)	122/484 (25.2)	0.72
T-ALL			
4-gene classifier			
favorable, N (%)	99/172 (57.7)	136/249 (54.6)	0.62
unfavorable, N (%)	73/172 (42.4)	113/249 (45.4)	0.59

Abbreviations: ALL, acute lymphoblastic leukemia; CNS, central nervous system; WBC, white blood cell count; LH/NT, low hypodiploidy/near-triploidy

Table 3. Early response according to age range and trial

	GRAALL-2005			GRAALL-2014					
	Total N=787	**Age < 45y N=542**	**Age ≥ 45y N=245**	**Total N=743**	**Age < 45y N=504**	**Age ≥ 45y N=239**	**p**	**p (<45y)**	**P (≥45y)**
Early response									
D8 prednisone response, N (%)	602/785 (76.7)	400/542 (73.8)	202/245 (82.4)	565/742 (76.2)	371/503 (73.5)	194/239 (81.2)	0.80	1.00	0.73
CR after induction course, N (%)	705/787 (89.6)	501/542 (92.4)	204/245 (83.3)	663/743 (89.2)	458/504 (90.9)	205/239 (85.8)	0.87	0.37	0.45
Salvage, N (%)	32/787 (4.1)	21 (3.9)	11 (4.5)	49/743 (6.6)	27 (5.4)	22 (9.2)	**0.03**	0.30	**0.047**
CR after salvage, N (%)	18/32 (56.3)	12/21 (57.1)	6/11 (54.5)	30/49 (61.2)	16/27 (59.3)	14/22 (63.6)	0.82	1.00	0.71
Early death, N (%)	44/787 (5.6)	17/542 (3.1)	27/245 (11.0)	20/743 (2.7)	12/504 (2.4)	8/239 (3.3)	**0.005**	0.57	**0.001**
Resistant disease, N(%)	20/787 (2.5)	12/542 (2.2)	8/245 (3.3)	30/743 (4.0)	18/504 (3.6)	12/239 (5.0)	0.11	0.20	0.37
Overall CR, N (%)	723/787 (91.9)	513/542 (94.6)	210/245 (85.7)	693/743 (93.3)	474/504 (94.0)	219/239 (91.6)	0.33	0.69	**0.045**
Standard-risk*, N(%)	ND	ND	ND	259/693 (37.4)	330/474 (69.6)	126/219 (57.5)	NA	NA	NA
Very high-risk, N(%)**	467/677 (69.0)	330/483 (68.3)	137/194 (70.6)	213/693 (30.7)	130/474 (27.4)	83/219 (37.9)	**<0.0001**	**<0.0001**	**<0.0001**
AlloHSCT*, N (%)	278/723 (38.5)	206/513 (40.2%)	72/210 (34.3)	159/693 (22.9)	101/474 (21.3)	58/219 (26.5)	**<0.0001**	**<0.0001**	0.093

Abbreviations: CR, complete remission; ND, not defined; NA, not applicable; y, year; alloHSCT, allogeneic hematopoietic stem cell transplantation

* defined among patients in CR. Standard-risk was defined in GRAALL-2014 only. Criteria: B-ALL without KMT2A-r or IKZF1del, T-ALL with either NOTCH1 or FBXW7 mutation without RAS or PTEN alteration, MRD1 $<10^{-4}$

** defined among patients in CR. For the GRAALL-2005: central nervous system involvement, poor prednisone response, slow bone marrow blast clearance at D8 of induction, CR after salvage course. For BCP-ALL: WBC $\geq 30\times10^9$/L, *KMT2A*-r, low hypodiploidy or near-triploidy, t(1;19)/E2A::PBX1, complex karyotype, and $CD10^-$ immature immunophenotype. For the GRAALL-2014: CR after salvage course, MRD1$\geq10^{-4}$, or MRD2$\geq10^{-4}$

Table 4. Outcomes according to age range and trial

	GRAALL-2005			GRAALL-2014					
	Total **N=787**	Age < 45y **N=542**	Age ≥ 45y **N=245**	Total **N=743**	Age < 45y **N=504**	Age ≥ 45y **N=239**	**p**	**p (<45y)**	**P (≥45y)**
Events									
Relapse, N(%)	216/723 (30.0)	155/513 (30.2)	61/210 (29.0)	253/693 (36.5)	167/474 (35.2)	86/219 (39.3)	-	-	-
Death in CR, N(%)	89/723 (12.3)	44/513 (8.6)	45/210 (21.4)	38/693 (5.5)	20/474 (4.2)	18/219 (8.2)	-	-	-
Death, N(%)	321/787 (40.8)	190/542 (35.1)	131/245 (53.5)	246/743 (33.1)	143/504 (28.4)	103/239 (43.1)	-	-	-
Estimates									
4y-CIR, % (95%CI)	29.6 (26.3-33.0)	29.5 (25.6-33.6)	29.9 (23.7-36.5)	37.6 (33.9-41.3)	36.1 (31.6-40.6)	40.9 (34.1-47.6)	**0.016**	0.17	**0.026**

4y-NRM, % (95%CI)	11.6 (9.3-14.1)	8.2 (6.0-10.8)	20.0 (14.7-25.8)	5.3 (3.7-7.2)	4.5 (2.8-6.8)	6.9 (4.0-10.8)	**<0.0001**	**0.009**	**0.0005**
4y-DFS, % (95%CI)	58.8 (55.2-62.6)	62.2 (58.1-66.6)	50.0 (43.5-57.7)	57.1 (53.4-61.1)	59.4 (55.0-64.2)	52.2 (45.7-59.1)	0.89	0.96	0.73
4y-OS, % (95%CI)	61.0 (57.3-64.3)	65.5 (61.7-69.8)	49.6 (43.5-56.5)	67.8 (64.4-71.4)	71.7 (67.7-76.0)	59.5 (53.5-66.3)	**0.003**	**0.031**	**0.011**

Abbreviations: CR, complete remission; CIR, cumulative incidence of relapse; NRM, non-relapse mortality; DFS, disease-free survival; OS, overall survival; CI, confidence interval

Figure 1

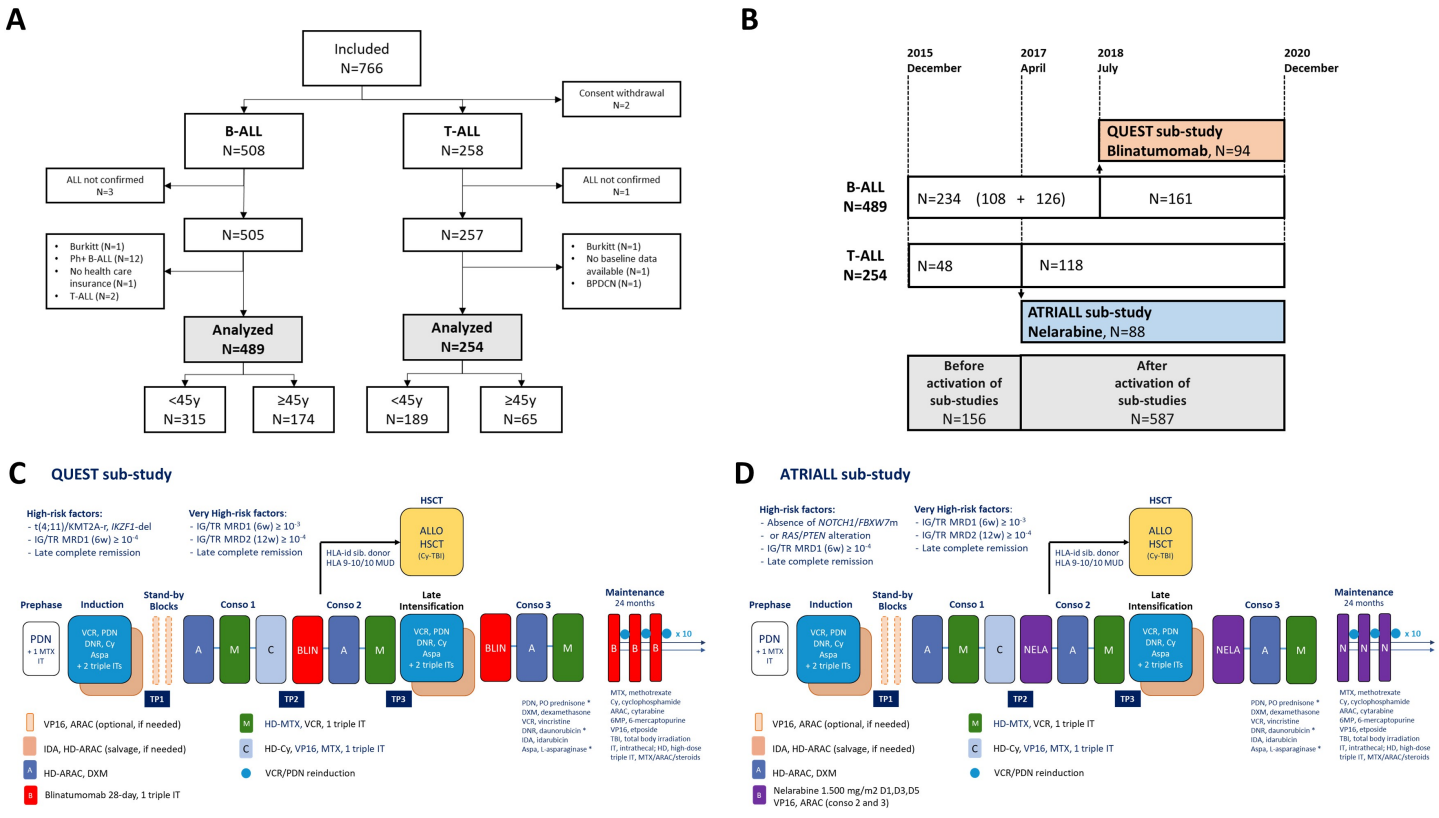

A
Included
N=766
Consent withdrawal
N=2
B-ALL
N=508
T-ALL
N=258
ALL not confirmed
N=3
ALL not confirmed
N=1
N=505
N=257
Burkitt (N=1)
Ph+ B-ALL (N=12)
No health care insurance (N=1)
T-ALL (N=2)
Burkitt (N=1)
No baseline data available (N=1)
BPDCN (N=1)
Analyzed
N=489
Analyzed
N=254
<45y
N=315
≥45y
N=174
<45y
N=189
≥45y
N=65
B
2015 December
2017 April
2018 July
2020 December
QUEST sub-study
Blinatumomab, N=94
B-ALL
N=489
N=234 (108 + 126)
N=161
T-ALL
N=254
N=48
N=118
ATRIALL sub-study
Nelarabine, N=88
Before activation of sub-studies
N=156
After activation of sub-studies
N=587
C
QUEST sub-study
High-risk factors:
- t(4;11)/KMT2A-r, IKZF1-del
- IG/TR MRD1 (6w) ≥ 10-3
- Late complete remission
Very High-risk factors:
- IG/TR MRD1 (6w) ≥ 10-3
- IG/TR MRD2 (12w) ≥ 10-4
- Late complete remission
HSCT
ALLO HSCT (Cy-TBI)
HLA-id sib. donor HLA 9-10/10 MUD
Prephase
Induction
Stand-by Blocks
Conso 1
Conso 2
Late Intensification
Conso 3
Maintenance 24 months
PDN + 1 MTX IT
VCR, PDN DNR, Cy Aspa + 2 triple ITs
A
M
C
BLIN
x 10
TP1
TP2
TP3
PDN, PO prednisone *
DXM, dexamethasone
VCR, vincristine
DNR, daunorubicin *
IDA, idarubicin
Aspa, L-asparaginase *
MTX, methotrexate
Cy, cyclophosphamide
ARAC, cytarabine
6MP, 6-mercaptopurine
VP16, etposide
TBI, total body irradiation
IT, intrathecal; HD, high-dose
triple IT, MTX/ARAC/steroids
VP16, ARAC (optional, if needed)
IDA, HD-ARAC (salvage, if needed)
HD-ARAC, DXM
Blinatumomab 28-day, 1 triple IT
HD-MTX, VCR, 1 triple IT
HD-Cy, VP16, MTX, 1 triple IT
VCR/PDN reinduction
D
ATRIALL sub-study
High-risk factors:
- Absence of NOTCH1/FBXW7m
- or RAS/PTEN alteration
- IG/TR MRD1 (6w) ≥ 10-4
- Late complete remission
Very High-risk factors:
- IG/TR MRD1 (6w) ≥ 10-3
- IG/TR MRD2 (12w) ≥ 10-4
- Late complete remission
HSCT
ALLO HSCT (Cy-TBI)
HLA-id sib. donor HLA 9-10/10 MUD
Prephase
Induction
Stand-by Blocks
Conso 1
Conso 2
Late Intensification
Conso 3
Maintenance 24 months
PDN + 1 MTX IT
VCR, PDN DNR, Cy Aspa + 2 triple ITs
A
M
C
NELA
x 10
TP1
TP2
TP3
PDN, PO prednisone *
DXM, dexamethasone
VCR, vincristine
DNR, daunorubicin *
IDA, idarubicin
Aspa, L-asparaginase *
MTX, methotrexate
Cy, cyclophosphamide
ARAC, cytarabine
6MP, 6-mercaptopurine
VP16, etposide
TBI, total body irradiation
IT, intrathecal; HD, high-dose
triple IT, MTX/ARAC/steroids
VP16, ARAC (optional, if needed)
IDA, HD-ARAC (salvage, if needed)
HD-ARAC, DXM
Nelarabine 1.500 mg/m2 D1,D3,D5
VP16, ARAC (conso 2 and 3)
HD-MTX, VCR, 1 triple IT
HD-Cy, VP16, MTX, 1 triple IT
VCR/PDN reinduction

Figure 2

Figure 2.

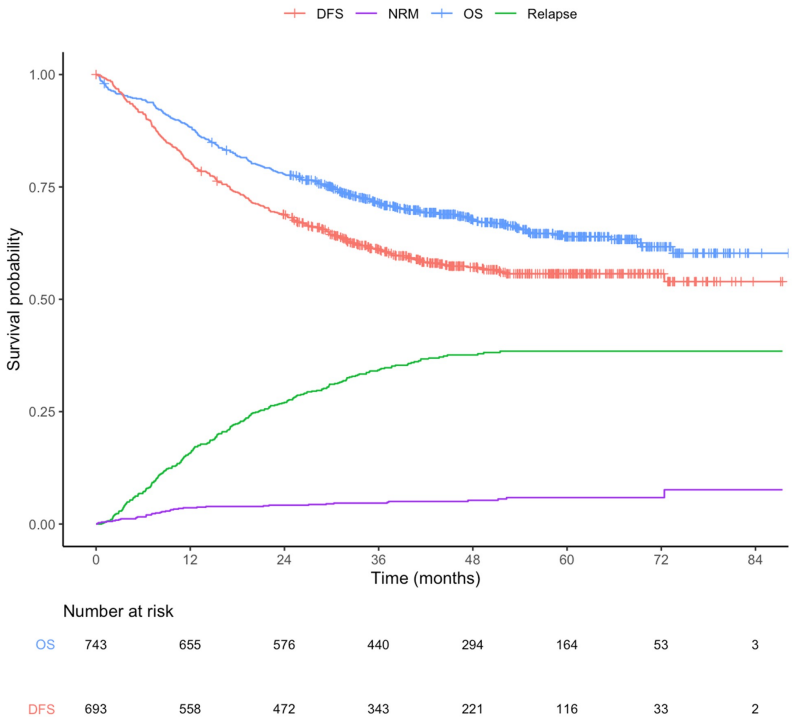


Figure 3.

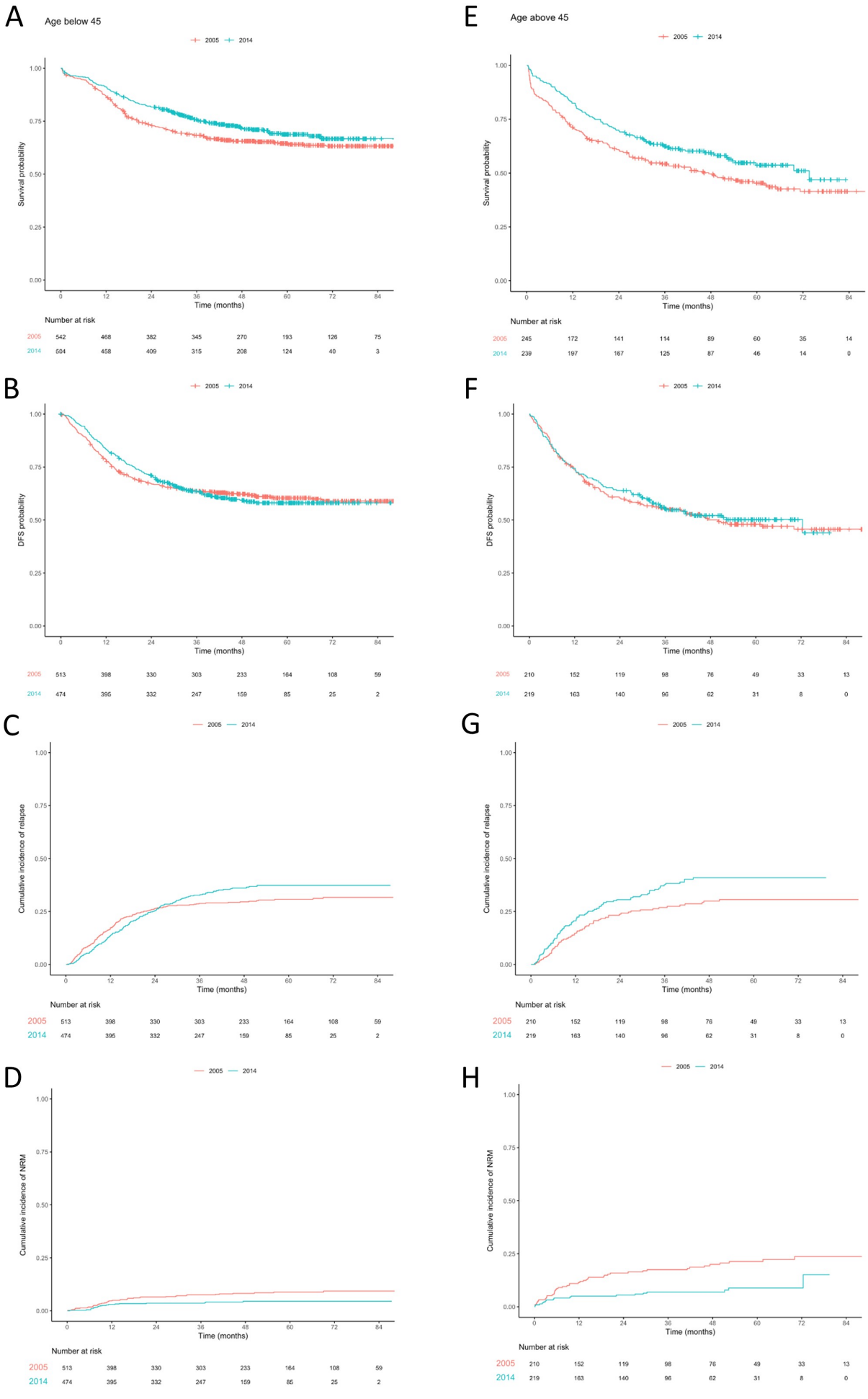
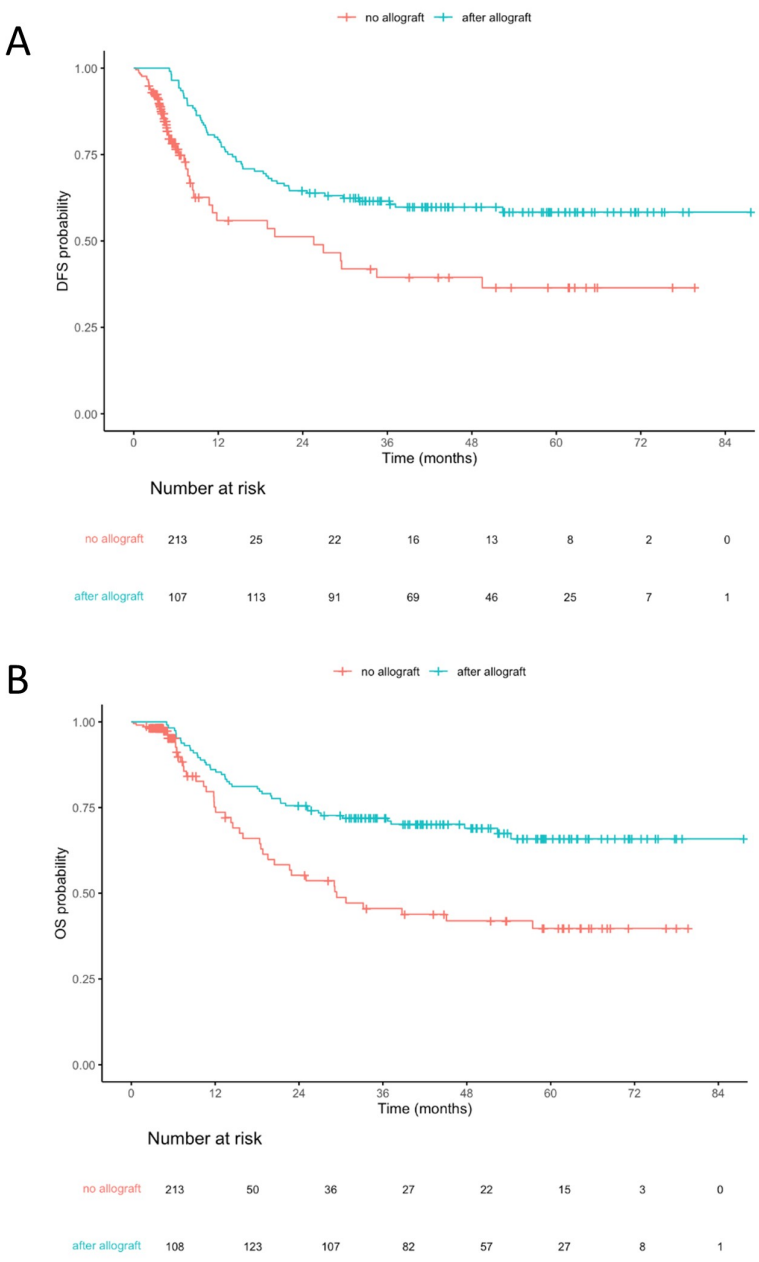

A
Age below 45
2005
2014
Survival probability
Time (months)
Number at risk
2005 542 468 382 345 270 193 126 75
2014 504 458 409 315 208 124 40 3
B
DFS probability
2005 513 398 330 303 233 164 108 59
2014 474 395 332 247 159 85 25 2
C
Cumulative incidence of relapse
D
Cumulative incidence of NRM
E
Age above 45
2005 245 172 141 114 89 60 35 14
2014 239 197 167 125 87 46 14 0
F
2005 210 152 119 98 76 49 33 13
2014 219 163 140 96 62 31 8 0
G
H

Figure 4.


A
no allograft
after allograft
DFS probability
1.00
0.75
0.50
0.25
0.00
0
12
24
36
48
60
72
84
Time (months)
Number at risk
no allograft 213 25 22 16 13 8 2 0
after allograft 107 113 91 69 46 25 7 1
B
no allograft
after allograft
OS probability
1.00
0.75
0.50
0.25
0.00
0
12
24
36
48
60
72
84
Time (months)
Number at risk
no allograft 213 50 36 27 22 15 3 0
after allograft 108 123 107 82 57 27 8 1

Age-adapted Chemotherapy and MRD-oriented AlloHSCT for Ph- ALL GRAALL-2014 trial

Rationale

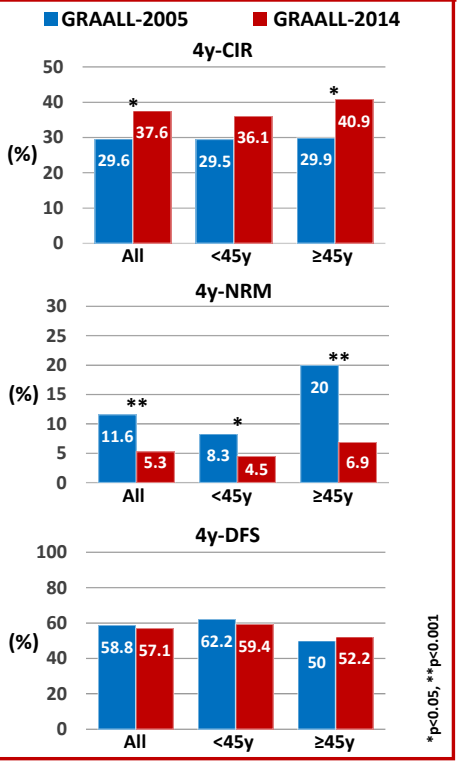
GRAALL-2005
Lessons

- Pediatric-inspired regimen improves the outcome of adults 18-59y with Ph- ALL
- NRM increases with age
- Allo-HSCT benefits to poor early responders

GRAALL-2014

- Age-adapted chemotherapy (reduced steroids, asparaginase, anthracycline for ≥ 45y)
- Reduced indication for alloHSCT, based on MRD only
- Objective = non-inferior DFS

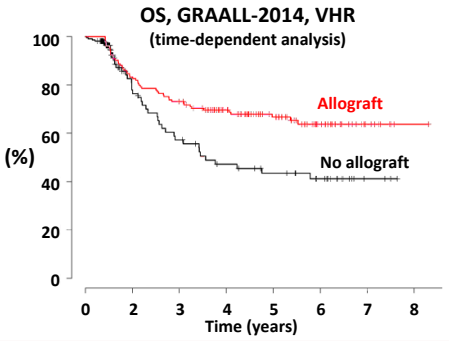
Outcomes



Allo-HSCT

Eligibility criteria

	GRAALL-2005	GRAALL-2014
Baseline	High WBC CNS-3 Pro-B ALL *TCF3::PBX1*, *KMT2A*-r Complex karyotype (≥5 abn.)	
Early response	Poor PB steroid response Poor D8 BM response Late CR	Late CR MRD1 ≥ 10^{-3} MRD2 ≥ 10^{-4}



- In patients over 45 years old, age-adapted protocol improved complete remission rate and decreased non-relapse mortality
- MRD-based transplant decisions reduced transplant indications and ensured the procedure was limited to patients most likely to benefit