Treatment Intensification With Either Fludarabine, AraC, G-CSF and Idarubicin, or Cladribine Plus Daunorubicin and AraC on the Basis of Residual Disease Status in Older Patients With AML: Results From the NCRI AML18 Trial

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ABSTRACT

PURPOSE To evaluate the survival benefit of chemotherapy intensification in older patients with AML who have not achieved a measurable residual disease (MRD)–negative remission.

METHODS Five hundred twenty-three patients with AML (median age, 67 years; range, 51-79) without a flow cytometric MRD-negative remission response after a first course of daunorubicin and AraC (DA; including 165 not in remission) were randomly assigned between up to two further courses of DA or intensified chemotherapy—either fludarabine, cytarabine, granulocyte colony-stimulating factor and idarubicin (FLAG-Ida) or DA with cladribine (DAC).

RESULTS Overall survival (OS) was not improved in the intensification arms (DAC *v* DA: hazard ratio [HR], 0.74 [95% CI, 0.55 to 1.01]; $P = .054$; FLAG-Ida *v* DA: HR, 0.86 [95% CI, 0.66 to 1.12]; $P = .270$); OS at 3 years was 34%, 46%, and 42% for DA, DAC, and FLAG-Ida, respectively. Early deaths and other adverse events were more frequent with FLAG-Ida (9% day 60 deaths *v* 4% after DA or DAC; $P = .032$). Of patients entering random assignment, 131 had MRD unknown status. In this subgroup of patients lacking evidence of residual leukemia by flow cytometry, there was no detectable survival advantage from intensification. A planned sensitivity analysis excluding these patients demonstrated a survival benefit for both DAC (HR, 0.66 [95% CI, 0.46 to 0.93]; $P = .018$) and FLAG-Ida (HR, 0.72 [95% CI, 0.53 to 0.98]; $P = .035$); OS at 3 years was 30%, 46%, and 46% for DA, DAC, and FLAG-Ida, respectively. There was a concordant reduction in relapse (DAC *v* DA: HR, 0.66 [95% CI, 0.45 to 0.98]; $P = .039$; FLAG-Ida *v* DA: HR, 0.70 [95% CI, 0.49 to 0.99]; $P = .042$). DAC benefit was maintained when survival was censored for transplant ($P = .042$).

CONCLUSION In this study of older patients with AML considered fit and with evidence of residual disease after first induction, chemotherapy intensification improved survival. DAC intensification was better tolerated than FLAG-Ida.

ACCOMPANYING CONTENT

Data Sharing Statement
Data Supplement
Protocol

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INTRODUCTION

In AML, either failure to achieve remission or measurable residual disease (MRD) positivity after induction chemotherapy identifies a poor risk group of patients. In the absence of randomized comparisons, the impact of MRD-directed chemotherapy intensification on outcomes of this group remains uncertain. In the NCRI AML16 trial for older patients (>60 years), those who were in remission but MRD+ by flow cytometry in their remission bone marrow (BM) after first induction had a significantly impaired survival (26% at 3 years compared with 42% if MRD−) because of a high risk of relapse.[1] These results suggested that in older adults, flow cytometric detection of MRD after course 1 acts as a surrogate marker for drug resistance, predicting an extremely poor outcome with

CONTEXT

Key Objective

Do older fit patients with AML benefit from chemotherapy intensification if they have not attained remission with measurable residual disease (MRD) negativity after the first induction?

Knowledge Generated

Older patients with AML, considered fit after first induction, had a survival benefit from an intensified second course compared with continuing with daunorubicin/AraC if they had residual disease detected by flow cytometry after their first cycle, including those not in remission. Although MRD-guided intensification by either cladribine with daunorubicin/AraC or fludarabine, AraC, granulocyte colony-stimulating factor, and idarubicin improved survival and reduced relapse risk, the former regimen was better tolerated.

Relevance *(S. Lentzsch)*

MRD assessment post induction is critical to optimize outcomes for older patients with AML. Older patients with MRD should be considered for treatment intensification with daunorubicin, cytarabine, and cladribine due to better outcome associated with better tolerance.*

*Relevance section written by *JCO* Associate Editor Suzanne Lentzsch, MD, PhD.

standard therapy and could be used to evaluate the benefit of targeted further treatment intensification compared with continuing standard chemotherapy.

In this context, fludarabine, AraC, G-CSF, and idarubicin (FLAG-Ida) has previously been reported to be effective salvage therapy for younger adults failing daunorubicin and AraC (DA) induction,[2] and is frequently applied in the setting of refractory disease.[3] Furthermore, in the MRC AML15 trial, which included 556 patients older than 60 years, FLAG-Ida induction offered the best antileukemic therapy when compared with AraC, daunorubicin, and etoposide (ADE) chemotherapy in all age groups including the over 60s.[4] The nucleoside analog cladribine appears to enhance the efficacy of agents commonly used in the treatment of AML, including anthracyclines and Ara-C. Furthermore, randomized studies from the Polish AML Group combining DA with cladribine (DAC) reported improved response rates and overall survival (OS) in younger patients compared with DA alone.[5,6] A subsequent study in older patients suggested a survival benefit in patients age 60-65 years without increased toxicity.[7] We therefore conducted a randomized study to evaluate the benefit of treatment intensification with these regimens in older adults who were not in an MRD− remission.

In NCRI AML18, patients who did not meet the criteria of an MRD− remission after their first course of intensive induction were eligible to be randomly assigned between continuing treatment with up to two further courses of standard treatment with DA as given in the AML16 trial[1] or receiving intensified chemotherapy with up to two courses of FLAG-Ida or DAC. Based upon results from our previous AML16 trial,[8] patients could be considered for allogeneic transplant in first remission if there was a suitably matched donor.

METHODS

Patients and Trial Treatments

The AML18 protocol (ISRCTN-31682779, EudraCT-2013-002730-21) was designed for patients age 60 years and older who were fit for intensive chemotherapy and did not have blast transformation of chronic myeloid leukemia or acute promyelocytic leukemia. Patients with high-risk myelodysplastic syndrome, which was defined as >10% marrow blasts at diagnosis, were eligible. The protocol permitted patients younger than 60 years (n = 23), who were not considered suitable for the concurrent AML19 trial for younger patients (which included high-dose Ara-C), to enter after discussion with a trial coordinator. Clinical secondary AML was defined as resulting from either antecedent hematologic disorder or previous chemotherapy for a nonhematologic malignancy.

Between November 2014 and January 2023, 1,631 patients entering AML18 were assigned course 1 comprising daunorubicin (60 mg/m^2 once daily day 1 [d1], 3, 5) and AraC (100 mg/m^2 twice a day d1-10) with zero, one, or two doses of gemtuzumab ozogamicin (GO) once daily.[9] After course 1, remission assessment taken at count recovery included flow cytometric measurable disease. Patients who failed to achieve a complete remission (CR) or CR with incomplete count recovery (CRi) or who were MRD+ or for whom MRD results were not available (because of no diagnostic

leukemia-associated immunophenotype or inadequate/missing samples) were eligible to be randomly assigned between DA, DAC, or FLAG-Ida for up to two further courses of therapy. The daily AraC dose in FLAG-Ida was limited to 1 g/m² and FLAG-Ida was further dose-reduced for patients older than 70 years and in course 3 for all patients (fludarabine from 30 mg/m² once daily days 1-5 to 25 mg/m² days 1-4, idarubicin from 8 mg/m² once daily days 3-5 to 5 mg/m² days 2-4) or patients age 60-70 years. In the DAC regimen, cladribine was given once daily on days 1-5 inclusive by subcutaneous injection (capped at a maximum of 10 mg per dose) with DA3+8 for course 2 and DA2+5 for course 3. The DAC random assignment was closed in May 2019 because of drug logistical issues. Full treatment schedules and trial schema are shown in the Data Supplement (Fig S1, online only). Patients could also enter a postcourse 1 random assignment to receive quizartinib or not, and the results of this random assignment have been reported elsewhere.[10]

Patients were enrolled from 81 centers in the United Kingdom and six in Denmark. The study was approved by the ethics committees (All Wales Research Ethics Committee, approved by Danish national and regional ethics bodies for sites in Denmark) and conducted in accordance with Good Clinical Practice guidelines and the Declaration of Helsinki. All patients provided written informed consent for trial entry and the random assignment.

Laboratory Studies

Cytogenetic analyses performed locally were reviewed and coded centrally according to the criteria by Grimwade et al.[11] Mutation analysis of *FLT3* and *NPM1* was performed in a single reference laboratory. Banked diagnostic DNA was analyzed for variants in 95 recurrently mutated myeloid genes (Data Supplement, Methods). AML with secondary-type mutations (myelodysplasia-related mutations) was defined by the presence of one or more mutations in *ASXL1*, *BCOR*, *EZH2*, *RUNX1*, *SF3B1*, *SRSF2*, *STAG2*, *U2AF1*, or *ZRSR2*.[3,12,13]

MRD was assessed by using flow cytometry in a single reference laboratory as previously described.[1,9,14,15] Details of sample logistics, processing, and analysis strategy are provided in the Data Supplement (Methods). Results were entered into the trial database within 24-48 hours of sample receipt, blinded to investigator-reported remission status to allow independent refinement of clinical remission assessments.[16,17] Postcourse 1 results were then issued immediately to investigators by the trials unit. Flow MRD testing combined detection of diagnostic leukemic aberrant immunophenotypes (LAIP) and different from normal aberrant immunophenotypes (DfN) as per consensus recommendations[18] with any measurable level of MRD considered positive (above sensitivity threshold of 0.02%-0.05%). An MRD− result required negativity in an adequate BM by both DfN and LAIP analysis (prerequisite of LAIP target(s) identified at baseline). Patients were categorized as MRD unassessable after course 1 in the absence of a baseline LAIP to confirm MRD negativity or if no adequate BM was received before course 2 assignment.

Statistical Considerations and End Points

The analyses are by intention to treat; the primary end point was OS for the intensification versus no intensification random assignments. Trial statistical design with further information is provided in the Data Supplement. Median follow-up for censored patients in each comparison is DA versus DAC, 5 versus 4.9 years, and DA versus FLAG-Ida, 4.1 versus 4.0 years.

End points were defined according to the revised International Working Group criteria.[19] CR and CRi were assessed up to 50 days after the random assignments. Survival outcomes were compared using log-rank tests and Cox regression. To account for nonproportional hazards effect observed by graphical analysis, supportive analyses of restricted mean survival time were performed (further information in the Data Supplement). For the exploratory analyses of key subgroups with forest plots, hazard ratios (HRs) were calculated by Cox proportional hazards models, with test for trend for heterogeneity across the subgroups wherever applicable. Competing-risk analysis was performed for relapse with adjustment for nonrelapse mortality using the Gray's test and Fine and Gray model. Relapse was also compared by the cause-specific Cox model. For comparison of transplant versus no transplant, to counteract the immortal time bias introduced by patients needing to have survived long enough to receive a transplant, Mantel-Byar methodology was used.

RESULTS

Patients

From November 2014 to January 2023, 1,015 patients assessed after induction course 1, comprising DA with zero, one, or two doses of GO, did not have an MRD− remission as either not in CR/CRi (excluding early deaths) or in CR/CRi but MRD+ or MRD unknown. Of these, 523 patients were randomly assigned between either continuing with DA or intensification with FLAG-Ida or DAC for up to two courses (courses 2 and 3; Data Supplement, Fig S1). The nonrandomized group were older, and had a worse performance status, with a higher frequency not in CR/CRi (Fig 1).

One hundred ninety-three randomly assigned patients were assigned DA, 191 FLAG-Ida, and 139 to DAC with median follow-up of 4.1, 3.9, and 4.9 years, respectively. Baseline characteristics along with induction chemotherapy given for course 1 were generally balanced between these treatment arms (Table 1). The median age of the overall population was 67 years (range, 51-79). 19.9% patients had adverse risk cytogenetics, and 9% and 46% with mutation results were *TP53*-mutated or had myelodysplasia-related gene

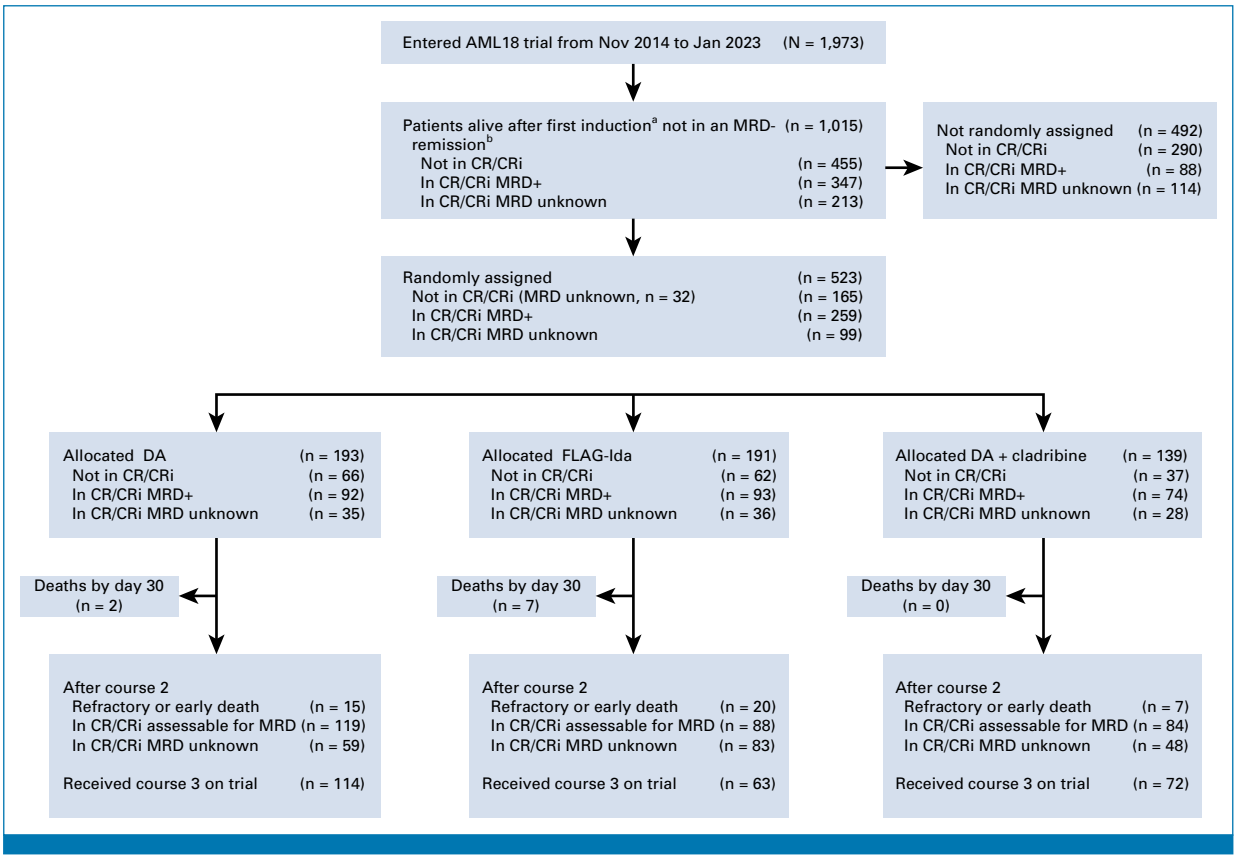

FIG 1. CONSORT diagram. Patients not randomly assigned had a higher frequency in >70 years age group (37% of 492 compared with 31% of 523 randomly assigned patients) and ECOG performance status (PS ≥2, 8.5% compared with 6% of randomly assigned patients, PS ≥1, 56% compared wth 49% of randomly assigned patients) at trial entry. [a]Alive at day 30. [b]By flow cytometric MRD. CR, complete remission; CRi, CR with incomplete count recovery; DA, daunorubicin, Ara-C; ECOG, Eastern Cooperative Oncology Group; FLAG-Ida, fludarabine, AraC, G-CSF, and idarubicin; MRD, measurable residual disease; PS, performance status.

mutations, respectively. Flow MRD was assessed independently of remission status. Disease status before course 2 in the 523 patients was 31.5% not in CR/CRi (including 35 with MRD unknown results), 49.5% CR/CRi MRD+, and 18.9% CR/CRi MRD unknown (Table 1; Data Supplement, Fig S2). Distribution of disease status by random assignment was generally balanced. Baseline MRD levels were higher within FLAG-Ida and DAC treatment arms compared with DA but this difference was not significant (Data Supplement, Fig S3A).

Toxicity and Resource Usage

As expected, there was greater hematologic toxicity with both DAC and FLAG-Ida compared with DA and greater requirement for supportive care and hospitalization (Data Supplement, Table S1). After course 2, the time to neutrophil recovery to 1×10^9/L was 25, 29, and 30 days for DA, DAC, and FLAG-Ida, respectively ($P < .001$). The time to platelet recovery to 100×10^9/L was 26, 33, and 34 days, respectively ($P < .001$). Patients receiving FLAG-Ida experienced more GI (nausea/vomiting and diarrhea) and liver toxicity (grade 2 and above); cardiac function adverse events grade 3 and above were more frequent with DAC (Data Supplement, Fig S4A). Day 60 mortality was increased in patients randomly assigned to FLAG-Ida (9%) compared with DA (4%; $P = .03$) but was not increased with DAC (4%). The distribution of causes of deaths were comparable between the two intensification treatment arms, although there were slightly more deaths from infection with FLAG-Ida (Data Supplement, Fig S4B). Early deaths rates in patients age 70 years and older were 3% DA, 0% DAC, and 7% FLAG-Ida.

A higher proportion of patients in the DA and DAC arms received course 3 compared with FLAG-Ida (DA, 59%; DAC 52%, FLAG-Ida 33%; Data Supplement, Fig S1).

Response and Outcome

After course 2, overall response rate within 50 days for patients not in remission after course 1 was 55%, 57%, and 34% for DA, DAC, and FLAG-Ida, respectively (DA *v* DAC: $P = .536$, DA *v* FLAG: $P = .02$; Table 2). Conversion to CR from CRi or refractory disease was attained by 30% (26/86), 35% (17/48), and 14% (11/77) for DA, DAC, and FLAG-Ida, respectively (Table 2).

Of all 282 patients providing MRD results after both course 1 and 2, 51% (60/117), 63% (50/79), and 58% (50/86)

TABLE 1. Patient Demographics and Clinical Characteristics

Characteristic	Overall (N = 523)	DA (n = 193)	FLAG-Ida (n = 191)	DA-Cladribine (n = 139)
Age, years, median (range)	67 (51-79)	67 (56-79)	67 (53-79)	67 (51-79)
≥65, No. (%)	370 (71)	135 (70)	136 (71)	99 (71)
≥70, No. (%)	160 (31)	59 (31)	61 (32)	40 (29)
Male, No. (%)	316 (60)	118 (61)	113 (59)	85 (61)
WBC × 10^9/L, median (range)	5.2 (0.5-394)	6.6 (0.6-242)	3.7 (0.5-366)	6.3 (0.5-394)
<10, No. (%)	328 (63)	119 (62)	129 (68)	80 (58)
≥50, No. (%)	53 (10)	23 (12)	20 (10)	10 (7)
Diagnosis, No. (%)				
Clinical de novo AML	413 (79)	152 (79)	152 (80)	109 (78)
Clinical secondary AML	59 (11)	21 (11)	20 (10)	18 (13)
High-risk MDS	51 (10)	20 (10)	19 (10)	12 (9)
Performance status (ECOG), No. (%)				
0	264 (50)	99 (51)	94 (49)	71 (51)
1	226 (43)	81 (42)	86 (45)	59 (42)
2	33 (7)	13 (7)	11 (6)	9 (6)
Disease status before course 2, No. (%)	N = 523	N = 193	N = 191	N = 139
Not in CR/CRi	165 (31.5)	66 (34.2)	62 (32.4)	37 (26.6)
MRD+	129 (24.7)	52 (26.9)	45 (23.5)	32 (23)
MRD unknown	32 (6.1)	12 (6.2)	15 (7.9)	5 (3.6)
MRD-negative	3 (0.6)	1 (0.5)	2 (1)	0
EMD (MRD-negative)	1	1[a]	0	0
In CR/CRi	358 (68.5)	127 (65.8)	129 (67.5)	102 (73.4)
MRD+	259 (49.5)	92 (47.7)	93 (48.7)	74 (53.2)
MRD+ ≥0.1%	160 (30.6)	55 (28.5)	59 (30.9)	46 (33.1)
MRD+ <0.1%	99 (18.9)	37 (19.2)	34 (17.8)	28 (20.1)
MRD unknown	99 (18.9)	35 (18.1)	36 (18.8)	28 (20.1)
Course 1 treatment				
DA	193 (37)	74 (38)	73 (38)	46 (33)
DA GO1	149 (28)	51 (26)	50 (26)	48 (35)
DA GO2	181 (34)	68 (35)	68 (36)	45 (32)
Small molecule from course 2				
Long quizartinib	69 (13)	24 (12)	24 (13)	21 (15)
Short quizartinib	71 (14)	26 (13)	23 (12)	22 (16)
No quizartinib	383 (73)	143 (74)	144 (75)	96 (69)
Genetic risk				
Cytogenetic (Grimwade et al[11])				
Favorable	11 (2.2)	5 (2.7)	3 (1.6)	3 (2.3)
Intermediate	367 (73.8)	134 (72.8)	137 (75.3)	96 (73.3)
Adverse	99 (19.9)	38 (20.7)	32 (17.6)	29 (22.1)
Failed	20 (4)	7 (3.8)	10 (5.5)	3 (2.3)
Not reported	26	9	9	8
TP53+	46 (9)	16 (8)	17 (9)	13 (9)
ELN 2017				
Favorable	82 (25)	34 (29)	23 (21)	25 (23)
Intermediate	74 (22)	26 (22)	24 (22)	24 (22)
Adverse	168 (50)	51 (44)	60 (55)	57 (53)
Unknown	9 (3)	5 (4)	2 (2)	2 (2)
Not reported	190	77	82	31

(continued on following page)

TABLE 1. Patient Demographics and Clinical Characteristics (continued)

Characteristic	Overall (N = 523)	DA (n = 193)	FLAG-Ida (n = 191)	DA-Cladribine (n = 139)
FLT3 mutations	89 (17)	35 (18)	29 (15)	25 (18)
NPM1 mutations	107 (25)	44 (24)	38 (22)	25 (19)
MDS-related mutations	154 (46)	53 (46)	49 (45)	52 (48)

Abbreviations: BPDCN, blastic plasmacytoid dendritic neoplasm; CR, complete remission; CRi, CR with incomplete count recovery; DA, daunorubicin and AraC; ECOG, Eastern Cooperative Oncology Group; ELN, European LeukemiaNet; EMD, extramedullary disease; FLAG-Ida, fludarabine, AraC, granulocyte colony-stimulating factor, and idarubicin; GO, gemtuzumab ozogamicin; MDS, myelodysplastic syndrome; MRD, measurable residual disease.
[a]BPDCN patient with extramedullary disease.

converted to MRD negativity after DA, DAC, and FLAG-Ida, respectively (DA *v* DAC: *P* = .16; DA *v* FLAG: *P* = .33; Data Supplement, Fig S3B). A similar increment in MRD conversion of approximately 10% after intensified chemotherapy compared with DA was apparent when results were restricted to those in CR/CRi after course 1 (DA, 56%; DAC, 69%; FLAG-Ida, 66%; Table 2). We also observed that upper quartile levels of % MRD after course 2 were lower in the DAC (upper quartile 0.07%) or FLAG-Ida (upper quartile 0.09%) treatment groups compared with DA (upper quartile 0.16%).

There was no significant OS benefit for intensification versus no intensification (HR, 0.82 [95% CI, 0.66 to 1.04]; *P* = .102; Data Supplement, Fig S5A) nor in the separate intensification arms of DAC or FLAG-Ida versus DA (DAC *v* DA: HR, 0.74 [95% CI, 0.55 to 1.01]; *P* = .054; FLAG-Ida *v* DA: HR, 0.86 [95% CI, 0.66 to 1.12]; *P* = .270); OS at 3 years was 34%, 46%, and 42% for DA, DAC, and FLAG-Ida, respectively (Fig 2; Table 2).

Three hundred eighty-eight patients entering the course 2 random assignment had an MRD result by flow cytometry after course 1 including 133 of the 165 not in CR/CRi. Remaining patients were categorized as MRD unknown (Table 1; Data Supplement, Table S2). There was no detectable survival benefit from intensification for MRD unknown patients (HR, 1.32 [95% CI, 0.81 to 2.14]) in stratified analyses (Data Supplement, Fig S6). A planned

TABLE 2. Comparison of Outcomes by Course 2 Random Assignment—All Patients

Course 2 Outcomes	DA	FLAG-Ida			DA-Cladribine		
Response Conversion After Course 2	No. (%)	No. (%)	OR (95% CI)	*P*	No. (%)	OR (95% CI)	*P*
Not in CR/CRi after course 1	n = 66	n = 62			n = 37		
ORR (CR + CRi) after course 2	36 (55)	21 (34)	0.42 (0.21 to 0.87)	.020	21 (57)	1.31 (0.55 to 3.11)	.536
CR after course 2	23 (35)	7 (11)	0.24 (0.09 to 0.61)	.003	13 (35)	1.19 (0.48 to 2.96)	.706
CRi after course 1	n = 20	n = 15			n = 11		
CR after course 2	3 (15)	4 (27)		.430	4 (36)		.381
CR/CRi after course 1 evaluable for MRD conversion[a]	n = 78	n = 58			n = 55		
MRD-negative after course 2	44 (56)	38 (66)	1.47 (0.73 to 2.96)	.284	38 (69)	1.18 (0.53 to 2.62)	.628

Outcomes	3-Year, %		HR (95% CI)	*P*	3-Year, %	HR (95% CI)	*P*
Overall survival	34	42	0.86 (0.66 to 1.12)	.270	46	0.74 (0.55 to 1.01)	.054
Cumulative incidence of relapse	52	47	0.85 (0.63 to 1.15)	.290	43	0.76 (0.53 to 1.08)	.128
Relapse-free survival	28	35	0.87 (0.67 to 1.12)	.281	37	0.78 (0.58 to 1.06)	.117

Transplantation	No. (%)	No. (%)	OR (95% CI)	*P*	No. (%)	OR (95% CI)	*P*
Allografts in CR1	70 (36)	62 (33)	0.84 (0.55 to 1.29)	.432	58 (42)	1.30 (0.80 to 2.11)	.288

NOTE. MRD conversion rates for all evaluable patients (including those not in CR/CRi after course 1) are shown in the Data Supplement (Fig S3B). Proportions of patients supplying postcourse 2 MRD data were lower in FLAG-Ida and DA-cladribine arms (62% and 74.5%, respectively) compared with DA (81%).
Abbreviations: CR, complete remission; CR1, first remission; CRi, CR with incomplete count recovery; DA, daunorubicin and AraC; FLAG-Ida, fludarabine, AraC, granulocyte colony-stimulating factor, and idarubicin; HR, hazard ratio; MRD, measurable residual disease; OR, odds ratio; ORR, overall response rate.
[a]Patients in CR/CRi after course 1 with MRD after post course 1 and 2.

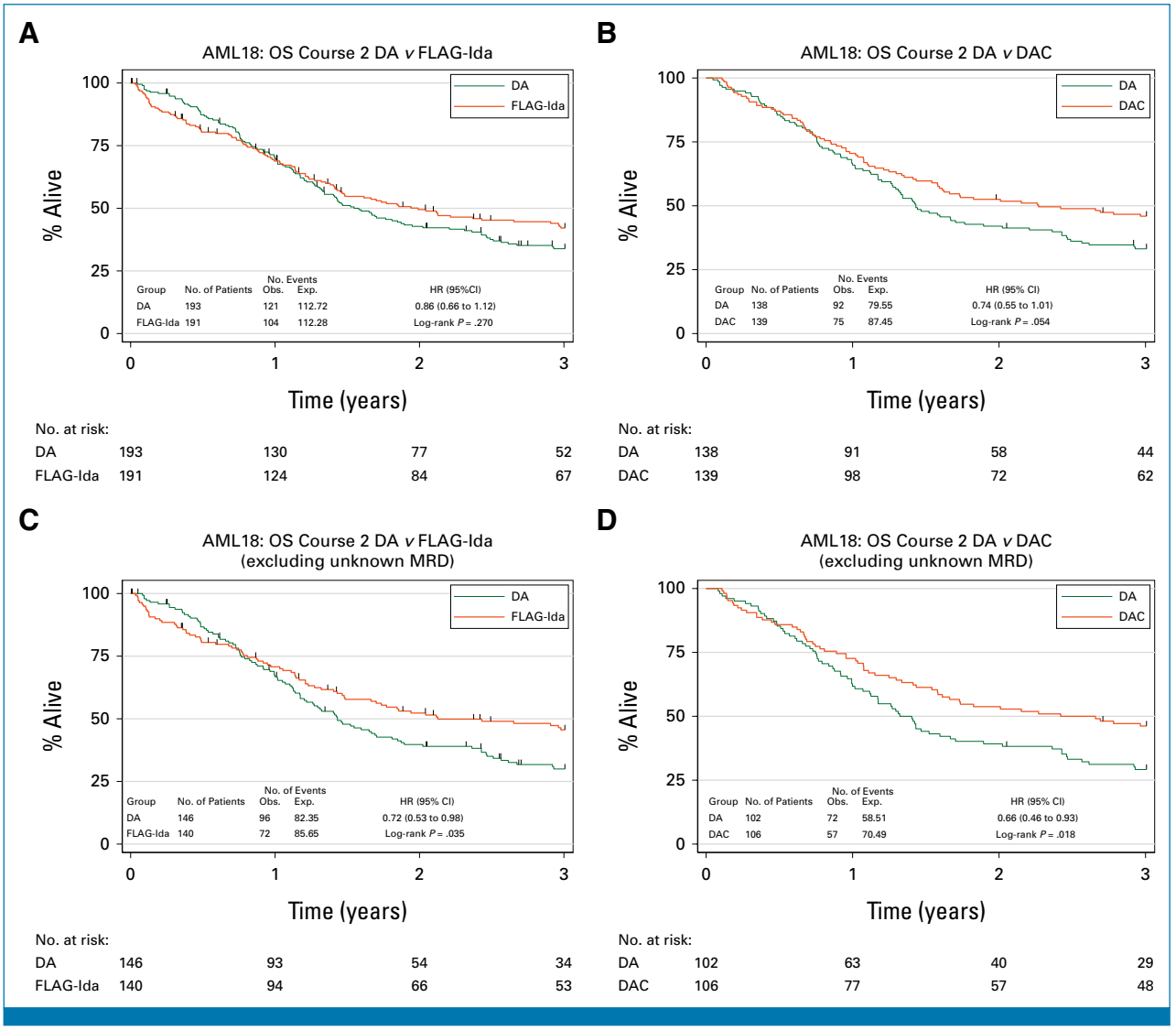

FIG 2. OS by random assignment. (A) DA versus FLAG-Ida. (B) DA versus DAC. (C) DA versus FLAG-Ida, excluding patients with unknown MRD status. (D) DA versus DAC, excluding patients with unknown MRD status. MRD unknown includes MRD unknown not in CR/CRi (Table 1; Data Supplement, Fig S2). CR, complete remission; CRi, CR with incomplete count recovery; DA, daunorubicin, Ara-C; DAC, DA with cladribine; FLAG-Ida, fludarabine/Ara-C/G-CSF/idarubicin; HR, hazard ratio; MRD, measurable residual disease; OS, overall survival.

sensitivity analysis excluded MRD unknown patients as these were without predefined measurable residual leukemia. The outcomes from this are summarized in Table 3, showing a significant OS benefit for both DAC (HR, 0.66 [95% CI, 0.46 to 0.93]; P = .018) and FLAG-Ida (HR, 0.72 [95% CI, 0.53 to 0.98]; P = .035; Figs 2C and 2D) in addition to intensification overall (HR, 0.71 [95% CI, 0.54 to 0.92]; P = .010; Data Supplement, Fig S5B). OS at 3 years was 30%, 46%, and 46% for DA, DAC, and FLAG-Ida, respectively.

There was no evidence that this improvement in survival from intensification depended on remission as there was no detectable interaction by baseline remission status (DAC v DA, test for heterogeneity P = .25; FLAG-Ida v DA, test for heterogeneity P = .92; Data Supplement, Fig S7). OS at 3 years for patients in MRD+ CR/CRi at baseline was 51% in the DAC arm (v 29% DA, HR, 0.58 [95% CI, 0.38 to 0.91]; P = .016) and 45% in the FLAG-Ida arm (v 29% DA, HR, 0.74 [95% CI, 0.51 to 1.08]; P = .122; Data Supplement, Figs S7C and S7D).

Concordant with the survival benefit, cumulative incidence of relapse (CIR) was significantly reduced by DAC and FLAG-Ida compared with DA in patients with flow measurable residual leukemia (DAC v DA: HR, 0.66 [95% CI, 0.45 to 0.98]; P = .039; FLAG-Ida v DA: HR, 0.70 [95% CI, 0.49 to 0.99]; P = .042). CIR at 3 years were 58%, 46%, and 46% for DA, DAC, and FLAG-Ida, respectively (Table 3; Fig 3).

The results for OS from the time of random assignment favored both intensification regimens compared with DA in

TABLE 3. Comparison of Outcomes by Course 2 Random Assignment Excluding Patients With Unknown MRD

Course 2 Outcomes	DA (N = 146)	FLAG-Ida (N = 140)			DA-Cladribine (N = 106)		
Response Conversion After Course 2	No. (%)	No. (%)	OR (95% CI)	*P*	No. (%)	OR (95% CI)	*P*
Not in CR/CRi after course 1	n = 53	n = 47			n = 32		
ORR (CR + CRi) after course 2	31 (59)	14 (30)	0.30 (0.13 to 0.69)	.005	19 (59)	1.39 (0.54 to 3.57)	.496
CR after course 2	20 (38)	5 (11)	0.20 (0.07 to 0.58)	.003	12 (38)	1.2 (0.45 to 3.19)	.715
CRi after course 1	n = 17	n = 11			n = 9		
CR after course 2	3 (18)	2 (18)		1	3 (33)		.643

Outcomes	3-Year, %		HR (95% CI)	*P*	3-Year, %	HR (95% CI)	*P*
Overall survival	30	46	0.72 (0.53 to 0.98)	.035	46	0.66 (0.46 to 0.93)	.018
Cumulative incidence relapse	58	46	0.70 (0.49 to 0.99)	.042	46	0.66 (0.45 to 0.98)	.039
Relapse-free survival	23	39	0.66 (0.49 to 0.90)	.008	36	0.66 (0.47 to 0.92)	.015

Transplantation	No. (%)	No. (%)	OR (95% CI)	*P*	No. (%)	OR (95% CI)	*P*
Allografts in CR1	52 (36%)	44 (31%)	0.83 (0.51 to 1.36)	.454	43 (41%)	1.37 (0.78 to 4.20)	.281

NOTE. MRD known patients include three MRD-negative patients (one DA and two FLAG-Ida).
Abbreviations: CR, complete remission; CR1, first remission; CRi, CR with incomplete count recovery; DA, daunorubicin and AraC; FLAG-Ida, fludarabine, AraC, granulocyte colony-stimulating factor, and idarubicin; HR, hazard ratio; MRD, measurable residual disease; OR, odds ratio; ORR, overall response rate.

most subgroups on the basis of clinically defined characteristics at trial entry (age, Eastern Cooperative Oncology Group performance status, cytogenetics, clinical disease type, and sex); this included secondary AML defined clinically but also by mutations.[12] Benefit appeared to be maintained in patients older than 70 years (Data Supplement, Fig S8).

Impact of Transplant

In total, 190 (36%) of randomly assigned patients underwent allogeneic stem-cell transplantation (ASCT) in first remission (CR1). By random assignment, this was DA 70/193 (36%), DAC 58/139 (42%), and FLAG-Ida 62/191 (33%;

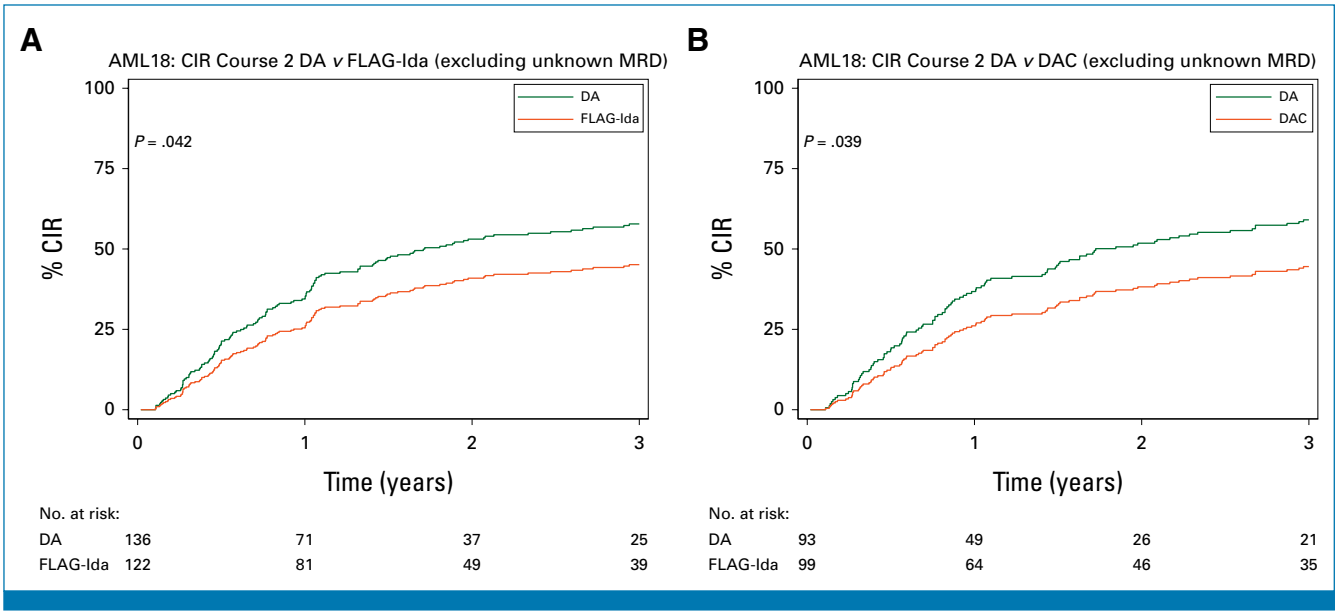

FIG 3. CIR by random assignment excluding patients with unknown MRD status. (A) DA versus FLAG-Ida. (B) DA versus DAC. MRD unknown includes MRD unknown not in CR/CRi (Table 1; Data Supplement, Fig S2). Results from comparison of relapse by cause-specific HRs are DAC versus DA: HR, 0.62 (95% CI, 0.47 to 0.93); *P* = .019; FLAG-Ida versus DA: HR, 0.67 (95% CI, 0.47 to 0.95); *P* = .024. CIR, cumulative incidence of relapse; CR, complete remission; CRi, CR with incomplete count recovery; DA, daunorubicin/Ara-C; DAC, DA with cladribine; FLAG-Ida, fludarabine/Ara-C/G-CSF/idarubicin; HR, hazard ratio; MRD, measurable residual disease.

Table 2). Only 24 were performed in patients older than 70 years. Fifty-two percent (98/190) CR1 transplants took place after course 3. The OS benefit from intensification observed among the group with known MRD was maintained for DAC when patients were censored at ASCT but was reduced for FLAG-Ida (Fig 4; Data Supplement, Fig S9). Of patients not proceeding to ASCT, 13, 11, and 14 received course 3 in the DA, DAC, and FLAG-Ida arms, respectively.

DISCUSSION

The outcomes for older patients with AML with intensive chemotherapy remain poor including in those who attain remission but have persisting measurable disease. Thus, in our NCRI AML16 trial, survival at 3 years was only 26% consequent to a very high relapse risk (83% CIR) if MRD+ after course 1. This compared with 42% survival at 3 years if MRD−. In an attempt to improve the outcome for patients without an MRD− remission, we undertook a randomized trial asking whether treatment intensification after first induction would improve their response and survival or would merely worsen toxicity. For intensification, we used the FLAG-Ida regimen or a second course of DA intensified by the addition of cladribine. Patients who were in remission but with MRD unknown status were also eligible for this random assignment. Importantly, patients have to be considered fit enough for this approach as exemplified by the selection of younger, fitter patients for the random assignment. Our results highlight that inclusion of patients without predefined MRD (no-target cohort) in MRD-directed strategies reduces efficiency. In this trial, we saw a significant long-term survival benefit for intensified therapy with both DAC and FLAG-Ida including in refractory patients but only in a preplanned exploratory sensitivity analysis that excluded patients with MRD unknown status. The absence of benefit when MRD was unassessable after first induction may be explained by an enrichment for patients with hypocellular BMs (categorized as inadequate BMs) and/or MRD negativity. The latter is supported by the observation that MRD was undetectable by different-from-normal analysis in the MRD unknown patients providing an adequate postcourse 1 BM (~60% of MRD unknown cohort).

In the group with residual leukemia by flow cytometry, survival advantage was despite the greater myelotoxicity and greater requirement for supportive care and hospitalization with intensification. This was particularly apparent with FLAG-Ida, which appeared less well-tolerated than DAC with higher early mortality despite dose reductions for older age. Furthermore, of patients who were refractory after first induction, fewer attained CR/CRi after FLAG-Ida intensification compared with DA and DAC, possibly because of greater myelosuppression. We would speculate that the efficacy of FLAG-Ida is more dependent on hematopoietic reserve than DAC. Overall, DAC intensification appeared better tolerated, was more consistently applied with subsequent consolidation including allogeneic transplant, and delivered the same long-term survival benefits as FLAG-Ida without the increase in early deaths. Notably, the survival benefit for intensification with DAC was not lost on censoring for transplant unlike FLAG-Ida, and was maintained in patients older than 70 years who are not normally considered transplant-eligible. We suggest that this may be because patients in the DAC arm were more likely to receive two intensification courses (52% received a second course of

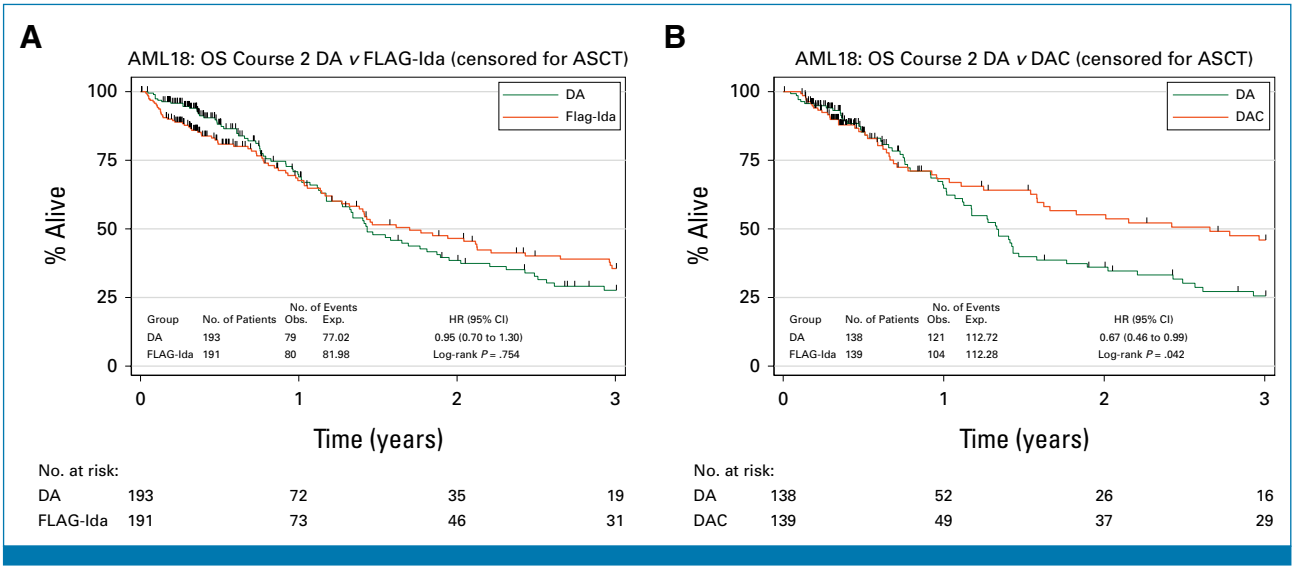

FIG 4. OS by random assignment censored for allogeneic transplant, all patients. (A) DA versus FLAG-Ida and (B) DA versus DAC. In transplanted patients, post-transplant NRM (with relapse considered as a competing risk) at 3 years was DA, 35%; FLAG-Ida, 26%; and DAC, 24% (OS by random assignment censored for allogeneic transplant, excluding patients with unknown MRD, is displayed in the Data Supplement, Fig S9). DA, daunorubicin/Ara-C; DAC, DA with cladribine; FLAG-Ida, fludarabine/Ara-C/G-CSF/idarubicin; HR, hazard ratio; MRD, measurable residual disease; NRM, nonrelapse mortality; OS, overall survival; ASCT, allogeneic stem-cell transplantation.

assigned intensification compared with 33% in the FLAG-Ida arm), thus possibly increasing the depth of response further. Encouragingly, treatment modification with DAC intensification in patients with post-first induction residual leukemia (below and above the morphologic remission threshold) is associated with a 5-year survival of 31%, approaching that observed in AML18 patients achieving MRD negativity after first induction (37% at 5 years). Limitations to the generalizability of these findings include the large number of potentially eligible patients who were not randomly assigned; this group was enriched for refractory patients, possibly because of the historically frequent use of FLAG-Ida in this setting as salvage therapy.[3] However, there was no significant heterogeneity by remission status for the observed benefit from intensification in patients with residual disease. We also note that small sample size precludes evidence in *TP53*-mutated patients. Furthermore, findings cannot be extrapolated to molecular MRD, particularly when only low levels are detected. Finally, residual disease was assessed by BM taken at count recovery rather than at day 14.

In summary, we have shown that MRD assessment after induction is critical to optimize outcomes for older patients with AML. Treatment intensification in patients with measurable persistent leukemia after induction can reduce the risk of relapse and improve OS. For this purpose, DAC was as effective and less toxic than FLAG-Ida.

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DATA SHARING STATEMENT

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AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

Treatment Intensification With Either Fludarabine, AraC, G-CSF and Idarubicin, or Cladribine Plus Daunorubicin and AraC on the Basis of Residual Disease Status in Older Patients With AML: Results From the NCRI AML18 Trial

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