The Clinical Role of microRNAs in Cytogenetically Normal Acute Myeloid Leukemia: miR-155 Upregulation Independently Identifies High-Risk Patients

Marcucci, et al

Supplemental Text

The Clinical Role of microRNAs in Cytogenetically Normal Acute Myeloid Leukemia: *miR-155* Upregulation Independently Identifies High-Risk Patients

Marcucci et al.

Participating institutions

The following CALGB/Alliance institutions participated in this study and contributed at least 1% of patents. For each of these institutions current principal investigators are listed.

The Ohio State University Medical Center, Columbus, OH: Clara D. Bloomfield (grant no. U10CA077658); Wake Forest University School of Medicine, Winston-Salem, NC: David D. Hurd; North Shore University Hospital, Manhasset, NY: Daniel R. Budman (grant no. U10CA035279); Duke University Medical Center, Durham, NC: Jeffrey Crawford (grant no. U10CA047577); Roswell Park Cancer Institute, Buffalo, NY: Ellis G. Levine (grant no. U10CA059518); University of Iowa Hospitals, Iowa City, IA: Daniel A. Vaena; Washington University School of Medicine, St. Louis, MO: Nancy L. Bartlett (grant no. U10CA077440); University of Chicago Medical Center, Chicago, IL: Hedy L. Kindler (grant no. U10CA041287); Vermont Cancer Center, Burlington, VT: Steven M. Grunberg; University of Massachusetts Medical Center, Worcester, MA: William V. Walsh; Dana Farber Cancer Institute, Boston, MA: Harold J. Burstein (grant no. U10CA032291); Ft. Wayne Medical Oncology/Hematology, Ft. Wayne, IN: Sreenivasa Nattam; Dartmouth Medical School, Lebanon, NH: Konstantin Dragnev; Eastern Maine

Medical Center, Bangor, ME: Thomas H. Openshaw; University of North Carolina, Chapel Hill, NC: Thomas C. Shea (grant no. U10CA047559); Weill Medical College of Cornell University, New York, NY: John Leonard; Mount Sinai School of Medicine, New York, NY: Lewis R. Silverman; Rhode Island Hospital, Providence, RI: William Sikov; SUNY Upstate Medical University, Syracuse, NY: Stephen L. Graziano; Moores University of California San Diego Cancer Center, San Diego, CA: Barbara A. Parker; University of Missouri/Ellis Fischel Cancer Center, Columbia, MO: Carl E. Freter; University of Maryland Cancer Center, Baltimore, MD: Martin J. Edelman; Christiana Care Health Services, Inc., Newark, DE: Stephen S. Grubbs; Western Pennsylvania Hospital, Pittsburgh, PA: John Lister.

Patients' Treatment

All patients received cytarabine-daunorubicin-based induction chemotherapy. Patients younger than 60 years were treated on Cancer and Leukemia Group B (CALGB) trials 9621[1,2] or 19808.[3] These patients received induction chemotherapy with cytarabine, daunorubicin, and etoposide with or without PSC-833 (valspodar), a multidrug resistance protein inhibitor. On achievement of complete remission (CR), the patients were assigned to intensification with high-dose cytarabine and etoposide for stem-cell mobilization followed by myeloablative treatment with busulfan and etoposide supported by autologous stem-cell transplantation (SCT). Older patients (≥60 years) were treated with less intense regimens on CALGB protocols 8525,[4] 8923,[5] 9420,[6] 9720,[7,8] or 10201.[9] Patients treated on protocols CALGB 8525[4] and 8923[5] received induction therapy with cytarabine and daunorubicin. All patients achieving CR were randomly

assigned to postremission therapy consisting of four cycles of cytarabine in standard, intermediate, or high dose, followed by four cycles of maintenance therapy with cytarabine and daunorubicin on CALGB 8525,[4] and four cycles of standard-dose cytarabine or two cycles of mitoxantrone plus intermediate-dose cytarabine on CALGB 8923.[5] Protocols CALGB 9420, 9720, and 10201 included investigational agents other than chemotherapy. CALGB 9720 was initiated as a phase III trial in untreated acute myeloid leukemia (AML) patients 60 years and older evaluating multidrug resistance (MDR) modulation by valspodar during induction with cytarabine, daunorubicin and etoposide, and consolidation therapy, which consisted of a single course of postremission chemotherapy similar to the initial induction regimen, except that cytarabine was given for 5 rather than 7 days and there were 2 rather than 3 doses of daunorubicin and etoposide.[7,8] Similar consolidation therapy, ie, one course of attenuated induction chemotherapy was also received by patients on CALGB 9420.[6] The valspodar arm of CALGB 9720 was closed after randomized assignment of 120 patients because of excessive early deaths. Enrollment on this protocol continued on the chemotherapy-only control arm. CALGB 9420 and CALGB 9720 also evaluated a subcutaneous interleukin-2 regimen as maintenance therapy, which was demonstrated to induce no clear benefit.[7,8] CALGB 10201 evaluated the *BCL2* antisense, oblimersen sodium (Genasense; G3139) administered with induction (cytarabine and daunorubicin) and consolidation (two cycles of higher dose cytarabine) chemotherapy; preliminary results showed no impact of the antisense on outcome.[9] Per the protocols, no younger or older patients enrolled on these studies received allogeneic SCT during first CR.

Definition of clinical endpoints

Clinical endpoints were defined according to generally accepted criteria.[10] Per protocol, all patients were to receive at least one induction cycle. For patients with residual leukemia present in a bone marrow biopsy after one induction cycle, a second cycle of induction was administered. CR required a bone marrow (BM) aspirate with cellularity >20% with maturation of all cell lines, <5% blasts and undetectable Auer rods; no circulating leukemic blasts, blood neutrophil count ≥1.5 x 10^9/L and platelets >100 x 10^9/L, and no evidence of extramedullary leukemia, all of which had to persist ≥4 weeks. Relapse was defined by >5% blasts in BM aspirates or the development of extramedullary leukemia in patients with previously documented CR.[10] Disease-free survival (DFS) was measured from the date of CR until the date of relapse or death (from any cause); patients alive and in CR were censored at last follow-up. Overall survival (OS) was measured from the date of study entry until the date of death (from any cause), and patients alive at last follow-up were censored.

Multivariable models

Variables that were significant at $P < .2$ in the univariable models and considered for multivariable model inclusion were as follows. In the model for achievement of CR in all patients: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-type), *FLT3*-ITD (positive *v* negative), *WT1* (mutated *v* wild-type), *TET2* (mutated *v* wild-type), *ASXL1* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *ERG* (high *v* low) , *BAALC* (high *v* low), hemoglobin (continuous), WBC (continuous, 50-unit increase) and age group (≥60 *v* <60). In the model for DFS in all patients: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-

type), *FLT3*-ITD (positive *v* negative), *WT1* (mutated *v* wild-type), *DNMT3A* (mutated *v* wild-type), *MLL*-PTD (positive *v* negative), *RUNX1* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), WBC (continuous, 50-unit increase), race (white *v* nonwhite) and age group (≥60 *v* <60). In the model for OS in all patients: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-type), *FLT3*-ITD (positive *v* negative), *WT1* (mutated *v* wild-type), *ASXL1* (mutated *v* wild-type), *DNMT3A* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *IDH1* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), WBC (continuous, 50-unit increase), race (white *v* nonwhite) and age group (≥60 *v* <60). In the model for achievement of CR in patients aged <60 years: *miR-155* (high *v* low), *FLT3*-ITD (positive *v* negative), *CEBPA* (mutated *v* wild-type), *TET2* (mutated *v* wild-type), *ASXL1* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *IDH2* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), hemoglobin (continuous), WBC (continuous, 50-unit increase) and age (continuous, 10-year increase). In the model for DFS in patients aged <60 years: *miR-155* (high *v* low), FLT3-ITD (positive *v* negative), *FLT3*-TKD (positive *v* negative), *WT1* (mutated *v* wild-type), *MLL*-PTD (positive *v* negative), *RUNX1* (mutated *v* wild-type), *IDH1* (mutated *v* wild-type), *IDH2* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), WBC (continuous, 50-unit increase) and race (white *v* nonwhite). In the model for OS in patients aged <60 years: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-type), *FLT3*-ITD (positive *v* negative), *CEBPA* (mutated *v* wild-type), *WT1* (mutated *v* wild-type), *TET2* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *IDH1* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), hemoglobin (continuous), WBC (continuous, 50-unit increase) and race (white *v* nonwhite). In the model for achievement of CR in patients aged ≥60 years: *miR-155*

(high *v* low), *NPM1* (mutated *v* wild-type), *WT1* (mutated *v* wild-type), *ASXL1* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low), WBC (continuous, 50-unit increase) and age (continuous, 10-year increase). In the model for DFS in patients aged ≥60 years: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-type), *FLT3*-ITD (positive *v* negative), *FLT3*-TKD (positive *v* negative), *ASXL1* (mutated *v* wild-type), *ERG* (high *v* low) and *BAALC* (high *v* low). In the model for OS in patients aged ≥60 years: *miR-155* (high *v* low), *NPM1* (mutated *v* wild-type), *FLT3*-ITD (positive *v* negative), *WT1* (mutated *v* wild-type), *ASXL1* (mutated *v* wild-type), *RUNX1* (mutated *v* wild-type), *ERG* (high *v* low), *BAALC* (high *v* low) and WBC (continuous, 50-unit increase).

Information on missing samples

The patients included in our study were compared with those who were not included because of lack of available material. The two groups were similar for age (P=.27), sex (P=.89), race (P=.33), hemoglobin (P=.86), and platelets (P=.55). The patients included in the present study had higher WBC (P<.001), blood blasts (P<.001), and BM blasts (P<.001) compared with the patients that were not included. This was not unexpected, as patients with higher WBC and blood and BM blasts usually have more material that can be banked and utilized for correlative studies.

SUPPLEMENTAL REFERENCES

1. Kolitz JE, George SL, Dodge RK, et al: Dose escalation studies of cytarabine, daunorubicin, and etoposide with and without multidrug resistance modulation with PSC-833 in untreated adults with acute myeloid leukemia younger than 60 years: Final induction results of Cancer and Leukemia Group B Study 9621. J Clin Oncol 22:4290-4301, 2004
2. Kolitz JE, George SL, Barrier R, et al: A novel post-remission consolidation regimen for patients with acute myeloid leukemia (AML) < 60 years old with normal or unfavorable cytogenetics: Results from CALGB 9621. Blood 102:175a, 2003 (abstr 609)
3. Kolitz JE, George SL, Marcucci G, et al: P-glycoprotein inhibition using valspodar (PSC-833) does not improve outcomes for patients under age 60 years with newly diagnosed acute myeloid leukemia: Cancer and Leukemia Group B study 19808. Blood 116:1413-1421, 2010
4. Mayer RJ, Davis RB, Schiffer CA, et al: Intensive postremission chemotherapy in adults with acute myeloid leukemia. N Engl J Med 331:896-903, 1994
5. Stone RM, Berg DT, George SL, et al. Postremission therapy in older patients with de novo acute myeloid leukemia: A randomized trial comparing mitoxantrone and intermediate-dose cytarabine with standard-dose cytarabine. Blood 98:548-553, 2001
6. Lee EJ, George SL, Caligiuri M, et al: Parallel phase I studies of daunorubicin given with cytarabine and etoposide with or without the multidrug resistance

modulator PSC-833 in previously untreated patients 60 years of age or older with acute myeloid leukemia: Results of Cancer and Leukemia Group B study 9420. J Clin Oncol 17:2831-2839, 1999

7. Baer MR, George SL, Dodge RK, et al: Phase 3 study of the multidrug resistance modulator PSC-833 in previously untreated patients 60 years of age and older with acute myeloid leukemia: Cancer and Leukemia Group B study 9720. Blood 100:1224-1232, 2002

8. Baer MR, George SL, Caligiuri MA, et al: Low-dose interleukin-2 immunotherapy does not improve outcome of patients age 60 years and older with acute myeloid leukemia in first complete remission: Cancer and Leukemia Group B study 9720. J Clin Oncol 26:4934-4939, 2008

9. Marcucci G, Moser B, Blum W, et al: A phase III randomized trial of intensive induction and consolidation chemotherapy ± oblimersen, a proapoptotic Bcl-2 antisense oligonucleotide in untreated acute myeloid leukemia patients >60 years old. J Clin Oncol 25:360s, 2007 (suppl; abstr 7012)

10. Cheson BD, Cassileth PA, Head DR, et al: Report of the National Cancer Institute-sponsored workshop on definitions of diagnosis and response in acute myeloid leukemia. J Clin Oncol 8:813-819, 1990

SUPPLEMENTAL TABLES

Supplemental Table 1. Comparison of Clinical and Molecular Characteristics Associated With *miR155* expression in younger (<60 years) Patients With Primary Cytogenetically Normal Acute Myeloid Leukemia

Characteristic	Low *miR155*[†] (n=77)	High *miR155*[†] (n=76)	*P*
Age, years			.84
Median	45	46	
Range	19-59	18-59	
Male sex, no. (%)	35 (45)	38 (50)	.63
Race, no. (%)			.31
White	66 (86)	70 (92)	
Nonwhite	11 (14)	6 (8)	
WBC, $x10^9$/L			.01
Median	20.4	32.3	
Range	0.9-295.0	1.4-273.0	
Blood blasts, %			.09
Median	55	66	
Range	0-97	0-97	
Bone marrow blasts, %			.11
Median	63	70	
Range	10-94	21-95	
Hemoglobin, g/dL			.38
Median	9.4	9.1	
Range	4.9-13.4	4.6-13.3	
Platelet count, $x10^9$/L			.24
Median	70	57	
Range	11-445	8-380	
Extramedullary involvement, no. (%)	22 (29)	21 (28)	1.00
NPM1, no. (%)			.24
Mutated	46 (60)	53 (70)	
Wild-type	31 (40)	23 (30)	
FLT3-ITD, no. (%)			<.001
Present	14 (18)	45 (59)	
Absent	63 (82)	31 (41)	
CEBPA, no. (%)			.02
Mutated	21 (27)	9 (12)	
Single mutated	4	7	
Double mutated	17	2	
Wild-type	56 (73)	67 (88)	
ELN Genetic Group, no. (%)*			<.001
Favorable	55 (71)	25 (33)	
Intermediate-I	22 (29)	51 (67)	

Characteristic	Low *miR155*[†] (n=77)	High *miR155*[†] (n=76)	*P*
TET2, no. (%)			.28
Mutated	10 (13)	15 (20)	
Wild-type	67 (87)	61 (80)	
ASXL1, no. (%)			.21
Mutated	1 (1)	4 (5)	
Wild-type	75 (99)	71 (95)	
DNMT3A, no. (%)			
Mutated	28 (38)	26 (36)	.87
R882	22	19	
Non-R882	6	7	
Wild-type	46 (62)	46 (64)	
RUNX1, no. (%)			.09
Mutated	4 (5)	10 (15)	
Wild-type	67 (95)	58 (85)	
IDH1, no. (%)			.30
Mutated	6 (8)	10 (13)	
Wild-type	70 (92)	64 (87)	
IDH2, no. (%)			.12
IDH2 mutated	12 (16)	5 (7)	
Codon R140 mutation	9	5	
Codon R172 mutation	3	0	
Wild-type	64 (84)	69 (93)	
FLT3-TKD, no. (%)			.009
Present	12 (16)	2 (3)	
Absent	65 (84)	74 (97)	
WT1, no. (%)			.25
Mutated	8 (10)	13 (17)	
Wild-type	69 (90)	63 (83)	
MLL-PTD, no. (%)			.37
Present	4 (5)	7 (9)	
Absent	73 (95)	69 (91)	
ERG expression group, no. (%)[†]			.02
High	31 (40)	45 (59)	
Low	46 (60)	31 (41)	
BAALC expression group, no. (%)[†]			.26
High	35 (45)	42 (55)	
Low	42 (55)	34 (45)	

Abbreviations: ELN, European LeukemiaNet; *FLT3*-ITD, internal tandem duplication of the *FLT3* gene; *FLT3*-TKD, tyrosine kinase domain mutation in the *FLT3* gene; *MLL*-PTD, partial tandem duplication of the *MLL* gene; WBC, white blood count.

* Within CN-AML patients, the ELN Favorable Genetic Group comprises patients with mutated *CEBPA*, and/or mutated *NPM1* without *FLT3*-ITD. All remaining CN-AML patients (ie, those with wild-type *CEBPA*, and *NPM1*-mutated with *FLT3*-ITD or with wild-type *NPM1* with or without *FLT3*-ITD) belong to the ELN Intermediate-I Genetic Group.

† The median expression value was used as a cut point.

Supplemental Table 2. Comparison of Clinical and Molecular Characteristics Associated With *miR155* expression in Older (≥60 years) Patients With Primary Cytogenetically Normal Acute Myeloid Leukemia

Characteristic	Low *miR155*[†] (n=105)	High *miR155*[†] (n=105)	*P*
Age, years			.15
Median	70	68	
Range	60-83	60-81	
Male sex, no. (%)	59 (56)	52 (50)	.41
Race, no. (%)			.63
White	94 (92)	94 (90)	
Nonwhite	8 (8)	11 (10)	
WBC, $x10^9$/L			.007
Median	22.2	39.1	
Range	0.8-249.3	1.0-450.0	
Blood blasts, %			.02
Median	42	62	
Range	0-97	0-99	
Bone marrow blasts, %			<.001
Median	60	76	
Range	4-97	6-97	
Hemoglobin, g/dL			.70
Median	9.3	9.4	
Range	5.4-14.5	6.0-15.0	
Platelet count, $x10^9$/L			.55
Median	70	67	
Range	4-481	11-850	
Extramedullary involvement, no. (%)	21 (21)	30 (29)	.20
NPM1, no. (%)			.88
Mutated	63 (62)	64 (63)	
Wild-type	39 (38)	37 (37)	
FLT3-ITD, no. (%)			<.001
Present	23 (22)	52 (51)	
Absent	80 (78)	49 (49)	
CEBPA, no. (%)			.09
Mutated	17 (17)	8 (8)	
Single mutated	10	5	
Double mutated	7	3	
Wild-type	85 (83)	93 (92)	
ELN Genetic Group, no. (%)*			<.001
Favorable	64 (63)	30 (30)	
Intermediate-I	38 (37)	70 (70)	
TET2, no. (%)			1.00
Mutated	32 (32)	31 (31)	
Wild-type	67 (68)	68 (69)	

Characteristic	Low *miR155*[†] (n=105)	High *miR155*[†] (n=105)	*P*
ASXL1, no. (%)			.84
Mutated	15 (15)	13 (13)	
Wild-type	85 (85)	86 (87)	
DNMT3A, no. (%)			
Mutated	29 (29)	37 (39)	.13
R882	14	25	
Non-R882	15	12	
Wild-type	72 (71)	58 (61)	
RUNX1, no. (%)			<.001
Mutated	5 (5)	23 (25)	
Wild-type	90 (95)	70 (75)	
IDH1, no. (%)			.37
Mutated	9 (9)	13 (13)	
Wild-type	93 (91)	87 (87)	
IDH2, no. (%)			.02
IDH2 mutated	32 (31)	17 (17)	
Codon R140 mutation	27	15	
Codon R172 mutation	5	2	
Wild-type	70 (69)	83 (83)	
FLT3-TKD, no. (%)			1.00
Present	12 (12)	11 (11)	
Absent	90 (88)	93 (89)	
WT1, no. (%)			.05
Mutated	3 (3)	10 (10)	
Wild-type	99 (97)	90 (90)	
MLL-PTD, no. (%)			<.001
Present	6 (6)	7 (7)	
Absent	97 (94)	98 (93)	
ERG expression group, no. (%)[†]			.27
High	48 (46)	57 (54)	
Low	57 (54)	48 (46)	
BAALC expression group, no. (%)[†]			.002
High	41 (39)	64 (61)	
Low	64 (61)	41 (39)	

Abbreviations: ELN, European LeukemiaNet; *FLT3*-ITD, internal tandem duplication of the *FLT3* gene; *FLT3*-TKD, tyrosine kinase domain mutation in the *FLT3* gene; *MLL*-PTD, partial tandem duplication of the *MLL* gene; WBC, white blood count.

* Within CN-AML patients, the ELN Favorable Genetic Group comprises patients with mutated *CEBPA*, and/or mutated *NPM1* without *FLT3*-ITD. All remaining CN-AML patients (ie, those with wild-type *CEBPA*, and *NPM1*-mutated with *FLT3*-ITD or with wild-type *NPM1* with or without *FLT3*-ITD) belong to the ELN Intermediate-I Genetic Group.

[†] The median expression value was used as a cut point.

Supplemental Table 3. Complete Remission Rates, Relapse Rates, Death Rates, and Disease-Free and Overall Survival in High *versus* Low *miR-155* Expressers

End Point	Low *miR-155**	High *miR-155**	*P*
Younger patients (n=153)			
No. of patients	77	76	
Complete remission rate, no. (%)	69 (90)	58 (76)	.03
Relapse rate, no. (%)	31 (45)	42 (72)	.002
Number expired, no. (%)	35 (45)	56 (74)	<.001
Disease-free survival			<.001
Median, years	NR	0.7	
% disease-free at 3 years (95% CI)	58 (45 to 69)	31 (20 to 43)	
% disease-free at 5 years (95% CI)	55 (43 to 66)	27 (17 to 39)	
Overall survival			<.001
Median, years	NR	1.2	
% alive at 3 years, (95% CI)	64 (52 to 73)	32 (22 to 42)	
% alive at 5 years, (95% CI)	57 (45 to 67)	28 (19 to 39)	
Older patients (n=210)			
No. of patients	105	105	
Complete remission rate, no. (%)	81 (77)	62 (59)	.008
Relapse rate, no. (%)	69 (85)	55 (89)	.62
Number expired, no. (%)	91 (87)	100 (95)	.05
Disease-free survival			.26
Median, years	1.0	0.6	
% disease-free at 3 years (95% CI)	19 (11 to 28)	18 (9 to 28)	
Overall survival			<.001
Median, years	1.5	0.8	
% alive at 3 years (95% CI)	25 (17 to 33)	15 (9 to 23)	

Abbreviations: CI, confidence interval; NR, not reached.

* The median expression value was used as a cut point. Gene expression was measured using NanoString.

Supplemental Table 4. Multivariable Model Limited to Patients With Measured Expression Level of *miR-181a* (n=298)

Group	Overall Survival		
	HR	95% CI	*P*
miR-155 expression, high *v* low	1.95	1.46 to 2.61	<.001
miR-181a expression, continuous	0.80	0.71 to 0.90	<.001
FLT3-ITD, positive *v* negative	1.43	1.06 to 1.94	.02
WT1, mutated *v* wild-type	2.03	1.26 to 3.26	.004
IDH1, mutated *v* wild-type	1.58	1.05 to 2.39	.03
ERG expression, high *v* low	1.47	1.09 to 1.98	.01
BAALC expression, high *v* low	1.92	1.41 to 2.60	<.001
WBC, each 50 units	1.12	1.02 to 1.22	.02
Age group, older *v* younger	3.66	2.67 to 5.01	<.001

NOTE: Hazard ratios greater than (less than) 1.0 indicate higher (lower) risk for death for the higher values of the continuous variables and the first category listed for the categorical variables.

Abbreviations: OR, odds ratio; HR, hazard ratio; CI, confidence interval; WBC, white blood count.

Supplemental Table 5. Complete Remission Rates and Disease-Free and Overall Survival in High *versus* Low *miR-155* Expressers According to the European LeukemiaNet Genetic Groups

End Point	Favorable*			Intermediate-I*		
	Low *miR-155*†	High *miR-155*†	*P*	Low *miR-155*†	High *miR-155*†	*P*
Younger patients (n=153)						
No. of patients	55	25		22	51	
Complete remission rate, no. (%)	53 (96)	20 (80)	.03	16 (73)	38 (75)	.99
Disease-free survival			.04			.41
Median, years	NR	3.5		1.1	0.7	
% disease-free at 3 years (95% CI)	68 (54 to 79)	50 (27 to 69)		25 (8 to 47)	21 (10 to 35)	
% disease-free at 5 years (95% CI)	66 (52 to 77)	45 (23 to 65)		19 (5 to 41)	18 (8 to 32)	
Overall survival			.02			.20
Median, y	NR	3.4		1.4	0.9	
% alive at 3 years, (95% CI)	75 (61 to 84)	52 (31 to 69)		36 (17 to 56)	22 (12 to 34)	
% alive at 5 years, (95% CI)	71 (57 to 81)	48 (28 to 66)		23 (8 to 41)	17 (8 to 29)	
Older patients (n=202)						
No. of patients	64	30		38	70	
Complete remission rate, no. (%)	53 (83)	21 (70)	.18	26 (68)	38 (54)	.22
Disease-free survival			.68			.83
Median, years	1.1	1.1		0.6	0.6	
% disease-free at 3 years (95% CI)	25 (14 to 37)	24 (9 to 43)		8 (1 to 22)	13 (5 to 26)	
Overall survival			.06			.05
Median, years	1.8	1.2		1.1	0.6	
% alive at 3 years (95% CI)	34 (23 to 46)	23 (10 to 39)		11 (3 to 23)	11 (5 to 20)	

Abbreviations: CI, confidence interval; NR, not reached.

* Within CN-AML patients, the ELN Favorable Genetic Group comprises patients with mutated *CEBPA*, and/or mutated *NPM1* without *FLT3*-ITD. All remaining CN-AML patients (ie, those with wild-type *CEBPA*, and *NPM1*-mutated with *FLT3*-ITD or with wild-type *NPM1* with or without *FLT3*-ITD) belong to the ELN Intermediate-I Genetic Group.

† The median expression value was used as a cut point. Gene expression was measured using NanoString.