Supplementary Appendix

**Acute Megakaryoblastic Leukemia Shows High Frequency of Chromosome 1q Aberrations and Dismal Outcome**

# Supplementary Methods

## Ethics

Written informed consent for inclusion in the clinical trials AMLCG 99, AMLCG 04 and AMLCG 08 or the AMLCG registry (clinical AML registry and biobank sampling approved by the Ethics Committee of the Ludwig Maximilian university until 2026, project number 204-11) were provided by patients treated in those trials or the registry. All study protocols were in accordance with the Declaration of Helsinki and approved by the institutional review boards of each participating centers. Patients in the AMLCG 99 and 08 trials as well as patients in the AMLCG registry, but not patients treated in the AMLCG 04 trial provided written informed consent for genetic analyses. Therefore, genetic analyses were not performed in patients treated in the AMLCG 04 trial. Patients treated at the local center provided written informed consent for genetic analyses. This retrospective analysis of AMKL patients treated in the AMLCG trials, AMLCG registry or local center has been approved by the ethics committee of the Ludwig-Maximilians University Munich.

## Treatment

21 patients within the final cohort of 38 patients with AMKL were treated in the AMLCG 99 study (clinicaltrials.gov identifier: NCT00266136), 3 patients were treated in the AMLCG 04 study (European Leukemia Trial Registry Nr. LN\_AMLINT\_2004\_230), and 2 patients were treated in the AMLCG 08 study (clinicaltrials.gov identifier: NCT01382147). Details of all three protocols have been published (1-4).

In short, patients in the AMLCG 99 study <60 years either received TAD-9 (thioguanine 2x 100mg/m2/d days 3-9, cytarabine 100mg/m2/d days 1-2 and 2x 100mg/m2/day days 3-8 and daunorubicin 60mg/m2/d days 3-5) or HAM (Cytarabine 2x 3g/m2/d days 1-3 and mitoxantrone 10mg/m2/d days 3-5) followed each by HAM on day 21 for induction chemotherapy. Patients were further randomized to either TAD-9 consolidation followed by autologous stem cell transplantation or TAD-9 followed by 3 years of cytarabine-based maintenance therapy. Patients with a suitable sibling-donor had the possibility of allogeneic stem cell transplantation in first CR. Patients ≥ 60 years were randomized between TAD-9 or HAM induction chemotherapy and only received another course of HAM if bone marrow blast were above 5% on day 16. Patients ≥ 60 years received TAD-9 as consolidation therapy, followed by cyarabine-based maintenance therapy.

In the AMLCG 04 trial patients received sequential HAM (sHAM) induction (1 g/m2 in patients >60 years). Consolidation chemotherapy was TAD-9 followed by a cytarabine-based maintenance therapy.

In the AMLCG 08 trial patients < 60 years and fit patients <70 years were randomized between induction chemotherapy with TAD-9 and HAM or dose dense induction with sHAM. Patients with good response to induction therapy and favourable risk profile received TAD-9 consolidation followed by 3-year cytarabine-based maintenance therapy. For patients with CRi only or intermediate/high risk of relapse an allogeneic transplantation in first CR was recommended. Patients ≥ 60 years were randomized between induction therapy with HAM (1g/m2 cytarabine) followed by another cycle of HAM if blasts in the bone marrow were above 5% on day 16 or dose-dense induction with sHAM (1g/m2 cytarabine). Consolidation therapy was TAD-9 followed by 3-year cytarabine-based maintenance therapy.

12 of 38 patients with AMKL were treated at our local center or other centers in Germany enrolling patients in the AMLCG registry (DRKS00020816). 4/12 patients received an induction chemotherapy with sHAM analog AMLCG 08 protocol with 1g/m2 cytarabine. Of these, one patient received TAD-9 consolidation chemotherapy and had a relapse afterwards. One patient had a relapse before consolidation therapy and for 2 patients we have no data on further treatment following sHAM induction. 1/12 patient received TAD-9/HAM Induction and had refractory disease. 2/12 patients received induction chemotherapy “7+3” and had refractory disease. 3/12 patients had a transition from MDS to AMKL and received cytoreductive therapy in context of planned allogeneic transplantation. 2/12 patients were treated with low-dose cytarabine only.

## Definition of clinical endpoints

Clinical end points were defined in accordance with commonly accepted criteria:

Complete remission (CR): bone marrow (BM) aspirate with a cellularity of 2+ and less than 5% blasts, in the peripheral blood a neutrophil count ≥ 1.5 G/µl and a platelet count ≥ 100 G/µl as well as absence of extramedullary leukemia.

Complete remission with incomplete hematologic recovery (CRi): BM aspirate with a cellularity below 2+ and less than 5% blasts, peripheral blood with no blast cells and either less than 1.5 G/µl neutrophils or less than 100 G/µl platelets.

Relapse: ≥ 5% blasts in the bone marrow or extramedullary leukemia manifestations.

Overall survival (OS): measured from day of study entry or diagnosis until the date of death. Patients alive were censored at last date of follow up.

Relapse free survival (RFS): measured from date of CR until date of relapse or death. Patients alive without relapse were censored at last date of follow up.

## Supplemental References

1. Buchner T, Berdel WE, Schoch C, Haferlach T, Serve HL, Kienast J, et al. Double induction containing either two courses or one course of high-dose cytarabine plus mitoxantrone and postremission therapy by either autologous stem-cell transplantation or by prolonged maintenance for acute myeloid leukemia. J Clin Oncol. 2006 Jun 1;24(16):2480-9.

2. Krug U, Berdel WE, Gale RP, Haferlach C, Schnittger S, Müller-Tidow C et al. Increasing intensity of therapies assigned at diagnosis does not improve survival of adults with acute myeloid leukemia. Leukemia. 2016 Jun;30(6):1230-6.

3. Braess J, Spiekermann K, Staib P, Gruneisen A, Wormann B, Ludwig WD, et al. Dose-dense induction with sequential high-dose cytarabine and mitoxantone (S-HAM) and pegfilgrastim results in a high efficacy and a short duration of critical neutropenia in de novo acute myeloid leukemia: a pilot study of the AMLCG. Blood. 2009 Apr 23;113(17):3903-10.

4. Braess J, Amler S, Kreuzer KA, Spiekermann K, Lindemann HW, Lengfelder E, et al. Sequential high-dose cytarabine and mitoxantrone (S-HAM) versus standard double induction in acute myeloid leukemia-a phase 3 study. Leukemia. 2018 Dec;32(12):2558-71.

5. Grimwade D, Hills RK, Moorman AV, Walker H, Chatters S, Goldstone AH, et al. Refinement of cytogenetic classification in acute myeloid leukemia: determination of prognostic significance of rare recurring chromosomal abnormalities among 5876 younger adult patients treated in the United Kingdom Medical Research Council trials. Blood. 2010 Jul 22;116(3):354-65.

6. Metzeler KH, Herold T, Rothenberg-Thurley M, Amler S, Sauerland MC, Gorlich D, et al. Spectrum and prognostic relevance of driver gene mutations in acute myeloid leukemia. Blood. 2016 Aug 4;128(5):686-98.

# Supplementary Figures

**Figure S1: Flow chart of AMKL patient selection**

Diagram

Description automatically generatedWe screened 53 patients with possible AMKL from the AMLCG trails, the AMLCG registry and 8 patients from our local center. After review of clinical documentation, laboratory results and pathological reports we excluded 11 patients. 6 patients had a history of CML with megakaryocytic features, 4 patients had MDS and 1 patient had an AML without megakaryoblastic differentiation. Bone marrow biopsies for histological review were available in 25 patients. 3 patients without megakaryoblastic differentiation of myeloid blasts and one patient with a mastocytosis (MC) were excluded. The final cohort encompassed 38 patients.

**Figure S2: Patient-based overview of results from targeted sequencing and corresponding cytogenetic abnormalities.**

**A screenshot of a computer

Description automatically generated with low confidence**

Data of targeted amplicon sequencing of 26 AMKL patients and corresponding cytogenetic data are depicted (grey: no mutation/aberration identified; blue: mutation respectively chromosomal abnormality identified; white: no data available). Adverse and intermediate karyotype are defined as by Grimwade et al (5). Each column represents one patient.

**Figure S3: Overview of ELN classification for 34 AMKL patients with available cytogenetics data, next generation sequencing (NGS) data or both.**

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Additional: compared to number within same ELN group in n=22 patients with cytogenetics and NGS data available

**Figure S4: Overall survival according to AMKL origin**

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Overall survival of 36 intensively treated patients with AMKL according to origin of AML (blue: de novo AMKL; red: secondary/therapy-related-AMKL (s-/t-AMKL)). 2 patients treated with low-dose cytarabine only are not shown. Of note, 5 patients remain alive after day 1000.

HR: Hazard ratio, CI: confidence interval.

**Figure S5: Landmark analysis of survival in intensively treated patients comparing allo-HSCT with conventional intensive chemotherapy**

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(A) \*Overall survival (OS) was calculated from start of treatment to death or last follow up. Patients with OS times <120 days (=median time from start of treatment to transplantation) were excluded from this analysis, except for censored patients. Patients treated with low-dose cytarabine only were not included. Of note, 5 patients treated with allo-HSCT remained alive after day 1000.

(B) \*\*In patients receiving an allo-HSCT, post transplant survival was calculated from date of allo-HSCT to death or last follow up. In patients receiving chemotherapy only, an analogue of post transplant survival was calculated from start of treatment to death or last follow up minus 120 days (the median expected time from start of treatment to transplant in transplanted patients). Patients with observation times <120 days (=median time from start of treatment to transplantation) were excluded from this analysis.

**Figure S6: Estimated cumulative incidence of relapse and death without relapse post allo-HSCT**

Chart, histogram

Description automatically generated

Estimated cumulative incidence curves with relapse and death without relapse as competing events. Cumulative incidence of relapse was 62% (95% CI 32.8-90.6) and cumulative incidence of death was 15% (95% CI NA-34.9).

**Figure S7: cytogenetic aberrations on chromosome 1q**

Chart, bar chart

Description automatically generated

Ideogram of chromosome 1 showing structural gains (rectangular) and breakpoints (triangles) identified in AMKL patients on the long arm q. Whole arm gains were found in 4 patients and duplications of 1q11-32 and 1q21-42 in one patient each. 1q21 was the break-point in 2 translocations and one insertion. 1q23 was the breakpoint of one translocation. Ideogram was generated using NCBI Genome Decoration Page (https://www.ncbi.nlm.nih.gov/genome/tools/gdp).

# Supplementary Tables

**Table S1: Overview of analyzed target regions**

|  |  |  |  |
| --- | --- | --- | --- |
| **Target name** | **Coordinates** | **Target name** | **Coordinates** |
| ASXL1\_EX14 | 20:31022226-31024720 | RAD21\_EX5 | 8:117870592-117870697 |
| BCOR\_EX10 | X:39922000-39922324 | RAD21\_EX6 | 8:117869507-117869712 |
| BCOR\_EX11 | X:39921393-39921646 | RAD21\_EX7 | 8:117868886-117869010 |
| BCOR\_EX12 | X:39916409-39916574 | RAD21\_EX8 | 8:117868406-117868527 |
| BCOR\_EX13 | X:39914622-39914766 | RAD21\_EX9 | 8:117866485-117866707 |
| BCOR\_EX14 | X:39913510-39913586 | RUNX1\_EX4B | 21:36259141-36259393 |
| BCOR\_EX15 | X:39913140-39913295 | RUNX1\_EX5 | 21:36252855-36253010 |
| BCOR\_EX16 | X:39911363-39911653 | RUNX1\_EX6 | 21:36231772-36231875 |
| BCOR\_EX3 | X:39937098-39937182 | RUNX1\_EX7 | 21:36206708-36206898 |
| BCOR\_EX4 | X:39935708-39935785 | RUNX1\_EX8 | 21:36171599-36171759 |
| BCOR\_EX5 | X:39931603-39934433 | RUNX1\_EX9 | 21:36164433-36164907 |
| BCOR\_EX6 | X:39930891-39930943 | SETBP1 | 18:42531710-42532150 |
| BCOR\_EX7 | X:39930227-39930412 | SF1\_EX10 | 11:64535044-64535316 |
| BCOR\_EX8 | X:39923590-39923852 | SF1\_EX11A | 11:64534665-64534723 |
| BCOR\_EX9 | X:39922862-39923205 | SF1\_EX12 | 11:64534373-64534551 |
| BCORL1\_EX3 | X:129146916-129150199 | SF1\_EX13 | 11:64533291-64533627 |
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| BRAF\_EX12 | 7:140481377-140481493 | SF1\_EX1A | 11:64545835-64545864 |
| BRAF\_EX15 | 7:140453076-140453193 | SF1\_EX2 | 11:64543888-64544098 |
| CALR\_EX9 | 19:13054517-13054737 | SF1\_EX3B | 11:64540903-64540977 |
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| CBL\_EX8 | 11:119148877-119149007 | SF1\_EX5 | 11:64537437-64537525 |
| CBL\_EX9 | 11:119149221-119149423 | SF1\_EX6 | 11:64536899-64537081 |
| CEBPA | 19:33792235-33793330 | SF1\_EX7 | 11:64536696-64536810 |
| CSF3R\_EX14 | 1:36933416-36933573 | SF1\_EX8 | 11:64536495-64536601 |
| CSF3R\_EX15 | 1:36933152-36933262 | SF1\_EX9 | 11:64535579-64535758 |
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| KIT\_EX8 | 4:55589741-55589874 | TP53\_EX11 | 17:7572928-7573008 |
| KIT\_EX9B | 4:55592014-55592226 | TP53\_EX2 | 17:7579840-7579912 |
| KRAS\_EX2 | 12:25398199-25398328 | TP53\_EX3 | 17:7579701-7579721 |
| KRAS\_EX3 | 12:25380159-25380356 | TP53\_EX4 | 17:7579313-7579590 |
| KRAS\_EX4 | 12:25378527-25378733 | TP53\_EX5 | 17:7578372-7578554 |
| MPL\_EX10 | 1:43814924-43815040 | TP53\_EX6 | 17:7578178-7578289 |
| MPL\_EX11 | 1:43817877-43817984 | TP53\_EX7 | 17:7577500-7577608 |
| MPL\_EX12 | 1:43818179-43818453 | TP53\_EX8 | 17:7577020-7577155 |
| MPL\_EX9 | 1:43814504-43814683 | TP53\_EX9 | 17:7576854-7576926 |
| NOTCH1\_EX26 | 9:139399116-139399490 | U2AF1\_EX2 | 21:44524426-44524512 |
| NOTCH1\_EX27 | 9:139397625-139397792 | U2AF1\_EX7 | 21:44514766-44514898 |
| NOTCH1\_EX28 | 9:139396715-139396950 | U2AF2\_EX1 | 19:56166462-56166529 |
| NOTCH1\_EX34 | 9:139390514-139391458 | U2AF2\_EX10 | 19:56180440-56180557 |
| NPM1\_EX11 | 5:170834695-170834788 | U2AF2\_EX11 | 19:56180801-56181068 |
| NPM1\_EX12 | 5:170837522-170837579 | U2AF2\_EX12 | 19:56185291-56185444 |
| NRAS\_EX\_2 | 1:115258662-115258791 | U2AF2\_EX2 | 19:56170567-56170721 |
| NRAS\_EX\_3 | 1:115256412-115256609 | U2AF2\_EX3 | 19:56171534-56171597 |
| NRAS\_EX4 | 1:115252173-115252373 | U2AF2\_EX4 | 19:56171873-56171995 |
| PHF6\_EX10B | X:133559232-133559360 | U2AF2\_EX5 | 19:56172395-56172565 |
| PHF6\_EX2 | X:133511649-133511785 | U2AF2\_EX6 | 19:56173859-56173994 |
| PHF6\_EX3 | X:133512036-133512136 | U2AF2\_EX7 | 19:56174963-56175120 |
| PHF6\_EX4 | X:133527532-133527664 | U2AF2\_EX8 | 19:56179864-56179962 |
| PHF6\_EX5 | X:133527940-133527982 | U2AF2\_EX9 | 19:56180027-56180168 |
| PHF6\_EX6B | X:133547519-133547687 | WT1\_EX1 | 11:32456243-32456600 |
| PHF6\_EX7 | X:133547854-133547996 | WT1\_EX10B | 11:32413519-32413610 |
| PHF6\_EX8A | X:133549047-133549252 | WT1\_EX11 | 11:32410603-32410729 |
| PHF6\_EX9 | X:133551200-133551337 | WT1\_EX3 | 11:32450044-32450165 |
| PTEN\_EX1 | 10:89624228-89624305 | WT1\_EX4 | 11:32449503-32449604 |
| PTEN\_EX2 | 10:89653783-89653866 | WT1\_EX5 | 11:32439124-32439200 |
| PTEN\_EX3 | 10:89685271-89685314 | WT1\_EX6 | 11:32438033-32438090 |
| PTEN\_EX4 | 10:89690804-89690846 | WT1\_EX7 | 11:32421495-32421590 |
| PTEN\_EX5 | 10:89692771-89693008 | WT1\_EX8 | 11:32417804-32417953 |
| PTEN\_EX6 | 10:89711876-89712016 | WT1\_EX9 | 11:32414213-32414301 |
| PTEN\_EX7 | 10:89717611-89717776 | ZRSR2\_EX1 | X:15808620-15808659 |
| PTEN\_EX8 | 10:89720652-89720875 | ZRSR2\_EX10 | X:15838331-15838439 |
| PTPN11\_EX13 | 12:112926829-112926979 | ZRSR2\_EX11 | X:15840855-15841365 |
| PTPN11\_EX3 | 12:112888123-112888316 | ZRSR2\_EX2 | X:15809058-15809136 |
| RAD21\_EX10 | 8:117864789-117864947 | ZRSR2\_EX3 | X:15817996-15818076 |
| RAD21\_EX11 | 8:117864188-117864335 | ZRSR2\_EX4 | X:15821812-15821919 |
| RAD21\_EX12 | 8:117862858-117863006 | ZRSR2\_EX5 | X:15822235-15822320 |
| RAD21\_EX13 | 8:117861186-117861268 | ZRSR2\_EX6 | X:15826357-15826394 |
| RAD21\_EX14 | 8:117859740-117859930 | ZRSR2\_EX7 | X:15827324-15827441 |
| RAD21\_EX2 | 8:117878826-117878968 | ZRSR2\_EX8 | X:15833801-15834013 |
| RAD21\_EX3 | 8:117875370-117875498 | ZRSR2\_EX9 | X:15836711-15836765 |
| RAD21\_EX4 | 8:117874081-117874179 |  |  |

**Table S2: Karyotypes of 38 AMKL**

|  |  |
| --- | --- |
| UPN | Karyotype |
| 1 | 47,XY,+i(1)(q10),dup(3)(q21q26),der(19)t(3;19)(q25;p13)[9]/  47,XY,+i(q10)(dup1)(q11q32),dup(3)(q21q26),der(19)t(3;19)(q25;p13)[6] |
| 2 | 46,XX,inv(3)(q21q26)[17]/45,XX,inv(3)(q21q26),-7[3] |
| 3 | 46,XX,del(7)(q22)[20] |
| 4 | 45,XY,der(7)del(7)(p15)del(7)(q11)[5]/45,XY,-7[8] |
| 5 | 45,XY,der(1)t(1;3)(q23;?),der(3)t(3;5)(p11;?)t(1;5)(?;?)t(1;16)(?;q15),  der(4)t(4;11)(q21;?),del(5)(q13q34),der(6)t(6;11)(p25;?)t(6;11)(q11;?),  del(11)(q14q23),der(11)t(2;11)(?;p15),der(13)i(13)(q10)t(4;13)(q25;q34)  t(1;13)(?;q34),der(15)t(6;15)(q11;q15),-17[13]/46,XY[3] |
| 6 | 45,XY,der(2)t(2;15)(q37;q11),i(5)(p10),inv(7)(p11q31),  der(11)t(11;17)(q23;q21),-15,-17,+mar[18]/46,XY[2] |
| 7 | 45,XX,t(3;8)(q26,q24),-7[20] |
| 8 | 45,XX,inv(3)(q21q26),-7[20] |
| 9 | 46,XY[23] |
| 10 | 46,XY,der(11)t(1;11)(q21;q25)[12]/46,XY,der(11)t(1;11)(q21;q25)dup(1)(q21q42)[8] |
| 11 | 46,XY[15] |
| 12 | *no cells in division* |
| 13 | *missing* |
| 14 | 46,XY,+1,der(1;7)(q10;p10)[20] |
| 15 | 46,XY |
| 16 | *missing* |
| 17 | 44~45,XX,-4,-5,+6,der(7)t(5;7)(q22;?),+der(8)t(8;14)(q22;q?),-14, der(14)t(14;18)(p11;q11)t(5;14)(?;q32)t(4;5)(?;?),der(17)t(5;17)(?;p11)t(5;8)(?;?),-18,  der(20)t(20;22)(q13;q?),der(21)amp(21)(q22),der(22)t(4;22)(?;q13)[cp6]/46,XX[4] |
| 18 | 46,XY |
| 19 | 46,XX,+1,der(1;7)(q10;p10)[13]/47,XX,+1,der(1;7)(q10;p10),+21[7] |
| 20 | 46,XY |
| 21 | 46,XX |
| 22 | 46,XY,+1,der(1;7)(q10;p10)[4]/46,XY[8] |
| 23 | *missing* |
| 24 | 46,XY[20] |
| 25 | *no cells in division* |
| 26 | 46,XX,+16,der(7;16)(q10;p10)[6]/46,XX[17] |
| 27 | 48,XY,+8,+10,der(17)t(1;17)(?;p1?2)[3]/38~48,XY,sl,-X,-3,-4,+4,der(4)(4pter  ->4q21::7q11->7q22::10q11->10qter),der(4)t(4;6)(q3?;?p23)x2, der(6)t(6;7)(q13;?q22),-7,-7,der(7;10)(p10;p10),der(9)t(4;9)(?;p2?),-12,  -13,der(13)(q?),-17,der(17)(1?ter->1?::17p13->17q2?::6?->6?ter),-20,-20,-21,-22[cp4]/46,XY[6] |
| 28 | *no cells in division* |
| 29 | 46,Y,der(X)t(X;8)(q1?;p2?1),ins(1;7)(q21;?),t(3;21)(q26;q22),t(4;6)(q?21;p21),  del(7)(q11),r(7)(?),der(8)(Xpter->Xp2?::8p2?1->8q21::X?->X?::8q2?3->8qter)[10]/45,Y,der(X)t(X;8)(q1?;p2?1),t(4;6)(q?21;p21),-7,r(7)(?),der(8)(Xpter->Xp2?::8p2?1->8q21::X?->X?::8q2?3->8qter),del(20)(q11)[cp2]/46,XY[2] |
| 30 | 49,XY,der(1)(9?qter->9?::1p2?->1q2?::7?->7?pter),  der(7)t(1;7)(p2?;p1?3),+8,der(9)t(1;9)(q2?;q?13),+19,+21[18] |
| 31 | 46,XY,t(9;22)(q34;q11) |
| 32 | 43-44,XX,add(1)(p11),add(1)(q11),del(3)(q?),del(4)(p11),-5,-7,+8[9]/  add(11p),der(12)-15,add(16)(q?),-17,-20,+mar[22] |
| 33 | 47,XX,t(3;3)(q21;q26),+8[13]/46,XX[9] |
| 34 | *missing* |
| 35 | 45,XX,-7[16]/47,XY,+8[3]/46,XY[1] |
| 36 | 47,XY,t(1;8)(q21;q21),dup(17)(q21),+19[8]/48,sl,+8[3]/46,XY[3] |
| 37 | *missing* |
| 38 | 46,XY[20] |

**Table S3: Distribution of FAB types in 637 patients treated within the AMLCG99 and AMLCG2008 trials and available information for ELN 2017 risk\*, age and FAB type that were used for multivariable Cox regression.**

|  |  |  |
| --- | --- | --- |
| **FAB type** | **n** | **%** |
| 0 | 34 | 5.3 |
| 1 | 154 | 24.2 |
| 2 | 173 | 27.2 |
| 4 | 160 | 25.1 |
| 5 | 81 | 12.7 |
| 6 | 17 | 2.7 |
| 7 | 18 | 2.8 |
| total | 637 | 100% |

\*The dataset of Metzeler et al (6) was used for this analysis (patients treated within the AMLCG99 and AMLCG2008 trials in whom ELN 2017 classification was available). In addition, AML M7 patients treated within AMLCG99 and AMLCG2008 trials and included in our study were added to this cohort if they were not already present in the dataset published by Metzeler et al (6).