**Supplementary Materials**

**Pharmacokinetics**

Plasma samples were analyzed for concentrations of glasdegib at Covance Bioanalytical Services, LLC (Indianapolis, IN) using a validated, sensitive, and specific high-performance liquid chromatography–tandem mass spectrometric. Samples were stored at ~20°C or colder for a maximum of 372 days until analysis. Calibration standard responses were linear over the range of 0.200 ng/mL to 200 ng/mL for glasdegib, using a l/concentration2 weighted linear regression. The glasdegib lower limit of quantification was 0.200 ng/mL.

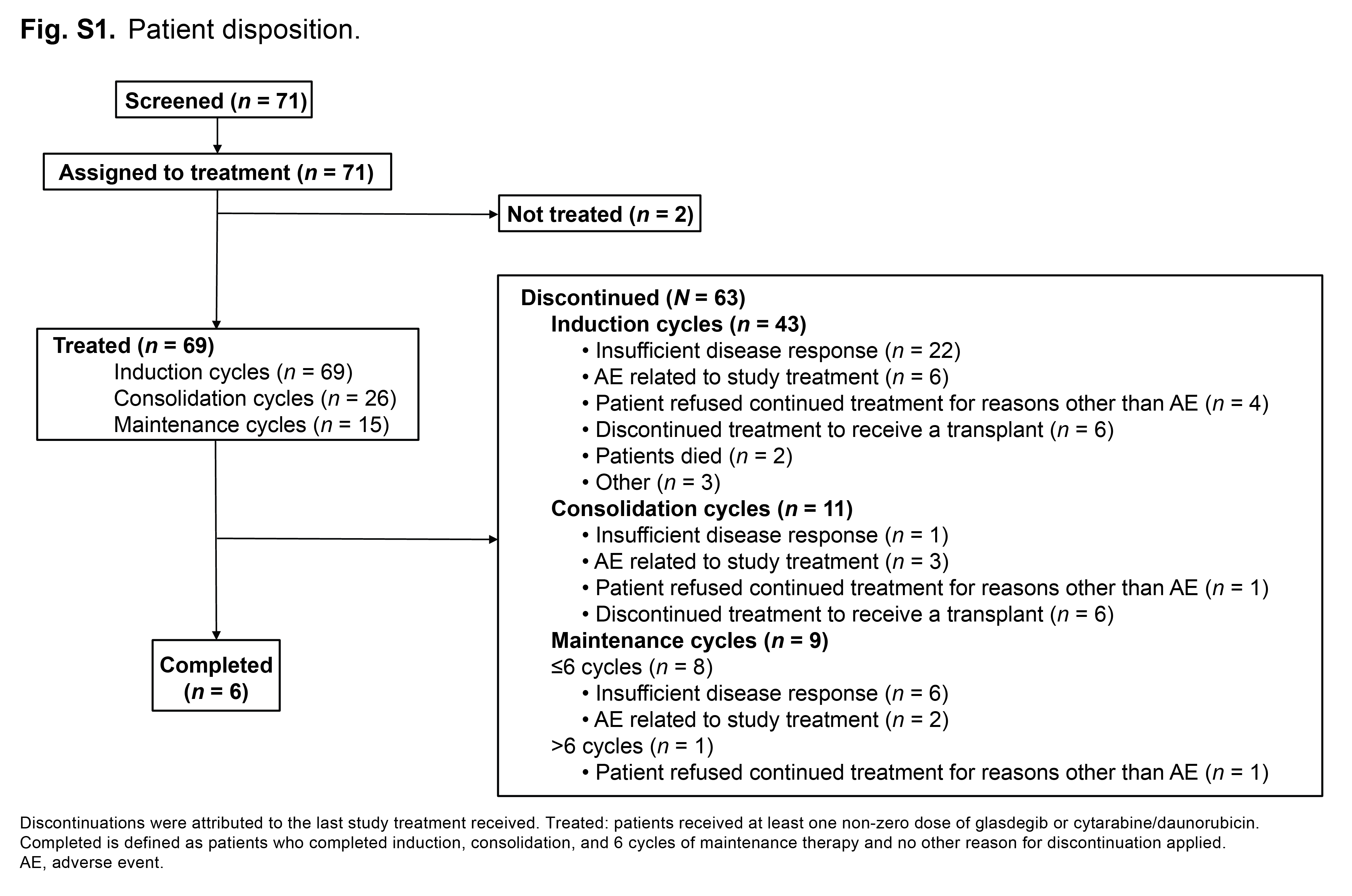
The inter-batch assay accuracy, expressed as percent relative error of the mean glasdegib quality control (QC) sample concentrations, ranged from ~8.1% to 1.3% across the low (0.600 ng/mL), medium (15.0 ng/mL), high (150 ng/mL), and diluted (150 and 1000 ng/mL) QC samples. Inter-batch assay precision, expressed as percent coefficient of variation of the glasdegib QC sample concentrations, was ≤8.5% across the low, medium, high, and diluted QC samples.

Pharmacokinetics parameters were calculated using noncompartmental analysis of plasma concentration–time data, including: minimum and maximum observed plasma concentration (Cmin and Cmax); Time to first occurrence of Cmax, terminal plasma half-life, area under the plasma concentration–time profile from time 0 to infinity; and predose plasma concentration.

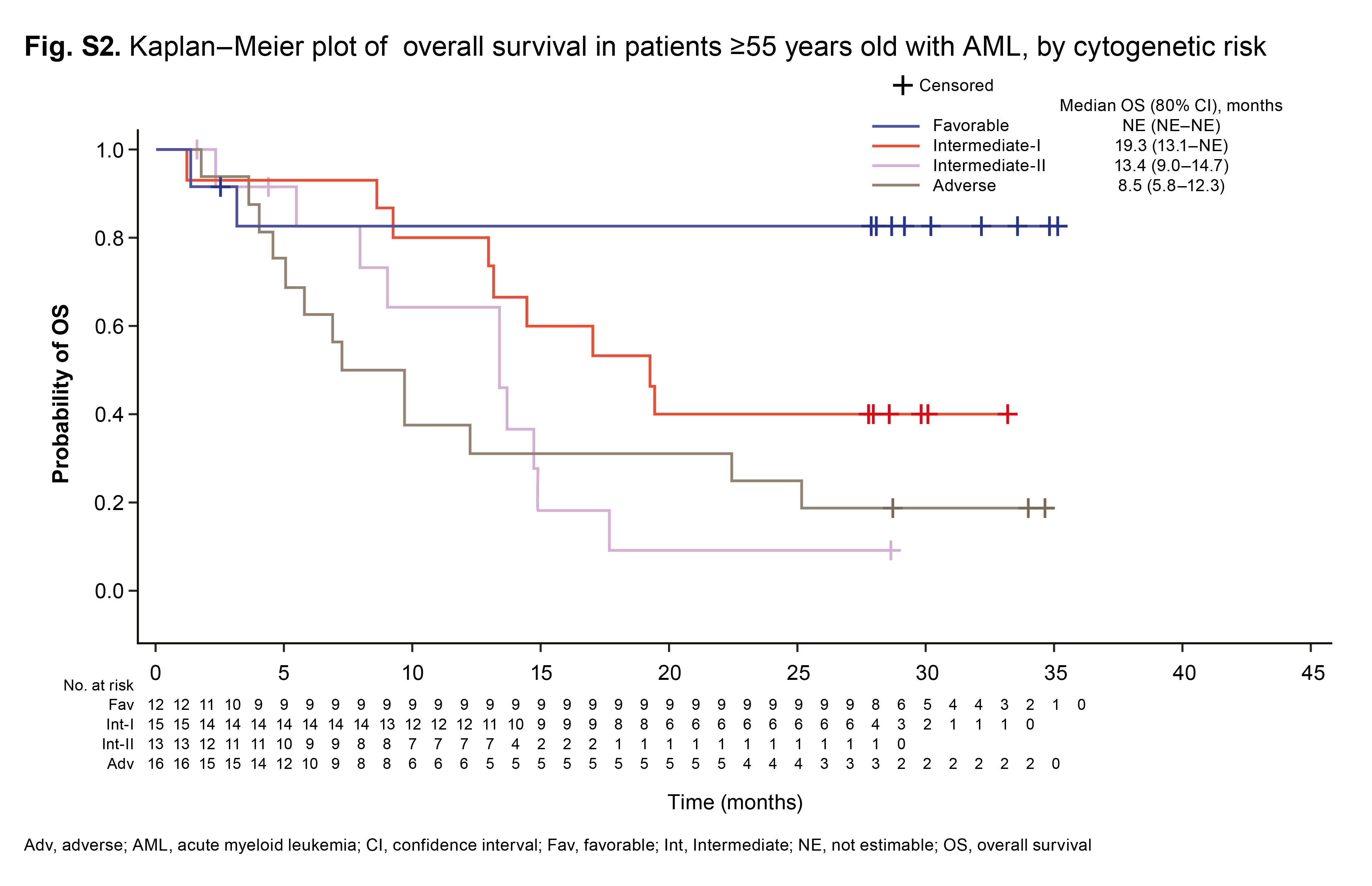
**Mutational Analyses**

DNA samples extracted from frozen peripheral blood or bone marrow aspirates were analyzed using next-generation DNA sequencing validated to Good Clinical Practice guidelines of a panel of 12 genes performed using the Illumina® MiSeq instrument (San Diego, CA). In a secondary assay, an amplicon-based approach was used to further characterize the *FLT3* gene for the presence of internal tandem duplication mutations.

**Supplementary Figure S1.** Patient disposition



**Supplementary Figure S2.** Kaplan–Meier plot of overall survival in patients ≥55 years old with AML, by cytogenetic risk



**Supplementary Table 1. Patient demographics and baseline characteristics, by age group**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Total**  **N = 71** | **≥55 years**  **n = 61** | **<55 years**  **n = 10** | |
| Sex, n (%) |  |  |  | | |
| Female | 28 (39.4) | 22 (36.1) | 6 (60.0) | | |
| Male | 43 (60.6) | 39 (63.9) | 4 (40.0) | | |
| Age, years |  |  |  | | |
| Mean (SD) | 61.9 (9.6) | 65.0 (5.0) | 42.8 (8.8) | | |
| Median (range) | 64.0 (27–75) | 65.0 (55–75) | 44.0 (27–54) | | |
| Race, n (%) |  |  |  | | |
| White | 60 (84.5) | 51 (83.6) | 9 (90.0) | | |
| Black | 7 (9.9) | 7 (11.5) | 0 | | |
| Asian | 2 (2.8) | 2 (3.3) | 0 | | |
| Other | 2 (2.8) | 1 (1.6) | 1 (10.0) | | |
| ECOG performance status, n (%) |  |  |  | | |
| 0 | 25 (35.2) | 22 (36.1) | 3 (30.0) | | |
| 1 | 44 (62.0) | 38 (62.3) | 6 (60.0) | | |
| 2 | 2 (2.8) | 1 (1.6) | 1 (10.0) | | |
| Cytogenetic risk, n (%) |  |  |  | | |
| Good/intermediate | 50 (70.4) | 42 (68.9) | 8 (80.0) | | |
| Poor | 19 (26.8) | 17 (27.9) | 2 (20.0) | | |
| Not evaluated | 2 (2.8) | 2 (3.3) | 0 | | |
| Prognostic risk factors for AML\*, n (%) | n = 66 | n = 59 | n = 7 | | |
| Favorable | 13 (19.7) | 12 (20.3) | 1 (14.3) | | |
| Intermediate-I | 21 (31.8) | 16 (27.1) | 5 (71.4) | | |
| Intermediate-II | 13 (19.7) | 13 (22.0) | 0 | | |
| Adverse | 17 (25.8) | 16 (27.1) | 1 (14.3) | | |
| Not evaluated | 2 (3.0) | 2 (3.4) | 0 | | |
| Prognostic factors for MDS†, n (%) | n = 5 | n = 2 | n = 3 | | |
| Good risk | 1 (20.0) | 0 | 1 (33.3) | | |
| Intermediate risk | 2 (40.0) | 1 (50.0) | 1 (33.3) | | |
| Poor risk | 2 (40.0) | 1 (50.0) | 1 (33.3) | | |
| MDS IPSS score, n (%) | n = 5 | n = 2 | n = 3 | | |
| 1.5-2 (Intermediate-2) | 2 (40.0) | 1 (50.0) | 1 (33.3) | | |
| ≥2.5 (High) | 3 (60.0) | 1 (50.0) | 2 (66.7) | | |
| Patients with AML | n = 66 | n = 59 | n = 7 | | |
| Duration since histopathologic diagnosis, months |  |  |  | | |
| Mean | 0.3 | 0.3 | 0.3 | | |
| Median | 0.2 | 0.2 | 0.2 | | |
| Range | 0.00–2.53 | 0.00–2.53 | 0.13–0.62 | | |
| Disease history, n |  |  |  | | |
| *De novo* | 47 | 40 | 7 | | |
| Secondary disease | 19 | 19 | 0 | | |
| Patients with MDS | n = 5 | n = 2 | n = 3 | | |
| Duration since histopathologic diagnosis, months |  |  |  | | |
| Mean | 3.3 | 7.5 | 0.6 | | |
| Median | 0.6 | 7.5 | 0.6 | | |
| Range | 0.03–14.95 | 0.03–14.95 | 0.26–0.89 | | |
| Disease history, n |  |  |  | | |
| *De novo* | 5 | 2 | 3 | | |
| \* For AML: good/intermediate cytogenetic risk = favorable, intermediate-I and intermediate-II risk groups; poor cytogenetic risk = adverse risk group.  † For MDS, good/intermediate cytogenetic risk = good and intermediate risk groups; poor cytogenetic risk = poor risk group.  N = number of patients evaluable for the parameter; n = number of patients in the category; SD = standard deviation; ECOG = Eastern Cooperative Oncology Group; AML = acute myeloid leukemia; MDS = myelodysplastic syndromes; IPSS = International Prognostic Scoring System. | | | |

**Supplementary Table 2.** Follow-up systemic therapies in all patients who received study treatment (N = 69)

|  |  |
| --- | --- |
| **Follow-up systemic treatment** | **n (%)** |
| n | 50 (72.5) |
| Transplant | 8 (11.6) |
| Chemotherapy | 39 (56.5) |
| Biologic | 0 (0) |
| TKI | 1 (1.4) |
| Investigational | 2 (2.9) |

Each subject is counted only once and placed into one category using the hierarchy shown above.

TKI = tyrosine kinase inhibitor.

**Supplementary Table S3.** Best overall response (investigator-assessed) for patients with AML (n = 64)

|  |  |  |  |
| --- | --- | --- | --- |
|  | **n** | **%** | **80% Exact CI\*** |
| Objective response |  |  |  |
| Disease status |  |  |  |
| Morphologic CR | 30 | 46.9 | 38.3–55.6 |
| Morphologic CRi | 5 | 7.8 | 3.8–14.0 |
| MLFS | 4 | 6.3 | 2.8–12.1 |
| PR | 1 | 1.6 | 0.2–5.9 |
| PRi | 1 | 1.6 | 0.2–5.9 |
| Minor response | 7 | 10.9 | 6.2–17.7 |
| Stable disease | 2 | 3.1 | 0.8–8.1 |
| Indeterminate | 0 | 0.0 | 0.0–3.5 |
| Cytogenetic response |  |  |  |
| Cytogenetic CR | 23 | 35.9 | 27.9–44.7 |
| Molecular response |  |  |  |
| Molecular CR | 24 | 37.5 | 29.4–46.2 |
| Objective disease progression |  |  |  |
| Disease status |  |  |  |
| Treatment failure | 13 | 20.3 | 13.9–28.2 |
| Resistant disease | 13 | 20.3 | 13.9–28.2 |
| Not evaluable | 1 | 1.6 | 0.2–5.9 |
| Further endpoints of interest† |  |  |  |
| CR/CRi | 35 | 54.7 | 46.7–62.7 |
| Disease-modifying response = CR, CRi, MLFS, PR | 40 | 62.5 | 54.7–70.3 |
| Clinically beneficial response = CR, CRi, MLFS, PR, PRi | 41 | 64.1 | 56.4–71.7 |

\* Using exact method based on binomial distribution.

† The CI for further endpoints is 80% CI using normal approximation.

AML = acute myeloid leukemia; CI = confidence interval; CR = complete remission; CRi = complete response with incomplete blood count recovery; MLFS = morphologic leukemia-free state; PR = partial remission; PRi = partial response with incomplete blood count recovery.

**Supplementary Table S4.** Best overall response (investigator-assessed) for patients with MDS (n = 5)

|  |  |  |  |
| --- | --- | --- | --- |
|  | **n** | **%** | **80% Exact CI\*** |
| Objective response |  |  |  |
| Disease status |  |  |  |
| CR | 2 | 40.0 | 11.2–75.3 |
| PR | 0 | 0.0 | 0.0–36.9 |
| mCR | 1 | 20.0 | 2.1–58.4 |
| Stable disease | 1 | 20.0 | 2.1–58.4 |
| Indeterminate | 0 | 0.0 | 0.0–-36.9 |
| Cytogenetic response |  |  |  |
| Complete cytogenetic response | 3 | 60.0 | 24.7–88.8 |
| Partial cytogenetic response | 0 | 0.0 | 0.0–36.9 |
| Objective disease progression |  |  |  |
| Disease status |  |  |  |
| Disease progression | 1 | 20.0 | 2.1–58.4 |
| Failure | 0 | 0.0 | 0.0–36.9 |
| Not evaluable | 0 | 0.0 | 0.0–36.9 |
| Further endpoints of interest† |  |  |  |
| CR/CRi | 2 | 40.0 | 11.9–68.1 |
| Disease-modifying response = CR, mCR, PR | 3 | 60.0 | 31.9–88.1 |
| Clinically beneficial response = CR, mCR, PR, SD | 4 | 80.0 | 57.1–100.0 |

\* Using exact method based on binomial distribution.

­† The CI for further endpoints is 80% CI using normal approximation.

MDS = myelodysplastic syndromes; CI = confidence interval; CR = complete remission; PR = partial remission; mCR = marrow complete response; CRi = complete response with incomplete blood count recovery; SD = stable disease.

**Supplementary Table S5.** Overall survival by diagnosis and cytogenetic risk

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Acute myeloid leukemia** | | | | | | | | |
|  | **Total** | **AML Cytogenetic Risk** | | | | | | |
|  | **Favorable** | **Intermediate I** | | **Intermediate II** | **Adverse** | | **Not evaluated** |
| n | 64 | 13 | 19 | | 13 | 17 | | 2 |
| Deaths\*, n (%) | 37 (57.8) | 2 (15.4) | 10 (52.6) | | 10 (76.9) | 13 (76.5) | | 2 (100.0) |
| Disease under study | 30 (46.9) | 1 (7.7) | 8 (42.1) | | 10 (76.9) | 10 (58.8) | | 1 (50.0) |
| Unknown | 4 (6.3) | 0 | 2 (10.5) | | 0 | 1 (5.9) | | 1 (50.0) |
| Other | 11 (17.2) | 1 (7.7) | 3 (15.8) | | 2 (15.4) | 4 (23.5) | | 1 (50.0) |
| Number censored, n (%) | 27 (42.2) | 11 (84.6) | 9 (47.4) | | 3 (23.1) | 4 (23.5) | | 0 |
| Probability of survival at month 12 (80% CI)† | 65.5 (57.0–72.7) | 83.9 (64.7–93.2) | 78.9 (63.8–88.3) | | 64.2 (42.8–79.3) | 38.0 (22.8–53.1) | | 50.0 (7.7–82.9) |
| Median OS (80% CI)‡, mo | 16.3 (13.4–19.4) | NE (NE–NE) | 19.4 (14.5–NE) | | 13.4 (9.0–14.7) | 9.7 (5.8–12.3) | | 8.4 (0.5–16.3) |
| **Myelodysplastic syndromes** | | | | | | | | |
|  | **Total** | **MDS Cytogenetic Risk** | | | | | | |
|  | **Good** | | **Intermediate** | | | **Poor** | |
| n | 5 | 1 | | 2 | | | 2 | |
| Deaths\*, n (%) | 4 (80.0) | 1 (100.0) | | 1 (50.0) | | | 2 (100.0) | |
| Disease under study | 4 (80.0) | 1 (100.0) | | 1 (50.0) | | | 2 (100.0) | |
| Unknown | 0 | 0 | | 0 | | | 0 | |
| Other | 0 | 0 | | 0 | | | 0 | |
| Number censored, n (%) | 1 (20.0) | 0 | | 1 (50.0) | | | 0 | |
| Probability of survival at month 12 (80% CI)† | 80.0 (44.7–94.0) | 100.0 (100.0–100.0) | | 100.0 (100.0–100.0) | | | 50.0 (7.7–82.9) | |
| Median OS (80% CI)‡, mo | 13.0 (11.0–15.6) | 12.1 (NE–NE) | | NE (13.0–NE) | | | 13.3 (11.0–15.6) | |

\* Patients may have multiple reasons for cause of death.  
† Calculated from the product-limit method.

‡ Based on the Brookmeyer and Crowley Method.

AML = acute myeloid leukemia; CI = confidence interval; MDS = myelodysplastic syndromes; NE = not estimable.

**Supplementary Table S6.** Baseline gene mutations in responding and nonresponding patients (50 evaluable patients)

| Gene name | Mutation Status | Responding  n (%) | Non-Responding  n (%) | *P* |
| --- | --- | --- | --- | --- |
| *CEBPA* | mutated | 6 (12.0) | 3 (6.0) | 1.0000 |
|  | non-mutated | 26 (52.0) | 15 (30.0) |  |
| *DNMT3A* | mutated | 12 (24.0) | 6 (12.0) | 1.0000 |
|  | non-mutated | 20 (40.0) | 12 (24.0) |  |
| *FLT3* | mutated | 3 (6.0) | 2 (4.0) | 1.0000 |
|  | non-mutated | 29 (58.0) | 16 (32.0) |  |
| *FLT3-ITD\** | mutated | 2 (4.0) | 1 (2.0) | 1.0000 |
|  | non-mutated | 30 (60.0) | 17 (34.0) |  |
| *IDH1* | mutated | 2 (4.0) | 1 (2.0) | 1.0000 |
|  | non-mutated | 30 (60.0) | 17 (34.0) |  |
| *IDH2* | mutated | 5 (10.0) | 4 (8.0) | 0.7049 |
|  | non-mutated | 27 (54.0) | 14 (28.0) |  |
| *KIT* | mutated | 2 (4.0) | 1 (2.0) | 1.0000 |
|  | non-mutated | 30 (60.0) | 17 (34.0) |  |
| *KRAS* | mutated | 0 | 1 (2.0) | 0.3600 |
|  | non-mutated | 32 (64.0) | 17 (34.0) |  |
| *NPM1* | mutated | 12 (24.0) | 3 (6.0) | 0.1990 |
|  | non-mutated | 20 (40.0) | 15 (30.0) |  |
| *NRAS* | mutated | 5 (10.0) | 1 (2.0) | 0.3991 |
|  | non-mutated | 27 (54.0) | 17 (34.0) |  |
| *RUNX1* | mutated | 7 (14.0) | 7 (14.0) | 0.3251 |
|  | non-mutated | 25 (50.0) | 11 (22.0) |  |
| *TET2* | mutated | 7 (14.0) | 5 (10.0) | 0.7349 |
|  | non-mutated | 25 (50.0) | 13 (26.0) |  |
| *WT1* | mutated | 0 | 1 (2.0) | 0.3600 |
|  | non-mutated | 32 (64.0) | 17 (34.0) |  |

\* In a secondary assay, an amplicon-based approach was used to further characterize the *FLT3* gene for the presence of an internal tandem duplication mutation.

Baseline gene mutational status was identified based on the combined results from bone marrow and blood: bone marrow results used if available, blood results if bone marrow results not available, with results unknown if neither available.

For AML, investigator-reported best overall response responders: CR, CRi, MLFS, PR, or PRi.

For MDS, investigator-reported best overall response responders: CR, mCR, PR, or SD.

Statistical significance in comparison of responders with non-responders was determined using Fisher’s exact test.

AML = acute myeloid leukemia; CR = complete remission; CRi = complete response with incomplete blood count recovery; MLFS = morphologic leukemia‑free state; PR = partial remission; PRi = partial remission with incomplete blood count recovery; MDS = myelodysplastic syndromes; mCR = marrow complete response; SD = stable disease.