# *SUPPLEMENTARY APPENDIX*

## Panobinostat and decitabine prior to donor lymphocyte infusion in allogeneic stem cell transplantation recipients with poor-risk acute myeloid leukemia and high-risk MDS

**LEGENDS**

**Supplementary Figure 1.** Dose limiting toxicities decision diagram.

Schematic overview interim analyses for selection highest feasible dose level

Abbreviations: Nr, number of patients; and DLT, dose limiting toxicities; PNB mono, Panobinostat monotherapy; PNB/DAC20, Panobinostat combined with Decitabine 20 mg/m$^2$; PNB/DAC10, Panobinostat combined with Decitabine 20 mg/m$^2$.

**Supplementary Figure 2.** Overall survival, by risk classification

Kaplan-Meier estimates progression-free survival by risk classification

Abbreviations: F, number of failure; and N, number of patients.

**Supplementary Figure 3.** Cumulative incidence of relapse, by MRD status

Kaplan-Meier estimates of cumulative incidence of relapse by MRD-negative and –positive patients.

Abbreviations: F, number of failures; N, number of patients; MRD, minimal residual disease; MRD-pos, MRD-positive; and MRD-neg, MRD-negative

**Supplementary Figure 4.** Overall survival, by eligibility for alloHSCT

Kaplan-Meier estimates of overall survival (OS) by eligibility for alloHSCT, with starting point at registration.

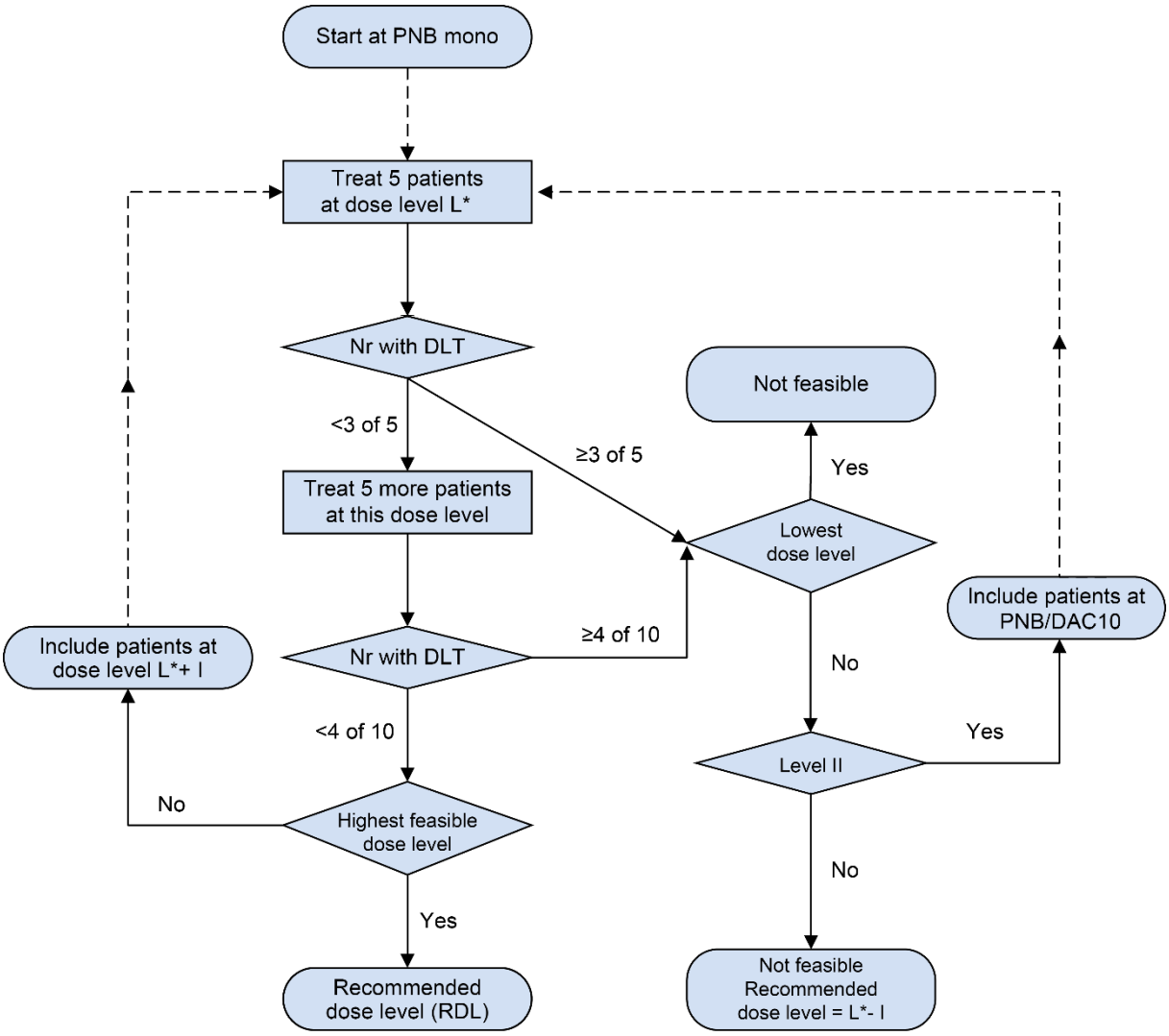
Abbreviations: F, number of failures; and N, number of patients.

**Supplementary Figure 5.** Graft versus host free/relapse free survival

Kaplan-Meier estimates of graft versus host free/relapse free survival, is defined as the time from transplantation to the date of; aGVHD grade 3 or 4, cGVHD requiring systemic therapy, relapse or death whichever comes first. For the cGVHD requiring systemic treatment the following treatments were considered as systemic: prednisone, cyclosporine, myfortic.

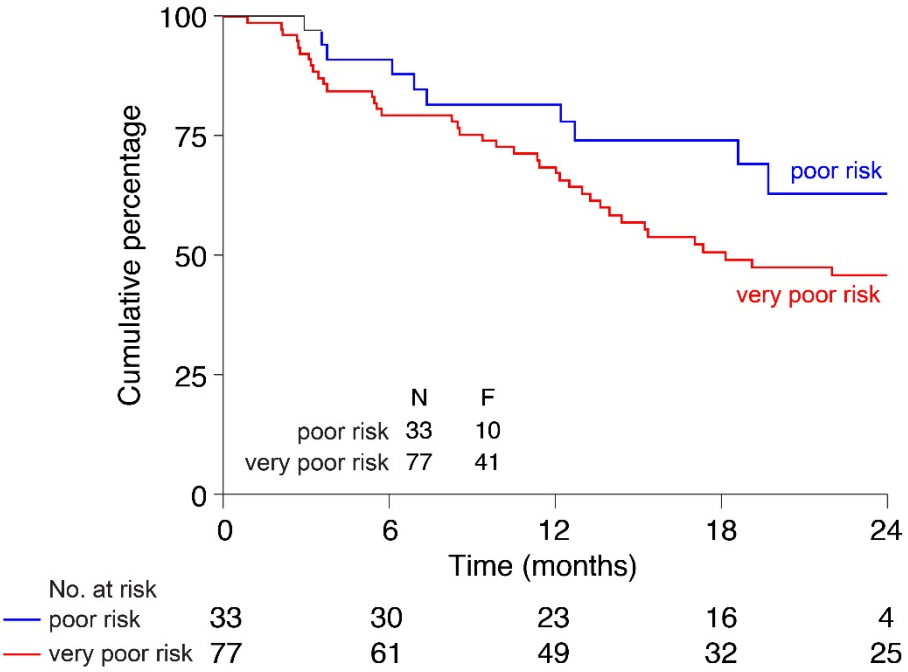
Abbreviations: F, number of failures; and N, number of patients

**Supplementary Figure 1.** Dose limiting toxicities decision diagram

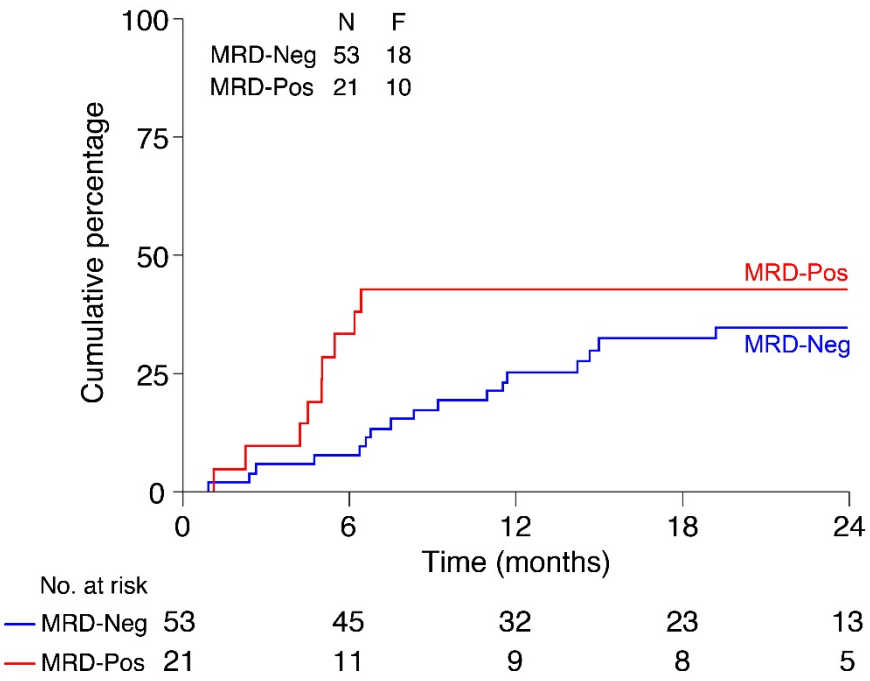



L* should be read as `I ' or `II', whichever applicable

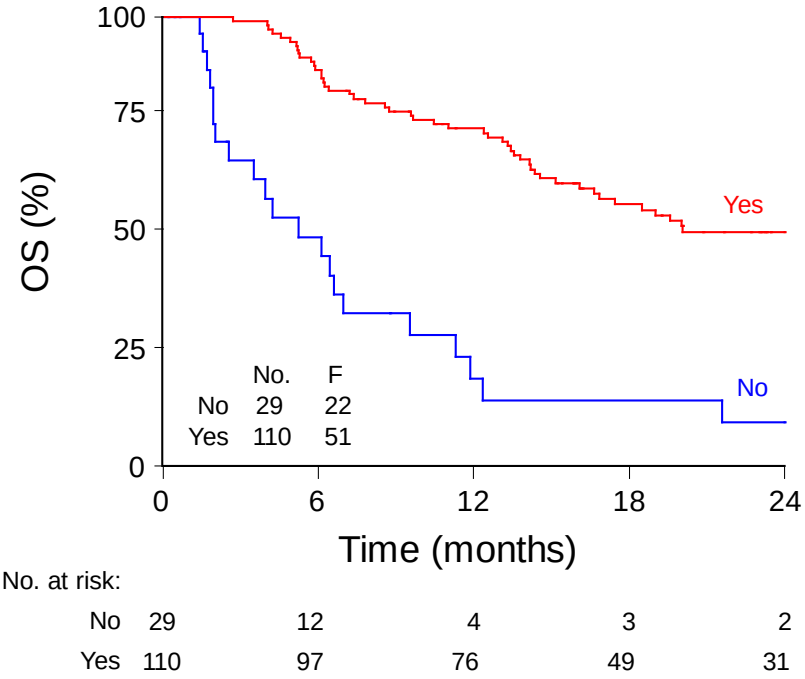
**Supplementary Figure 2.** Overall survival by risk classification



**Supplementary Figure 3.** Cumulative incidence of relapse, by MRD status



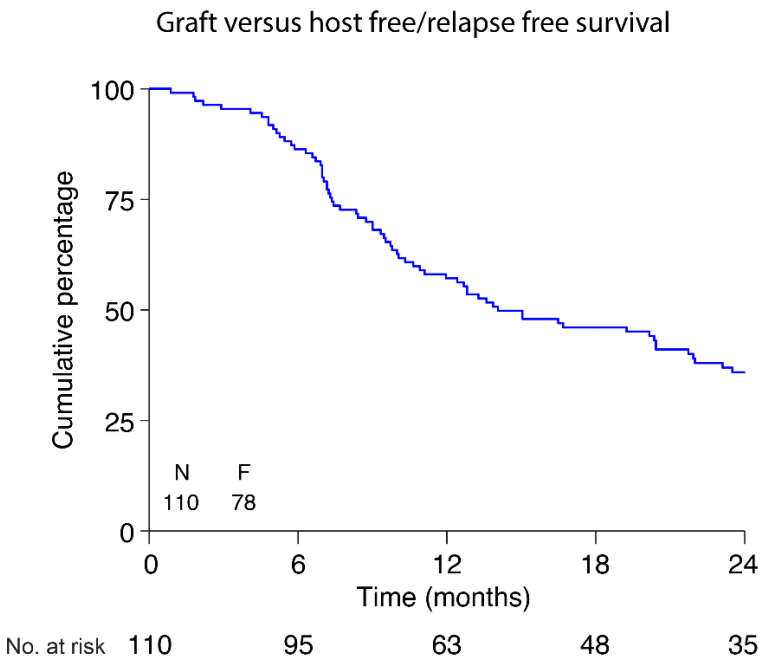
**Supplementary Figure 4** Overall survival, by eligibility for transplant




No. at risk:

| | | | | | |
|---|---|---|---|---|---|
| No | 29 | 12 | 4 | 3 | 2 |
| Yes | 110 | 97 | 76 | 49 | 31 |

**Supplementary figure 5** Graft versus host free/relapse free survival



## DETAILS OF HOVON 116 AML STUDY

### 1. HOVON-SAKK AML risk classification

| Risk | | Definition | % pts at baseline | % pts with CR & consolidation |
|---|---|---|---|---|
| Good | GR1 | t(8;21) or AML1-ETO, WBC≤20 | 5 % | 7 % |
| | GR2 | inv(16)/t(16;16) or CBFB-MYH11 gene | 6 % | 7 % |
| | GR3 | MK-, CEBPA+ | 7 % | 8 % |
| | GR4 | MK-, FLT3ITD-/NPM1+, CRe | 11 % | 13 % |
| Intermediate | IR1 | t(8;21) or AML1-ETO, WBC>20 | 2 % | 2 % |
| | IR2 | CN –X –Y, WBC≤100, CRe | 17 % | 21 % |
| Poor | PR1 | CN –X –Y, WBC≤100, not CRe | 10 % | 8 % |
| | PR2 | CN –X –Y, WBC>100 | 5 % | 4 % |
| | PR3 | CA, non CBF, MK-, no abn3q26, EVI1- | 16 % | 15 % |
| Very Poor | VPR1 | Non CBF, MK+ | 9 % | 5 % |
| | VPR2 | Non CBF, abn3q26 | 2 % | 1 % |
| | VPR3 | Non CBF, EVI1+ | 9 % | 9 % |

Abbreviations: CN –X-Y: cytogenetically normal or only loss of X or Y chromosome; CA: cytogenetically abnormal; CRe: attainment of early CR, ie after cycle I; MK-: monosomal karyotype negative; MK+: monosomal karyotype positive

The table gives the % distribution of each risk subgroup of all patients at diagnosis and of all patients that have reached CR and have received consolidation treatment.

- The core-binding factor (CBF) leukemias involve AML's with cytogenetic abnormality t(8;21)(q22;q22) or the *AML1-ETO* fusion gene and the cytogenetic abnormalities inv(16)(p13q22) or t(16;16)(p13;q22) or the related fusion gene *CBFB-MYH11*.
- If cytogenetics unknown, consider as CN
- Monosomal karyotype (MK) refers to AML with two or more autosomal monosomies or a single autosomal monosomy in the presence of one or more structural cytogenetic abnormalities
- EVI1+ refers to high EVI1 mRNA expression

- FLT3-ITD-/NMP1+ : FLT3-ITD mutant negative (FLT3ITD-) but NPM1-mutant positive (NPM1+): Fms-like tyrosine kinase receptor-3 internal tandem duplications (FLT3-ITD) and nucleophosmin-1 (NPM!) mutations often go together as dual genetic anomalies in the same AML.
- *To exclude ambiguities in the classification patients should be classified in the following hierarchical order: first patients with CBF abnormalities in GR1, GR2 or IR1, of the remaining patients the MK+ patients in VPR1, followed by the abn3q26 patients in VPR2 subsequently the CEBPA+ patients in GR3 and the FLT3ITD-/NPM1+ patients in GR4, subsequently the EVI1+ patients in VPR3. The remaining patients are classified in PR1, IR2, PR2 and PR3.*

## 2. Eligibility for registration

### 2.1 Inclusion criteria

- Patients with poor-risk or very poor-risk AML or RAEB with IPSS ≥ 1.5
- Eligibility for continuation with intensive chemotherapy
- Eligible for allogeneic donor search (related/unrelated)
- 18-70 years, inclusive
- Negative serum pregnancy test for female patients of childbearing potential, at registration
- Female patients of childbearing potential must use an effective contraceptive method during the study and for a minimum of 6 months after study treatment
- Written informed consent

### 2.2 Exclusion criteria

- History of active malignancy during the past 2 years with the exception of basal carcinoma of the skin or carcinoma “in situ” of the cervix or breast
- Known HIV-positivity
- Pregnant or breast-feeding female patients

## 3. Eligibility criteria for alloHSCT

### 3.1 Inclusion criteria

- Responsive disease (< 10% blasts at 3 and/or 4 weeks after start of cycle II chemotherapy)
- Recovery of mucositis after preceding chemotherapy
- Absence of active opportunistic infections
- Absence of active CNS localisation
- HLA-compatible donor available (≥ 7/8 matched unrelated donor or fully matched sibling donor)
- WHO-performance status 0-2
- Written informed consent

### 3.2 Exclusion criteria

- Severe cardiac dysfunction (NYHA classification III-IV)
- Severe pulmonary dysfunction (CTCAE grade III-IV)
- Severe neurological or psychiatric disease
- Significant hepatic dysfunction (serum bilirubin or transaminases ≥ 5 times upper limit of normal)
- Significant renal dysfunction (creatinine clearance < 30 ml/min after rehydration)

- Any psychological, familial, sociological and geographical condition potentially hampering compliance with the study protocol and follow-up schedule
- Concurrent severe and/or uncontrolled medical condition (e.g. uncontrolled diabetes, infection, hypertension, cancer, etc.)

**4. Eligibility criteria for post-transplantation panobinostat and decitabine chemotherapy**

**4.1 Inclusion criteria**

- Absolute neutrophil count $\geq 1.0 \times 10^9$/L
- Platelet count $\geq 25 \times 10^9$/L
- Serum creatinine cleareance $\geq$ 30 ml/min
- Total bilirubin $\leq$30 μmol/l
- AST (SGOT) and ALT (SGPT) $\leq$ 3 x Upper Limit of Normal (ULN);

**4.2 Exclusion criteria**

- Severe cardiac dysfunction (NYHA classification III-IV)
- Severe pulmonary dysfunction (CTCAE grade III-IV)
- Severe neurological or psychiatric disease
- Significant renal dysfunction (creatinine clearance < 30 ml/min after rehydration)
- GvHD requiring the use of combination immunosupressive therapy
- Acute GvHD grades 3-4, chronic extensive GvHD
- Serious active infections
- CMV reactivation, which is not responsive to first line valganciclovir

N.B. CR after alloHSCT is NOT required to continue to panobinostat/decitabine treatment

**5. Definition of dose limiting toxicities**

- Grade 4 non-hematological toxicity between start cycle 1 panobinostat/decitabine and start cycle panobinostat/decitabine or day 35 (whichever occurs first)
- Hematological toxicity which results in delay of the start of cycle 2 after day 35
- TRM between start cycle 1 panobinostat/decitabine and start cycle 2 panobinostat/decitabine or day 35 (whichever occurs first)

A patient who did not receive cycle 1 panobinostat/decitabine is considered not evaluable for DLT and will be replaced.