nature portfolio

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Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Portfolio policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒ A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐ A description of all covariates tested
☐	☒ A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒ A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒ For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐ For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☐	☒ For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☐	☒ Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	FACSDiva ver8.0.1 (BD Biosciences) for flow cytometry data acquisition. NIS-Elements vD4.40.00 (Nikon) for imaging of cytological analyses.
Data analysis	Flowjo v10 (Tree Star) for analysis of flow cytometry. GraphPad Prism v8.0 (GraphPad) for statistical analysis and scientific graphing. Bulk RNA-seq/ATAC-seq analysis: FastQC (v0.11.8), Trim Galore! (v0.6.6), STAR aligner (v2.7.5b), Salmon (v1.2.1), Bowtie2 (v2.1.0), DESeq2 R package (v1.30.1), pheatmap (v1.0.12), ggplot2 (v.3.4.3), clusterProfiler (v3.16.0), SAMtools (v1.11), Picard (v2.2.4), Bedtools (v2.29.2), deepTools (v3.2.1), rtracklayer (v1.60.1), deepStats (v0.4), fgsea (v1.22), Homer (v.4.10) Single-cell RNA-Seq analysis: Cell Ranger (v7.1.0 and v5.0.1), Seurat (v4.0.3), Harmony (v0.1.0), ggplot2 (v3.3.5), destiny (v3.4.0), GLM (v4.3.1)

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

RNA-seq, ATAC-seq and scRNAseq data have been deposited in the NCBI Gene Expression Omnibus under the accession number GSE253715. Alignment was performed using the GRCh38 reference genome (v.36; https://www.gencodegenes.org/human/release_36.html).

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	Sex and gender were not considered in study design. None of our findings apply to only one sex or gender. Sex and gender are not expected to influence the analyses or conclusions. Sex information is included in a table describing patient characteristics in the Extended Data of the manuscript.
Reporting on race, ethnicity, or other socially relevant groupings	There is no reporting on race, ethnicity or other socially relevant groupings.
Population characteristics	Patients with AML who received frontline therapy with DEC and VEN on a prospective clinical trial at the University of Texas MD Anderson Cancer Center, Houston, TX were included (NCT03404193). Patient characteristics are presented in detail in Supplementary Tables of the manuscript.
Recruitment	Patients were included if they were 60 years old or older, or unfit to receive intensive chemotherapy. Patients with European LeukemiaNet (ELN) favorable risk cytogenetics and prior BCL2 inhibitor exposure were excluded.
Ethics oversight	All studies were conducted with informed consent in accordance with Declaration of Helsinki ethical guidelines and with approval by an Institutional Review Committee at the University of Texas MD Anderson Cancer Center, Houston, TX.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	For in vitro experiments, sample size calculation was based on power analysis and historical observations to detect a >2-fold increase (student's t-test, $p < 0.05$). For in vivo experiments, treatment group size was estimated based on biostatistics consultation, as well as historical observations. To reduce number of experimental animals used, the smallest sample size estimated to provide >80% power to detect differences in leukemic potential was used.
Data exclusions	No exclusions were made.
Replication	The experimental findings were replicated successfully 3 or more times. There were no replication data excluded.
Randomization	Animals were randomly assigned to the treatment and control groups at the onset of the experiments.
Blinding	Investigators were not blinded. Data collection for all experiments was automated (e.g. flow cytometry, sequencing etc.) and data interpretation was based on appropriate controls rather than subjective assessment by investigators.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☐	☒ Animals and other organisms
☐	☒ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used

Flow cytometry: CD34-PE (clone 563, 550761, BD Pharmingen), CD34-BV711 (clone 563, 740803, BD Biosciences), CD45-APC (clone HI30; 555485, BD Pharmingen), mCD45-PE-Cy7 (clone 30-F11, 552848, BD Pharmingen), CD33-BV421 (clone WM53, 562854, BD Biosciences), CD19-PE (clone HIB19, 561741, BD Biosciences), CD19-BV650 (clone HIB19, 740568, BD Biosciences), CD38-PE-Cy7 (clone HIT2, 980312, Biolegend), CD123-BV421 (clone 7G3, 563362, BD Biosciences), CD45RA-APC (clone MEM-56, MHCD45RA05, Thermo Fisher Scientific), CD68-PE-Cy7 (clone Y1/82A, 565595, BD Pharmingen), CD11b-BB515 (clone ICRF44, 564517, BD Biosciences), CD11b-BV650 (clone ICRF44, 301336, Biolegend), CD14-APC (clone M5E2, 555399, BD Biosciences), CD14-BV421 (clone M5E2, 565283, BD Biosciences) and CD271 (LNGFR)- APC-Cy7 (clone ME20.4; 345125, Biolegend).
CD45 MicroBeads (130-045-801, Miltenyi Biotec), Mouse Cell depletion kit (130-104-694, Miltenyi Biotec)

Western blotting: P-p44/42 MAPK (ERK1/2; 4370S, Cell Signaling Technologies), p44/42 MAPK (ERK1/2, 4696S, Cell Signaling Technologies), BCL2 (M0887, DAKO), MCL1 (5453S, Cell Signaling Technologies), BCL-xL (2764S, Cell Signaling Technologies), β-Actin (5125S, Cell Signaling Technologies).

Validation

All antibodies were purchased from commercial vendors and have been validated by the manufacturers for use in the species and assays utilized in this study. The validations statements and published references are on the manufacturer's websites. Further in-house validation was performed with appropriate negative and positive controls for each individual antibody.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)

All iPSC lines used as parental lines or for experiments described in the manuscript were previously generated in our lab and described in previous publications cited in the manuscript text (Kotini et al, Nat Biotech 2015; Kotini et al. Cell Stem Cell 2017; Kotini et al. Blood Cancer Discovery 2023).

Authentication

iPSC lines were genotyped and karyotyped after establishment and after each gene editing step. All cultured iPSC lines were genotyped every 6-8 weeks or after 20 passages.

Mycoplasma contamination

Cell lines were tested monthly for mycoplasma contamination. All cell lines tested negative.

Commonly misidentified lines
(See ICLAC register)

None of the cell lines used in this study are included in the commonly misidentified cell lines registry.

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals

Species: Mus musculus; Strains used: NSG (NOD.Cg-PrkdcscidIl2rgtm1Wjl/SzJ) and NSG-SGM3 (NOD.Cg-PrkdcscidIl2rgtm1WjlTg(CMV-IL3,CSF2,KITLG)1Eav/MloySzJ); Sex: female; Age: 6-8 weeks. Mice were maintained at specified pathogen-free (SPF) health status in individually ventilated cages at 21-22 degrees Celsius, 50% humidity and 12h light/dark cycle.

Wild animals

The study did not involve wild animals.

Reporting on sex

Only female mice were used. Sex is not expected to affect the conclusions.

Field-collected samples

The study did not involve samples collected from the field.

Ethics oversight	All mouse studies were performed in compliance with Icahn School of Medicine at Mount Sinai laboratory animal care regulations and approved by an Institutional Animal Care and Use Committee (IACUC).

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration	NCT03404193
Study protocol	The detailed study protocol has previously been published in DiNardo et al. NEJM 2020, referenced in the manuscript.
Data collection	The details of data collection has previously been described in DiNardo et al. NEJM 2020, referenced in the manuscript.
Outcomes	The detailed study protocol has previously been published in DiNardo et al. NEJM 2020, referenced in the manuscript.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	Up to 1 million hematopoietic cells were washed with PBS + 2% FBS (FACS Buffer), labeled in 100uL FACS Buffer containing the antibodies, and incubated at 4C in the dark for 30 minutes. Samples were washed 2X in 1mL FACS Buffer before data acquisition.
Instrument	Cells were assayed on a BD Fortessa or or BD Symphony A5 SE.
Software	Data were collected with BD FACSDiva and analyzed with FlowJo software (Tree Star).
Cell population abundance	A minimum of 10,000 single cell events were acquired for data analysis. Reported cell population abundance range was 0.01-99%.
Gating strategy	First gate: FSC/SSC. Second gate: FSC-A/FSC-H or FSC-H/FSC-W and SSC-H/SSC-W (selection of singlets). Third gate: DAPI (dead cell exclusion). Other markers were determined positive when signal was above FMO (fluorescence minus one) control.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.