Supplementary Material

Legends

Supplementary Figure 1

Supplementary Figure 2

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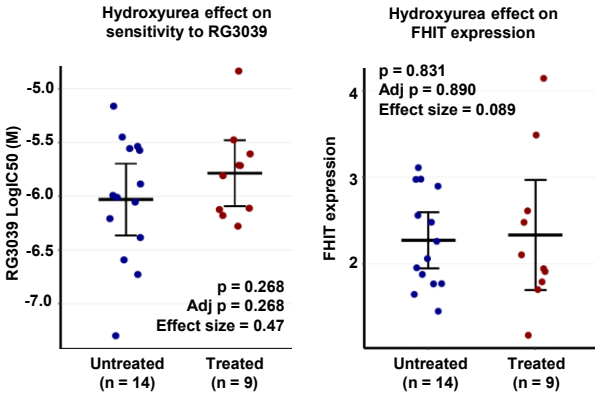
Supplementary References

Supplementary Figure 1. Validation of FHIT biomarker for prediction of DCPS inhibition response in AML and MDS A Dot plot visualization of RG3039 LogIC50 (left) and *FHIT* expression (log2(RPKM+1), right) in primary AML patients treated (n=9) or untreated (n=14) with hydroxyurea prior to collection. Two-sample t-test was used to assess differences between groups; p-values were adjusted using the Benjamini-Hochberg method and effect size is indicated. Mean and 95% CI are shown. **B** Statistical analysis of phenotypic markers in hydroxyurea-treated and untreated samples. Standard or Welch's t-test was performed for each marker based on the variance equality; sample size (n) is indicated. **C** Dot plot visualization of FLT3 expression (log2(RPKM+1)) in primary AML patients treated (n=9) or untreated (n=13) with hydroxyurea. Two-sample t-test was used to assess differences between groups; p-values were adjusted using the Benjamini-Hochberg method and effect size is indicated. Mean and 95% CI are shown. **D** *FHIT* methylation status in adult (n=194, upper panel) and pediatric (n=184, lower panel) AML patients. Methylation z-scores were calculated for each cohort, with z-score >1 defining hypermethylation. Bars represent percentage of patients.

Supplementary Figure 2. Investigation of novel biomarkers in clinical datasets and primary AML cells A Comparison of *FHIT* expression between wild-type and mutant samples for genes in the molecular panel from the OHSU 2022 cBioPortal repository (n=943). Mann-Whitney U test was performed for each gene; p-values and FDR-adjusted p-values are reported. No significant differences were observed. **B** Spearman correlation between *FHIT* expression and selected genes from the AML panel in OHSU 2022 cBioPortal repository (n=943). Correlation coefficients, p-values, and FDR-adjusted p-values are reported. **C** Dot plot of *FHIT* expression in wild-type versus mutant samples for selected genes in primary AML patients. Two-sample t-test or Mann-Whitney U test was performed based on data distribution; sample size (n) is shown for each gene. Mean and 95% CI are shown. **D** Dot plot of RG3039 sensitivity (LogIC50) in primary AML samples with wild-type versus mutant status for selected genes. Two-sample t-test or Mann-Whitney U test was performed based on data distribution; sample size (n) is shown for each gene. Mean and 95% CI are shown. **E** Dot plot of *FHIT* expression (left) and RG3039 sensitivity (LogIC50, right) in primary AML patients with primary (n=17) versus relapsed/refractory (n=3) disease. Two-sample t-test was performed. Mean and 95% CI are shown.

Supplementary Figure 1

A

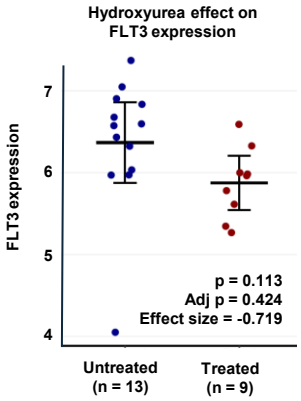


B

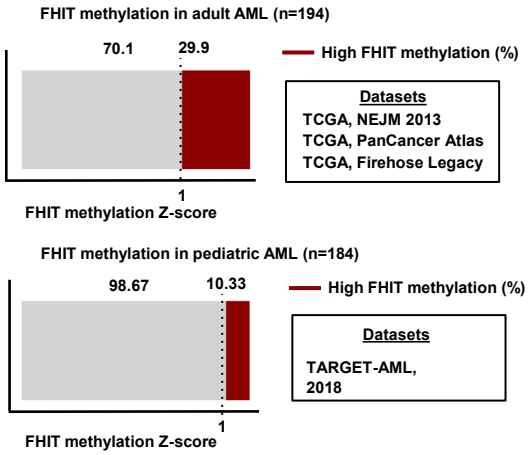
Top 10 hydroxyurea-affected phenotypic markers

Marker	N treated	N untreated	Effect size	Test used	P value	P adjusted
CD123	8	12	-0.574	Welch t test	0.152	0.774
CD13	8	12	-0.641	Standard t test	0.177	0.774
CD64	8	12	-0.506	Welch t test	0.203	0.774
CD7	8	12	0.782	Welch t test	0.209	0.774
Monoblasts	8	12	-0.567	Standard t test	0.230	0.774
CD56	8	12	-0.459	Welch t test	0.249	0.774
T Cells	8	9	-0.568	Standard t test	0.261	0.774
NK Cells	7	8	-0.557	Welch t test	0.285	0.774
Monoblast CD11b	8	12	-0.419	Welch t test	0.303	0.774
LSC	8	12	-0.404	Welch t test	0.304	0.774

C



D



Supplementary Figure 2

A

Comparison of FHIT expression in wild-type and mutant AML gene panel – Non-significant genes

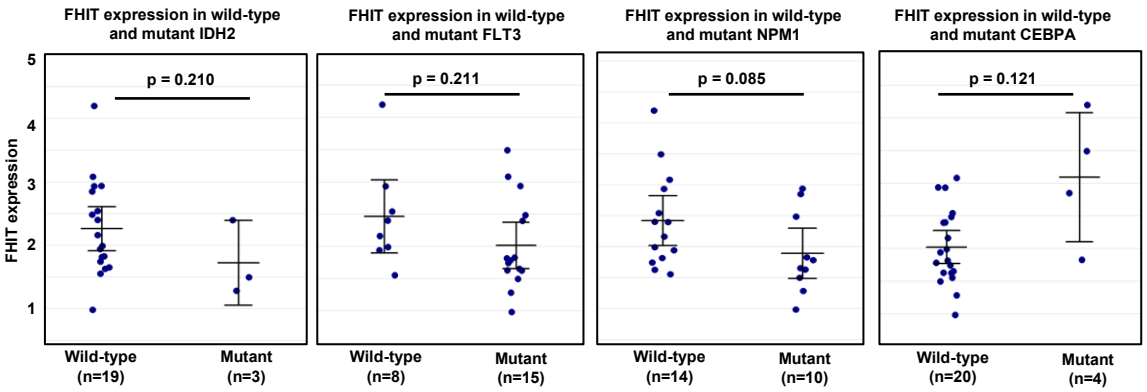
Gene	N mutated	N wild-type	Mean FHIT mutated	Mean FHIT wild type	Mean difference	Test used	P value	FDR
IDH1	49	573	1.941	2.273	-0.332	Mann-Whitney U test	2.35E-02	5.10E-02
KRAS	34	588	1.940	2.264	-0.324	Mann-Whitney U test	2.78E-02	5.10E-02
KIT	16	606	2.622	2.237	0.385	Mann-Whitney U test	1.67E-01	2.63E-01
TP53	57	565	2.187	2.253	-0.066	Mann-Whitney U test	2.96E-01	4.07E-01
RUNX1	75	547	2.176	2.256	-0.080	Mann-Whitney U test	4.65E-01	5.68E-01
NRAS	96	526	2.214	2.253	-0.039	Mann-Whitney U test	5.61E-01	6.17E-01
DNMT3A	138	484	2.265	2.241	0.024	Mann-Whitney U test	7.18E-01	7.18E-01

B

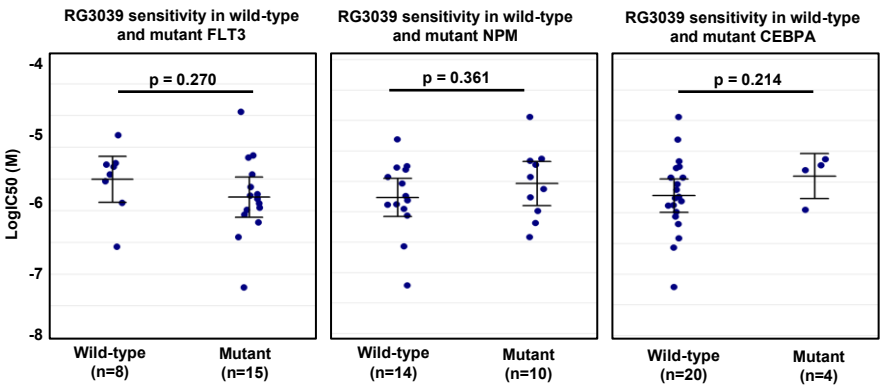
Correlation of FHIT expression and AML gene panel expression – All other genes

Gene	Correlation	P value	N samples	P adjusted
IDH1	-0.304	8.48E-16	671	**9.33E-15**
NRAS	0.197	2.71E-07	671	**9.93E-07**
RUNX1	-0.180	2.74E-06	671	**7.55E-06**
TP53	-0.174	5.47E-06	671	**1.20E-05**
KRAS	0.074	5.36E-02	671	6.55E-02
DNMT3A	0.021	5.88E-01	671	6.47E-01
KIT	0.015	6.93E-01	671	6.94E-01

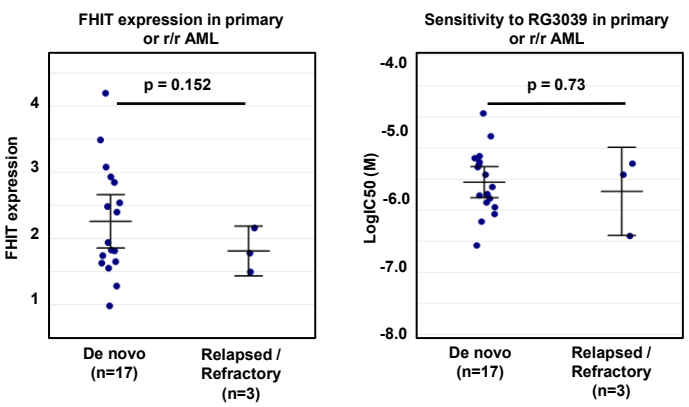
C



D



E



Supplementary Table 1

Data Availability Information for analyzed databases

Database name	Accession number / ID	Direct web link	Publication reference (if available)
TCGA, NEJM 2013	n/a	https://gdc.cancer.gov/about-data/publications/laml_2012	1
TCGA, PanCancer Atlas	n/a	https://gdc.cancer.gov/about-data/publications/PanCan-CellOfOrigin	2
TCGA, Firehose Legacy	n/a	https://gdac.broadinstitute.org/runs/stddata__2016_01_28/data/LAML/20160128/	n/a
OHSU, Nature 2018	phs001657.v1.p1	https://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs001657.v1.p1	3
OHSU, Cancer Cell 2022	phs001657.v2.p1	https://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs001657.v2.p1	4
TARGET GDC 2025	phs000465.v23.p8	https://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs000465.v23.p8	n/a
TCGA GDC, 2025	phs000178.v11.p8	https://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs000178.v11.p8	n/a
TARGET, 2018	phs000465.v15.p7	https://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs000465.v15.p7	5

Supplementary References

1. Ley TJ, Miller C, Ding L, Raphael BJ, Mungall AJ, Robertson A, et al. Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. N Engl J Med. 2013 May 30;368(22):2059–74.

2. Hoadley KA, Yau C, Hinoue T, Wolf DM, Lazar AJ, Drill E, et al. Cell-of-Origin Patterns Dominate the Molecular Classification of 10,000 Tumors from 33 Types of Cancer. Cell. 2018 Apr 5;173(2):291-304.e6.

3. Tyner JW, Tognon CE, Bottomly D, Wilmot B, Kurtz SE, Savage SL, et al. Functional genomic landscape of acute myeloid leukaemia. Nature. 2018 Oct 25;562(7728):526–31.

4. Bottomly D, Long N, Schultz AR, Kurtz SE, Tognon CE, Johnson K, et al. Integrative analysis of drug response and clinical outcome in acute myeloid leukemia. Cancer Cell. 2022 Aug 8;40(8):850-864.e9.

5. Bolouri H, Farrar JE, Triche T, Ries RE, Lim EL, Alonzo TA, et al. The molecular landscape of pediatric acute myeloid leukemia reveals recurrent structural alterations and age-specific mutational interactions. Nat Med. 2018 Jan 1;24(1):103–12.