PHARMACODYNAMICS AND ANTI-TUMOR MECHANISM OF THE BRG1/BRM (SMARCA4/2) INHIBITOR FHD-286 IN A PHASE 1 STUDY IN SUBJECTS WITH AML OR MDS

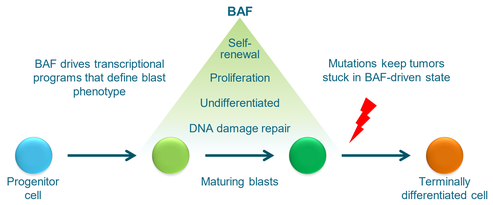
Mike Collins[1], Astrid Thomsen[1], GiNell Elliott[1], Jessica Wan[1], Kim Horrigan[1], Laure Delestre[2], Virginie Penard-Lacronique[2], Stephane De Botton[2], Warren Fiskus[3], Kapil N. Bhalla[3], Courtney DiNardo[3], Samuel Agresta[1], Jessica Piel[1], Martin Hentemann[1]

[1]Foghorn Therapeutics, Cambridge, MA; [2]Institut Gustave Roussy, Villejuif, France; [3]The University of Texas MD Anderson Cancer Center, Houston, TX

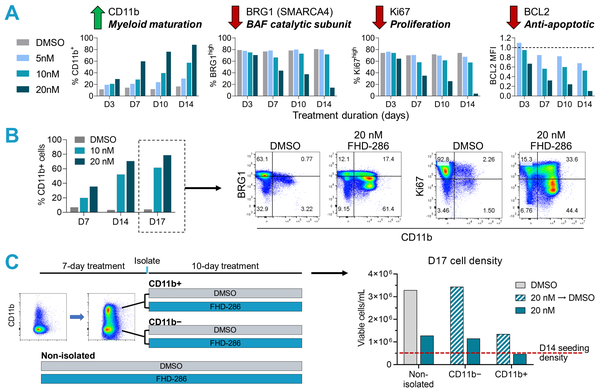
FOGHORN® THERAPEUTICS
www.foghorntx.com

INTRODUCTION

- The BAF complex is critical to the regulation of cellular differentiation and is thought to maintain cancer cells in an undifferentiated state.
- FHD-286 is a potent, selective, allosteric, small molecule inhibitor of the BAF catalytic subunits BRG1 (SMARCA4) and BRM (SMARCA2).
- FHD-286 pre-clinical data demonstrate broad-based differentiation across acute myeloid leukemia (AML) models.
- Biomarkers identified from these pre-clinical studies were analyzed in a Phase 1 dose escalation study of FHD-286 monotherapy in patients with relapsed or refractory (R/R) AML or myelodysplastic syndromes (MDS) (Study FHD-286-C-002).
- Flow cytometry for biomarkers of stemness (e.g. CD34), myeloid maturation (e.g. CD11b), and other relevant biomarkers was performed on patient bone marrow mononuclear cell (BMMC) and peripheral blood mononuclear cell (PBMC) samples. Single cell RNA sequencing (scRNA-seq) was also performed on matched screening and on-treatment bone marrow.



BACKGROUND



Background

A) Dose and time-dependent modulation of relevant biomarkers by flow cytometry in AML cells treated *in vitro*. Similar results were obtained in MOLM13, MV411, EOL1, OCI-AML2 and HL60 cells.

B) CD11b+ cells downregulate BRG1 and Ki67. Similar results were obtained in other AML cell lines.

C) CD11b+/− magnetic separation experiment in HL60 cells. CD11b+ cells have markedly reduced proliferation. CD11b− cells proliferate at approximately the same rate as parental cells.

D) AML primary patient-derived cells treated with FHD-286 *in vitro* show morphologic features of differentiation.

References and acknowledgements

1. Centore, R. C. *et al. Trends Genet.* **36**, 936–950 (2020)
2. Hentemann, M. Abstract ND14. *AACR Annual Meeting* (2022)
3. Collins, M. *et al.* Abstract 2122. *AACR Annual Meeting* (2023)
4. Fiskus, W. C. *et al.* Abstract 1140. *AACR Annual Meeting* (2023)
5. Elliott, G. *et al.* Abstract 35465. *AACR-NCI-EORTC International Conference on Molecular Targets and Therapeutics* (2023)

We wish to thank the patients, clinicians and investigators for their participation in the Phase 1 clinical trial of FHD-286

Study FHD-286-C-002 Overview

DESIGN

- Oral daily dosing of FHD-286 as monotherapy
- R/R AML and R/R MDS patients who exhausted all treatment options
- Doses tested: 2.5 mg, 5 mg, 7.5 mg, 10 mg once daily

PATIENTS

- 40 patients enrolled: 36 R/R AML and 4 R/R MDS
- 67.5% had 3+ prior lines
- Majority with abnormal karyotype (82.5%) and poor genetic risk factors (65% with adverse genetic status)
- Broad range of mutations

STUDY OBJECTIVES

- Safety and tolerability, MTD and/or RP2D
- Pharmacokinetics and pharmacodynamics, clinical activity, biomarker analysis

RESULTS

Phenotypic differentiation in patient bone marrow blasts measured by increased CD11b and decreased CD34

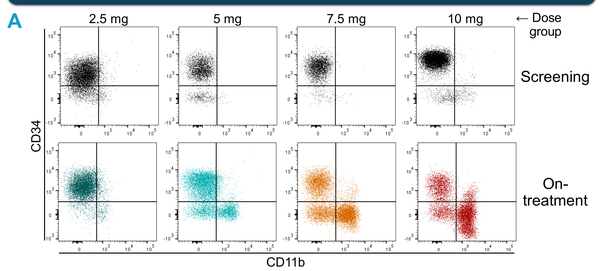


Figure 1. A) Representative flow cytometry plots of individual patient BMMC samples. Samples were live cell-gated, and blasts were identified by CD45 vs. side scatter. Quadrants illustrate positive/negative thresholds. **B)** Box and whisker plots of BMMC blasts analyzed by flow cytometry. Data points represents individual patient samples. MDS patients were excluded from this analysis. *Note: One 10 mg patient did not have a screening sample.

Differentiation in a broad range of genetic backgrounds

Starting dose	Diagnosis	Mutations	Cytogenetics	CD34+ blasts Screen	CD34+ blasts Min.	CD11b+ blasts Screen	CD11b+ blasts Max.
2.5 mg	AML	*NRAS, WT1*	Adverse				
5 mg	AML	*RUNX1, NRAS, ASLX1*	Adverse				
	AML	*TET2, WT1, GATA2, PLCG2, ARHGEF28, BRD4, CDK12, DDX41, KMT20, PARP1, ZRSR2*	Intermediate				
	AML	N/A	Adverse				
	MDS	*RUNX1, NRAS, KRAS, SF3B1, ASXL2, CSF3R, GATA2*	Adverse				
	AML	N/A	Adverse				
	MDS	*DNMT3a, TET2*	Intermediate				
7.5 mg	AML	*CBFB (locus at 16q22)*	Favorable				
	AML	*RUNX1, KRAS, ASXL1, JAK2, TET2, EZH2, ETNK1*	Adverse				
	AML	*ASXL1, TP53, U2AF1*	Adverse				
	AML	N/A	Adverse				
	AML	*KMT2A rearrangement*	Adverse				
10 mg	AML	N/A	Adverse				
	AML	*NRAS, SF3B1*	Intermediate				

CD34 % pos. 0 100 CD11b % pos. 0 100

Table 1. Genetic characteristics of subjects for which bone marrow biomarkers were evaluated.

Dose and time-dependent biomarker modulation in patient peripheral blasts

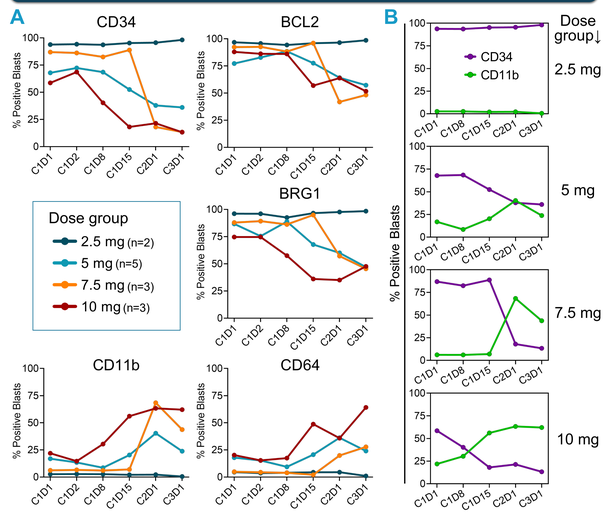


Figure 2. A) Patient PBMCs were analyzed by flow cytometry for the indicated markers. Samples were live cell-gated, and blasts were identified by CD45 vs. side scatter. MDS patients, patients who completed <1 cycle, and samples with <1000 viable blasts were excluded from this analysis. Data points represent group median. **B)** Percentage of PBMC blasts positive for CD34 and CD11b, separated by dose. Samples were analyzed as in **A**.

Biomarker changes correspond to FHD-286 exposure and peripheral blast reductions

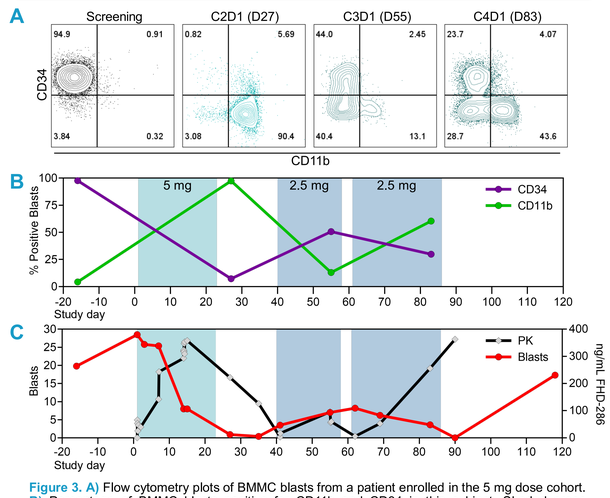


Figure 3. A) Flow cytometry plots of BMMC blasts from a patient enrolled in the 5 mg dose cohort. **B)** Percentage of BMMC blasts positive for CD11b and CD34 in this subject. Shaded areas indicate actual dose administered. White areas indicate periods when no drug was administered. **C)** Peripheral blast count and plasma concentration of FHD-286 (PK) in this subject.

FHD-286 reduces leukemogenic potential in an in vivo PDX transplant model

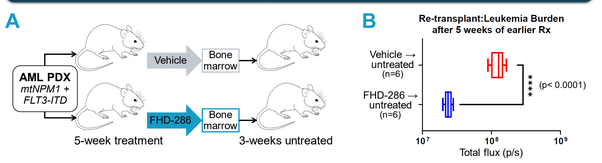


Figure 4. A) Mice were inoculated by tail vein injection with luciferase-expressing tumor cells and treated with either vehicle or 1.5 mg/kg FHD-286 QD on a 5 days on/2 days off schedule. After 5 weeks of treatment, tumor cells were isolated from bone marrow using a human antibody cocktail, and equal numbers of viable cells were transplanted in naïve mice. **B)** Total animal bioluminescence in untreated mice, 3 weeks after re-transplant.

scRNA-seq reveals shift from LSC to myeloid maturation signature in patient BMMCs

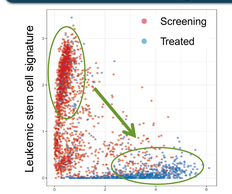


Figure 5

- Single-cell RNA-seq performed on paired screening and on-treatment patient bone marrow aspirates.
- Shown is an analysis of screening and C2D1 samples from a single patient in the 5 mg dose cohort.
- Similar effects observed across 5 mg, 7.5 mg and 10 mg dose levels.
- For more information, refer to Elliott, G., et al. *Leukemic stem cell differentiation visible at single-cell resolution in acute myeloid leukemia patients treated with FHD-286, an inhibitor of BRG1/BRM (SMARCA4/2)* **(Poster A013)**

In vitro combo benefit with cytarabine and decitabine

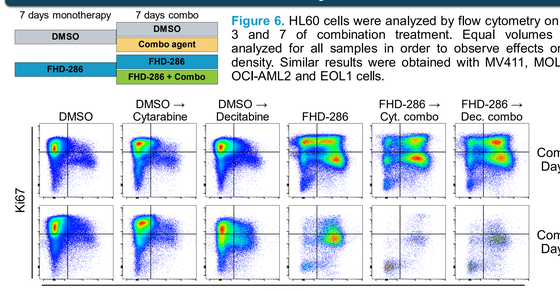


Figure 6. HL60 cells were analyzed by flow cytometry on days 3 and 7 of combination treatment. Equal volumes were analyzed for all samples in order to observe effects on cell density. Similar results were obtained with MV411, MOLM13, OCI-AML2 and EOL1 cells.

CONCLUSIONS

- FHD-286 treatment led to phenotypic differentiation in AML patient blasts
  - Downregulation of stem cell and blast markers CD34, BRG1 and BCL2
  - Upregulation of myeloid maturation markers CD11b and CD64
- Biomarker modulation deeper and more rapid at higher doses
- Differentiation observed in heavily pretreated patients, regardless of mutation status
- Correlation between biomarker modulation, FHD-286 plasma exposure, and blast reductions in patients
- Preclinical and clinical data suggest that FHD-286 may reduce LSC potential
- Combination with cytoreductive agents may be a promising strategy to target both LSCs and proliferative blasts