Tolerability and Single Agent Anti-Neoplastic Activity of the CD3xCD123 Bispecific Antibody APVO436 in Patients with Relapsed/Refractory AML or MDS

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ABSTRACT

APVO436 is a recombinant T-cell engaging humanized bispecific antibody designed to redirect host T-cell cytotoxicity in an MHC-independent manner to CD123-expressing blast cells from patients with hematologic malignancies and has exhibited single-agent anti-leukemia activity in murine xenograft models of acute myeloid leukemia (AML). This first in human clinical trial of APVO436 (NCT03647800) was designed as a multi-institutional, open-label, multiple-dose Phase 1B dose escalation study in patients with relapsed/refractory (R/R) AML and myelodysplastic syndrome (MDS). 58 R/R adult AML/MDS patients were screened; 12 patients were screen failures, and the remaining 46 eligible patients were enrolled in the study between 15/05/18 and 04/06/21. Thirty nine patients (84.8%) had R/R AML and 7 had R/R MDS. The median age was 68 years. Patients had failed 1-8 prior lines of therapy (Mean±SE: 3.2±0.3). A 3+3 design was used to guide the dose escalation. APVO436 was administered as weekly intravenous (IV) infusions at 10 different dose levels (Cohorts 1-10), ranging from a minimum anticipated biological effect level (MABEL) of 0.3 mcg to 60 mcg. In Cohorts 5-10, APVO436 was administered according to an intra-patient step-up strategy to reduce the risk for cytokine release syndrome (CRS). Response criteria of the International Working Group (IWG) were used for assessment of MDS patients. Standard European LeukemiaNet (ELN) 2017 criteria were used for response assessments in AML patients. The date of data cutoff was July 22, 2021. The primary endpoint of identifying a clinically active recommended phase 2 dose (RP2D) level for further clinical development of APVO436 was met. APVO436 exhibited a favorable safety profile with acceptable tolerability and manageable drug-related adverse events (AEs), and its maximum tolerated dose (MTD) was not reached at a weekly dose of 60 mcg. The most common APVO436-related AEs were infusion-related reactions (IRR) occurring in 13 (28.3%) patients and cytokine release syndrome (CRS) occurring in 10 (21.7%). No hematologic DLT was observed in any of the 10 dose cohorts.Ten patients experienced 12 episodes of Grade 3 febrile neutropenia and each one of these 12 episodes was reported as not related to APVO436. APVO436-related transient neurotoxicity occurred only in 5 of 46 patients (10.9%). It was mild with Grade 1 AEs including headache, tremor, dizziness, lethargy, insomnia, memory loss, and confusion. A single case of Grade 3 confusion was encountered on the first day of treatment and resolved within a day.The single dose RP2D level has been identified as 18 mcg flat dose (Cohort 6; ~0.2 mcg/kg based on the body weights of the patients enrolled). Stable disease (SD), partial remissions (PR) and complete remissions (CR) were observed in 8 R/R AML patients as best overall responses to APVO436 at the RP2D level. Seven of 8 had failed 2-4 prior lines of anti-AML therapy and one patient had relapsed after achieving a remission on frontline venetoclax plus decitabine therapy. One patient had clearance of peripheral blasts with >50% decrease in the BM blast percentage. Two primary AML patients with >25% BM blasts and unfavorable cytogenetics and/or adverse risk group genomic mutations achieved a PR at 58 days and 75 days, respectively, that deepened to a CR with full hematologic recovery at 92 and 113 days, respectively. Time-to-progression ranged from 87 to 238 days (Median: 177 days). Notably, the median overall survival OS was >300 days for the 8 R/R AML patients with a favorable response (prolonged SD and PRs/CRs). Five of the 8 patients remain alive at 110, 124, 323, 352, and 395 days, respectively. By contrast, the median OS for the remaining 31 AML patients in the intent to treat patient population (including 5 who were not evaluable for response) was 100 days (95% CI: 49.8-150.2) and 24 of 31 (77.4%) died (Log Rank c^2 = 5.298, P=0.021).There were too few MDS patients to accurately analyze the clinical activity of APVO436. The time to progression in 6 evaluable patients ranged from >78 days to 321 days. Three of these 6 patients had marrow CRs. In conclusion, the safety and preliminary evidence of efficacy of APVO436 in R/R AML and MDS patients warrant further investigation of its clinical impact potential.

Materials and Methods

Investigational Medicinal Product. APVO436 is a humanized bispecific antibody with an estimated molecular weight (MW) of 161 kDa that targets CD123 and CD3ε [23, 31, 32]. It is a glycosylated homodimeric antibody comprised of two sets of binding domains linked to a human immunoglobulin (Ig) G1 fragment crystallizable (Fc) domain (**Figure 1**).

Study Design and Eligibility Criteria. The clinical trial of APVO436 (Title: "Phase 1B Open-label, Dose Escalation and Dose Expansion Study of APVO436 in Patients with Relapsed or refractory (R/R) AML or High Grade MDS") was designed as a multiple-dose Phase 1B dose escalation study in patients with relapsed AML and high-risk MDS. It was registered in the clinical trial database ClinicalTrials.gov with the identifier number NCT03647800.

Patients and Patient Disposition. 58 R/R adult AML/MDS patients were screened; 12 patients were screen failures, and the remaining 46 eligible patients were enrolled in the study.

Study Conduct. The study was performed under an IND at 10 centers in the US as an open-label study sponsored by Aptevo Therapeutics.

The starting dose in Cohort 1 was 0.3 mcg (~0.005 mcg/kg for a 60-kg patient) which was the Minimum Anticipated Biological Effect Level (MABEL) based on T-cell activation assays [36]. The assigned weekly target dose levels for Cohorts 2-10 were 1 mcg, 3 mcg, 9 mcg, 18 mcg (Cohort 6A), 12 mcg (Cohort 6B), 24 mcg, 36 mcg, 48 mcg, and 60 mcg, respectively. A 3+3 design was used to guide the dose escalation.

Flow Cytometry. Immunophenotyping was performed on cryopreserved peripheral blood mononuclear cells from patients by standard flow cytometry using a BD LSR II flow cytometer and FACSDiva Software Version 8.0.2 fluorochrome-labeled monoclonal antibodies reactive with CD45 (anti-human CD45, Clone H130, V500, BD Biosciences #560777), CD34 (anti-human CD34, Clone REA1164, VioBright 515, Miltenyl Biotech #130-120-517), CD38 (anti-human CD38, clone HIT-2, BV605, Biolegend#303532), and CD123 (anti-human CD123, Clone 9F5, AF647, BD Biosciences #563599) antigens.

Ethics Statement and Study Approval. The study protocol was approved by the WCG-Central Institutional Review Board (IRB) (OHRP/FDA registration number: IRB00000533) and the local IRB at participating centers. The study was performed in compliance with the International Conference on Harmonization (ICH) guidelines for Good Clinical Practice (ICHE6/GCP). Each patient provided a written informed consent (ICF) prior to enrollment.

Statistical Analyses. Standard statistical methods were applied for the analysis of the clinical data. Survival data was analyzed by the Kaplan-Meier method using the GraphPad Prism 9 statistical program (GraphPad Software, LLC, San Diego, CA). Log-rank statistics was used to compare the differences between patient subgroup.

Results

Safety

- APVO436 exhibited a favorable safety profile with acceptable tolerability and manageable side effects. The MTD was not reached at a weekly flat dose of 60 mcg (Cohort 10), which was tolerated by all 4 patients enrolled without any DLTs or Grade 3-4 AEs. The single dose RP2D level has been identified as 18 mcg flat dose (Cohort 6; ~0.2 mcg/kg based on the body weights of the patients enrolled), which is 30% of the Cohort 10 dose level.
- The most common Grade ≥3 AEs suspected to be APVO436-related were Grade 3-4 cytokine release syndrome (CRS) occurring in 4 of 46 patients (8.7%), Grade 3-4 anemia occurring in 2 of 46 patients (4.3%) and infusion related reactions (IRR) occurring in 2 of 26 patients (4.3%). A single Grade 5 AE was encountered in a patient who developed Grade 2 CRS and subsequently a fatal (Grade 5) acute renal failure.
- No hematologic DLT was observed in any of the 10 dose cohorts.
- APVO436-related SAEs were encountered in 13 of the 46 patients (28.3%) and were most commonly related to CRS (7 of 13 SAE cases; affecting 15.2% of the safety population) and IRR (3 of 17 cases; affecting 6.5% of the safety population). The remaining 3 cases included a case of possibly related sepsis that resolved within 5 days, generalized weakness that resolved after 28 days, and Grade 1 neurotoxicity that resolved within 2 days.
- APVO436-related transient neurotoxicity occurred only in 5 of 46 patients (10.9%). It occurred during the first cycle in 4 of the 5 patients and in Cycle 8 in the remaining patient. It was mild with Grade 1 AEs including headache, tremor, dizziness, lethargy, insomnia, memory loss, and confusion. A single case of Grade 3 confusion (Unique patient number [UPN]31 in Cohort 7) was encountered on the first day of treatment and resolved within a day. Neurotoxicity did not show any dose-dependence. Gender, race, age, absolute lymphocyte count or percentage of lymphocytes in peripheral blood did not predict neurotoxicity. Neurotoxicity occurred in 3 patients who also experienced CRS who did not develop CRS. Conversely, of 10 patients who developed CRS, 7 did not experience any neurotoxicity.

Efficacy

- Of the 39 R/R AML patients, 34 were evaluable for surrogate response measurements.
- Twenty two of these 34 patients (64.7%) had stable disease (SD) as their best overall response.
- In 8 patients of these 22 patients corresponding to 23.5% of the evaluable 34 AML patients, SD was achieved between 31 and 75 days after study entry and lasted >3 months.
- Seven of 8 had failed 2-4 prior lines of anti-AML therapy. They were enrolled 7-39 days after documentation of progressive leukemia or leukemic relapse.
- The onset and duration of the SD, PR or CR in these 8 patients is illustrated by the Swimmer plot depicted in Figure 2. Time-to-progression ranged from 87 to 238 days (Median: 177 days).
- Of these 8 patients with a favorable response to APVO436, one (UPN31) had clearance of peripheral blasts with >50% decrease in the bone marrow (BM) blast percentage. Notably, 2 primary AML patients with >25% BM blasts and unfavorable cytogenetics (del 5q and monosomy 7 in UPN28) and/or adverse risk group genomic mutations (TP53 mutation in UPN28 and ZRSR2 in UPN21) achieved a PR at 58 days and 75 days, respectively, that deepened to a CR with full hematologic recovery at 92 and 113 days, respectively.
- UPN28 had developed PD after Venetoclax + Decitabine therapy, while UPN21 had failed 3 lines of prior anti-AML therapy. On Day 1 of Cycle 5 (C5D1), UPN28 achieved CR with full hematologic recovery, including an ANC of 2.8 x10^9/L, platelet count of 141,000/μL, hemoglobin of 9.8 g/dL with 0% blasts in the BM and no circulating blasts. He relapsed 85 days later with 10% BM blasts on Day 1 of Cycle 7 (C7D1), no circulating blasts, a WBC of 4.2x10^9/L, ANC of 3.0x10^9/L, platelet count of 139,000/μL, and Hgb of 10.0 g/dL (**Figure 2**). He continued to have SD until Cycle 10 when he started showing a gradual increase in BM blast percentage with a concomitant drop in ANC and platelet count.
- UPN21 who had 33% BM blasts at screening and 46% BM blasts on day 1 of Cycle 2, was in PR with 8% BM blasts on Day 1 of Cycle 3 along with an increase of ANC to 2.0x10^9/L, a platelet count of 106,000/μL and Hgb of 11.2 g/dL. On Day 1 of Cycle 5, he was in CR with 4% blasts, no circulating blasts, ANC of 2.8x10^9/L, platelet count of 134,000/μL, Hgb of 12.2 g/dL. He relapsed 57 days later with 66% BM blasts or a noticeable drop of peripheral counts, including an ANC of 1.6 x10^9L, platelet count of 113,000/μL, and Hgb of 11.7 g/dL (**Figure 2**).
- One patient (UPN31) had a complete clearance of peripheral blasts at 113 days (from 21% pre-treatment to 14% in cycle 2, 2% in cycle 3, and 0% in cycle 4) and a >50% decrease of pretreatment BM blast percentage (from 78% prior to treatment to 37% post-treatment) followed by a sustained SD (**Figure 2**).
- Notably, the median OS was >300 days for the 8 R/R AML patients with a favorable response (prolonged SD and PRs/CRs). Five of the 8 patients remain alive at 110, 124, 323, 352, and 395 days, respectively (**Figure 2**). By contrast, the median OS for the remaining 31 AML patients in the intent to treat patient population (including 5 who were not evaluable for response) was 100 days (95% CI: 49.8-150.2) and 24 of 31 (77.4%) died. This difference in survival outcome of favorable responders vs. non-responders was statistically significant (Log Rank χ^2 = 5.298, P=0.021, **Figure 3**). Likewise, the survival outcome of the favorable responders was significantly better than the OS of 26 non-responders who were evaluable for response determinations whose median OS was 121.0 days (95% CI: 85.2-21.0 days) (Log Rank χ^2 = 5.120, P=0.023).
- Of the 7 MDS patients enrolled, 6 were evaluable for response and they had SD without a significant hematological improvement as their best overall response. The time to progression intervals were 104 days, >106 days, 138 days, >147 days, 211 days, and 321 days, respectively. Of these, 3 achieved a marrow CR.

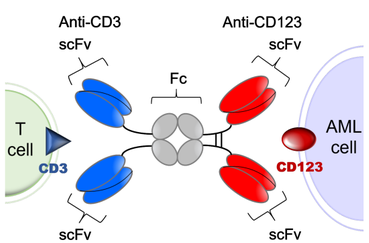


Figure 1. Composition and Mode of Action of APVO436. APVO436 targeting CD123 on AML cells and redirecting CD3+ T-cells to the close vicinity of the target leukemia cells. APVO436 is a humanized bispecific antibody that targets both CD123 and CD3. It is comprised of two sets of binding domains linked to a human IgG1 Fc domain. The CD123 binding domain is a fully human scFv directed against human CD123. The CD3 binding domain is a humanized scFv that binds human CD3. The Fc region has been engineered to minimize complement fixation and interaction with Fcγ receptors.

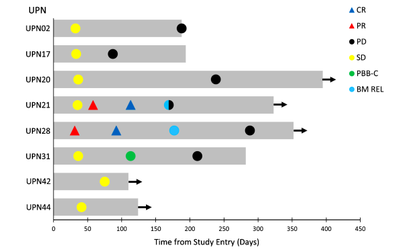


Figure 2A. Swimmer Plot of Best Overall Responses of the 8-Patient Favorable Response Population of R/R AML Patients. The onset and duration of SD, PR, CR, clearance of peripheral blasts (PBB-C), bone marrow relapse (BM REL) and onset of PD are indicated with specific symbols. Arrow: Alive.

Table 1. Patient Characteristics, Demographic Features and APVO436 Exposure for Safety Population (N=46)

Diagnosis	
AML	39 (84.8%)
Primary AML	26 (56.5%)
Secondary (s)-AML	9 (19.6%)
Treatment related (t)-AML	4 (8.7%)
MDS	7 (15.2%)
Age (years)	
Mean ± SE	65.4 ±2.0
Median	69
Range	18-82
Sex	
Female	22 (47.8%)
Male	24 (52.2%)
Ethnic origin	
Caucasian, not Hispanic or Latino	34 (73.9%)
Caucasian, Hispanic or Latino	6 (13.0)
Black or African American	3 (6.5%)
Hispanic or Latino	1 (2.2%)
Asian	2 (4.3 %)
Prior # of chemotherapy regimens	
1	9 (19.6%)
2	14 (30.4%)
3	6 (13.0%)
≥4	16 (34.8%)
Range	1-8
Not reported	1 (2.2%)
Mean±SE (Median)	3.2±0.3 (2.5)
Number of APVO436 Treatments Mean±SE (Median)	11±2 (7)

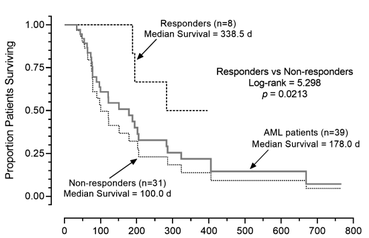


Figure 3. Survival Outcome of AML Patients According to Response to APVO436. Depicted are the overall survival curves of the 8 patients favorable responses, 31 patients who did not respond, and all 39 patients combined. Favorable responses of CR, PR or SD≥3 months is associated with improved overall survival in R/R AML patients treated with APVO436 monotherapy.

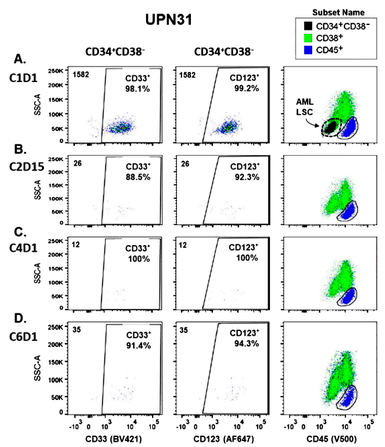


Figure 2B. Depletion of Circulating Putative AML-LSC Cells in VENAZA-resistant Relapsed AML Patient Receiving APVO436 Monotherapy. The numbers in the left upper corner in the first 2 columns represent the relative total LSC numbers in the analyzed samples of which the vast majority co-expressed both CD123 and CD33. Virtually all of the CD34+CD38- cells in the analyzed peripheral blood samples were CD123+ and CD33+ consistent with AML. The size of this CD123+CD33+CD34+CD38- AML LSC population indicated with the arrow in Panel A, 3rd column, was significantly reduced by APVO436 monotherapy. APVO436 was administered IV weekly with 4 weeks/cycle. C: Cycle number; D: Day number.

Summary

- Of the 34 relapsed AML patients evaluable for surrogate response measurements, 8 had clinically meaningful stabilization of their leukemia or a CR. 7 of these 8 had failed 2-4 prior lines of anti-AML therapy, and 1 patient had relapsed after frontline therapy with Venetoclax plus decitabine.
- The median OS was >300 days for the 8 R/R AML patients with a favorable response (prolonged SD and PRs/CRs). Five of the 8 patients remain alive at 110, 124, 323, 352, and 395 days, respectively. The OS of these 8 patients was significantly better than the survival outcome of the total population as well as non-responders.
- It is noteworthy that in mouse xenograft models of human AML, APVO436 exhibited anti-leukemic activity at HED levels of ≥0.08 mcg/kg with maximal activity obtained at HED of 0.4 mcg/kg. In agreement with the preclinical proof-of-concept data, CRs were observed as best overall responses to APVO436 as a single agent at the anticipated sub-MTD clinical dose levels of 0.15-0.19 mcg/kg (UPN28 in Cohort 6B, 12 mcg flat dose = ~0.2 mcg/kg [BW: 80.3 kg]; UPN21 in Cohort 6A, 18 mcg flat dose = ~0.2 mcg/kg [BW: 94.8 kg].
- The off-the-shelf availability of APVO436 combined with its single agent anti-leukemic activity, favorable safety profile, ease of administration not requiring continuous infusion but weekly short infusions (over 4 hours after Cycle 1) that can be administered in outpatient settings makes it an attractive option as a bispecific T-cell engager against AML.

Future Directions

In the open-label, multi-center, dose expansion phase of the study (Part 2), we will evaluate the safety and tolerability of APVO436 at the RP2D level when it is used as an adjunct to the standard of care and obtain a preliminary assessment of the anti-leukemia activity of APVO436-containing experimental monotherapy and combination therapy modalities.

Expansion Cohorts

In Cohorts 1-4, APVO436 will be administered at a fixed dosage of 18 mcg after a weekly ramp up during Cycle 1 (Cohorts 1, 3, 4) or Cycle 1-2 (Cohort 2). In Cohort 5, APVO436 will be administered at a fixed dose of 18 mcg twice weekly after a weekly ramp up during Cycle 1.

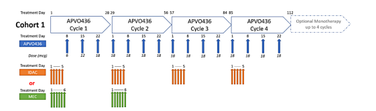
Endpoints

Primary: Safety

The cumulative incidence of Grade 3-4 AEs, and SAEs, and the incidence of AES of interest (≥Grade 2 CRS, ≥Grade 2 Infusion related reaction, ≥2 cardiac toxicity and ≥2 neurotoxicity as complications of CRS) for safety

Secondary: Efficacy

- Incidence of composite CR (CR + CRi + CRh) in relapsed patients as a measure of efficacy within the confines of a Phase 1B study. CR with partial hematological recovery (CRh) is used in the context of clinical studies [<5% blasts in the BM, without evidence of extramedullary disease, platelets ≥50 × 10^9/l and absolute neutrophil count (ANC) ≥0.5 × 10^9/l]. CR with incomplete hematological recovery (CRi) requires recovery of at least one lineage (either ANC ≥1.0 × 10^9/l or platelets ≥100 × 10^9/l). Morphological leukemia-free state (MLFS) consists of <5% BM blasts, absence of blasts with Auer rods, no extramedullary disease and a lack of hematological recovery of both neutrophils and platelets where the BM may not be merely aplastic and at least 200 cells should be counted or cellularity at trephine biopsy should be at least 10%.
- Incidence of patients who are able to achieve MRD-negative CR and undergo HSCT post protocol therapy



Cohort 1. Induction with Chemotherapy (ChT) plus APVO436. 1st or 2nd Early Relapse; or Primary Refractory Disease

Cohort 2. Induction with APVO436 + Venetoclax + Azacitidine - Frontline or 1st Relapse or Primary Refractory Disease.

Cohort 3: Consolidation post 7+3 – Frontline, 1st Relapse or Primary Refractory Disease.

Cohort 4: MRD-positive (MRD+) 1st Remission, APVO436 + Azacitidine (Oral) combination

Cohort 5: MRD+ 2nd Remission, Single Agent APVO436

References

Uckun FM and Watts J. CD123-Directed Bispecific Antibodies for Targeting MDS Clones and Immunosuppressive Myeloid-Derived Suppressor Cells (MDSC) in High-Risk Adult MDS Patients. ***Front. Aging*** 2021; 2:757276. doi: 10.3389/fragi.2021.757276

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