Phase I Study of APR-246 in Combination with Venetoclax and Azacitidine in *TP53*-Mutant Myeloid Malignancies

Protocol No.:	A19-11184
IND No.:	139738
Sponsor:	Aprea Therapeutics, Inc. [redacted]
Responsible Medical Officer:	Eyal C. Attar, M.D.
Medical Monitor:	[redacted]
Protocol Version:	5.0
Replaces:	4.0 / 28 September 2020
Date:	28 October 2021

CONFIDENTIAL

INVESTIGATOR'S STATEMENT

1. I have carefully read this protocol entitled "Phase I Study of APR-246 in Combination with Venetoclax and Azacitidine in *TP53*-Mutant Myeloid Malignancies" and agree that it contains all the necessary information required to conduct the study. I agree to conduct this study as outlined in the protocol.
2. I understand that this study will not be initiated without approval of the appropriate Institutional Review Committee/Independent Ethics Committee (IRB/IEC), and that all administrative requirements of the governing body of the Institution will be complied with fully.
3. Informed written consent will be obtained from all participants in accordance with institutional guidelines, FDA requirements as specified in Title 21 CFR, Part 50, the European Union Directive 2001/20/EC and its associated Detailed Guidances, European Union GCP Directive 2005/28/EC, the International Counsel for Harmonisation (ICH) Guideline for Good Clinical Practice (GCP), Section 4.8, and the terms of the Declaration of Helsinki (2013).
4. I will enroll participants who meet the protocol criteria for entry.
5. I understand that my signature on each completed Case Report Form (CRF) indicates that I have carefully reviewed the complete set of CRFs and accept full responsibility for the contents thereof.
6. I understand that the information presented in this study protocol is confidential, and I hereby assure that no information based on the conduct of the study will be released without prior consent from the Sponsor unless this requirement is superseded by the Food and Drug Administration (FDA), a Competent Authority of the European Union or another Regulatory Authority.

Protocol Version 5.0: 28 October 2021

Name: _____ Telephone: _____

_____ _____

Address: _____

_____

Signature: _____ Date: _____

Sponsor/Representative:

S[redacted] _ [redacted]

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CLINICAL STUDY SYNOPSIS

<table>
<tr><td>Title</td><td>Phase I of APR-246 in Combination with Venetoclax and/or Azacitidine in Myeloid Malignancies</td></tr>
<tr><td>Sponsor</td><td>Aprea Therapeutics</td></tr>
<tr><td>Monitor/Contract Research Organization (CRO)</td><td>[redacted]</td></tr>
<tr><td>Number of Study Centers</td><td>Multicenter</td></tr>
<tr><td>Clinical Phase</td><td>I</td></tr>
<tr><td>Investigational Agent</td><td>APR-246</td></tr>
<tr><td>Study Design</td><td>This clinical trial is a Phase I, open-label, dose-finding and cohort expansion study to determine the safety and preliminary efficacy of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies. The study includes a safety lead-in dose-finding portion followed by expansion portion (Table 1).
Table 1. Study Treatment and Prior Therapy by Cohort
<table>
<tr><th>Cohort</th><th>Disease</th><th>Study Treatment</th><th colspan="6">Prior Therapy Permitted</th></tr>
<tr><th></th><th></th><th></th><th>VEN</th><th>HMA</th><th>LDAC</th><th>APR-246</th><th>SCT</th><th>IC</th></tr>
<tr><td colspan="9">Safety Lead-In Cohorts</td></tr>
<tr><td>Cohort 1</td><td>F/L AML</td><td>APR-246 + VEN</td><td>no</td><td>≥1 prior for MDS required</td><td>no</td><td>no</td><td>yes for MDS</td><td>no</td></tr>
<tr><td>Cohort 2</td><td>F/L AML</td><td>APR-246 + VEN + AZA</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td></tr>
<tr><td colspan="9">Expansion Cohorts</td></tr>
<tr><td>Cohort 1</td><td>F/L AML</td><td>APR-246 + VEN</td><td>no</td><td>yes for MDS</td><td>no</td><td>no</td><td>yes for MDS</td><td>no</td></tr>
<tr><td>Cohort 2</td><td>F/L AML</td><td>APR-246 + VEN + AZA</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td></tr>
<tr><td>Cohort 3</td><td>R/R AML</td><td>APR-246 + VEN</td><td>yes</td><td>yes</td><td>yes</td><td>yes for MDS</td><td>yes</td><td>yes</td></tr>
<tr><td>Cohort 4</td><td>R/R AML</td><td>APR-246 + VEN + AZA</td><td>yes</td><td>no</td><td>no</td><td>yes for MDS</td><td>yes</td><td>≤1 prior</td></tr>
<tr><td>Cohort 5</td><td>F/L AML</td><td>APR-246 + AZA</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td><td>no</td></tr>
</table></td></tr>
</table>

Abbreviations: AZA = azacitidine; F/L = front line; HMA = hypomethylating agent; IC = intensive chemotherapy; LDAC = low-dose cytarabine; MDS = myelodysplastic syndrome; R/R = refractory or relapsed; SCT = stem cell transplant; VEN = venetoclax

During the safety lead-in portion of the study, two parallel cohorts will independently enroll patients following a 3 + 3 design, as outlined in Figure 1.

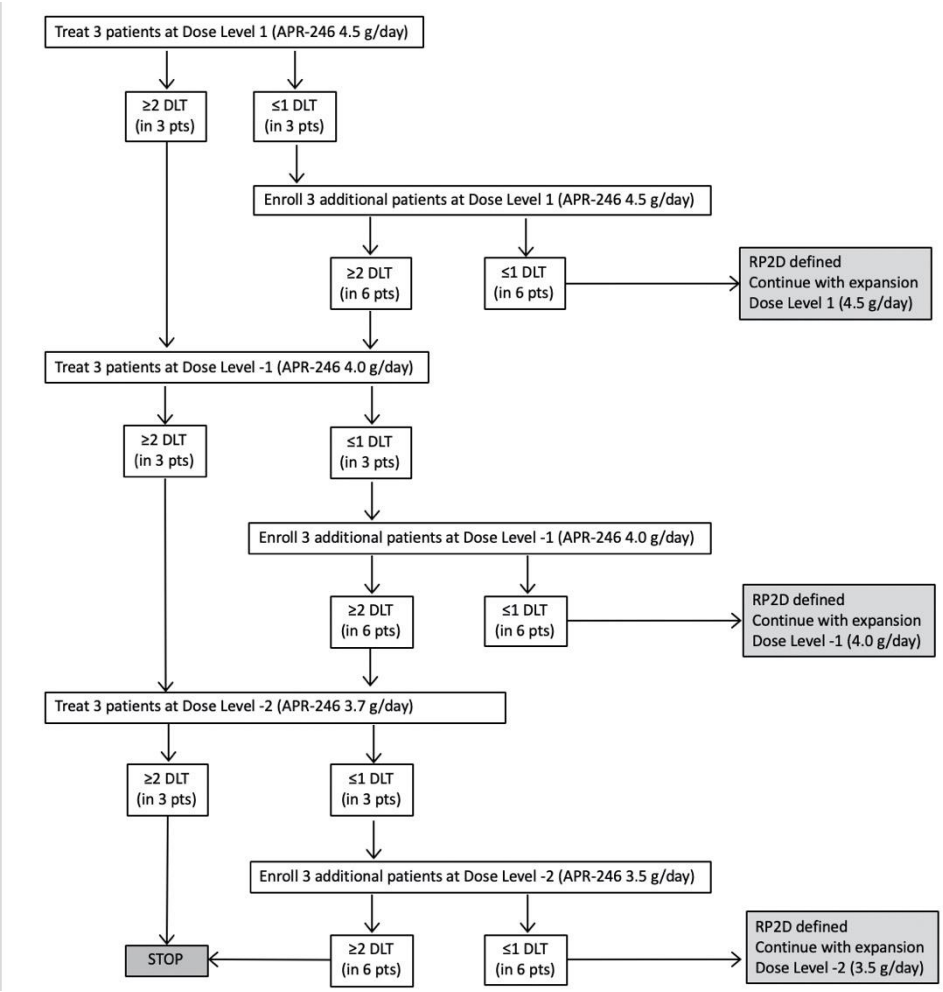

Figure 1. Dose-Finding Study Design

Dose-finding study design will be applied to each cohort of the safety lead-in portion (Figure 1). Each cohort will enroll up to a maximum of 6 patients. Dose-limiting toxicity (DLT) will be assessed after three patients have been enrolled in respective cohort and the last enrolled patient has completed the 4-week safety assessment period (i.e., one cycle of combination regimen). A patient that discontinues therapy during Cycle 1 without DLT is considered evaluable for the purpose of safety only if at least 80% of scheduled doses of APR-246 in combination with venetoclax with and without azacitidine were administered in the first cycle. At the first dose level of 4.5 g/day of APR-246, if ≤1 patient out of 3 experiences a DLT, 3 additional patients will be enrolled. If ≤1 patient out of 6 experiences DLT, the dose level (4.5 g/day of APR-246) will be deemed the recommended Phase II dose (RP2D) for that cohort. If ≥2 patients out of the total 3 – 6 patients in the cohort experience DLT, the study will

	continue enrollment at Dose Level -1 (4.0 g/day of APR-246). If ≤1 patient out of 6 experiences DLT at this dose level, the dose level (4.0 g/day of APR-246) will be deemed the RP2D for that cohort. If ≥2 patients out of the total 3 – 6 patients at that dose level experience DLT, the study will continue enrollment at Dose Level -2 (3.5 g/day of APR-246). If ≤1 patient out of 6 experiences DLT at this dose level, the dose level (3.5 g/day of APR-246) will be deemed the RP2D for that cohort. If ≥2 patients out of the total 3 – 6 patients at this dose level experience DLT, no additional patients will be enrolled at this dose. The trial will be halted, and the Data Review Team (DRT) will consider potential future dosing modifications. The expansion portion will begin once the recommended Phase II dose (RP2D) of APR-246 in combination with venetoclax and azacitidine has been determined in order to assess the antitumor activity of these combinations. Approximately 70 patients with *TP53*-mutant myeloid malignancies will be enrolled in up to five cohorts, as outlined in Table 1. The two Data Review Meetings (DRMs) to assess Safety Lead-in Cohort 2 were held on 27 February 2020 (first cohort of 3 patients in Cohort 2) and 26 June 2020 (second cohort of 3 patients at Dose Level 1 in Cohort 2). After review of the 6 patients with AML treated for at least 1 cycle with the combination of APR-246, venetoclax, and azacitidine, it was concluded by the DRT that no DLTs were observed, and that the RP2D of APR-246 in this combination is 4.5 g/day. The two DRMs to assess Safety Lead-in Cohort 1 were held on 17 March 2020 and 7 August 2020. After review of the 6 patients with AML treated for at least 1 cycle with the combination of APR-246 and venetoclax, it was concluded by the DRT that no DLTs were observed, and that the RP2D of APR-246 in this combination is 4.5 g/day.
Study Objectives	**Primary objective:** To evaluate the safety and tolerability of administration of APR-246 in combination with venetoclax and/or azacitidine in patients with *TP53*-mutant myeloid malignancies. **Secondary objectives:** 1. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of complete remission (CR) rate. 2. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of CR plus complete remission with incomplete hematologic recovery (CRi) rate. 3. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of CR plus complete remission with partial hematologic recovery (CRh) rate. 4. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of overall response rate (ORR). 5. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of progression-free survival (PFS).

	6. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of transition rate to hematopoietic stem cell transplant (HSCT). 7. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of overall survival (OS). 8. To assess clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determining the rate of red blood cell (RBC) and/or platelet transfusion independence (TI) for at least 56 days. 9. To determine the pharmacokinetic profile of APR-246 and venetoclax. **Exploratory objectives:** 1. To investigate molecular biomarkers for response prediction and disease monitoring in baseline and serial bone marrow or blood samples.
Study Endpoints	**Primary endpoints:** 1. DLTs, classified and graded according to the National Cancer Institute's Common Terminology Criteria for Adverse Events (NCI CTCAE, version 5.0). 2. Frequency of treatment-emergent adverse events (TEAEs), and serious adverse events (SAEs) related to APR-246 in combination with venetoclax and/or azacitidine during the trial. 3. The RP2D of APR-246 (the dose at which <2 out of 6 patients experience dose-limiting toxicity during the safety assessment period). **Secondary endpoints:** 1. CR rate, defined as the proportion of patients who achieve CR. 2. CR + CRi rate, defined as the proportion of patients who achieve CR or CRi. 3. CR + CRh rate, defined as the proportion of patients who achieve CR or CRh. 4. Overall response rate (ORR), defined as the proportion of patients achieving CR, complete remission with incomplete hematologic recovery (CRi), complete remission with incomplete platelet recovery (CRp), partial remission (PR) and morphologic leukemia-free state (MLFS) by the IWG 2003 criteria (APPENDIX I). The complete remission with partial hematologic recovery (CRh) rate will also be determined and is defined as bone marrow blasts <5%, ANC >0.5 × 10^9/L and platelets >50 × 10^9/L. 5. Overall survival (OS), measured from the date of initiating study treatment to the date of death. Patients who have not died by the analysis data cut-off date will be censored at their last date of contact. 6. Rate of red blood cell (RBC) and/or platelet transfusion independence (TI) for at least 56 days. 7. Progression-free survival (PFS), defined from the date of initiating study treatment to the date of disease progression or death as a result of any cause. 8. Proportion of patients who transition to HSCT. 9. Pharmacokinetic parameters: maximum concentration (C_{max}), area under curve (AUC), V_d and clearance (CL) of APR-246 and time of maximum concentration (T_{max}), C_{max} and AUC of venetoclax.

	Exploratory endpoints: 1. Exploratory molecular analyses may include, but are not limited to: *TP53* VAF by NGS, p53 immunohistochemistry (IHC), mutations in other genes by NGS, RNA expression.
Eligibility Criteria for Patients in Safety Lead-In Cohorts	**Inclusion Criteria**: 1. Signed informed consent form (ICF) and ability to comply with protocol requirements. 2. Documented diagnosis of initial or relapsed AML according to World Health Organization (WHO) classification. 3. Adequate organ function as defined by the following laboratory values: a. Creatinine clearance >30 mL/min (by Cockcroft-Gault method; APPENDIX II), b. Total serum bilirubin <1.5 × ULN unless due to Gilbert's syndrome, leukemic organ involvement, hemolysis or considered an effect of regular blood transfusions, c. Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) <3 × ULN, unless due to leukemic organ involvement. 4. Age ≥18 years at the time of signing the ICF. 5. At least one *TP53* mutation which is not benign or likely benign. 6. Eastern Cooperative Oncology Group (ECOG) performance status of 0, 1 or 2 (APPENDIX III). 7. Projected life expectancy of ≥12 weeks. 8. Negative serum or urine pregnancy test prior to study treatment initiation in female patients of childbearing potential. 9. Females of childbearing potential and males with female partners of childbearing potential must be willing to use an effective form of contraception such as latex condom, hormonal birth control, intrauterine device or double barrier method (APPENDIX IV) during chemotherapy treatment and for at least six months thereafter. **Exclusion Criteria:** 1. Prior treatment for *TP53*-mutant acute myeloid leukemia (AML) not permitted per Table 1. 2. Known history of human immunodeficiency virus (HIV) or active hepatitis B or active hepatitis C infection. 3. Any of the following cardiac abnormalities: a. Myocardial infarction within six months prior to registration; b. New York Heart Association Class III or IV (APPENDIX V) heart failure or known left ventricular ejection fraction (LVEF) <40%; c. A history of familial long QT syndrome; d. Symptomatic atrial or ventricular arrhythmias not controlled by medications; e. QTcF ≥470 msec, unless due to underlying bundle branch block and/or pacemaker and with approval of the medical monitor. 4. Concomitant malignancies for which patients are receiving active therapy at

	the time of signing consent. For example, patients with adequately treated basal or squamous cell carcinoma of the skin, adequately treated carcinoma *in situ* (e.g. cervix), or breast cancer receiving adjuvant endocrine therapy may enroll irrespective of the time of diagnosis with Medical Monitor approval. 5. Known active CNS involvement from AML. A diagnostic lumbar puncture is not required in the absence of suspicion for CNS disease. 6. Malabsorption syndrome or other condition that precludes enteral route of administration. 7. Pregnancy or lactation. 8. Active uncontrolled systemic infection (viral, bacterial or fungal).
Eligibility Criteria for Patients in Expansion Cohorts	**<u>Inclusion Criteria</u>**: 1. Signed ICF and ability to comply with protocol requirements. 2. Newly diagnosed or R/R AML according to WHO classification with permitted prior therapy, as per Table 1. For patients with newly diagnosed AML bone marrow and/or blood evidence of AML with ≥20% blasts is required. For patients with newly diagnosed AML, prior therapy with HMA and/or any chemotherapeutic agent for MDS is excluded. 3. Adequate organ function as defined by the following laboratory values: a. Creatinine clearance >30 mL/min (by Cockcroft-Gault method; APPENDIX II), b. Total serum bilirubin <1.5 × ULN unless due to Gilbert's syndrome, leukemic organ involvement, hemolysis or considered an effect of regular blood transfusions, c. ALT and AST <3 × ULN, unless due to leukemic organ involvement. 4. Age ≥18 years at the time of signing the ICF. 5. At least one *TP53* mutation which is not benign or likely benign, based on central *TP53* testing results. If local *TP53* mutation and/or p53 IHC results become available before central TP53 mutation results, enrollment may be made based on local results with Medical Monitor approval. 6. ECOG performance status of 0, 1 or 2 (APPENDIX III). 7. Projected life expectancy of ≥12 weeks. 8. Negative serum pregnancy test prior to study treatment initiation in female patients of childbearing potential. 9. Females of childbearing potential and males with female partners of childbearing potential must be willing to use an effective form of contraception such as latex condom, hormonal birth control, intrauterine device or double barrier method (APPENDIX IV) during chemotherapy treatment and for at least six months thereafter. **<u>Exclusion Criteria</u>:** 1. Known history of myeloproliferative neoplasm (MPN) (i.e., polycythemia vera, essential thrombocythemia and primary myelofibrosis are excluded). 2. Known history of HIV or active hepatitis B or active hepatitis C infection. 3. Any of the following cardiac abnormalities: a. Myocardial infarction within six months prior to registration,

	b. New York Heart Association Class III or IV (APPENDIX V) heart failure or known left ventricular ejection fraction (LVEF) <40%, c. A history of familial long QT syndrome, d. Symptomatic atrial or ventricular arrhythmias not controlled by medications, e. QTcF ≥470 msec, unless due to underlying bundle branch block and/or pacemaker and with approval of the medical monitor. 4. Concomitant malignancies for which patients are receiving active therapy at the time of signing consent. For example, patients with adequately resected basal or squamous cell carcinoma of the skin, adequately resected carcinoma *in situ* (e.g. cervix), or breast cancer receiving adjuvant endocrine therapy may enroll irrespective of the time of diagnosis with Medical Monitor approval. 5. Prior exposure to anticancer therapies including chemotherapy, radiotherapy or other investigational therapy, including targeted small molecule agents within 14 days of the first day of study treatment or within 5 half-lives prior to first dose of study treatment. 6. Prior exposure to biologic agents (e.g. monoclonal antibodies) for anti-neoplastic intent within 14 days prior to first dose of study drug. 7. Known active CNS involvement from AML. A diagnostic lumbar puncture is not required in the absence of suspicion for CNS disease. 8. Malabsorption syndrome or other condition that precludes enteral route of administration. 9. Pregnancy or lactation. 10. Active uncontrolled systemic infection (viral, bacterial or fungal).
Treatment Plan	Treatment may be administered on an outpatient basis. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's disease. Hydroxyurea may be used for control of leukocytosis. The study includes a safety lead-in dose-finding portion followed by the expansion portion, as outlined in Table 1. During the safety lead-in portion of the study, two cohorts will independently enroll patients following a 3 + 3 design. • Safety lead-in cohort 1 will enroll patients with *TP53*-mutant AML, who have received at least one prior HMA regimen for MDS. These patients will receive APR-246 at 4.5 g/day on days 1–4 of each 28-day cycle administered concurrently with venetoclax given orally at the dose of 400 mg daily. • Safety lead-in cohort 2 will enroll patients with previously untreated *TP53*-mutant AML. These patients will receive APR-246 at 4.5 g/day on days 1–4 administered concurrently with venetoclax given orally at the dose of 400 mg daily and with azacitidine at the standard dose of 75 mg/m^2 over 7 consecutive days as a subcutaneous injection or IV infusion, on days 1–7 of each 28-day cycle. Each cohort will enroll up to 6 patients. For dose-finding strategy please consult Study Design section of the Clinical Study Synopsis.

	The expansion portion will begin once the RP2D of APR-246 in combination with venetoclax and/or azacitidine has been determined in order to assess the antitumor activity of these combinations. Approximately 70 patients with *TP53*-mutant myeloid malignancies will be enrolled in up to five cohorts, as outlined in Table 1. In each portion, patients may continue treatment as long as toxicity remains acceptable and the patient has not withdrawn consent. Response will be assessed based on the International Working Group (IWG) AML response criteria (APPENDIX I) after every treatment cycle the first year, then every two cycles.
Duration of Follow-Up	Patients will be followed as per the Study Calendar. After a patient is removed or withdrawn from study treatment, the patient will be followed until death or withdrawal of consent for study participation. Off-treatment data on overall survival will be updated every month (i.e. every 28 days ±3 days) or until death or withdrawal of consent for study participation, whichever occurs first. If a patient is removed from the study due to unacceptable adverse events (AEs), the event(s) will be followed until resolution or stabilization of the adverse event. Patients who respond and discontinue study treatment for reasons other than PD should have response assessments and survival should be collected every month until relapse, death or withdrawal of consent for study participation, whichever occurs first. After relapse, data for survival should be collected every month (i.e. every 28 days ±3 days) until death or withdrawal of consent for study participation, whichever occurs first. *Criteria for Removal from Study Treatment* Study treatment can continue for patients receiving clinical benefit, unless one or more withdrawal criteria are met, or at the patient's discretion, or if the study is terminated. 1. Study Treatment Discontinuation Study treatment must be discontinued if: • Evidence of disease progression. Patients who have relapsed or progressive disease but who are continuing to derive clinical benefit in the opinion of the investigator may continue to receive study treatment. • A patient becomes pregnant. • A patient is significantly non-compliant with the requirements of the protocol. • A patient has an adverse experience that would, in the Investigator's judgment, make continued participation in the study an unacceptable risk. • The patient starts new treatment for their underlying disease. 2. Patient Withdrawal from Study Treatment If the patient is permanently withdrawn from study treatment, but does not withdraw consent from the Study, the Investigator must make every effort to have the patient complete all withdrawal assessments at the time of withdrawal and complete all scheduled follow-up visits.

	3. Study Completion Patient must be taken off the study if: • The patient dies during the study. • The patient is lost to follow-up. • The patient withdraws consent. 4. Patient Withdrawal from Study A patient may voluntarily withdraw from study treatment or withdraw consent from the study at any time. The investigator may also, at his or her discretion, discontinue a patient from participating in the Study Treatment at any time. The Investigator and/or designated staff will record the date and the reason for patient withdrawal from the study treatment and/or study participation, as applicable.
Statistics	This is a Phase I, open-label, dose-finding and cohort expansion study to determine the safety and preliminary efficacy of APR-246 in combination with venetoclax and/or azacitidine in patients with AML. The RP2D of APR-246 will be defined as the dose at which <2 out of 6 patients experience DLT during the 4-week safety assessment period after administration of APR-246 in combination with venetoclax and/or azacitidine. *Definition of Dose-Limiting Toxicity (DLT)* An event will be considered a DLT per NCI CTCAE version 5.0 criteria if it occurs within the 4-week safety assessment period (Cycle 1 of study treatment) and is not attributable to the underlying disease. DLT is defined as: • Absolute neutrophil count (ANC) not recovering to >0.5 × 10^9/L and/or platelets not recovering to >25 × 10^9/L by day 42 of study treatment in the absence of active leukemia or myelodysplasia; • Grade ≥ 3 nausea/vomiting/diarrhea or CNS toxicity that does not resolve to Grade ≤ 1 within 7 days despite treatment interruption and/or maximal medical therapy; • Treatment-related non-hematological Grade ≥ 3 toxicity that does not resolve to Grade ≤ 1. A DLT will be considered related to the study treatment unless there is a clear, alternative explanation for the AE. A ≥ Grade 3 metabolic or electrolyte abnormalities that is not clinically significant and is adequately controlled within 72 hours is not to be considered DLT. Tumor lysis syndrome (TLS) that responds to therapy and resolves within 72 hours is not considered DLT. Additionally, AEs that meet the above criteria, but occur after the DLT evaluation period will not be defined as DLTs, but will be reported as AEs/Serious Adverse Events (SAEs) and will be reviewed across all cohorts during the study to help determine the AE profile. A patient that discontinues therapy during Cycle 1 without DLT is considered evaluable for the purpose of safety only if at least 80% of scheduled Cycle 1 dose of APR-246 in combination with venetoclax and/or azacitidine were administered in the first cycle.

DRT consisting of the Medical Monitor, Site Principal Investigators, and other clinical research personnel that the Sponsor may deem appropriate, will hold DRM on an interim basis at a frequency dependent on study accrual. At these meetings, the DRT will review AEs and DLT and make recommendations regarding the RP2D. In the expansion portion of the study, the DRT will evaluate safety and tolerability after 5 patients have completed 1 cycle of treatment in each cohort. All accumulated safety data will be discussed during DRMs.

Determination of Sample Size

This trial assumes a sample size of 12 – 36 patients (6 – 18 patients for each cohort) in the safety lead-in portion of the study and approximately 70 patients in the expansion portion of the study.

Analysis Populations

Safety population: Patients will be evaluable for safety if they receive at least one dose of APR-246 with venetoclax and/or azacitidine. The safety population will be used to summarize exposure and safety parameters.

DLT-evaluable population: All patients who either experienced a DLT during first the 4 weeks (Cycle 1) of the study treatment or received ≥80% of scheduled Cycle 1 dose of APR-246 in combination with venetoclax and/or azacitidine and did not experience a DLT. Any individual patient who is not evaluable for DLT will be replaced by a new patient through additional patient enrollment.

Efficacy evaluable (EE) population: All patients who complete at least one treatment cycle of APR-246 in combination with venetoclax and/or azacitidine and who have at least one post-treatment clinical response assessment. Patients who fail to complete one treatment cycle will also be considered EE if they show clear evidence of clinically significant disease progression. The EE population will be the secondary analysis population for efficacy.

Pharmacokinetics (PK) population: Patients will be evaluable for pharmacokinetics if at least one post-dose sample for PK evaluation has been obtained.

Efficacy Analyses

CR will be summarized for all enrolled patients as the proportion (%) of patients with CR. In addition to presenting the CR rate, its associated exact 95% confidence intervals (CI) for each treatment arm will also be presented. CR rate will not be formally compared between treatment arms.

CR + CRh rate will be summarized as the proportion of patients who achieve CR or CRh. CRh is defined as bone marrow blasts <5%, ANC >0.5 × 10^9/L and platelets >50 × 10^9/L. CR + CRi rate will be summarized as the proportion of patients who achieve CR or CRi. The rate of TI will be summarized as the proportion of patients with RBC and/or platelet TI for at least 56 days.

Duration of response (DoR) is defined as the time from the date when criteria for response are met to the date of relapse or progressive disease (PD) or death due to

	any cause, whichever occurs first. Patients alive with no progressive disease will have their DOR censored at the date of the last clinical assessment. The duration of CR will be summarized in each treatment arm by providing duration of complete response (DOCR) and duration of overall response (DOR), using Kaplan-Meier methodology. DoR endpoints will not be formally compared between treatment arms. Overall response will be summarized in number (%) of patients in each category of responses and ORR will be analyzed by using the similar method as CR rate. Survival data are collected at treatment and follow-up periods. Patients will be followed until death or withdrawal of consent for study participation. Overall survival (OS) is defined as the number of days from the first day of treatment to the date of death. Kaplan-Meier methodology will be utilized. Progression-free survival (PFS) is defined as the time from the first day of treatment to disease progression or death from AML, whichever occurs first. If neither event occurs, PFS will be censored at the date of the last clinical assessment. Kaplan-Meier methodology will be utilized. *Safety Analyses* Safety data including AEs, vital signs, laboratory data, electrocardiogram (ECG), physical exam will be tabulated for the safety population. AEs will be tabulated by System Organ Class (SOC), preferred term, severity, and relationship to treatments. The tabulation of laboratory parameters will include the normal ranges for each parameter. Each value will be classified as falling above, below, or within the normal range. Laboratory parameters will also be tabulated by maximum NCI CTCAE v5.0 severity grade. *Pharmacokinetic Analysis* PK sampling for APR-246 are performed in Cycle 1, on Days 1, 2 and 4, and on Day 4 of every subsequent cycle. PK sampling for venetoclax is performed on Days 1, 2, 22 and 23 of Cycle 1 and on Day 4 of Cycles 2 – 5. Non-compartmental or population pharmacokinetic methods will be used to derive APR-246 and venetoclax PK parameters (C_{max}, T_{max}, AUC, V_d and CL). The pharmacokinetics of APR-246 and venetoclax will be summarized using descriptive statistics (mean, standard deviation, CV% mean, geometric mean, CV% geometric mean) and compared with historical control data. APR-246 AUC and C_{max} will then be tested for association with signs of efficacy and safety. If an observable trend exists, a PK/PD model will be developed to evaluate the exposure-response relationship between APR-246 plasma exposure and outcome measures in the presence of venetoclax. Demographic and clinical data (ethnicity, current age, body weight, sex, disease status, etc.) will be utilized to assess interpatient variability in the PK and PK/PD relationships.

LIST OF ABBREVIATIONS

Abbreviation	Definition
AE	Adverse Event
ALT	Alanine Aminotransferase
AML	Acute Myeloid Leukemia
ANC	Absolute Neutrophil Count
AST	Aspartate Aminotransferase
AUC	Area Under Curve
BUN	Blood Urea Nitrogen
CHF	Congestive Heart Failure
CI	Confidence Interval
C_{max}	Maximum Concentration
CR	Complete Remission
CRF	Case Report Form
CRh	Complete Remission with Partial Hematologic Recovery
CRO	Contract Research Organization
CRp	Complete Remission with Incomplete Platelet Recovery
DLT	Dose-Limiting Toxicity
DOCR	Duration of Complete Response
DOR	Duration of Overall Response
DoR	Duration of Response
DRM	Data Review Meeting

DRT	Data Review Team
EC	European Commission
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDTA	Ethylenediaminetetraacetic Acid
EMD	Extramedullary Disease
EOI	End of Infusion
FDA	Food and Drug Administration
GCP	Good Clinical Practice Guidelines
HIV	Human Immunodeficiency Virus
HSCT	Hematopoietic Stem Cell Transplantation
HMA	Hypomethylating Agent
ICF	Informed Consent Form
ICH	International Council for Harmonisation
IEC	Institutional Ethics Committee
IHC	Immunohistochemistry
IMP	Investigational Medicinal Product
IPCW	Inverse Probability of Censoring Weights
IRB	Institutional Review Board
IV	Intravenous
IWG	International Working Group

LBM	Lean Body Mass
LVEF	Left Ventricular Ejection Fraction
µg	Microgram
MDS	Myelodysplastic Syndrome
MLFS	Morphologic Leukemia-Free State
MTD	Maximum Tolerated Dose
NCI CTCAE	National Cancer Institute – Common Terminology Criteria for Adverse Events
NGS	Next-Generation Sequencing
OS	Overall Survival
ORR	Overall Response Rate
PFS	Progression-Free Survival
PK	Pharmacokinetic(s)
PLD	Pegylated Liposomal Doxorubicin
PR	Partial Remission
QTcF	Corrected QT Interval by Fridericia's formula
RA	Refractory Anemia
RAEB	Refractory Anemia with Excess Blasts
RAEB-T	Refractory Anemia with Excess Blasts in Transformation
RP2D	Recommended Phase II Dose
SAE	Serious Adverse Event

SAR	Suspected Adverse Reaction
SCT	Stem Cell Transplantation
SOC	System Organ Class
TEAE	Treatment-Emergent Adverse Event
TI	Transfusion Independence
tid	Three Times Daily (*Ter in Die*)
TLS	Tumor Lysis Syndrome
TrxR1	Thioredoxin Reductase 1
ULN	Upper Limit of Normal
USPI	US Prescribing Information
VAF	Variant Allele Frequency
V_d	Volume of Distribution
WHO	World Health Organization

1.0 GENERAL INFORMATION

1.1 Protocol Number and Title of the Study

Protocol No. A19-11184
Protocol Title: A Phase I Study to Evaluate Safety and Tolerability of APR-246 in Combination with Venetoclax and Azacitidine in Patients with Myeloid Malignancies

1.2 Sponsor

Aprea Therapeutics, Inc.
[redacted]

1.3 Monitor

[redacted]

1.4 Signature Authorization

The protocol will be signed by Aprea Therapeutics, Inc.

2.0 BACKGROUND INFORMATION

2.1 Introduction

2.1.1 Acute Myeloid Leukemia

AML is a group of hematopoietic neoplasms characterized by a clonal proliferation of myeloid precursors with a reduced capacity to differentiate into mature cells. AML is the most common type of acute leukemia in adults, with an incidence of over 20,000 cases per year in the United States[1–4].

Cytogenetic and mutation testing remain critical prognostic tools to assess prognosis and plan induction and post-remission treatment strategies[5]. Testing peripheral blood and bone marrow samples using next generation sequencing (NGS) has developed into a standard of care for determining molecular mutations. Mutations in the tumor suppressor gene *TP53* are found in approximately 20% of patients with AML and are associated with an adverse prognosis. In addition, approximately 80% of patients with *TP53*-mutant AML have a complex karyotype, which also confers an adverse prognosis[6].

Despite increased understanding of the biology and development of new therapeutic targets, AML treatment remains relatively unchanged, with therapies limited to intensive chemotherapy comprised of cytarabine and anthracycline and non-intensive therapies consisting of a HMA, sometimes incorporating the BCL-2 inhibitor venetoclax. Allogeneic stem cell transplantation is recommended for patients with high risk and relapsed or refractory (R/R) AML who achieve a suitable response and are eligible for SCT.

Non-intensive therapies are utilized to treat patients who are considered ineligible for intensive chemotherapy on the basis of advanced age, comorbidities, and low likelihood of achieving a response based on adverse cytogenetic/molecular features and/or a history of an antecedent hematologic disorder. Non-intensive therapies consist of the HMAs decitabine and azacitidine and low-dose cytarabine (LDAC)[7–10]. Both agents have activity in AML as initial induction therapy and in the relapsed setting[8,9,11]. Recently, HMAs[12] and LDAC[13] have been combined with venetoclax in the initial treatment of AML. In addition, HMAs have been combined with venetoclax to treat patients with R/R AML[14].

2.1.2 *TP53* Mutations in Myeloid Malignancies

TP53 is a tumor suppressor protein that plays a pivotal role in maintaining genomic stability in response to DNA damage. It is mutated in over half of human cancers[15,16], and such mutations represent an important mechanism of resistance to DNA-damaging chemotherapeutic agents[17,18]. A significant proportion of mutations recurrently target 'hot-spot' codons. They either directly disrupt the DNA-binding domain of *TP53* or cause conformational changes of the *TP53* protein, thus leading to severely impaired *TP53* function.

Mutated *TP53* is predominantly observed in therapy-related AML[19,20] and/or in patients with complex karyotype (70-80%)[21]. The reported frequency of *TP53* mutations is 5-10% in *de-novo* AML[22–24]. It has been demonstrated that gain of function mutations in hot spot regions can promote a more aggressive AML[25–28]. In multivariate analysis, the presence of *TP53* mutations predicted for poor overall survival and inferior response to treatment[27,29–32].

In a retrospective analysis of 55 patients who received azacitidine therapy for first line treatment of *TP53*-mutant AML, the CR + CRi rate was 21.8%[33]. Though the remission rate was not significantly different from patients with wild type *TP53*, the median survival was significantly shorter (7.9 vs 12.6 months, p<.001) for patients with *TP53* mutations. A similar median OS of 7.9 months was observed in patients with *TP53*-mutant AML who received initial therapy with azacitidine[34]. Regarding patients with *TP53* mutations who received venetoclax with HMAs, the CR/CRi rate was 47%[12]. In 10 patients who received venetoclax with LDAC, there were no CRs and 3 patients achieved CRi[13]. Responses in later lines are even poorer[14].

Together, these data highlight the poor outcomes of *TP53*-mutant patients, even with recently approved venetoclax-containing therapies, and the important need for the development of novel therapeutic strategies, particularly in this patient population.

2.2 APR-246

[redacted]

2.3 Venetoclax

Venetoclax is a BCL-2 inhibitor that approved by the U.S. FDA for use in combination with HMAs for the treatment of patients with newly diagnosed AML who are 75 years or older or who have comorbidities that preclude use of intensive induction chemotherapy. Accelerated

approval was granted based on the results of the Phase Ib study with HMAs for patients 65 years or older with treatment-naïve AML, ineligible for intensive chemotherapy by age or comorbidities[35]. Sixty-seven percent of patients in the study achieved CR or CR with incomplete count recovery (CRi). The median DoR was 11.3 months, and median overall survival was 17.5 months in all patients. It was shown that treatment with venetoclax in combination with an HMA leads to a decrease in oxidative phosphorylation via disruption of electron transport chain in a glutathione-dependent fashion and selective targeting of the leukemia stem cell population[36].

Per venetoclax US Prescribing Information (USPI)[37] dated May 2020, the most common adverse reactions (≥30%) of any grade in AML patients receiving venetoclax in combination with azacitidine are nausea, diarrhea, constipation, neutropenia, thrombocytopenia, hemorrhage, peripheral edema, vomiting, fatigue, febrile neutropenia, rash, and anemia. Serious adverse reactions were reported in 75% of AML patients receiving venetoclax in combination with azacitidine, with the most frequent serious adverse reactions in ≥5% of patients being febrile neutropenia, pneumonia (excluding fungal), sepsis (excluding fungal), respiratory failure, and multiple organ dysfunction syndrome.

Following multiple doses under fed conditions, maximum plasma concentration of venetoclax are reached 5–8 hours after dose. It is recommended that venetoclax be administered with a meal as food increases the absorption after oral administration. Venetoclax is cleared from systemic circulation via hepatic elimination. The terminal elimination half-life is approximately 26 hours. The pharmacokinetics of venetoclax do not change over time. Venetoclax is predominantly metabolized by CYP3A4/5. CYP3A4 inhibitors and inducers as well as P-gp inhibitors cause clinically relevant drug-drug interactions with venetoclax. For the same reason certain food must be avoided: grapefruit, grapefruit juice, Seville oranges (often used in marmalades), starfruit (CYP3A inhibiting), and St John's wort (CYP3A inducing). Based on their metabolic profiles, a pharmacokinetic drug-drug interaction of venetoclax with APR-246 is considered unlikely.

2.4 Azacitidine

Azacitidine is a nucleoside metabolic inhibitor indicated by the U.S. FDA for the treatment of patients with the following French-American-British (FAB) MDS subtypes: refractory anemia (RA) or refractory anemia with ringed sideroblasts (if accompanied by neutropenia or thrombocytopenia or requiring transfusions), refractory anemia with excess blasts (RAEB), refractory anemia with excess blasts in transformation (RAEB-T), and chronic myelomonocytic leukemia (CMML).

In 2008, azacitidine was approved by the European Medicines Agency (EMA) for the treatment of AML patients with 20–30% bone marrow blasts, older than 64 years and who are ineligible for HSCT. EMA approval of azacitidine was expanded on 30 October 2015 to include AML patients with >30% bone marrow blasts.

The recommended starting dose for the first treatment cycle, for all patients regardless of baseline hematology laboratory values, is 75 mg/m^2 subcutaneously or intravenously, daily for 7 days. Cycles should be repeated every 28 days. Treatment may be continued as long as the patient derives clinical benefit.

Azacitidine is rapidly absorbed after subcutaneous or IV administration with a V_d of 76 ± 26 L and a half-life of 41 ± 8 minutes. C_{max} is reached after about 30 minutes. Azacitidine and its metabolites are known to be substantially excreted by the kidney, and the risk of adverse reactions to this drug may be greater in patients with impaired renal function. Urinary excretion is the primary route of elimination; the cumulated urinary excretion is 85% of radioactive dose with less than 1% recovered in feces over 3 days.

Azacitidine undergoes spontaneous hydrolysis in aqueous solution and is deaminated by cytidine deamidases, whereas APR-246 is glucuronidated. Drug-drug interaction is not likely although both compounds and their degradation products and metabolites are primarily eliminated by urinary excretion. The pharmacokinetic interaction between APR-246 and azacitidine will be investigated in an ongoing parallel trial.

2.5 Combination of Venetoclax with Azacitidine

Venetoclax has demonstrated synergistic activity in combination with azacitidine in preclinical models[38]. This relationship was supported by clinical data from a large, multicenter, Phase Ib study (NCT02203773) in elderly patients with treatment-naive AML, ineligible for intensive chemotherapy[12]. Promising efficacy and a tolerable safety profile of this novel combination, with a high CR + CRi rate of 73% in the 400-mg venetoclax in combination with HMA (azacitidine or decitabine) cohort (P=0.35), low early mortality rates, and OS extending beyond 17 months. The median duration of CR + CRi for the venetoclax 400 mg in combination with HMA was 12.5 months (95% CI). Patients with *TP53* mutations achieved a CR/CRi rate of 47%[22,39]. Importantly, the best response of CR + CRi has been attained at median of 1.8 months compared with 3.5 months with azacitidine alone[40]. Response rates were similar between the azacitidine and decitabine cohorts (76% vs 71%, respectively). A Phase III, randomized, double-blind, placebo-controlled study of venetoclax 400 mg combined with azacitidine in treatment-naïve elderly and adult patients with AML ineligible for standard induction therapy is currently underway (NCT02993523) and aims to evaluate the potential impact of venetoclax with azacitidine compared to azacitidine alone [redacted]

2.6 APR-246 Preclinical Studies

2.6.1 Pharmacology and Mode of Action

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]

2.6.3 Pharmacokinetics and Metabolism in Animals

[redacted]

[redacted]

[redacted]

[redacted]

2.7 APR-246 Clinical Studies

[redacted]

2.7.1 Phase I/II Study in Solid and Hematological Malignancies

[redacted]

2.7.2 Phase I/II Study in *TP53*-Mutant Advanced Ovarian Cancer

[redacted]

[redacted]

2.7.3 Phase I/Ib Study in Refractory Hematologic Malignancies

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]

2.7.5 Phase III Multicenter, Randomized, Open Label Study of APR-246 in Combination with Azacitidine

[redacted]

[redacted]

[redacted]

2.7.6 APR-246 CNS Safety Overview

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]

2.8 Dose Rationale for APR-246 in Combination with Venetoclax and/or Azacitidine

[redacted]

2.8.1 APR-246 and Azacitidine

[redacted]

2.8.2 Venetoclax and Azacitidine

The doses of venetoclax and azacitidine for patients receiving doublet therapy (APR-246 and venetoclax) or triplet therapy (APR-246, venetoclax and azacitidine) are consistent with the doses recommended in the venetoclax USPI[48] for patients with AML.

2.8.3 Venetoclax and APR-246

[redacted]

[redacted]

[redacted]

[redacted]

2.8.4 Expansion Cohorts in Front Line AML

[redacted]

[redacted]

[redacted]

[redacted]

2.9.2 Potential Benefits

Patients with *TP53*-mutant myeloid malignancies, including AML, have a poor prognosis and unmet medical need. Azacitidine and venetoclax represent standard of care agents for *TP53*-mutant AML. APR-246 in combination with azacitidine has demonstrated compelling clinical efficacy and a manageable safety profile in patients with *TP53*-mutant MDS/AML, and APR-246 has demonstrated *in vitro* synergy with azacitidine and venetoclax. The purpose of this clinical trial is to define the safety and tolerability of APR-246 in combination with venetoclax and/or azacitidine in patients with *TP53*-mutant AML and to determine preliminary efficacy. Given that *TP53*-mutant MDS and AML patients have a limited life expectancy and lack of effective treatments, the benefit/risk assessment supports the use of APR-246 in this trial.

2.10 Characteristics of a Well-Conducted Trial

The following characteristics of an adequate and well-conducted trial will be implemented:

1. The Investigators will be well qualified by scientific training and experience.
2. Detailed CRFs will be completed for every patient.
3. Requirements for institutional ethics review as set forth by the appropriate Institutional Review Board/Independent Ethics Committee (IRB/IEC), Title 21 Code of Federal Regulations (CFR) Part 56, the European Union Directive 2001/20/EC and its associated Detailed Guidances, European Union GCP Directive 2005/28/EC, the ICH Guideline for GCP, Sections 3 and 4, and the terms of the Declaration of Helsinki

(2013), will be followed.

4. Requirements for informed consent in accordance with institutional guidelines, FDA requirements as specified in Title 21 CFR, Part 50, the European Union Directive 2001/20/EC and its associated Detailed Guidances, European Union GCP Directive 2005/28/EC, the ICH Guideline for GCP, Section 4.8, and the terms of the Declaration of Helsinki (2013), will be followed.
5. Safety data will be recorded and evaluated.
6. Routine monitoring visits will be conducted by the Sponsor's representative to ensure data accuracy.
7. Drug accountability will be strictly maintained.
8. This trial will be conducted according to GCP, the protocol and applicable regulatory requirements.

2.11 Patient Population

This study will enroll adult male and female patients of age ≥ 18 years with documented diagnosis of AML, according to WHO classification, and documented *TP53* mutation, which is not benign or likely benign, who also meet the eligibility requirements of this protocol.

3.0 TRIAL OBJECTIVES AND PURPOSE

Primary objective:
To evaluate the safety and tolerability of administration of APR-246 in combination with venetoclax and/or azacitidine in patients with *TP53*-mutant myeloid malignancies.

Secondary objectives:

1. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of CR rate.
2. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of CR plus CRi rate.
3. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of CR plus CRh rate.
4. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with myeloid malignancies by determination of ORR.
5. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of PFS.
6. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of transition rate to HSCT.
7. To assess the clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determination of OS.

8. To assess clinical activity of APR-246 in combination with venetoclax and/or azacitidine in patients with AML by determining the rate of RBC and/or platelet TI for at least 56 days.
9. To determine the pharmacokinetic profile of APR-246 and venetoclax.

Exploratory objectives:

1. To investigate molecular biomarkers for response prediction and disease monitoring in baseline and serial bone marrow or blood samples.

4.0 TRIAL DESIGN

4.1 Overview of Trial Design

Study treatment may be administered on an outpatient basis. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's disease. Hydroxyurea may be used for control of leukocytosis.

The study includes a safety lead-in dose-finding portion followed by expansion portion. During the safety lead-in portion of the study, two cohorts will independently enroll patients following a 3 + 3 design.

Safety lead-in cohort 1 will enroll patients with AML or MDS who have received at least one prior HMA regimen for MDS. These patients will receive APR-246 at 4.5 g/day on days 1 – 4 of each 28-day cycle administered concurrently with venetoclax that is given orally at the dose of 400 mg daily once the ramp-up phase is completed. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax. Safety lead-in cohort 2 will enroll patients with previously untreated *TP53*-mutant AML (prior MDS is allowed, but not treatment with HMA), who will receive APR-246 at 4.5 g/day on days 1 – 4 administered concurrently with venetoclax given orally at the dose of 400 mg daily and with azacitidine at the standard dose of 75 mg/m^2 over 7 consecutive days as a subcutaneous injection or IV infusion on days 1 – 7 of each 28-day cycle.

Each cohort will enroll up to 6 patients. DLT will be assessed after three patients have been enrolled in respective cohort and the last enrolled patient has completed the 4-week safety assessment period (i.e., one cycle of combination regimen). A patient that discontinues therapy during Cycle 1 without DLT is considered evaluable for the purpose of safety only if at least 80% of scheduled doses of APR-246 in combination with venetoclax and/or azacitidine were administered in the first cycle. At the first dose level of 4.5 g/day of APR-246, if ≤1 patient out of 3 experiences a DLT, 3 additional patients will be enrolled. If ≤1 patient out of 6 experiences DLT, the dose level (4.5 g/day of APR-246) will be deemed the RP2D for that cohort. If ≥2 patients out of the total 3–6 patients in the cohort experience DLT, the study will continue enrollment at Dose Level -1 (4.0 g/day of APR-246). If ≤1 patient out of 6 experiences DLT at this dose level, the dose level (4.0 g/day of APR-246) will be deemed the

RP2D for that cohort. If ≥2 patients out of the total 3 – 6 patients at that dose level experience DLT, the study will continue enrollment at Dose Level -2 (3.5 g/day of APR-246). If ≤1 patient out of 6 experiences DLT at this dose level, the dose level (3.5 g/day of APR-246) will be deemed the RP2D for that cohort. If ≥2 patients out of the total 3–6 patients at this dose level experience DLT, no additional patients will be enrolled at this dose. The trial will be halted, and the DRT will consider potential future dosing modifications.

Table 2. APR-246 Dose Levels

Dose Modification	APR-246 Dose
Starting Dose Level (DL)	APR-246 4.5 g/day 1.5 g (for first 45 minutes) + 3.0 g (for 5 hours 15 minutes)
Dose Level Reduction -1 (DL-1)	APR-246 4.0 g/day 1.33 g (for first 45 minutes) + 2.67 g (for 5 hours 15 minutes)
Dose Level Reduction -2 (DL-2)	APR-246 3.5 g/day 1.17 g (for first 45 minutes) + 2.33 g (for 5 hours 15 minutes)

The expansion portion will begin once the RP2D of APR-246 in combination with venetoclax and/or azacitidine have been determined in order to assess the antitumor activity of these combinations. Approximately 70 patients will be enrolled in up to five cohorts, as outlined in Table 3:

Table 3. Study Treatment and Prior Therapy by Cohort

Cohort	Disease	Study Treatment	Prior Therapy Permitted					
			VEN	HMA	LDAC	APR-246	SCT	IC
Safety Lead-In Cohorts								
Cohort 1	F/L AML	APR-246 + VEN	no	≥ 1 prior for MDS required	no	no	yes for MDS	no
Cohort 2	F/L AML	APR-246 + VEN + AZA	no	no	no	no	no	no
Expansion Cohorts								
Cohort 1	F/L AML	APR-246 + VEN	no	yes for MDS	no	no	yes for MDS	no

Cohort 2	F/L AML	APR-246 + VEN + AZA	no	no	no	no	no	no
Cohort 3	R/R AML	APR-246 + VEN	yes	yes	Ye s	yes for MDS	yes	yes
Cohort 4	R/R AML	APR-246 + VEN + AZA	yes	no	no	yes for MDS	yes	≤ 1 prior
Cohort 5	F/L AML	APR-246 + AZA	no	no	no	no	no	no

Abbreviations: AZA = azacitidine; F/L = front line; HMA = hypomethylating agent; IC = intensive chemotherapy; LDAC = low-dose cytarabine; MDS = myelodysplastic syndrome; R/R = refractory or relapsed; SCT = stem cell transplant; VEN = venetoclax

Cytopenia and related adverse events (thrombocytopenia, anemia, neutropenia, and febrile neutropenia) are common in patients with AML, particularly in patients with baseline neutropenia or those with secondary AML.

For patients receiving venetoclax in combination with azacitidine, in a case of CRi or a morphologic leukemia-free bone marrow (MLFS) after completion of Cycle 1, administration of venetoclax and APR-246 may be interrupted to allow for ANC recovery from Day 29 until ANC $\geq 0.5 \times 10^9$/L. Blood counts can be assessed within 4 weeks of bone marrow biopsy for response assessments. Cycle 2 administration of azacitidine will also be delayed until ANC $\geq 0.5 \times 10^9$/L. Both venetoclax, azacitidine, and APR-246 will resume on the same day after the interruption. If a patient presents with new onset Grade 4 neutropenia for more than 1 week during subsequent cycles, unless it is thought to be due to the underlying disease, venetoclax dosing may be interrupted until ANC is $\geq 0.5 \times 10^9$/L. For subjects in CR/CRi who required interruption or delay of study drug administration for cytopenia (neutropenia or thrombocytopenia) venetoclax may be administered for 14-21 days out of 28 days during each of the subsequent cycles.

If hematologic recovery is achieved within 14 days after completion of the cycle, the duration of venetoclax is reduced to 21 days of the cycle. If a 25% increase has not been achieved within 14 days after the completion of a cycle, azacitidine dose adjustment can be made based on bone marrow cellularity. In the setting of bone marrow hypocellularity, the dose of azacitidine in the next treatment cycle should be reduced to 50% for 15-50% hypocellularity and to 33% for <15% hypocellularity.

In each portion of the study, patients may continue treatment as long as toxicity remains acceptable and the patient has not withdrawn consent. Response and progressive disease will be assessed based on the IWG AML response criteria (APPENDIX I) after every treatment cycle the first year, then every two cycles thereafter.

4.2 End of Study

The end of the study is defined as the date of the last visit of the last patient undergoing the trial.

4.3 Drug Products

4.3.1 APR-246

[redacted]

[redacted] *Administration*: Venetoclax should be taken orally, with food, at a dose of 400 mg daily once the ramp-up phase is completed. If no prophylactic antifungal agent is administered concurrently, the ramp-up phase should consist of 100, 200, and 400 mg of venetoclax once daily given on days 1, 2, and 3 of Cycle 1, respectively. For day 4 of Cycle 1 and beyond, the venetoclax dose is 400 mg once daily given in combination with APR-246 with and without azacitidine. If a prophylactic antifungal agent is administered concurrently, the ramp-up phase and maintenance dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax (see APPENDIX VIII). The dose of venetoclax should be modified accordingly to account for changes in a patient's antifungal medication use.

4.3.3 Azacitidine

Chemical Name: 4-amino-1-β-D-ribofuranosyl-s-triazin-2(1H)-one

Formulation, preparation, storage and stability: Please see commercial package insert approved by regulatory agencies[56].

Route of Administration: Subcutaneous injection, or IV infusion for 7 consecutive days (Days 1-7). Subcutaneous route is preferred but IV administration is acceptable at the Investigator's discretion. The same route should be maintained over the 7-day treatment period.

4.4 Duration of Therapy

Patients may remain on study treatment to the end of the trial while deriving clinical benefit, unless unacceptable toxicity, progression, death or patient withdrawal requires discontinuation. For patients without disease progression or unacceptable toxicity, study treatment should proceed for a minimum of 6 months to allow time for clinical response. Patients may remain on protocol therapy after relapse or progression if they are continuing to derive clinical benefit in the opinion of the Investigator.

4.5 Trial Discontinuation

For reasonable cause, either the Investigator or the Sponsor may terminate this study prematurely. Written notification of the termination is required. Conditions that may warrant termination include, but are not limited to:

- The discovery of an unexpected, significant, or unacceptable risk to the patients enrolled in the study.
- Failure of the Investigator to enter patients at an acceptable rate.
- Insufficient adherence to protocol requirements (non-compliance).
- Lack of evaluable and/or complete data.
- Decision to modify the developmental plan of the drug.
- A decision on the part of the Sponsor to suspend or discontinue development of the drug.

4.6 Drug Accountability/Disposition of Clinical Trial Supplies

Drug accountability records will be maintained for all clinical trial supplies.

All unused clinical trial supplies will be destroyed per the institution's standard operating procedure. Destruction of drug and trial supplies must be documented, and the documentation will be reviewed by/sent to the Sponsor or their Designee.

4.7 Registration

Prior to registration and any study-specific evaluations being performed, all patients must have given written informed consent for the study and must have completed the pre-study evaluations (see Section 7.1). Patients must meet all of the eligibility requirements listed in Section 5.0. Patients will be registered on the study by using the [redacted] Interactive Web Response System (IWRS) automated patient registration system.

5.0 SELECTION AND WITHDRAWAL OF PATIENTS

5.1 Inclusion Criteria for Patients in Safety Lead-In Cohorts

1. Signed ICF and ability to comply with protocol requirements.
2. Documented diagnosis of initial or relapsed AML according to World Health Organization (WHO) classification.
3. Adequate organ function as defined by the following laboratory values:
   a. Creatinine clearance >30 mL/min (by Cockcroft-Gault method; APPENDIX II),
   b. Total serum bilirubin <1.5 × ULN unless due to Gilbert's syndrome, leukemic organ involvement, hemolysis or considered an effect of regular blood transfusions,
   c. ALT and AST <3 × ULN, unless due to leukemic organ involvement.
4. Age ≥18 years at the time of signing the ICF.
5. At least one *TP53* mutation which is not benign or likely benign.
6. ECOG performance status of 0, 1 or 2 (APPENDIX III).
7. Projected life expectancy of ≥12 weeks.
8. Negative serum or urine pregnancy test prior to study treatment initiation in female patients of childbearing potential.
9. Females of childbearing potential and males with female partners of childbearing potential must be willing to use an effective form of contraception such as latex condom, hormonal birth control, intrauterine device or double barrier method (APPENDIX IV) during chemotherapy treatment and for at least six months thereafter.

5.2 Exclusion Criteria for Patients in Safety Lead-In Cohorts

1. Prior treatment for *TP53*-mutant AML not permitted per Table 3.
2. Known history of HIV or active hepatitis B or active hepatitis C infection.
3. Any of the following cardiac abnormalities:
   a. Myocardial infarction within six months prior to registration,
   b. New York Heart Association Class III or IV (APPENDIX V) heart failure or known LVEF <40%;
   c. A history of familial long QT syndrome,
   d. Symptomatic atrial or ventricular arrhythmias not controlled by medications,
   e. QTcF ≥470 msec, unless due to underlying bundle branch block and/or pacemaker and with approval of the Medical Monitor.
4. Concomitant malignancies for which patients are receiving active therapy at the time of signing consent. For example, patients with adequately treated basal or squamous cell carcinoma of the skin, adequately treated carcinoma *in situ* (e.g. cervix), or breast cancer

receiving adjuvant endocrine therapy may enroll irrespective of the time of diagnosis with Medical Monitor approval.
5. Known active CNS involvement from AML. A diagnostic lumbar puncture is not required in the absence of suspicion for CNS disease.
6. Malabsorption syndrome or other condition that precludes enteral route of administration.
7. Pregnancy or lactation.
8. Active uncontrolled systemic infection (viral, bacterial or fungal).

5.3 Inclusion Criteria for Patients in Expansion Cohorts

1. Signed ICF and ability to comply with protocol requirements.
2. Newly diagnosed or R/R AML according to WHO classification with permitted prior therapy, as per Table 3. For patients with newly diagnosed AML bone marrow and/or blood evidence of AML with ≥20% blasts is required. For patients with newly diagnosed AML, prior therapy with HMA and/or any chemotherapeutic agent for MDS is excluded.
3. Adequate organ function as defined by the following laboratory values:
   a. Creatinine clearance >30 mL/min (by Cockcroft-Gault method; APPENDIX II),
   b. Total serum bilirubin <1.5 × ULN unless due to Gilbert's syndrome, leukemic organ involvement, hemolysis or considered an effect of regular blood transfusions,
   c. ALT and AST <3 × ULN, unless due to leukemic organ involvement.
4. Age ≥18 years at the time of signing the ICF.
5. At least one *TP53* mutation which is not benign or likely benign, based on central *TP53* testing results. If local *TP53* mutation and/or p53 IHC results become available before central *TP53* mutation results, enrollment may be made based on local results with Medical Monitor approval.
6. ECOG performance status of 0, 1 or 2 (APPENDIX III).
7. Projected life expectancy of ≥12 weeks.
8. Negative serum or urine pregnancy test prior to study treatment initiation in female patients of childbearing potential.
9. Females of childbearing potential and males with female partners of childbearing potential must be willing to use an effective form of contraception such as latex condom, hormonal birth control, intrauterine device or double barrier method (APPENDIX IV) during chemotherapy treatment and for at least six months thereafter.

5.4 Exclusion Criteria for Patients in Expansion Cohorts

1. Known history of MPN (i.e., polycythemia vera, essential thrombocythemia and primary myelofibrosis are excluded).
2. Known history of HIV or active hepatitis B or active hepatitis C infection.
3. Any of the following cardiac abnormalities:
   a. Myocardial infarction within six months prior to registration,

b. New York Heart Association Class III or IV (APPENDIX V) heart failure or known LVEF <40%,
c. A history of familial long QT syndrome,
d. Symptomatic atrial or ventricular arrhythmias not controlled by medications,
e. QTc ≥470 msec calculated from a mean of 3 ECG readings using Fridericia's correction ($QTcF = QT/RR^{0.33}$).

4. Concomitant malignancies for which patients are receiving active therapy at the time of signing consent. For example, patients with adequately resected basal or squamous cell carcinoma of the skin, adequately resected carcinoma *in situ* (e.g. cervix), or breast cancer receiving adjuvant endocrine therapy may enroll irrespective of the time of diagnosis with Medical Monitor approval.
5. Prior exposure to anticancer therapies including chemotherapy, radiotherapy or other investigational therapy, including targeted small molecule agents within 14 days of the first day of study drug treatment or within 5 half-lives prior to first dose of study drug.
6. Prior exposure to biologic agents (e.g. monoclonal antibodies) for anti-neoplastic intent within 14 days prior to first dose of study drug.
7. Known active CNS involvement from AML. A diagnostic lumbar puncture is not required in the absence of suspicion for CNS disease.
8. Malabsorption syndrome or other condition that precludes enteral route of administration.
9. Pregnancy or lactation.
10. Active uncontrolled systemic infection (viral, bacterial or fungal).

5.5 Inclusion of Women, Minorities and Children

Both men and women and members of all races and ethnic groups are eligible for this study. Children are not eligible for this study because the safety and tolerability of the proposed dosing schedule has not been determined in adults.

5.6 Withdrawal Criteria

Protocol therapy can continue for patients receiving clinical benefit, unless one or more study treatment discontinuation criteria are met, or at the patient's discretion, or if the study is terminated.

5.6.1 Study Treatment Discontinuation

Study treatment must be discontinued if:

- Evidence of disease progression. Patients who have relapsed or progressive disease but who are continuing to derive clinical benefit in the opinion of the investigator may continue to receive study treatment.

- A patient becomes pregnant.
- A patient is significantly non-compliant with the requirements of the protocol.
- A patient has an adverse experience that would, in the Investigator's judgment, make continued participation in the study an unacceptable risk.
- The patient starts new treatment for their underlying disease.

5.6.2 Study Completion

Patient must be taken off the study if:

- The patient dies during the study.
- The patient is lost to follow-up.
- The patient withdraws consent.

5.6.3 Withdrawn Patients

When a patient is removed from the study treatment, the Investigator will clearly document the reason in the medical record and complete the appropriate CRF page describing the reason for discontinuation. In addition, every effort should be made to complete the appropriate assessments listed in Section 7.3.

5.7 Noncompliance

All instances of protocol deviations will be recorded according to guidelines per Sponsor's Representative.

6.0 TREATMENT OF PATIENTS

6.1 Drug Preparation and Administration

Study treatment may be administered on an outpatient basis. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's disease.

6.1.1 APR-246

APR-246 is administered as a 6-hour IV infusion daily on Days 1–4 of each 28-day cycle. APR-246 fixed dose is 4.5 g. APR-246 is administered in a 2-step infusion:

Step 1: Loading dose of 1.5 g for the first 45 minutes (± 2 min)
Step 2: Maintenance dose of 3 g over 5 hours 15 minutes (± 30 min)

In each portion of the study, if patients develop intolerable toxicity(ies) to APR-246 in combination with venetoclax and azacitidine, the dose of APR-246 may be reduced (please consult Section 4.1 and Table 2).
APR-246 vials are to be stored at 2-8°C (35.6-46.4°F). At the pharmacies and at the study centers, the prepared APR-246 study product (diluted in sodium chloride solution) is to be stored at not more than 25°C. The infusion should be completed within 24 hours from the time of preparation.

Detailed instructions on vial concentration, preparation and dispensing can be found in the Pharmacy Binder. The infusion timing, including start/stop times and the time of rate change, must be recorded.

6.1.2 Venetoclax

After ramp-up (see below), venetoclax tablets at the dose 400 mg (4 tablets) should be taken orally once daily with a meal and water, at approximately the same time every day. On days 1–4 of Cycle 1 it is recommended that venetoclax is taken approximately one hour after start of APR-246 infusion. On these days' patients will consume a low-fat meal prior to venetoclax consumption. Starting at cycle 2 venetoclax can be taken before or after start of APR-246 infusion (± 2 hours).

During Cycle 1, treatment with venetoclax should start with a ramp-up phase. If no prophylactic antifungal agent is administered concurrently, the ramp-up phase should consist of 100, 200, and 400 mg of venetoclax once daily given on days 1, 2, and 3 of Cycle 1, respectively. For day 4 of Cycle 1 and beyond, the venetoclax dose is 400 mg once daily given in combination with APR-246 with and without azacitidine. If a prophylactic antifungal agent is administered concurrently, the ramp-up and maintenance dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax (please see APPENDIX VIII).

If required, any dosage modifications of venetoclax should be done based on clinical judgement and institutional practice.

Detailed instructions on preparation and administration can be found in the Pharmacy Binder and current package insert.

6.1.3 Azacitidine

Azacitidine is given at the standard dose of 75 mg/m^2 subcutaneously or intravenously over 7 consecutive days, Days 1–7. On Days 1–4 azacitidine is administered immediately after the APR-246 infusion. SC method is preferred but IV is allowed at the Investigator's discretion. The same route should be maintained over the 7-day treatment period (whichever route is used on Day 1, the other days should follow the same route of administration). Detailed

instructions on preparation and administration can be found in the Pharmacy Binder, current package insert, and in Section 6.2, below. The administration of azacitidine may be interrupted by up to 3 days to account for institutional administration practices and policies in addition to holidays.

6.1.3.1 Administration on the Days of PK Blood Sampling

On Cycle 1 Day 1, in subjects receiving APR-246 in combination with venetoclax and azacitidine, APR-246 is the first agent to be administered after the baseline blood sample for PK analysis is collected. Fifteen (15) minutes after the start of the infusion, the subject is asked to have breakfast. One (1) hour after the start of APR-246, the subject is to take venetoclax. Azacitidine is to be administered after the infusion of APR-246.

After C1, venetoclax can be taken before or after start of APR-246 (±2 hrs) and after the baseline PK sample is obtained, as indicated in the Schedule of Study Evaluations.

On Day 1 of Cycle 1 patients will come to the clinic for PK blood sampling without taking the venetoclax in the morning. Patients will receive a low-fat meal and take the venetoclax within 30 min after the meal is finished and a baseline blood sample is taken.

6.1.4 Dose-Limiting Toxicity

An event will be considered a DLT per NCI CTCAE version 5.0 criteria if it occurs within 4-week safety assessment period (Cycle 1 of study treatment) and meets at least one of the criteria below:

- Absolute neutrophil count (ANC) not recovering to >0.5 × 10^9/L and/or platelets not recovering to >25 × 10^9/L by day 42 of study treatment in the absence of active leukemia/myelodysplasia;
- Grade ≥ 3 nausea/vomiting/diarrhea or CNS toxicity that does not resolve to Grade ≤ 1 within 7 days despite treatment interruption and maximum medical therapy;
- Treatment-related non-hematological Grade ≥ 3 toxicity that does not resolve to Grade ≤ 1.

A DLT will be considered related to the study treatment unless there is a clear, alternative explanation for the AE. A ≥ Grade 3 metabolic or electrolyte abnormalities that is not clinically significant and is adequately controlled within 72 hours is not to be considered DLT. A tumor lysis syndrome that responds to therapy and resolves within 72 hours is not to be considered DLT.

Additionally, AEs that meet the above criteria, but occur after the DLT evaluation period will not be defined as DLTs, but will be reported as AEs/Serious Adverse Events (SAEs) and will be reviewed across all cohorts during the study to help determine the AE profile. A patient

that discontinues therapy during Cycle 1 without DLT is considered evaluable for the purpose of safety only if at least 80% of doses of APR-246 in combination with venetoclax with and without azacitidine were administered in the first cycle.

Patients who are tolerating APR-246 in combination with venetoclax and/or azacitidine, will not have to discontinue dosing prematurely due to the occurrence of DLTs in another patient in the same cohort, unless decided by the DRT. If requested by the Investigator, depending on the nature of the DLT and patient status, the DRT and the Sponsor may allow a patient to continue with study treatment at a reduced dose.

6.1.5 Recommended Phase II Dose (RP2D)

The RP2D of APR-246 will be defined as the dose at which <2 out of 6 patients experience DLT during the 4-week safety assessment period after administration of APR-246 in combination with venetoclax and/or azacitidine.

DRT consisting of the Medical Monitor, Site Principal Investigators, and other clinical research personnel that the Sponsor may deem appropriate, will hold DRM on an interim basis at a frequency dependent on study accrual. At these meetings, the DRT will review AEs and DLTs and make recommendations regarding the RP2D. In the expansion portion of the study, the DRT will evaluate safety and tolerability after 5 patients have completed 1 cycle of treatment in each cohort. All accumulated safety data will be discussed during DRMs.

6.2 Dose Interruptions/Withholding

Study treatment may be withheld from a patient based on the Investigator's decision in the event of intercurrent illness, AE, administrative reasons, or other reasons. If the patient's condition subsequently improves, or the situation that resulted in withholding study drug rectifies itself, the Investigator may resume dosing as soon as possible, unless the delay is more than 4 weeks.

Dosing should be delayed for any DLT-equivalent toxicity and possible NCI CTCAE > Grade 2 AEs considered related to study medication. At the Investigator's discretion, dosing may recommence when the toxicity has resolved to Grade 2 or less. Immune-related reactions or allergy should resolve to ≤ Grade 1.

Treatment will be discontinued if a TEAE has not resolved (to acceptable grade) after ≤4 weeks.

6.3 Supportive Management

6.3.1 APR-246

This section outlines the requirements for proceeding with treatment with APR-246, and the protocol rules for APR-246 dose modification due to toxicity.

At screening, 12-lead ECGs should be collected in triplicate to confirm QT interval does not exceed 470 msec in patients without pre-existing bundle branch block (BBB) and/or pacemaker. For patients with pre-existing BBB and/or pacemaker, QTcF at screening should not exceed baseline reading known per medical history. QT interval must be calculated from a mean of all three ECG readings using Fridericia's correction ($QTcF = QT/RR^{0.33}$).

During Cycle 1 ECG should be collected in triplicate prior to infusion of APR-246 and at the end of infusion (EOI) of APR-246 (6 hours after start of infusion, ±30 min) on Days 1–4. QTcF must be calculated from a mean of all three ECG readings to confirm it does not exceed 470 msec in patients without pre-existing BBB and/or pacemaker. For patients with pre-existing BBB and/or pacemaker, QTcF measurements in Cycle 1 should be at or below values recorded during screening period.

If a pre-dose ECG shows QTcF ≥470 msec, the QTc reading should be confirmed by manual assessment using Fridericia's correction ($QTcF = QT/RR^{0.33}$). Serum concentrations of electrolytes should be monitored and corrected, if necessary. Additionally, concomitant medication should be reviewed and adjusted, if necessary. ECG may be repeated at any time, including the same day. APR-246 may only be administered when QTcF has returned to <470 msec. If APR-246 is given on the same day, procedures outlined in the Schedule of Study Evaluations (Table 8) must be followed. If APR-246 cannot be administered on the same day, that dose must be omitted from the cycle.

If there is a significant change in QTcF, defined as either: a) increase >60 msec from baseline (or pre-dose), or b) increase to an absolute value ≥501 msec, i.e. consistent with NCI CTCAE Grade 3 QTc prolongation, QTc prolongation must be confirmed by a manual assessment of the ECG, and using Fridericia's correction ($QTcF = QT/RR^{0.33}$). If confirmed, the APR-246 therapy should be interrupted until a cause (electrolyte disorders or an effect of a concomitant medication) has been identified and addressed, and QTcF has returned to <470 msec. If all other causes for clinically significant QT interval prolongation are excluded, APR-246 must be permanently discontinued.

During subsequent cycles ECG should be collected in triplicate prior to infusion of APR-246 on Day 1 of each cycle. QTcF must be calculated from a mean of all three ECG readings to confirm it does not exceed 449 msec in patients without pre-existing BBB and/or pacemaker. For patients with pre-existing BBB and/or pacemaker, QTcF measurements in subsequent cycles should be at or below values recorded during the screening period.

If pre-dose QTcF is 450-469 msec in patients with and without pre-existing BBB and/or pacemaker, APR-246 may be administered, and additional triplicate ECG should be performed at the EOI (6 hours after start of infusion, ±30 min).

If post-dose ECG shows a significant change in QTcF, defined as either: a) increase >60 msec from baseline (or pre-dose), or b) increase to an absolute value ≥501 msec, i.e. consistent with NCI CTCAE Grade 3 QTc prolongation, QTc prolongation must be confirmed by a manual assessment of the ECG, and using Fridericia's correction ($QTcF = QT/RR^{0.33}$). If confirmed, the therapy should be interrupted until a cause (electrolyte disorders or an effect of a concomitant medication) has been identified and addressed, and QTcF has returned to <470 msec. If all other causes for clinically significant QT interval prolongation are excluded, APR-246 must be permanently discontinued. If QTcF is unchanged or there is no significant change, additional ECG is not required during that cycle.

If repeated QTcF measurements show a stable QTcF <450 msec, or if QTcF remains stable within the interval of 450–469 msec with no significant change at the EOI during several cycles of treatment, reducing the number of ECGs performed in the study may be discussed with the Medical Monitor.

If a patient starts treatment with another medication known to prolong QT interval at any time during the study therapy, an additional pre- and post-dose (6 hours after start of infusion, ±30 min) ECG should be performed on the next treatment day.

6.3.2 Management of CNS Adverse Events

If a patient reports any clinical AE of any grade during the administration period of APR-246 that could be considered to originate from the CNS (e.g. dizziness, vertigo, nausea) then the patient will be given a rescue medication as per the institutional standard of care.

Dose modifications of APR-246 have been successfully used to manage potential CNS effects occurring during the infusion (Table 4). For any clinical AE Grade ≥ 3, the infusion of APR-246 should immediately be stopped, and if all symptoms resolve to CTCAE ≤ Grade 1 within 2 hours, the infusion of APR-246 may be resumed at the same infusion rate. If the same symptoms do occur or increase in severity during re-challenge the infusion of APR-246 should be stopped.

If the event lasts longer than 2 hours, then the APR-246 infusion should be discontinued for that day, the remaining drug should be discarded.

Table 4. Management of CNS Adverse Events (Dizziness, Dyskinesia and Ataxia)

Worst toxicity	Dose Modifications for APR-246
Grade 1	Maintain dose level
Grade 2	If resolved to ≤ Grade 1 with medical therapy, continue same dose level If not resolved despite treatment interruption and maximal medical therapy, stop infusion and ↓ 1 dose level for subsequent dose
Grade ≥ 3	Stop infusion and give medical therapy. If resolved (to ≤ Grade 1) with medical therapy, infusion may continue at the Investigator's discretion. ↓ 1 dose level for subsequent dose
Grade 4	Permanently discontinue patient from APR-246.

In prior studies, prochlorperazine 10 mg orally tid (three times daily) has been reported to be an effective treatment that may also be used prophylactically. When prochlorperazine is used prophylactically, start the day prior to the Day 1 and continue 10 mg tid to day 4 (as needed). The US label for prochlorperazine does not list QT interval prolongation as a known risk associated with use of this drug.

6.3.3 Management of Nausea and Vomiting

Patients who experience nausea and vomiting in association with APR-246 infusion will be prescribed appropriate rescue treatment and prophylaxis (e.g., anti-nausea or anti-emetics medication) as per institutional guidelines. Patients who receive additional drugs that are known to cause QT interval prolongation must be monitored for any signs of QT interval prolongation via an ECG before and after the APR-246 infusion.

- If QTc >501 msec is observed in a patient concomitantly treated with another QT interval prolonging drug, this drug should be stopped and treatment with APR-246 may be restarted when QTc <470 msec or baseline measurement in patients with pre-existing BBB and/or pacemaker (refer to Section 6.3.1).

A list of suggested rescue medications is provided below in Table 5.

Table 5. Medications for Managements of Nausea and Vomiting

Drug	Dosage	QT Interval Prolongation[a]
Ondansetron	8 mg PO administered 30 minutes before the start of infusion or per label	Yes
Dolasetron	100 mg PO administered within one hour before start of infusion or per label	Yes
Palonosetron	0.25 mg IV administered over 30 seconds beginning approximately 30 min prior to the start of infusion on Day 1 only	No

Prochlorperazine	10 mg PO three times daily. Continue until the end of Day 4 of the cycle. When used prophylactically in subsequent cycles, start the day prior to Day 1 administration of APR-246	No

[a] Please refer to Section 6.3.1 for details on concurrent administration of medications known to cause QTc interval prolongation.

6.3.4 Management of Infusion Reactions

If a patient experiences an infusion reaction during the study, the infusion will be stopped and appropriate medical care (e.g., epinephrine, oxygen, H1 and H2 antagonists, and/or corticosteroids) will be administered[57].

[redacted]

The following recommendations were taken from National Comprehensive Cancer Network Guidelines for Prevention and Treatment of Cancer-Related Infections (Version 1.2021).

Table 6. Recommendations for prevention and treatment of cancer-related infections

Organism	Recommendations
Bacterial	Consider fluoroquinolone or other suitable anti-bacterial prophylaxis during neutropenia
Fungal	Consider prophylaxis during neutropenia and for anticipated mucositis. Assess risk for Pneumocystis jirovecii pneumonia (PJP) and select agent(s) as clinically indicated.
Viral	Start treatment with anti-viral medication in the setting of neutropenia and continually assess the risk for viral infection during treatment with anti-cancer therapy.

Please refer to Section 6.2 Dose Interruptions/Withholding,for guidance on APR-246 dose modification in the setting severe infection and/or infestation event(s).

Refer to Section 6.4 Concomitant Treatment, for a description of drugs that are not allowed to be used while a patient is on study treatment.

6.3.6 Management of Neutropenia with or without Fever

Monitor complete blood counts at the time of study enrollment and throughout the course of study treatment. Supportive measures such as antimicrobials for prophylaxis or in the setting of prolonged myelosuppression, and/or at the first signs of infection are recommended to reduce the risk of a serious or severe infection that may lead to a fatal outcome in the setting of neutropenia with or without fever (see Section 6.3.5).

Myeloid growth factors may be used in the setting of severe or prolonged neutropenia. Refer to Section 6.4 Concomitant Treatment, for a description of drugs that are not allowed to be used while a patient is on study treatment.

Please refer to Section 6.2 Dose Interruption/Withholding for guidance on APR-246 dose modification in the setting neutropenia with or without fever.

6.3.7 APR-246 Dose Modification in the Setting of Moderate Renal Impairment

APR-246 is partially eliminated via the kidney and moderate renal impairment, defined by a creatinine clearance, or estimated glomerular filtration rate value of >30 to <60 mL/min, can lead to increases in plasma levels of approximately 30%. Therefore, for patients with moderate renal impairment, the dose of APR-246 should be reduced by 33% from the current dose (Table 7).

Table 7: APR-246 Dose Modification in the Setting of Moderate Renal Impairment

APR-246 Dose	33% Reduced Dose	Loading Dose (over 45 min)	Maintenance Dose (over 5 hr and 15 min)
4.5 g	3.0 g	1.0 g	2.0 g
4.0 g	2.7 g	0.9 g	1.8 g
3.5 g	2.3 g	0.8 g	1.5 g
3.0 g	2.0 g	0.7 g	1.3 g
<3.0 g	Please consult with Medical Monitor		

Monitoring renal function by assessment of serum creatinine prior to infusion of APR-246 is recommended in all patients.

6.3.8 Venetoclax Dose Adjustments

Please refer to venetoclax USPI [37] for recommended dose modifications.

6.3.9 Azacitidine Dose Adjustments

Missed doses (for any reason but toxicity) may, at the Investigator's decision, be compensated by adding an additional dosing day for azacitidine (e.g. Day 8) so that the patient receives the total 7 days of treatment per cycle.

If unexplained reductions in serum bicarbonate levels to less than 20 mEq/L occur during treatment with azacitidine, contact the study medical monitor to discuss if a reduction in azacitidine dose by 50% should occur with the next scheduled cycle. Similarly, if unexplained elevations in BUN or serum creatinine occur, contact the study medical monitor to discuss if a delay in the next cycle of azacitidine is warranted and/or a reduction in azacitidine dose by 50% should occur with the next scheduled cycle.

Please refer to azacitidine USPI[58] for recommended dose modifications.

6.3.10 Antifungal Prophylaxis

Antifungal agents may be used at the investigator's discretion and with appropriate monitoring of the QTc and adjustment of the venetoclax dose.

6.4 Concomitant Treatment

Patients must not receive any other concurrent anti-cancer therapy, including investigational agents while on study treatment. Please also see Section 2.3, regarding known drug-drug interactions for venetoclax, and the importance of avoiding certain food such as grapefruits. This is important both for patient safety and for enabling a correct assessment of an unlikely drug-drug interaction between the study drug APR-246 and venetoclax. For patients concomitantly treated with strong or moderate CYP3A inhibitors, or P-gp inhibitors, the dose of venetoclax should be determined based on institutional practice and current prescribing information. Strong or moderate CYP3A inducers should be avoided (see APPENDIX VIII).

Patients may continue their baseline medication(s) as long as they are not prohibited. Palliative and supportive care (e.g., anti-emetics, bisphosphonates) for disease-related symptoms will be offered to all patients in the study per institutional practices. AEs will be treated as clinically indicated. All concomitant medications that are currently in use or that become necessary during the study should be recorded.

If the patient develops an acute infusion reaction (≥ Grade 2), the infusion must be stopped until the reaction is resolved to ≤ Grade 1. Premedication (e.g., systemic corticosteroids) may be used after the first cycle.

Use of myeloid growth factors is allowed at the discretion of the investigator per institutional practices. If patients experience prolonged myelosuppression, they should be placed on infection prophylaxis per standard of care.

Hydroxyurea may be used for control of leukocytosis.

6.5 Monitoring Patient Compliance

This study will be monitored by Aprea Therapeutics, Inc. or its CRO according to ICH E6 guidelines of GCP. The study site monitor will regularly visit the study sites to ensure that the study is conducted according to the protocol and GCP principles. All instances of protocol deviations will be entered and reviewed by the Investigator, Sponsor and appropriate [redacted] designee.

7.0 STUDY EVALUATIONS

7.1 Schedule of Study Evaluations

Study evaluations are summarized in Table 8 and described in Sections 7.2 through 7.4.

Table 8: Study Calendar

Study Calendar Evaluation [a]	Screening[a]	Cycle 1									Cycle 2 and Subsequent Cycles									End of Odd Cycles	End of Even Cycles	End of Treat-ment [r]	Long-Term Follow-Up [s]
		D1	D2	D3	D4	D 5-7	D8 [b]	D 15[b]	D 22[b]	D 23	D1 [c]	D 2	D3	D4	D 5-7	D8 [b]	D15 [b]	D22 [b]	D23 [b]				
Informed consent	×																						
TP53 mutation by central lab [d]	×																						
Medical history [e]	×																						
Physical examination [f]	×	×									×											×	
Height	×																						
Weight	×	×									×											×	
Vital signs [f]	×	×	×	×	×						×	×	×	×								×	
ECOG PS	×	×									×											×	
APR-246 [g]		×	×	×	×						×	×	×	×									
Venetoclax [h]		×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×	×				
Azacitidine [i]		×	×	×	×	×					×	×	×	×	×								
Bone marrow/peripheral blood collection [j]	×								×												×	×	×
Response assessment [k]									×											×	×		
Hematology [l]	×	×					×	×	×		×[j]					×	×	×				×	×
Blood chemistry [m]	×	×					×	×	×		×					×	×	×				×	
Pregnancy test [n]	×																						
ECG [o]	×	×[o]	×[o]	×[o]	×[o]						×												
TLS lab monitoring [p]		×	×																				
APR-246 PK sample [q]		×	×		×									×									
Venetoclax PK sample [q]		×	×						×	×				×									
Survival																						×	×
Clinical toxicity assessment		Starting at the time of study treatment initiation through 30 days after last dose [t]																					

Study Calendar Evaluation [a]	Screening[a]	Cycle 1									Cycle 2 and Subsequent Cycles									End of Odd Cycles	End of Even Cycles	End of Treat-ment [r]	Long-Term Follow-Up [s]
		D1	D2	D3	D4	D 5-7	D8 [b]	D 15[b]	D 22[b]	D 23	D1 [c]	D 2	D3	D4	D 5-7	D8 [b]	D15 [b]	D22 [b]	D23 [b]				
Concomitant medications	×	← Reviewed throughout study →																					

Footnotes to Study Calendar

a. All screening/baseline evaluations are performed within 28 days prior to the start of study treatment. In the event that a visit or test cannot be scheduled on the exact visit day, a window of ±3 days is allowable.

b. A window of ±3 days applies to this study visit.

c. After the first cycle, Day 1 evaluations of subsequent cycles are to be done within 3 days prior to next cycle drug administration.

d. A bone marrow aspirate in ethylenediaminetetraacetic acid (EDTA) and peripheral blood sample in EDTA is required at the time of consent and screening for central lab *TP53* mutation determination. If the bone marrow aspirate cannot be obtained due to a dry tap, peripheral blood alone can be used for *TP53* mutation status for enrollment. If local *TP53* mutation and/or p53 IHC results become available before central *TP53* mutation results, enrollment may be made based on local results with Medical Monitor approval.

e. Full medical history is obtained at screening for safety and eligibility purposes; this will include any clinically significant findings from 28 days prior to screening date.

f. Physical exam and vital signs (including blood pressure, heart rate, respiration rate and temperature) are completed for safety purposes and clinically significant items are recorded as AEs where appropriate. Vital signs are collected prior to APR-246 infusion, 2 hours into infusion and at EOI (±30 minutes at all time points).

g. APR-246 is administered IV on days 1–4 of each 28-day cycle.

h. *Only in cohorts with venetoclax:* Venetoclax is given orally at the dose of 400 mg (4 tablets) once daily at approximately the same time every day, with a meal and water. On days 1 – 4 of Cycles 1 and 2 it is recommended that venetoclax is taken approximately one hour after start of APR-246 infusion. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax.

i. *Only in cohorts with azacitidine.* Azacitidine is administered subcutaneously over 7 consecutive days, on Days 1-7 of each 28-day cycle. On Days 1–4 azacitidine is administered immediately after the APR-246 infusion. Subcutaneous route is preferred but azacitidine may be administered intravenously at the Investigator's discretion. The same route should be maintained over the 7-day treatment period.

j. Bone marrow aspirate sample is collected at baseline and when bone marrow is sampled for disease assessment. If no aspirate is available due to a dry tap, then peripheral blood is acceptable..

k. Initial response assessment (blasts percentage) is performed on Day 22 (±3 days). In patients with bone marrow hypocellularity, venetoclax may need to be held and restarted after recovery of blood counts (ANC $\geq 0.5 \times 10^9$/L, and platelets $\geq 25 \times 10^9$/L), confirmed by repeat bone marrow biopsy on approximately Day 35 and following discussion with Medical Monitor. For subjects who

require a delay in study treatment for peripheral blood count recovery after a bone marrow evaluation, hematology values for up to 4 weeks from the bone marrow analysis can be used to determine the IWG response. Ongoing response assessments should be performed at the end of each cycle (every 4 weeks) starting at Cycle 2, based on peripheral blood and bone marrow sample (at the end of even cycles), and peripheral blood sample alone (at the end of odd cycles). After the first year of the study treatment response assessment will be performed every two months. Following a documented CR, a bone marrow aspirate/biopsy is not required if the response assessment can be made based on the peripheral blood, except at the time of suspected relapse at which time a bone marrow aspirate/biopsy is required.

l. Hematology, including complete blood count with white blood cell differential, weekly. Additional peripheral blood sample should be collected on Day 1 of each cycle starting at Cycle 2 for disease assessment, if bone marrow aspirate is not available.

m. Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, blood urea nitrogen (BUN), creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase weekly. At screening, the creatinine clearance may be calculated from the serum creatinine. A 24-hour urine for creatinine clearance may be performed at Investigator's discretion.

n. Serum or urine βhCG must be performed within 7 days prior to study treatment initiation in female patients of childbearing potential.

o. Standard 12-lead ECGs in triplicate at screening, on Days 1–4 of Cycle 1 and on Days 1 of each subsequent cycle with patient in a semi-recumbent position. Please consult Table 12 for ECG collection schedule.

p. *Only in cohorts with venetoclax:* Potassium, phosphorus, calcium and uric acid should be evaluated approximately 6-8 hr and 24 hr (± 4 hr) after the initial administration of venetoclax in order to monitor for potential development of TLS.

q. Please consult Section 8.4 for PK collection schedule.

r. Patients discontinuing treatment should complete their end of treatment visit within approximately 28 days of their last dose of APR-246. Physical exam, vital signs, clinical toxicity assessment, hematology, serum chemistry and bone marrow aspirate and biopsy with NGS analysis should be performed, if feasible.

s. Long-term follow up can be done remotely (e.g. via telephone, via local practitioner or via review of medical records). Assuming there is no withdrawal of consent, patients who stop study treatment for any reason (e.g. toxicity, transition to SCT, PD) will continue long-term follow-up (see Section 7.4). If a patient is removed from the study due to unacceptable AEs, the event(s) will be followed until resolution or stabilization of the AE. Off-treatment data on overall survival will be updated every month (i.e. every 28 days ±3 days) or until death or withdrawal of consent for study participation, whichever occurs first. If a patient is removed from the study due to unacceptable adverse events (AEs), the event(s) will be followed until resolution or stabilization of the adverse event. Patients who respond and discontinue study treatment for reasons other than PD should have response assessments and survival should be collected every month until relapse, death or withdrawal of consent for study participation, whichever occurs first. After relapse, data for

survival should be collected every month (i.e. every 28 days ±3 days) until death or withdrawal of consent for study participation, whichever occurs first.

t. AE description, grade and start date and resolution date should be documented.

7.2 Pre-Study Assessments

Prior to performing any procedures or assessments, the nature of the study and the potential risks associated with the trial will be explained to all patient candidates and written informed consent will be obtained. Patients who choose to participate will have to consent to the biobanking program and will be asked to sign the mandatory section in the main study consent form related to biobank samples. Evaluations obtained as part of routine medical care and performed during the screening period may be used in place of the study specific evaluations. Patients will acknowledge and agree to the possible use of this information for the study by giving informed consent.

7.2.1 Screening

All screening evaluations are to be performed within approximately 28 days of study treatment initiation, unless otherwise noted.

- Signed written informed consent
- *TP53* mutation determination by central lab. Bone marrow aspirate in EDTA and peripheral blood samples in EDTA submitted to central lab. The peripheral blood sample may be submitted alone in cases where the aspirate is not obtainable due to a dry tap. If local *TP53* mutation and/or p53 IHC results become available before central *TP53* mutation results, enrollment may be made based on local results with Medical Monitor approval.
- Medical history
- Physical examination
- Height
- Weight
- ECOG performance status (APPENDIX III)
- Vital signs, including blood pressure, heart rate, respiration rate and temperature
- Hematology, including complete blood count with white blood cell differential.
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase.
- Creatinine clearance (Cockcroft-Gault method; APPENDIX II)
- Serum or urine βhCG must be performed within 7 days prior to study treatment initiation for female patients of childbearing potential.
- ECG: standard 12-lead ECGs with patient in a semi-recumbent position in triplicate.
- Concomitant medication review
- Baseline bone marrow aspirate for exploratory objectives. If no aspirate is available, then peripheral blood is acceptable.

7.2.2 Cycle 1

7.2.2.1 Cycle 1 Days 1-7

Day 1 examinations marked * do not need to be repeated if already performed within 3 days prior to day 1 cycle 1.

- Clinical toxicity assessment
- Concomitant medications review
- Physical examination: Day 1*
- Weight: Day 1*
- ECOG performance status (Day 1*; APPENDIX III)
- Vital signs, including blood pressure, heart rate, respiration rate and temperature, collected 2 hours into APR-246 infusion (± 30 min) and at EOI (± 30 min)
- Hematology (Day 1*), including complete blood count with white blood cell differential
- Serum chemistry (Day 1*), including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- ECG: standard 12-lead ECG with patient in a semi-recumbent position; pre-dose (prior to the APR-246 PK blood draw before the infusion) and at the EOI, per Table 12
- APR-246 administration: Days 1 – 4
- Blood sample for APR-246 pharmacokinetics: on Days 1, 2 and 4, per Table 9
- *Only in cohorts with venetoclax.* Venetoclax administration: once daily, at approximately the same time every day. On days 1 – 4 it is recommended that venetoclax is taken with a meal and water approximately one hour after start of APR-246 infusion. If no prophylactic antifungal agent is administered concurrently, the ramp-up phase should consist of 100, 200, and 400 mg of venetoclax once daily given on days 1, 2, and 3 of Cycle 1, respectively. If a prophylactic antifungal agent is administered concurrently, the ramp-up and maintenance dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax
- *Only in cohorts with venetoclax.* Blood sample for venetoclax pharmacokinetics in the presence of APR-246 (Table 10)
- *Only in cohorts with venetoclax.* TLS lab monitoring: potassium, phosphorus, calcium and uric acid should be evaluated 6 – 8 hr and 24 hr (± 4 hr) after the initial administration of venetoclax in order to monitor for potential development of TLS
- Azacitidine administration, *only in cohorts with azacitidine*. Administered daily for 7 consecutive days starting on Day 1 (Days 1 to 7 inclusive). The same route should be maintained over the 7-day treatment period. On Days 1-4 azacitidine is given immediately after APR-246 infusion

7.2.2.2 Cycle 1 Day 8 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax

7.2.2.3 Cycle 1 Day 15 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax

7.2.2.4 Cycle 1 Day 22 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- *Only in cohorts with venetoclax.* Blood sample for venetoclax pharmacokinetics in the presence of APR-246 (Table 10)
- Bone marrow sample for response assessment (blasts percentage). In patients with bone marrow hypocellularity, venetoclax may need to be held and restarted after recovery of blood counts (ANC ≥0.5 × 10^9/L, and platelets ≥25 × 10^9/L), confirmed by repeat bone marrow biopsy on Day 35 and following discussion with Medical Monitor.

7.2.2.5 Weekly Assessments

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase

7.2.3 Cycle 2 Day 1 ±3 Day and Onwards (Cycle 3+)

These tests should be performed prior to first dose

- Clinical toxicity assessment
- Concomitant medications review
- Physical examination: Day 1*
- Weight
- ECOG performance status (Day 1; APPENDIX III)
- Vital signs
- Hematology (Day 1*), including complete blood count with white blood cell differential
- Serum chemistry (Day 1*), including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- ECGs: see Table 12

7.2.3.1 Cycle 2 Days 1-7

- Clinical toxicity assessment
- Concomitant medications review
- Vital signs, including blood pressure, heart rate, respiration rate and temperature, collected 2 hours into APR-246 infusion (± 30 min) and at EOI (± 30 min)
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax. *Note*: Day 4 of Cycles 2–5 only: Blood sample for venetoclax pharmacokinetics in the presence of APR-246 (Table 10)
- APR-246 administration: Days 1 – 4
- Azacitidine administration, *only in cohorts with azacitidine*. Administered daily for 7 consecutive days starting on Day 1 (Days 1 to 7 inclusive). The same route should be maintained over the 7-day treatment period. On Days 1-4 azacitidine is given immediately after APR-246 infusion
- Blood sample for APR-246 pharmacokinetics: on Day 4, per Table 9. Samples for APR-246 to be taken 1-3 hours after completion of APR-246 administration.

- ECG: standard 12-lead ECGs with patient in semi-recumbent position prior to the PK blood draw before APR-246 infusion on Day 1, per Table 12

7.2.3.2 Cycle 2 Day 8 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential, weekly
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase, weekly
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax

7.2.3.3 Cycle 2 Day 15 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential, weekly
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase, weekly
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of venetoclax should be determined based on institutional practice and current prescribing information for venetoclax

7.2.3.4 Cycle 2 Day 22 (±3 Days)

- Clinical toxicity assessment
- Concomitant medications review
- Hematology, including complete blood count with white blood cell differential, weekly
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase, weekly
- *Only in cohorts with venetoclax.* Venetoclax administration: 400 mg (4 tablets) once daily, at approximately the same time every day, with a meal and water. If a prophylactic antifungal agent is administered concurrently, the daily dose of

venetoclax should be determined based on institutional practice and current prescribing information for venetoclax

7.2.4 End of Odd Numbered Cycles

- Response assessment, based on peripheral blood (blasts percentage)

7.2.5 End of Even Numbered Cycles

- Bone marrow sample for response assessment (blasts percentage)
- Bone marrow aspirate for exploratory objectives (if no aspirate is available, then peripheral blood is acceptable)

7.3 End of Treatment Visit

This visit should take place within 28 days of the last dose of study treatment, if treatment is stopped early for any reasons.

- Physical examination
- Weight
- Vital signs
- Disease assessment
- Bone marrow aspirate for exploratory objectives. If no aspirate is available, then peripheral blood is acceptable
- ECOG performance status (APPENDIX III)
- Hematology, including complete blood count with white blood cell differential
- Serum chemistry, including sodium, potassium, chloride, magnesium, CO_2, BUN, creatinine, glucose, calcium, albumin, total protein, total bilirubin, AST, ALT, alkaline phosphatase
- Clinical toxicity assessment up to 30 days after the last dose

7.4 Long-Term Follow-Up

Long-term follow-up can be done remotely (e.g. via telephone, via local practitioner or via review of medical records). After a patient is removed or withdrawn from the study, the patient will be followed until death or withdrawal of consent for study participation. Off-treatment data on overall survival will be updated every month (i.e. every 28 days ±3 days) or until death, whichever occurs first. If a patient is removed from the study due to unacceptable AEs, the event(s) will be followed until resolution or stabilization of the AE. Patients who respond and discontinue study treatment for reasons other than PD should have response assessments and survival should be collected every month until relapse or death, whichever occurs first. After relapse, data for survival should be collected every month until death or withdrawal of consent for study participation, whichever occurs first.

8.0 STUDY ASSESSMENTS

8.1 Safety Assessments

8.1.1 Safety Analysis

Safety data will be tabulated for all patients and include vital signs, laboratory parameters, and AEs.

8.1.2 Reporting of Adverse Events

8.1.2.1 Adverse Events

An AE is any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product that does not necessarily have a causal relationship with the treatment. An AE can be any unfavorable and unintended sign (including a laboratory finding), symptom or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

The treatment-emergent AE reporting period starts at study treatment initiation. All treatment-emergent AEs will be collected for up to 30 days after the last dose of study treatment. At each evaluation, patients should be interviewed in a non-directed manner to elicit potential adverse reactions from the patient. The occurrence of an AE will be based on changes in the patient's physical examination, laboratory results, and/or signs and symptoms.

On Cycle 1, Day 1, any events observed from the time of signing informed consent but prior to initial study drug administration will be recorded as a serious or non-serious adverse event, if considered by the Investigator to be causally related to a study-required procedure or activity that was conducted prior to Cycle 1, Day 1 (i.e., screening period). Events observed from the time of signing informed consent, but prior to initial study drug administration that are not considered to be causally related to a study-required procedure or activity should be recorded on the medical history CRF page.

All AEs (except Grade 1 and 2 laboratory abnormalities that do not require an intervention), regardless of causal relationship, are to be recorded in the CRF and source documentation. The Investigator must determine the intensity of any AEs according to the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 and their causal relationship. Those AEs not covered by these criteria will be graded as follows:

1. Mild: Discomfort noticed, but no disruption of normal daily activity. Prescription drug not ordinarily needed for relief of symptom but may be given because of personality of patient.
2. Moderate: Discomfort sufficient to reduce or affect normal daily activity. Patient is able to continue in study; treatment for symptom may be needed.
3. Severe: Incapacitating, severe discomfort with inability to work or to perform normal daily activity. Severity may cause cessation of treatment with test drug; treatment for symptom may be given and/or patient hospitalized.
4. Life-Threatening: Symptom(s) place the patient at immediate risk of death from the reaction as it occurred; it does not include a reaction that, had it occurred in a more serious from, might have caused death.
5. Fatal: Event caused the death of the patient.

AEs will be followed until resolution or stabilization while the patient remains on-study. Once the patient is removed from study, events thought to be related to the study medication will be followed until resolution or stabilization, unless, in the Investigator's opinion the event is unlikely to resolve due to the patient's underlying disease, or until the patient starts a new treatment regimen or the patient is lost to follow-up.

8.1.2.2 Attribution Definitions

An AE is considered to be associated with the use of the study treatment if the attribution is determined as possible, probable or definite. Attribution of AEs will be recorded in the CRF as:

- Unrelated: The AE is clearly *not* related to the study treatment.
- Unlikely: The AE is doubtfully related to the study treatment.
- Possible: The AE may be related to the study treatment.
- Probable: The AE is likely related to the study treatment.
- Definite: The AE is clearly related to the study treatment.

8.1.2.3 Definition of an Unexpected Adverse Event

An unexpected AE is defined as any adverse drug experience, the specificity or severity of which is not consistent with the current Investigator Brochure; or, if an Investigator Brochure is not available, the specificity or severity of which is not consistent with the risk information described in this protocol or in the regulatory agency study authorization application.

For study treatments used in this protocol that have been approved for use in the US by the FDA (i.e., venetoclax, and azacitidine), the reference safety information that will be used for making expectedness decisions is the most current version of the product USPI that can be found on the FDA website.

Unexpected, as used in this definition, refers to an adverse drug experience that has not been previously observed (e.g., included in the Investigator's Brochure or USPI, as applicable) rather than from the perspective of such experience not being anticipated from the pharmacological properties of the pharmaceutical product.

8.1.2.4 Serious Adverse Event (SAE)

A serious adverse event is defined as any untoward medical occurrence that at any dose:

1. Results in death,
2. Is life-threatening (i.e., the patient was at risk of death at the time of the event. It does not refer to an event which hypothetically might have caused death if it was more severe),
3. Requires in-patient hospitalization or prolongation of existing hospitalization excluding that for pain management, disease staging/re-staging procedures, or catheter placement unless associated with other serious events,
4. Results in persistent or significant disability/incapacity, or
5. Is a congenital anomaly/birth defect.

Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious adverse drug events when, based on appropriate medical judgment, they may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed above.

8.1.2.5 Pregnancy

Any pregnancy detected during the study, or that occurs within 30 days after stopping study medication, must be reported immediately to the Investigator. Pregnancy, in and of itself, is not regarded as an AE, unless there is suspicion that study medication may have interfered with the effectiveness of a contraceptive medication. If the patient becomes pregnant while on-study, the study drug should be immediately discontinued. Pregnancy information about a female patient or a female partner of a male patient should be reported immediately from the time the Investigator first becomes aware of a pregnancy or its outcome. This will be performed by the Investigator per instructions from the Sponsor's monitoring CRO.

Any pregnancy complication, spontaneous abortion, elective termination of a pregnancy for medical reasons, outcome of stillbirth, congenital anomaly/birth defect, or serious adverse event in the mother will be recorded as an SAE and will be reported as described in Section 8.1.2.6.

8.1.2.6 Reporting of Serious Adverse Events

AEs classified as serious require expeditious handling and reporting to Sponsor's monitoring CRO to comply with regulatory requirements.

For any serious adverse event (SAE) that occurs while a patient is on-study; within 30 days of the last study treatment administration, regardless of any opinion as to the relationship of the SAE to the study treatment; or if any SAE that the Investigator feels is related to the study treatment occurs later than 30 days after the last study treatment administration, the Sponsor's monitoring CRO must be notified immediately (within 24 hours of becoming aware of the event) by electronic data capture (EDC) system, email, fax or telephone. Notification by email is preferred. The primary mechanism for reporting an SAE to the sponsor will be the electronic data collection tool. An SAE eCRF should be completed in the electronic data capture (EDC) system within 24 hours of site becoming aware of the event. EDC Alerts will be sent to Safety desk for all SAEs.

If the EDC system is inaccessible/unavailable, then the site will use the paper SAE form in order to report the SAE within 24 hours. Emailing the paper SAE forms is the preferred method of notification.

SAEs will be reported to: Email: [redacted]

8.1.2.7 Safety Monitoring Plan

The medical monitor is responsible for ongoing safety monitoring for the study per the detailed safety plan. This monitoring will include a review of all serious AEs as they are reported by the study site. The medical monitor will also be in contact with site monitors and will be available to discuss any issues concerning safety with site staff. Safety data will be reviewed periodically by [redacted] and the Sponsor Medical Officer.

DRT consisting of the Medical Monitor, Site Principal Investigators, and other clinical research personnel that the Sponsor may deem appropriate, will hold DRM on an interim basis at a frequency dependent on study accrual. At these meetings, the DRT will review AEs and DLTs and make recommendations regarding the RP2D. In the expansion portion of the study, the DRT will evaluate safety and tolerability after 5 patients have completed 1 cycle of treatment in each cohort. All accumulated safety data will be discussed during DRMs.

The DRT will produce summaries or minutes of these meetings. These summaries will be available for inspection when requested by any of the regulatory bodies charged with the safety of human subjects and the integrity of data.

8.2 Efficacy Assessments

8.2.1 Complete Remission Rate

CR rate will be defined as the proportion of patients who achieve CR.

8.2.2 Duration of Response

DoR will be measured from the time of initial response to disease relapse/progression or death.

8.2.3 Overall Response Rate

Overall response rate (ORR) will be defined as the proportion of patients achieving CR, CRi, complete remission with CRp, partial remission (PR) and MLFS by the IWG 2003 AML criteria (APPENDIX I).

8.2.4 Overall Survival

Overall survival (OS) is defined for all enrolled patients, as measured from the date of enrollment until the date of death.

8.2.5 Stem Cell Transplant

Patients may go on to receive SCT and this is likely to be a reflection of a positive outcome to treatment. The primary approach will be to use a treatment policy strategy (ICH E9 (R1) addendum) where patients who receive SCT will continue to be followed for OS and relapse from any response with their date of death/relapse used in the analysis. If for any reason a patient receives SCT prior to a documented CR they will not be counted as a response. Sensitivity analyses may be performed as counting post-SCT CR/PR as a response.

8.3 Progression-Free Survival (PFS)

Progression-free survival (PFS) is defined for each patient as the time from start of treatment to the date of the first documented progression or death due to any cause. Patients who die without a reported prior progression will be considered to have progressed on the date of their death. Patients who did not progress or die will be censored on the date of their last evaluable tumor assessment. Patients who started any subsequent therapy without a prior reported progression will be censored at the last evaluable tumor assessment prior to the initiation of the subsequent anticancer therapy.

8.4 Pharmacokinetics

Plasma concentrations of APR-246 will be measured in Cycle 1, at the timepoints provided in Table 9. Please refer to sections 2.3 and 6.4, regarding the risk of certain drugs and foods influencing the PK of venetoclax such as grapefruits (see APPENDIX VII). This is important both for patient safety and for enabling a correct assessment of an unlikely drug-drug interaction between the study drug APR-246 and venetoclax from PK samples.

Table 9. PK Blood Sampling Timepoints for APR-246

APR-246 plasma collection timepoints [a]	**Cycle 1**			**Subsequent Cycles**
	D1	**D2**	**D4**	**D4**
Prior to APR-246 infusion	×	×	×	×
At the end of APR-246 infusion	×		×	×
1 hour after the end of APR-246 infusion	×		×	× [c]
3 hours after the end of APR-246 infusion [b]	×			

[a] A window of ±15 min applies to each time point.

[b] Samples are collected as part of the venetoclax PK sampling scheme below (Table 10). A single PK sample should be drawn and split into 2 samples for analysis of both drugs at these timepoints.

[c] Sample collected between 1 and 3 hours after the end of infusion.

Only in cohorts with venetoclax, plasma concentrations of venetoclax will be measured in Cycle 1, at the timepoints provided in Table 10, and then prior to venetoclax administration on Day 4 of Cycles 2 – 5 (Table 11).

Table 10. PK Blood Sampling Timepoints for Venetoclax in Cycle 1

Venetoclax plasma collection timepoints	**D1**	**D2**	**D22**	**D23**
Prior to venetoclax administration [a]	×	×	×	×
2 hours following venetoclax administration [b]	x		x	
3 hours following venetoclax administration [c]	×		×	
4 hours following venetoclax administration [d]	×		×	

6 hours following venetoclax administration [e]	×		×	
8 hours following venetoclax administration [f]	×		×	

[a] Baseline sample collected prior to venetoclax administration. On Day 1, it is recommended that venetoclax is given 1 hour after the start of APR-246 infusion (±10 min), following a meal that will start approximately 15 minutes after initiation of APR-246 infusion. On Day 1, the baseline sample prior to APR-246 infusion can be split into two samples and used as pre-dose sample for venetoclax as well.

[b] 3 hours following initiation of APR-246 infusion on Day 1 (±15 min)

[c] 4 hours following initiation of APR-246 infusion on Day 1 (±15 min)

[d] 5 hours following initiation of APR-246 infusion on Day 1 (±15 min)

[e] 7 hours following initiation of APR-246 infusion on Day 1 (±15 min). This blood draw is also used for the 3rd sample for APR-246 measurements (Table 10; 1 hour after EOI).

[f] 9 hours following initiation of APR-246 infusion on Day 1 (±30 min). This blood draw is also used for the 4th sample for APR-246 measurements (Table 10; 3 hours after EOI)

Table 11. PK Blood Sampling Timepoints for Venetoclax in Cycles 2-5

Venetoclax plasma collection timepoints	**Day 4**
Prior to venetoclax administration [a]	×

8.5 Electrocardiographic Assessment

Table 12 describes the routine ECG requirements from screening through Cycle 4:

Table 12. ECG Assessment Requirements

Time Point	**ECG, number**	**Timing**
Baseline/Screening	Triplicate	Must be within 28 days of C1D1
Cycle 1, Days 1-4	Triplicate	Pre-dose; Post dose (6 hrs. after start of infusion; ± 30 min)
Cycles 2+, Day 1	Triplicate	Pre-dose

If repeated QTcF measurements show a stable QTcF < 450 msec, or if QTcF remains stable within the interval of 450–469 msec with no significant change at the EOI during several cycles of treatment, reducing the number of ECGs performed in the study may be discussed with the Medical Monitor.

If a patient starts treatment with another medication known to prolong QT interval at any time during the study therapy, an additional pre- and post-dose (6 hours after start of infusion, ±30 min) ECG should be performed on the next treatment day.

Please consult Section 6.3.1 for additional requirements for proceeding with treatment with APR-246.

8.6 *TP53* Testing

Patients are enrolled based on the central lab result confirming at least one *TP53* mutation not defined as benign or likely benign (see Laboratory Manual for mutation algorithm) in the bone marrow aspirate sample. The central peripheral blood sample may be used for enrollment in the event a TP53 mutation is not detected in the bone marrow aspirate sample or if the bone marrow aspirate cannot be obtained due to a dry tap.

If local *TP53* mutation and/or p53 IHC results become available before central *TP53* mutation results, enrollment may be made based on local results with Medical Monitor approval.

9.0 STATISTICS

Demographic data and disease-related characteristics will be summarized using descriptive statistics (count and percent, mean, median, standard deviation, minimum, maximum). Continuous variables will be presented by *n*, mean, median, standard deviation and range (minimum and maximum), and categorical variables will be presented by count and percentage of patients as appropriate. Data will be presented by each dose cohort in safety lead-in dose-finding portion and by each treatment arm and dose cohort in the expansion portion. All patient data, efficacy and safety data will be summarized.

9.1 Sample Size

This trial assumes a sample size of 12–36 patients (6–18 patients for each cohort) in the safety lead-in portion of the study and approximately 70 patients in the expansion portion of the study.

In expansion cohorts 2 and 5, Simon's two-stage "minimax" design will be used with alpha level of 0.05 and power of 80%. The null response rate will be set to 20% and the target response rate will be 40%. At the first stage if more than 4 responders in 18 patients observed, then the cohort will be continued to enroll another 15 patients in the second stage. A total of 11 responders or more in 33 patients will indicate the response rate is higher than the null response rate.

9.2 Analysis Populations

Safety population: Patients will be evaluable for safety if they receive at least one dose of APR-246 with venetoclax and/or azacitidine. The safety population will be used to summarize exposure and safety parameters.

DLT-evaluable population: All patients who either experienced a DLT during first 4 weeks (Cycle 1) of the study treatment or received ≥80% of scheduled Cycle 1 dose of APR-246 in combination with venetoclax and/or azacitidine and did not experience a DLT. Any individual patient who is not evaluable for DLT will be replaced by a new patient through additional patient enrollment.

Efficacy evaluable (EE) population: All patients who complete at least one treatment cycle of APR-246 in combination with venetoclax and/or azacitidine and who have at least one post-treatment clinical response assessment. Patients who fail to complete one treatment cycle will also be considered EE if they show clear evidence of clinically significant disease progression. The EE population will be the secondary analysis population for efficacy.

Pharmacokinetics (PK) population: Patients will be evaluable for pharmacokinetics if at least one post-dose sample for PK evaluation has been obtained.

9.3 Endpoints

9.3.1 Primary

- DLTs, classified and graded according to the NCI CTCAE, version 5.0.
- Frequency of treatment-emergent adverse events (TEAEs), and serious adverse events (SAEs) related to APR-246 in combination with venetoclax and/or azacitidine during the trial.
- The RP2D of APR-246 (the dose at which <2 out of 6 patients experience dose-limiting toxicity during the safety assessment period).

9.3.2 Secondary

1. CR rate, defined as the proportion of patients who achieve CR.
2. CR + CRi rate, defined as the proportion of patients who achieve CR or CRi.
3. CR + CRh rate, defined as the proportion of patients who achieve CR or CRh.
4. ORR, defined as the proportion of patients achieving CR, CRi, CRp, PR and MLFS by the IWG 2003 AML criteria (APPENDIX I). The CRh rate will also be determined and is defined as bone marrow blasts <5%, ANC >0.5 × 10^9/L and platelets >50 × 10^9/L.
5. OS, measured from the date of initiating study treatment to the date of death. Patients who have not died by the analysis data cut-off date will be censored at their last date of contact.

6. Rate of RBC and/or platelet TI for at least 56 days
7. PFS, defined from the date of initiating study treatment to the date of disease progression per IWG 2003 AML criteria (APPENDIX I) or death as a result of any cause.
8. Proportion of patients who transition to HSCT.
9. Pharmacokinetic parameters: C_{max}, AUC, V_d and CL of APR-246 and T_{max}, C_{max} and AUC of venetoclax.

9.3.3 Exploratory

- Exploratory molecular analyses may include, but are not limited to: *TP53* VAF by NGS, p53 IHC, mutations in other genes by NGS, RNA expression.

9.4 Safety

Safety data will be summarized for the safety population. These data will include AEs, ECGs and laboratory parameters. AE terms will be coded using the Medical Dictionary for Drug Regulatory Activities (MedDRA)®, version 22 or higher. AEs will be summarized by System Organ Class (SOC), preferred term, severity, and relationship to treatment. Serious adverse events, deaths, and AEs leading to dose modifications including early discontinuation of study treatment will be summarized. Laboratory parameters will be summarized by maximum NCI CTCAE version 5.0 severity grade and also by change from pre-treatment to scheduled time points using descriptive statistics. Laboratory parameter listings will include the normal ranges for each parameter. Each value will be classified as falling above, below, or within the normal range.

Only AEs related to study screening procedures are to be collected prior to the initiation of study treatment. Data summaries will include only treatment-emergent adverse events (TEAEs), defined as events occurring at the start of infusion on Day 1, Cycle 1 up to and including 30 days after last dose.

9.5 Efficacy

CR will be summarized for all enrolled patients as the proportion of patients (%) with CR. In addition to presenting the CR rate, its associated exact 95% CI for each treatment arm will also be presented. CR rate will not be formally compared between treatment arms.

DoR will be defined as the time from the date when criteria for response are met to the date of PD or death due to any cause, whichever occurs first. Patients that are alive without relapse or progression will have their DOR censored at the date of the last clinical assessment. The duration of CR will be summarized in each treatment arm by providing DOCR and DOR, using Kaplan-Meier methodology. DoR endpoints will not be formally compared between treatment arms.

Overall response will be summarized in number (%) of patients in each category of responses and ORR will be analyzed by using the similar method as CR rate.

Survival data will be collected at treatment and follow-up periods. Patients will be followed until death, or withdrawal of consent for study participation. OS is defined as the number of days from the date of enrollment to the date of death. Kaplan-Meier methodology will be utilized.

PFS will be defined as the time from the date of enrollment to disease progression or death from AML whichever occurs first. If neither event occurs, PFS will be censored at the date of the last assessment. Kaplan-Meier methodology will be utilized.

Transition rate to SCT will be analyzed using the similar methods as CR.

9.6 Pharmacokinetic Analysis

The pharmacokinetics of APR-246 and venetoclax will be summarized using descriptive statistics (mean, standard deviation, CV% mean, geometric mean, CV% geometric mean) and compared with historical control data.

APR-246 and venetoclax concentrations will be determined, and PK parameters (C_{max}, T_{max}, AUC, V_d and CL) will be derived using population pharmacokinetic or non-compartmental methods.

APR-246 concentrations will be determined by a validated high-performance liquid chromatography (HPLC) tandem mass spectrometry (LC/MS/MS) method. A population PK model will be used to estimate PK parameters C_{max}, AUC and CL for each individual patient. APR-246 AUC and C_{max} will then be tested for association with signs of efficacy and safety in the presence of venetoclax. If an observable trend exists, a PK/PD model will be developed to evaluate the exposure-response relationship between APR-246 plasma exposure and outcome measures. Demographic and clinical data (ethnicity, current age, body weight, sex, disease status, etc.) will be utilized to assess interpatient variability in the PK and PK/PD relationships.

APR-246 PK parameters will be compared to historical controls for the assessment of drug-drug interactions.

9.7 Exploratory analyses

Descriptive statistics/results from exploratory molecular analyses will be written and may include, but are not limited to: TP53 VAF by NGS, p53 IHC, mutations in other genes by NGS, RNA expression. Please consult Laboratory Manual regarding details on the exploratory biomarker sample processing.

10.0 QUALITY CONTROL AND QUALITY ASSURANCE PROCEDURES

10.1 Monitoring of the Study and Regulatory Compliance

The project manager, or designee, will make an initiation site visit to each institution to review the protocol and its requirements with the Investigator(s), inspect the drug storage area, fully inform the Investigator of his/her responsibilities and the procedures for assuring adequate and correct documentation. During the initiation site visit the CRFs will be reviewed. Other pertinent study materials will also be reviewed with the Investigator's research staff. During the course of the study, the monitor will make regular site visits in order to review protocol compliance, examine CRFs and individual patient's medical records and assure that the study is being conducted according to pertinent regulatory requirements. All CRF entries will be verified with source documentation. The review of medical records will be done in a manner to assure that patient confidentiality is maintained.

10.2 Curricula Vitae and Financial Disclosure of Investigators

All Principal Investigators will be required to provide a current signed and dated curriculum vitae, a completed FDA Form 1572 and a financial disclosure statement to Sponsor's monitoring CRO. All Sub-Investigators will be required to provide a current curriculum vitae and a financial disclosure statement to Sponsor's monitoring CRO.

10.3 Protocol Modifications

No modification of the protocol should be implemented without the prior written approval of the Sponsor or the Sponsor's representative. Any such changes which may affect a patient's treatment or informed consent, especially those increasing potential risks, must receive prior approval by the IRB/IEC. The exception to this is where modifications are necessary to eliminate an immediate hazard to trial patients, or when the change involves only logistical or administrative aspects of the trial (e.g., change in monitor, change in telephone number). Other administrative revisions which may impact the clinical portion of a study will be duly reported to the IRB/IEC by the Principal Investigator.

10.4 Publication Policy

The publication of the results of the study will be subject to the terms and conditions of the clinical trial agreement between the Sponsor and Investigators. Sponsor approval is required for publication of any data from this trial.

11.0 ETHICAL CONSIDERATIONS

11.1 Informed Consent

The Investigator will obtain written informed consent from each patient, or their authorized representative, participating in the study. The form must be signed, witnessed and dated. The informed consent form will contain all the Essential Elements of Informed Consent set forth in 21 CFR, Part 50, the European Union Directive 2001/20/EC and its associated Detailed Guidances, European Union GCP Directive 2005/28/EC, the ICH Guideline for GCP, Section 4.8, and the terms of the Declaration of Helsinki (2013). Copies of the signed document should be given to the patient and filed in the Investigator's study file, as well as the patient's medical record if in conformance with the institution's Standard Operating Procedures.

11.2 Institutional Review Board/Independent Ethics Committee

The study will not be initiated without approval of the appropriate IRB/IEC and compliance with all administrative requirements of the governing body of the institution. This protocol, consent procedures, and any amendments must be approved by the IRB/IEC in compliance with current regulations of the FDA and the European Union as applicable and in accordance with ICH/GCPs. A letter of approval will be sent to the Sponsor prior to initiation of the study and when any subsequent modifications are made. The IRB/IEC will be kept informed by the Investigator, Sponsor's monitoring CRO or the Sponsor, as required by national regulations, as to the progress of the study as well as to any serious and unexpected AEs.

11.3 Patient Privacy

In order to maintain patient confidentiality, all CRFs, study reports and communications relating to the study will identify patients by initials and assigned patient numbers; patients should not be identified by name. In accordance with local, national or federal regulations, the Investigator will allow the Sponsor or designee personnel access to all pertinent medical records in order to verify the data gathered on the CRFs and to audit the data collection process. Regulatory agencies such as the U.S. FDA may also request access to all study records, including source documentation for inspection. Clinical information will not be released without the written permission of the patient as outlined in the patient consent form.

12.0 DATA HANDLING AND RECORD KEEPING

12.1 Data to be Entered Directly in the Case Report Form

The CRF will be the source record.

12.2 Recording of Data

Data collected during the study will be entered in the patient's CRF by the investigational site staff. The staff will keep records of the patient's visit in the files considered as source documents for the site, e.g., hospital chart, research chart. The Investigator will be responsible for the recording of all data on the CRF and for submitting the data to the Sponsor or their designee in a timely manner. Should any value be significantly different from normal, the Investigator will comment in the appropriate sections provided in the CRF.

The Investigator will provide access to his/her original records to permit a representative from the Sponsor to verify the proper transcription of data. To facilitate photocopying, entries must be recorded legibly in black ink only. Erroneous entries will be crossed out with a single line, so as to remain legible. The correct value will be entered above the error and then initialed and dated by the person authorized to make the correction.

12.3 Study Records

U.S. Federal laws require that an Investigator maintain all study records for the indication under investigation for two years following the date a Product Licensing Application is approved or, if no application is to be filed or if the application is not approved for such indication, until two years after the investigation is discontinued and the FDA is notified.

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APPENDIX I – Response Criteria Based on Modified Recommendations of the International Working Group for Diagnosis, Standardization of Response Criteria, Treatment Outcomes, and Reporting Standards in Acute Myeloid Leukemia[59]

Response Criterion	Neutrophils, $\times 10^9$/L	Platelets, $\times 10^9$/L	Bone Marrow Aspirate Blasts (%) (by biopsy acceptable if aspirate not available)
CR	>1.0	>100	<5
CRi	ANC ≥1,000 OR platelets ≥100,000		<5
MLFS	NA	NA	<5
PR	>1.0	>100	>50% decrease from baseline to level of 5-25%
SD (for patients without a prior best response of CR, CRi, MLFS, or PR)	Absence of CR, CRi, PR, MLFS; and criteria for PD not met		
Hematologic relapse (for patients with a prior best response of CR, CRi, MLFS)	Bone marrow blasts ≥5%, or reappearance of blasts in the blood, on two consecutive assessments separated by ≥28 days. Development of new extramedullary disease.		
PD (for patients with a prior best response of SD or PR)	Evidence for an increase in bone marrow blast percentage and/or increase of absolute blast counts in the blood, on two consecutive assessments at least 28 days apart: >50% increase in marrow blasts over baseline (a minimum 15% point increase is required in cases with <30% blasts at baseline; or persistent marrow blast percentage of >70% over at least 3 months; without at least a 100% improvement in ANC to an absolute level (>0.5 × 10^9/L [500/mL], and/or platelet count to >50 × 10^9/L [50,000/mL] non-transfused); or >50% increase in peripheral blasts (WBC × % blasts) to >25 × 10^9/L (>25,000/mL) (in the absence of differentiation syndrome); or New extramedullary disease		
Additional Responses to be Assessed in Addition to Above Responses These responses in addition to other responses and do not have specific hematologic criteria.			
Cytogenetic CR	Normal karyotype in a patient with abnormal cytogenetics at baseline.		

TP53 Molecular CR	*TP53* mutation not detected by NGS based on local testing in a patient with detectible *TP53* VAF by NGS at baseline.
Additional Responses to be Assessed in Addition to Above Responses These responses in addition to other responses and do not have specific hematologic criteria.	
Cytogenetic CR	Cytogenetic CR
TP53 Molecular CR	*TP53* Molecular CR

APPENDIX II - Cockcroft-Gault Equation

Males:

$$\text{Creatinine CL (mL/min)} = \frac{\text{Weight (kg)} \times (140 - \text{Age})}{72 \times \text{serum creatinine (mg/dL)}}$$

Females:

$$\text{Creatinine CL (mL/min)} = \frac{\text{Weight (kg)} \times (140 - \text{Age})}{72 \times \text{serum creatinine (mg/dL)}} \times 0.85$$

APPENDIX III - ECOG Performance Status

Grade	
0	Fully active, able to carry on all pre-disease performance without restriction.
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light housework, office work.
2	Ambulatory and capable of all self-care but unable to carry out any work activities. Up and about more than 50% of waking hours.
3	Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.
4	Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.
5	Dead.

APPENDIX IV - Acceptable Contraceptive Methods

● Male or female condom with or without spermicide ● Cervical cap, diaphragm or sponge with spermicide
Highly Effective Contraceptive Methods That Are User Dependent [a] *Failure rate of <1% per year when used consistently and correctly.*
● Combined (estrogen- and progesterone-containing) hormonal contraception [b] ○ Oral ○ Intravaginal ○ Transdermal ○ Injectable
● Progestogen-only hormonal contraception [b] ○ Oral ○ Injectable
Highly Effective Methods That Have Low User Dependency *Failure rate of <1% per year when used consistently and correctly.*
● Progesterone-only contraceptive implant [b, c] ● Intrauterine hormone-releasing system (IUS) [b] ● Intrauterine device (IUD) ● Bilateral tubal occlusion
● **Vasectomy** Vasectomy is a highly effective contraception method provided that the partner is the sole male sexual partner of the female of childbearing potential and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.
● **Sexual abstinence** Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study treatment. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.)
Notes: Use should be consistent with local regulations regarding the use of contraceptive methods for participants of clinical studies. a) Typical use failure rates are lower than perfect-use failure rates (i.e., when used consistently and correctly). b) If hormonal contraception efficacy is potentially decreased due to interaction with study treatment, condoms must be used in addition to the hormonal contraception during the treatment period and for at least 120 days, (corresponding to time needed to

eliminate study treatment plus 30 days for study treatments with genotoxic potential) after the last dose of the study treatment.
c) If locally required, in accordance with Clinical Trial Facilitation Group (CTFG) guidelines, acceptable hormonal contraceptives are limited to those which inhibit ovulation.

APPENDIX V - New York Heart Association (NYHA) Classification

NYHA Grading		MET*
Class I	No limitations. Ordinary physical activity does not cause undue fatigue, dyspnea or palpitations (asymptomatic LV dysfunction)	>7
Class II	Slight limitation of physical activity. Ordinary physical activity results in fatigue, palpitation, dyspnea or angina pectoris (mild Congestive Heart Failure (CHF)).	5
Class III	Marked limitation of physical activity. Less than ordinary physical activity leads to symptoms (moderate CHF)	2 – 3
Class IV	Unable to carry on any physical activity without discomfort. Symptoms of CHF present at rest (severe CHF).	1.6

*MET (metabolic equivalent) is defined as the resting VO_2 for a 40-year-old 70kg man.
1 MET = 3.5 mL O_2/min/kg body weight.

APPENDIX VI - Study Drug Diaries

VENETOCLAX DRUG DIARY

Instructions:
You will use this diary to record each dose of venetoclax that you take. You should also use this diary to record any side effects that you experience and medications that you take other than the venetoclax. Please be sure to bring this diary with you to your next clinic visit.

Venetoclax:

- Venetoclax is taken once a day every day, approximately 24 hours apart.
- Venetoclax should be taken with food. On days 1 – 4 of cycle 1 it is recommended that venetoclax is taken approximately one hour after start of APR-246 infusion. Starting at cycle 2 venetoclax can be taken before or after start of APR-246 infusion (± 2 hours).
- If you forget to take a dose of venetoclax and it is within 12 hours of when it should have been taken, you should take the dose. If it has been more than 12 hours, you should skip that dose and take the next dose at the next regular time.
- If you vomit after taking a dose of venetoclax, you should not make up the dose or take additional pills, you should take the next dose at the next regular time.
- Always bring your pill bottles (including those that are empty) and your diary with you to each clinic visit.

If you have any questions, please ask a study team member or your doctor.

Patient Number: _____

Patient Initials: _____

Cycle Number: _____

Start Date: _____

STUDY STAFF USE ONLY
Staff member reviewing diary at end of study cycle:

Venetoclax tablets to take at each dose: 100 mg = _____

Other information/Comments:

_____

_____

VENETOCLAX DRUG DIARY (continued)

Patient Number/Initials: _____ Cycle: _____

Cycle Day	**Date Taken**	**Time Taken**	**Number of 100mg venetoclax tablets taken**	**Comments** (Side effects, complaints, other medications)
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
11				
12				
13				
14				
15				
16				
17				
18				
19				
20				
21				
22				
23				
24				
25				
26				
27				
28				

* NOTE: Add additional days in case needed due to scheduling delay or dose modifications.

APPENDIX VII - Patient Handout: Prohibited Medications (For Patients Enrolled in Cohorts with Venetoclax)

PROHIBITED MEDICATIONS

One of the medications you are receiving during this clinical trial, venetoclax, interacts with some drugs that are processed by your liver. Because of this, it is very important to tell your study doctors about all of your medicine before you start this study. It is also very important to tell them if you stop taking any regular medicine, or if you start taking a new medicine while you take part in this study. When you talk about your medicine with your study doctor, include medicine you buy without a prescription at the drug store (over-the counter remedy), or anything that you buy from the health food store or grocery store (herbal supplement). Many health care prescribers can write prescriptions. You must also tell your other prescribers (doctors, physicians' assistants or nurse practitioners) that you are taking part in a clinical trial.

Bring this paper with you.

- Venetoclax is processed by a certain enzyme in the liver called CYP3A4. Drugs that increase the activity of this enzyme are called "inducers", and drugs that decrease the activity of this enzyme are called "inhibitors". Venetoclax must be used very carefully with other medicines that are inducers or inhibitors of CYP3A4. Venetoclax may also interact with other drugs that are processed by the liver.
- You and healthcare providers who prescribe drugs for you must be careful about adding or removing any drug in this category.

Before you start the study, your study doctor will work with your regular prescriber to switch the following medications if you are taking them:

Avoid strong CYP3A inhibitors, i.e. clarithromycin, indinavir, itraconazole, ketoconazole, lopinavir/ritonavir, nefazodone, nelfinavir, posaconazole, ritonavir, saquinavir, telaprevir, telithromycin, and voriconazole. Avoid grapefruits and grapefruit juice. If a strong CYP3A inhibitor must be coadministered, the dose of venetoclax should be reduced. If the strong inhibitor is discontinued, the dose of venetoclax should be increased (after 3 to 5 half-lives of the inhibitor) to the dose used prior to initiation of the strong inhibitor.

Avoid strong CYP3A inducers, i.e. phenytoin, rifampin, enzalutamide, and St John's wort.

Avoid P-gp inhibitors, i.e. amiodarone, carvedilol, clarithromycin, dronedarone, itraconazole, lapatinib, lopinavir and ritonavir, propafenone, quinidine, ranolazine, ritonavir, saquinavir and ritonavir, telaprevir, tipranavir and ritonavir, verapamil.

Avoid P-gp substrates with narrow therapeutic index, i.e. amitriptyline, carbamazepine, clonidine, ciclosporin, digitoxin, digoxin, imipramine, phenobarbital, phenytoin, quinidine.

Your regular prescribers should look at these websites: https://www.crediblemeds.org http://medicine.iupui.edu/clinpharm/ddis/table.asp to see if any medicine they want to prescribe is on a list of drugs to avoid. Your study doctor may also have a list of medications for you to show your regular prescribers instead of, or in addition to, this website.

Eating grapefruit, grapefruit containing products, Seville oranges (including marmalade made with Seville oranges), and starfruit is prohibited while you are taking venetoclax as these may increase the amount of venetoclax in your blood. Please discuss any questions about prohibited foods with your study doctor.

Other medicines can be a problem with your study drugs.

- You should check with your doctor or pharmacist whenever you need to use an over-the-counter medicine or herbal supplement.
- Your regular prescriber should check a medical reference or call your study doctor before prescribing any new medicine for you. Your study doctor's name is _____ and he or she can be contacted at _____.

APPENDIX VIII - Dose Modifications for Use with CYP3A and P-gp Inhibitors

Concomitant use of venetoclax with *strong* CYP3A inhibitors at initiation and during ramp-up phase is contraindicated. Concomitant use of venetoclax with strong CYP3A inhibitors increases venetoclax exposure (i.e., C_{max} and AUC) and may increase the risk for TLS at initiation and during ramp-up phase. For patients who have completed the ramp-up phase and are on a steady daily dose of venetoclax, reduce the venetoclax dose by at least 75% when *strong* CYP3A inhibitors must be used concomitantly.

Avoid concomitant use of venetoclax with *moderate* CYP3A inhibitors or P-gp inhibitors. Consider alternative treatments. If a moderate CYP3A inhibitor or a P-gp inhibitor must be used, reduce the venetoclax dose by at least 50%. Monitor these patients more closely for signs of toxicities.

Resume the venetoclax dose that was used prior to initiating the CYP3A inhibitor or P-gp inhibitor 2 to 3 days after discontinuation of the inhibitor.

The recommendations for managing drug-drug interactions are summarized in the table below.

Co-administered drug	**Initiation and Ramp-Up Phase**	**Steady Daily Dose (After Ramp-Up Phase)**
Posaconazole	Day 1 – 10 mg Day 2 – 20 mg Day 3 – 50 mg Day 4 – 70 mg	Reduce venetoclax dose to 70 mg.
Other strong CYP3A inhibitor	Day 1 – 10 mg Day 2 – 20 mg Day 3 – 50 mg Day 4 – 100 mg	Reduce venetoclax dose to 100 mg.
Moderate CYP3A inhibitor	Reduce the venetoclax dose by at least 50%	
P-gp inhibitor		

Warfarin
Concomitant use of venetoclax increases warfarin C_{max} and AUC_{inf}, which may increase the risk of bleeding. Closely monitor international normalized ratio (INR) in patients using warfarin concomitantly with venetoclax.

P-gp Substrates
Concomitant use of venetoclax increases C_{max} and AUCinf of P-gp substrates, which may increase toxicities of these substrates. Avoid concomitant use of venetoclax with a P-gp

substrate. If a concomitant use is unavoidable, separate dosing of the P-gp substrate at least 6 hours before venetoclax.

Please refer to the venetoclax USPI[37] for additional details on drug-drug interactions.

Below (Table 13) are examples of CYP3A inhibitors and inducers (strong and moderate); however, this is not an exhaustive list.

Table 13. CYP3A Inhibitors and Inducers [1]

CYP3A Inhibitors	
Strong CYP3A Inhibitors	**Moderate CYP3A Inhibitors**
boceprevir, cobicistat, danoprevir and ritonavir, elvitegravir and ritonavir, indinavir and ritonavir, itraconazole, ketoconazole, lopinavir and ritonavir, paritaprevir and ritonavir and (ombitasvir and/or dasabuvir), posaconazole, ritonavir, saquinavir and ritonavir, telaprevir, tipranavir and ritonavir, telithromycin, troleandomycin, voriconazole	aprepitant, ciprofloxacin, conivaptan, crizotinib, cyclosporine, diltiazem, dronedarone, erythromycin, fluconazole, fluvoxamine, imatinib, tofisopam, verapamil
CYP3A Inducers	
Strong CYP3A Inducers	**Moderate CYP3A Inducers**
apalutamide, carbamazepine, enzalutamide, mitotane, phenytoin, rifampin, St. John's wort	bosentan, efavirenz, etravirine, phenobarbital, primidone

Note: In addition to the medications listed above, patients taking venetoclax should not consume grapefruit, grapefruit products, Seville oranges, (including marmalade containing Seville oranges), or star fruit.