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TITLE: A Phase 2 Study of Acalabrutinib, Venetoclax, and Obinutuzumab (AVO) for Initial Therapy of Chronic Lymphocytic Leukemia

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IND #: 139022
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SYNOPSIS

Study Title: A Phase 2 Study of Acalabrutinib, Venetoclax, and Obinutuzumab (AVO) for Initial Therapy of Chronic Lymphocytic Leukemia

Study Overview: This is an open-label, phase 2 study of acalabrutinib in combination venetoclax and obinutuzumab (AVO) in previously untreated patients with CLL in need of therapy. Patients will start on cycle 1, day 1 with one month of acalabrutinib monotherapy. Obinutuzumab will be added on cycle 2, day 1 and will administered at standard dosing for six monthly cycles, with acalabrutinib continued. During the third cycle of obinutuzumab, venetoclax will be added for triplet combination therapy. Venetoclax will be initiated in a ramp-up, stepwise dosing strategy with frequent monitoring to mitigate the risk of tumor lysis syndrome. After completion of obinutuzumab, venetoclax and acalabrutinib combination therapy will continue through cycle 15. Efficacy assessments will be performed at baseline and after cycles 3, 8, and 15, and will include CT scan, bone marrow (BM) biopsy (after cycles 8 and 15), and minimal residual disease (MRD) analysis by flow cytometry. At the conclusion of cycle 15, patients who have achieved a complete remission (CR) or complete remission with incomplete count recovery (CRi) with MRD-negativity in the peripheral blood and bone marrow will have the option to discontinue acalabrutinib and venetoclax and will be monitored for disease recurrence with peripheral blood MRD testing by flow cytometry every three cycles. Patients who have detectable recurrence in the peripheral blood will restart acalabrutinib and venetoclax. If subjects do not achieve an MRD-negative CR/CRi after cycle 15, they will continue acalabrutinib and venetoclax through cycle 24, with formal efficacy assessments after cycles 19 and 24, including CT scan, BM biopsy (after cycle 24 only), and MRD testing. Subjects who achieve undetectable MRD in both blood and marrow at this time point will also have the option to discontinue therapy and be monitored for recurrence. Should patients recur, they will have the option to resume acalabrutinib and venetoclax at that time. Subjects with detectable MRD at the 24 cycle mark will continue acalabrutinib and venetoclax indefinitely until progression or unacceptable toxicity.

Primary Objective
The primary objective of the study is to assess the rate of bone marrow MRD-negative complete response after 15 cycles of treatment with AVO in CLL patients.

Secondary Objectives
Secondary objectives are to assess the following:

- Safety and tolerability of the AVO combination
- Rate of partial response after 15 cycles of therapy, as defined by 2018 IW-CLL criteria
- Rate of complete remission (including complete remission with incomplete count recovery, CRi) after 15 cycles of therapy
- Median progression free survival (PFS), 2-year and 3-year PFS
- Median overall survival (OS), 2-year and 3-year OS
- Rates of complete remission with bone marrow MRD negativity at 8 cycles and 24 cycles
- Rates of best overall response, best CR/CRi, best peripheral blood and bone marrow MRD
- Rates of therapy discontinuation after 15 cycles and corresponding reasons for therapy

discontinuation (e.g. disease progression, achievement of MRD-negative CR, or intolerability)
- Time to MRD-positive disease recurrence, in patients who have achieved MRD negativity after 8, 15, or 24 cycles
- Time to clinical progression as defined by IWCLL criteria
- Association between established CLL prognostic factors (including FISH cytogenetics and *IGHV* mutation status) and rates of MRD-negativity, CR, PR, recurrence, PFS, and OS
- Rate of peripheral blood and bone marrow MRD-negativity at 8, 15, and 24 cycles
- Correlation between MRD-negativity as measured in the blood with MRD-negativity as measured in the bone marrow after 8, 15, and 24 cycles of treatment
- Rate of infusion reactions with obinutuzumab
- Rate of clinical and laboratory tumor lysis syndrome with venetoclax administration, as defined by the Cairo-Bishop Classification system
- To perform the above efficacy and safety analyses on cohort 1, cohort 2 plus the patients with TP53 aberrant disease in cohort 1, and in the entire group of cohort 1 plus cohort 2

Exploratory Objectives
Exploratory objectives are to assess the following:
- Association between novel CLL prognostic factors (including BH3 profiling and somatic mutations such as *SF3B1, TP53, NOTCH1,* and other mutations in the BCR/NFκB pathway) and rates of MRD-negativity, CR, PR, PFS, and OS
- The development and kinetics of *BTK*, *PLCG2*, *CARD11*, and *BCL2* acquired somatic resistance mutations
- MRD measurements by four-color flow cytometry as compared to those of the Adaptive ClonoSEQ sequencing technology at the primary response (15 cycle) endpoint

Schedule of Administration
Acalabrutinib will be administered orally twice daily during each 28-day cycle. Obintuzumab will be given intravenously at the standard dosing of six monthly cycles (with loading doses administered weekly during the first month). Venetoclax will be given orally daily with ramp-up stepwise dosing during the first month of therapy until the standard dose is achieved. After the six cycles of obinutuzumab are completed, venetoclax and acalabrutinib will continue as combination therapy until cycles 15, 24, or indefinitely (as outlined in Study Overview, above).

Definition of Unexpected Toxicities
Unexpected toxicities would include the following events that occur during AVO treatment, unless they are clearly due to extraneous causes: any grade 3 or higher clinically significant nonhematologic toxicities related to study treatment, grade 4 or higher infusion reaction, grade 4 or higher tumor lysis syndrome, grade 4 or higher infection, and grade 4 or higher neutropenia. Grade 3 or 4 drug-related toxicities occurring after the first cycle of treatment will be discussed with the Principal Investigator and will be considered in decisions regarding the recommended dose for future studies. The IWCLL Grading Scale for Hematologic Toxicity in CLL Studies will be used to grade hematologic toxicities and the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI-CTCAE) Version 5.0 will be used to grade non-hematologic toxicities during the trial unless otherwise specified.

Study Drug
Acalabrutinib is administered orally twice daily as a tablet formulation. The acalabrutinib drug product is supplied as 100mg tablets. Obinutuzumab is administered intravenously. Venetoclax is administered orally daily as a tablet formulation, with tablet strengths varying based on dosing.

Key Eligibility Criteria (for complete eligibility criteria, see section 3.1)

Inclusion Criteria

- Subjects must have CLL, SLL, or B-prolymphocytic leukemia (B-PLL)
- Subjects must have *TP53*-aberrant disease, defined as the detection of del(17p) and/or the presence of a *TP53* mutation
- Participants must have measurable disease (lymphocytosis > 5,000 / µl, or palpable or CT measurable lymphadenopathy ≥ 1.5 cm, or bone marrow involvement ≥30%).
- Subjects must not have received any prior systemic therapy for CLL or SLL due to meeting IWCLL 2018 guidelines, and must currently have an indication for treatment as defined by IWCLL 2018 Guidelines below, or must not have received therapy for B-PLL and must currently have an indication for treatment:
  - Massive or progressive or symptomatic splenomegaly; OR
  - Massive lymph nodes, nodal clusters, or progressive lymphadenopathy; OR
  - Significant fatigue (i.e. ECOG PS 2 or worse; cannot work or unable to perform usual activities); OR
  - Fever ≥ 100.5°F for 2 or more weeks without evidence of infection; OR
  - Night sweats for ≥ 1 month without evidence of infection; OR
  - Presence of weight loss ≥ 10% over the preceding 6 months; OR
  - Progressive lymphocytosis with an increase of ≥ 50% over a 2-month period or lymphocyte doubling time of less than 6 months; OR
  - Evidence of progressive marrow failure as manifested by the development of or worsening of anemia and/or thrombocytopenia; OR
  - Autoimmune anemia and/or thrombocytopenia that is poorly responsive to corticosteroids and another standard therapy such as rituximab
  - Symptomatic or functional extranodal involvement (e.g. skin, kidney, lung, spine)
- Age greater than or equal to 18 years.
- ECOG performance status ≤2 (see Appendix A)
- Participants must have adequate organ and marrow function as defined below:
  - total bilirubin ≤1.5 times upper limit of normal
  - AST and ALT ≤ 2.5 times the upper limit of normal
  - creatinine clearance (CrCl) ≥ 50 mL/min using 24-hour urine collection for creatinine clearance or calculated CrCl OR calculated glomerular filtration rate (GFR) ≥ 50 mL/min/1.73m^2 using the CKD-EPI equation
  - PT/INR ≤2 times the upper limit of normal and PTT ≤2 times the upper limit of normal
  - Absolute neutrophil count ≥750 cells/mm^3 (0.75 x 10^9/L) or ≥500 cells/mm^3 in subjects with documented bone marrow involvement
  - Platelet count without transfusional support must be ≥50,000 cells/mm^3 or ≥ 30,000 cells/mm^3 in subjects with documented bone marrow involvement
- Women of child-bearing potential must agree to remain abstinent or use highly effective

contraception during the treatment period and for at least 2 days after the last dose of acalabrutinib, 30 days after the last dose of venetoclax, or 18 months after the last dose of obinutuzumab, whichever is longer. Men with female sexual partners of childbearing potential should agree to remain abstinent or use contraceptive measures which include a condom plus an additional contraceptive for at least 30 days after the last dose of venetoclax or 6 months after the last dose of obinutuzumab, whichever is longer.

- Ability to understand and the willingness to sign a written informed consent document.

Exclusion Criteria

- Participants who have a history of other malignancies, with certain exceptions including early stage malignancies treated with curative intent or low risk neoplasms managed with active surveillance
- Participants who are receiving any other investigational agents
- History of severe allergic reactions attributed to compounds of similar chemical or biologic composition to obinutuzumab, venetoclax, or acalabrutinib. Patients with reactions to other CD20 monoclonal antibodies (e.g. rituximab, ofatumumab) are not excluded.
- Unable to receive prophylactic treatment for pneumocystis, herpes simplex virus (HSV), and herpes zoster (VZV) at start of treatment
- Known or suspected Richter's transformation or known CNS involvement
- Concurrent systemic immunosuppressant therapy (e.g., cyclosporine A, tacrolimus, etc., within 28 days of the first dose of study drug or chronic administration of >20 mg/day of prednisone within 7 days of the first dose)
- Vaccinated with live, attenuated vaccines within 4 weeks of first dose of study drug.
- Ongoing or recent infection requiring intravenous antimicrobials at time of screening
- Known bleeding disorders (e.g. von Willebrand's disease) or hemophilia.
- Presence of a gastrointestinal ulcer diagnosed by endoscopy within 3 months before screening.
- History of stroke or intracranial hemorrhage within 6 months prior to enrollment.
- Major surgery within 4 weeks of first dose of study drug. If a subject had major surgery greater than 4 weeks prior, they must have recovered adequately from any toxicity and/or complications from the intervention before the first dose of study drug.
- Currently active, clinically significant cardiovascular disease, such as uncontrolled arrhythmia or Class 3 or 4 congestive heart failure as defined by the New York Heart Association Functional Classification; or a history of myocardial infarction, unstable angina, or acute coronary syndrome within 6 months prior to randomization.
- Baseline QTcF >480 ms.
- Patients who require warfarin or other vitamin K antagonists for anticoagulation (other anticoagulants are allowed).
- Patients who require treatment with proton pump inhibitors (eg, omeprazole, esomeprazole, lansoprazole, dexlansoprazole, rabeprazole, or pantoprazole). Subjects receiving proton pump inhibitors who switch to H2-receptor antagonists or antacids are eligible for enrollment to this study.
- Patients who require treatment with strong CYP3A inhibitors or inducers
- Patients who require P-gp inhibitors or narrow therapeutic index P-gp substrates

- Unable to swallow tablets or malabsorption syndrome, disease significantly affecting gastrointestinal function, or resection of the stomach or small bowel if thought by the investigator to compromise systemic absorption, active, symptomatic inflammatory bowel disease or ulcerative colitis, or partial or complete bowel obstruction.
- Lactating or pregnant.
- Patients with human immunodeficiency virus (HIV), human T-cell leukemia virus (HTLV), active hepatitis C virus (HCV) or active hepatitis B virus (HBV) infection with detectable HBV DNA
- Unwilling or unable to participate in all required study evaluations and procedures.
- Unable to understand the purpose and risks of the study and to provide a signed and dated informed consent form (ICF) and authorization to use protected health information (in accordance with national and local subject privacy regulations)
- Significant co-morbid condition or disease which in the judgment of the Principal Investigator would place the patient at undue risk or interfere with the study
- Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements, compromise the subject's safety, or put the study outcomes at undue risk.

Statistical Methodology
Primary Endpoint: Rate of bone marrow minimal residual disease (MRD) negative complete response (CR) after 15 cycles of AVO therapy.

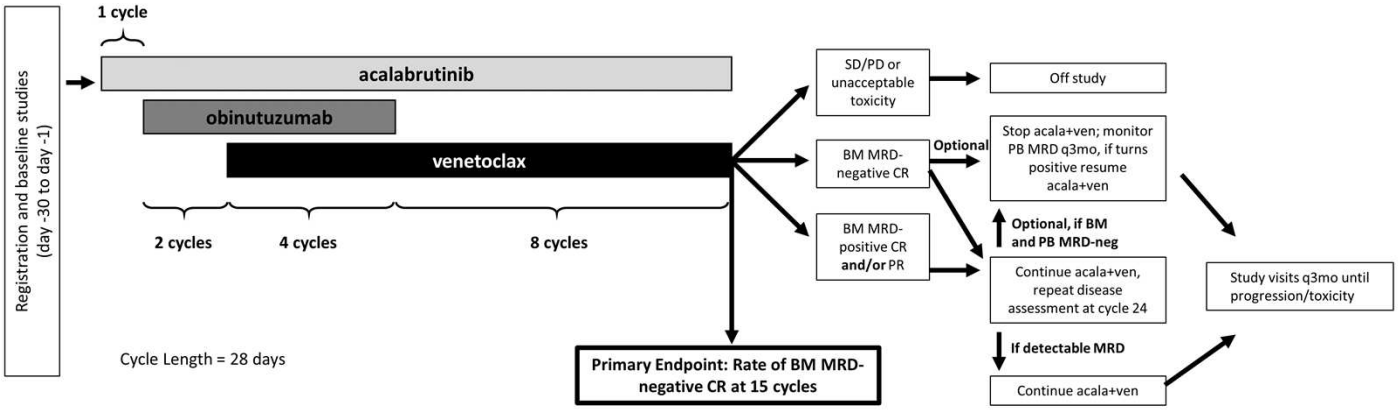
- *Secondary Endpoints*: The analysis of secondary endpoints will be primarily descriptive, including rate of PR after 15 cycles of therapy, rate of CR/CRi after 15 cycles of therapy, progression free survival (including 2-year and 3-year landmark rate), overall survival (including 2-year and 3-year landmark rate), rate of MRD negativity in the bone marrow at 8 and 24 cycles, rate of peripheral blood MRD-negativity at 8, 15, and 24 cycles, correlation between MRD-negativity in the peripheral blood and bone marrow, time to MRD-positive disease recurrence, time to clinical disease progression, rate of infusion related reactions, and rate of tumor lysis syndrome. The Kaplan Meier method will be used to estimate the median progression-free and overall survival. Association between clinical response and established CLL prognostic factors (ZAP70, FISH cytogenetics, *IGHV* mutation status, *TP53* mutation status) will be tested using Fisher's exact test. A similar descriptive analysis will be performed for laboratory correlative studies of BH3 profiling and genomic markers such as *SF3B1, NOTCH1,* and *BCR/NFKB* pathway somatic mutations. If feasible, association between clinical response and laboratory endpoints will be explored. The secondary endpoints on this study will be evaluated on cohort 1, cohort 2 plus the patients with TP53 aberrant disease in cohort 1, and in the entire group of cohort 1 plus cohort 2

Study Design/Sample Size:

This is a one-stage study to determine the MRD-negative CR/CRi rate in the bone marrow after 15 cycles of AVO combination therapy. In a previous study of venetoclax plus rituximab in the relapsed/refractory population, the CR/CRi rate was 51%, and 80% of these responses were

bone-marrow MRD-negative, for an MRD-negative CR/CRi rate of 41% (Seymour et al., 2017). Furthermore, 21 out of 25 CRs in this study occurred after 18 months of therapy, thus we will assume that ~80% of all marrow MRD-negative CRs will have occurred after 15 months of therapy here, leading to an expected marrow MRD-negative CR/CRi rate of 33%. Of note, this study was done in a relapsed/refractory population. Given that the depth of response is likely to be greater in the frontline setting, we are using an MRD-negative CR/CRi rate of 40% as the null hypothesis. With the addition of acalabrutinib, we hypothesize that the MRD-negative CR rate will be 60% or higher (alternative hypothesis). Initially, we planned to enroll 37 patients, and assuming approximately 1 screening failure this meant that 36 patients would be evaluable for the primary endpoint. This cohort has now completed enrollment and will be referred to as cohort 1. We have now expand the study to include a second cohort of 35 patients with high-risk disease (cohort 2), defined as 17p deletion and/or *TP53* mutation. In cohort 2, we have 80% power and one-sided type I error of 0.07 for an exact one-sample binomial test comparing an expected 55% response rate to the assumed 35% response rate for standard-of-care treatment in this high risk group.

SCHEMA



Note: Patients will have two opportunities to electively discontinue therapy: (1) if they reach MRD-negative CR/CRi after 15 cycles, or (2) if they reach undetectable MRD after 24 months of therapy.

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1. OBJECTIVES

1.1 Study Design

This is a Phase 2, open label, single arm, investigator-initiated study. Patients with treatment-naïve chronic lymphocytic leukemia (CLL) in need of therapy will begin with one 28-day cycle of acalabrutinib. Obinutuzumab is then given in combination with acalabrutinib during cycles 2 and 3 to help reduce the disease burden in preparation for starting venetoclax. After these initial 3 cycles, the first response assessment with a CT scan and bone marrow biopsy will occur.

Venetoclax will then be started at cycle 4, with a stepwise dose escalation over 4 weeks to minimize the risk of TLS. All three drugs will be given together during cycles 4-7, and another response assessment will occur at the end of cycle 7, this time including a CT scan,bone marrow biopsy, and flow cytometric MRD testing in the bone marrow and peripheral blood with a sensitivity of at least 10^{-4}. Acalabrutinib and venetoclax will be continued for cycles 8-15. After 15 cycles of therapy, the primary re-staging evaluation will occur, including a CT scan, bone marrow assessment, and flow cytometric MRD testing in the bone marrow and peripheral blood. The primary objective of this study is to assess the rate of complete response (CR) with bone marrow MRD negativity after 15 cycles of treatment.

At the end of 15 cycles, patients who achieve a CR with peripheral blood and bone marrow MRD-negativity will have the option to stop all therapy and those who do will be monitored by serial peripheral blood MRD testing for disease recurrence. If they become MRD positive again, they can resume acalabrutinib and venetoclax, continuing treatment until disease progression or unacceptable toxicity. Patients who are MRD-positive or in a partial remission (PR) at the end of the 15 cycles will continue acalabrutinib and venetoclax until the completion of 24 cycles of therapy. Another response evaluation will occur at that time, and patients who have undetectable MRD in the blood and marrow (regardless of iwCLL response status) have the option to discontinue therapy. Those who remain MRD-positive will continue acalabrutinib and venetoclax treatment until time of progression or unacceptable toxicity.

1.2 Primary Objectives

The primary objective of the study is to assess the rate of complete response with bone marrow MRD-negativity, as determined by four color flow cytometry, after 15 cycles of treatment with AVO in previously untreated CLL patients.

1.3 Secondary Objectives

Secondary objectives are to assess the following:

- Safety and tolerability of the AVO combination
- Rate of partial response after 15 cycles of therapy, as defined by 2018 IW-CLL criteria
- Rate of complete remission (including complete remission with incomplete count recovery, CRi) after 15 cycles of therapy
- Median progression free survival (PFS), 2-year and 3-year PFS
- Median overall survival (OS), 2-year and 3-year OS

- Rates of complete remission with bone marrow MRD negativity at 8 cycles and 24 cycles
- Rates of best overall response, best CR/CRi, best peripheral blood and bone marrow MRD
- Rates of therapy discontinuation after 15 cycles and corresponding reasons for therapy discontinuation (e.g. disease progression, achievement of MRD-negative CR, or intolerability)
- Time to MRD-positive disease recurrence, in patients who have achieved MRD negativity after 8, 15, or 24 cycles
- Time to clinical progression as defined by IWCLL criteria
- Association between established CLL prognostic factors (including FISH cytogenetics and *IGHV* mutation status) and rates of MRD-negativity, CR, PR, recurrence, PFS, and OS
- Rate of peripheral blood and bone marrow MRD-negativity at 8, 15, and 24 cycles
- Rate of infusion reactions with obinutuzumab
- Rate of clinical and laboratory tumor lysis syndrome, as defined by the Cairo-Bishop classification system
- Correlation between MRD-negativity as measured in the blood with MRD-negativity as measured in the bone marrow after 8, 15, and 24 cycles of treatment
- To perform the above efficacy and safety analyses on cohort 1, cohort 2 plus the patients with TP53 aberrant disease in cohort 1, and in the entire group of cohort 1 plus cohort 2

1.4 Exploratory Objectives

Exploratory objectives are to assess the following:

- Association between novel CLL prognostic factors (including BH3 profiling and somatic mutations such as *SF3B1, TP53, NOTCH1,* and other mutations in the BCR/NFκB pathway) and rates of MRD-negativity, CR, PR, PFS, and OS
- The development and kinetics of *BTK*, *PLCG2*, *CARD11*, and *BCL2* acquired somatic resistance mutations
- MRD measurements by four-color flow cytometry as compared to those of the Adaptive ClonoSEQ sequencing technology at the primary response (15 cycle) endpoint

2. BACKGROUND

2.1 Chronic Lymphocytic Leukemia Therapy

Overview of CLL
Chronic lymphocytic leukemia is a neoplastic disease characterized by the accumulation of monoclonal lymphocytes in the blood, bone marrow, and lymphoid tissues. It is the most common leukemia of adults. In the U.S. alone, on a yearly basis, about 20,000 new cases are diagnosed, with approximately 4,500 patients dying from the disease (Siegel, Miller, & Jemal, 2017). The natural history of CLL is heterogeneous, with a subset of patients having very indolent disease that may never require therapy, while others have steadily progressive or aggressive disease. Indications for the treatment of CLL are defined by consensus guidelines and include progressive cytopenias, compromised organ function due to lymphadenopathy, massive lymphadenopathy (e.g. >10cm), and constitutional symptoms due to the disease.

Current Approach to CLL Therapy
Current standard treatment options for CLL generally fall into one of two categories: combination chemoimmunotherapy or targeted therapy with novel agents. Like other low grade lymphomas, CLL has historically been thought of as an incurable disease for which treatment is palliative and not curative. Thus, as noted above, therapy is initiated only if the disease is causing symptoms or organ dysfunction, and outside of those instances, it can be monitored with active surveillance.

With regard to standard chemoimmunotherapy, over the decades, the paradigm has shifted from single agent to combination therapy. This disease was initially treated with alkylating agents like chlorambucil, and increased complete remission rates and progression-free survival (PFS) were later observed with the nucleoside analogue, fludarabine (Rai et al., 2000). Fludarabine-based combinations were then developed, demonstrating superior complete remission rates and PFS with the addition of either cyclophosphamide and/or rituximab to fludarabine alone (Byrd et al., 2005; S. M. O'Brien et al., 2001). More recently, an overall survival benefit has been demonstrated by the inclusion of rituximab with fludarabine and cyclophosphamide (FCR) over fludarabine and cyclophosphamide alone (FC) in patients with previously untreated CLL (Hallek et al., 2010). Furthermore, in a subset of patients with *IGHV*-mutated disease, combination chemoimmunotherapy with FCR can provide very longterm progression free survival, with 53.9% of *IGHV*-mutated patients without progression after a median follow-up of 12.8 years in a recent study (Thompson et al., 2016). Nevertheless, despite the efficacy of combination chemoimmunotherapy, there remain significant drawbacks to this approach. The standard FCR regimen commonly causes severe neutropenia and an increased risk of infections, and many older patients are not candidates for such a therapy. Additionally, there may be long-term complications of chemoimmunotherapy, including prolonged immune dysfunction and resultant late infections, pancytopenia, and therapy-related MDS/AML.

Given the possibility of long-term disease-free survival, chemoimmunotherapy remains a reasonable frontline treatment in fit patients without high-risk genetic features. However, for the majority of CLL patients, who fall outside of these criteria, the recent discovery and FDA approval of targeted therapies have revolutionized the approach to treatment. Specifically, since

2013, obinutuzumab, ibrutinib, idelalisib, and venetoclax have all been approved for the treatment of CLL in various settings, with additional next-generation agents (e.g. acalabrutinib) likely to be approved in the coming years. More detailed efficacy data on acalabrutinib and venetoclax are reviewed below, but in summary, experience with these agents has led to the following conclusions:

- Single agent use of targeted therapies (or their use in combination with anti-CD20 monoclonal antibodies) leads to overall response rates that are comparable to those seen with chemoimmunotherapy for most patients, and superior to those seen in patients with high-risk (e.g. 17p-deleted) disease.
- Targeted therapies each have their own spectrum of unique toxicities, but are generally well-tolerated and can be safely given to most older patients with other medical comorbidities.
- Single agent use of targeted therapies generally leads to partial responses rather than complete responses with an MRD-negative state. Given the low rate of MRD-negativity and the correlation between an MRD-negative state and long-term disease-free survival at least with chemoimmunotherapy (Kovacs et al., 2016), the current treatment paradigm is to continue a novel agent indefinitely until disease progression.

Thus, each category of therapy has its own particular benefits and limitations. By investigating time-limited therapy of a combination of targeted agents, this trial hopes to address some of the limitations of current treatment options (reviewed further in section 2.3).

Targeting Bruton Tyrosine Kinase in CLL
The B cell receptor (BCR) is a transmembrane protein located on the outer surface of B cells. It consists of an antibody, including the antigen recognition site, as well as a signal transduction moiety, CD79. Ultimately these two components work to transduce signals across the membrane when a cognate antigen is bound. In addition to its central role in the immune response, it is now clear that signaling through the B cell receptor pathway is important throughout the life of a B-cell, and in general promotes proliferation and survival. Furthermore, although not activated by somatic mutation, the BCR pathway is constitutively active in CLL and in fact upregulation of the BCR pathway is thought to be a hallmark of the pathophysiology underlying the disease (reviewed in (M. S. Davids & Brown, 2012)).

A key protein in the BCR pathway is Bruton Tyrosine Kinase (BTK). The role of BTK in BCR signal transduction is demonstrated by the human genetic immunodeficiency disease X-linked agammaglobulinemia and a murine X-linked immunodeficiency disease, both caused by a mutation in the *BTK* gene. These genetic diseases are characterized by reduced BCR signaling and a failure to generate mature B-cells. The BTK protein is expressed in most hematopoietic cells with the exception of T-cells and natural killer cells, but the selective effect of *BTK* mutations suggests that its primary functional role is in antigen receptor signaling in B-cells (Satterthwaite & Witte, 2000). *In vitro* inhibition of BTK results in abrogation of survival signaling downstream of the BCR, a decrease in pro-survival cytokines, and modest induction of CLL cell apoptosis (Herman et al., 2011). Moreover, inhibition of BTK in CLL cells *in vitro* with siRNA promotes apoptosis, and BTK inhibition in the TCL1 mouse model of CLL significantly delays the development of CLL, suggesting that BTK is a critical kinase for CLL

development and expansion (Woyach et al., 2014).

These preclinical data supporting BTK as an important target in CLL have been subsequently translated into the clinic with practice-changing results seen with the BTK inhibitor ibrutinib. A brief summary of some of the key results is as follows. In the randomized, double-blind phase III RESONATE study, ibrutinib monotherapy demonstrated both a PFS and OS benefit over the single agent therapy with the CD20 monoclonal antibody ofatumumab in relapsed/refractory CLL (Byrd et al., 2014). The complete response rate to ibrutinib monotherapy in this study was only 9% after 44 months of follow-up (Montillo et al., 2017). Ibrutinib has also shown efficacy in a phase II single arm study limited to patients with previously treated 17p-deleted disease, with an impressive ORR of 83% in this difficult-to-treat population (S. O'Brien et al., 2016). Finally, ibrutinib has demonstrated efficacy in the front-line setting. Compared to chlorambucil, first-line ibrutinib monotherapy demonstrated a significantly higher ORR (86% vs. 35%), longer PFS (not reached vs. 18.9 months), and longer OS (estimated survival at 24 months of 98% versus 85%) in patients over the age of 65, although again only 4% of patients demonstrated a complete response (Burger et al., 2015).

Despite ibrutinib's efficacy, the drug does have limitations. Resistance mutations in *BTK* are found in up to 85% of patients who relapse while on the drug (Woyach, Ruppert, et al., 2017). Ibrutinib use is associated with cardiac arrhythmias. There is a 5-6% incidence of atrial fibrillation after 18 months on therapy (Byrd et al., 2015), and ventricular arrhythmias and sudden cardiac death have also been reported (Lampson et al., 2017). Bruising and bleeding, including life-threatening intracranial bleeding, have been reported with ibrutinib (Burger et al., 2015; Byrd et al., 2014; Byrd et al., 2015). Both the cardiac arrhythmias and bleeding may be due to off-target inhibition of other kinases, as ibrutinib inhibits 19 other kinases with an IC_{50} of <100nM (Honigberg et al., 2010). Thus, more selective drugs such as the next-generation BTK inhibitor acalabrutinib may be safer while still retaining efficacy. Additionally, novel approaches to the use of BTK inhibitors, such as time-limited therapy, may reduce the cumulative toxicity of these agents.

Targeting the BCL-2 Pathway in CLL

Proteins in the BCL-2 family reside on the outer surface of the mitochondrial membrane and are responsible for regulating the process of mitochondrial outer membrane permeabilization (MOMP). Anti-apoptotic proteins in this family, including BCL-2, MCL-1, BCL-X_L, BCL-w and others, block the pro-death signaling cascade and thus promote cellular survival. Targeting this signaling cascade leads to tumor cell apoptosis. This is particularly true for CLL, as CLL cells are generally highly primed for apoptosis and are dependent on BCL-2 for survival (Del Gaizo Moore et al., 2007). Supporting this finding, BCL-2 protein expression levels are typically higher in CLL cells than in peripheral blood mononuclear cells. Despite this dependence on BCL-2 for survival, genetic mutations or translocations of BCL-2 family members have not been identified in CLL. This mirrors the functional dependence of CLL cells on the BCR signaling pathway, where activating mutations of key proteins such as BTK are also not present.

These preclinical data supporting BCL-2 as an important target in CLL have translated in the clinic with the oral, selective BCL-2 inhibitor venetoclax. As a single agent used for the treatment of relapsed/refractory CLL, it induces a response in 80% of patients, including a

complete response 20% (Roberts et al., 2016). When given in combination with rituximab in a phase Ib trial in the setting of relapsed/refractory disease, the complete response rate increased to 51% (Seymour et al., 2017). Notable adverse events with venetoclax include tumor lysis syndrome, neutropenia, and gastrointestinal symptoms. Tumor lysis syndrome at the time of treatment initiation was reported in early experience with the drug; this can be avoided with a careful stepwise dose escalation of venetoclax when beginning therapy. Additionally, it is hypothesized that tumor debulking prior to venetoclax administration may further decrease the incidence of tumor lysis syndrome. Venetoclax causes a grade ≥3 neutropenia in about 35-45% of patients (Roberts et al., 2016; Seymour et al., 2017), presumably due to on-target BCL-2 inhibition in neutrophil precursors. Gastrointestinal side effects such as nausea, vomiting, constipation, and diarrhea impact approximately half of all patients receiving the drug and are typically mild. Shortening the duration of exposure to venetoclax would presumably lessen the incidence of side effects such as neutropenia and gastrointestinal symptoms. Thus, these toxicities make time-limited therapy with venetoclax, such as that explored in this trial, an attractive goal.

Targeting CD20 in CLL
CD20 is a cell surface protein on B cells. The first monoclonal anti-CD20 antibody to be utilized for the treatment of CLL was rituximab, a type 1 chimeric antibody. The addition of rituximab to chemotherapy increased the response rate, progression free, and overall survival in a randomized, phase III study, the German CLL 8 study (Hallek et al., 2010). Recently, the combination of ibrutinib with rituximab was found to be safe and well-tolerated (Burger et al., 2014). In 2009, a second anti-CD20 antibody, ofatumumab, was FDA-approved for the treatment of relapsed/refractory CLL. As detailed in section 2.2.2, obinutuzumab is a type II, glycoengineered, fully humanized anti-CD20 monoclonal antibody with the ability to induce potent antibody dependent cellular cytotoxicity as well as directly inducing CLL cell death. Obinutuzumab was FDA approved in combination with chlorambucil for the initial therapy of CLL in 2014 (Goede et al., 2014) and was FDA approved in a fixed-duration combination with venetoclax for the initial therapy of CLL in 2019 (K. Fischer et al., 2019).

2.2 IND Agents

Acalabrutinib

2.2.1.1 Structure of Acalabrutinib and Preclinical Comparison to Ibrutinib

Acalabrutinib (ACP-196) is an imidazopyrazine analogue specifically designed to be a more potent and selective inhibitor of BTK to avoid off-target side effects seen with ibrutinib. Acalabrutinib binds covalently to cysteine 481 of BTK. Nonclinical studies with acalabrutinib indicate the following improvements over ibrutinib:

- Ibrutinib is a potent covalent inhibitor of EGFR (EC_{50} = 50-70nM). Acalabrutinib does not inhibit EGFR even at the highest concentration tested (10μM) (Byrd et al., 2016; Covey et al., 2015). The inhibition of EGFR may be the cause of some ibrutinib-related adverse events including diarrhea and rash.
- At physiologic concentrations, ibrutinib – but not acalabrutinib – reduced NK cell-

mediated lysis of CLL tumor cells and significantly inhibited rituximab-induced NK cell cytokine secretion. Ibrutinib's inhibition of antibody-dependent cell-mediated cytotoxicity is probably due to effects on ITK kinase, which acalabrutinib does not target. Thus, these results suggest that acalabrutinib may be a better agent than ibrutinib to use in combination with ADCC-dependent antibodies.

- Ibrutinib is associated with an increased risk of bleeding and inhibits thrombus formation in a murine model. Acalabrutinib does not have any significant effect on thrombus formation in the preclinical setting (Wu, Zhang, & Liu, 2016).

Thus, the preclinical results of acalabrutinib studies suggest that the drug may have an improved therapeutic window relative to ibrutinib; it may also be more readily combined with other agents (particularly anti-CD20 antibodies) for the treatment of CLL.

2.2.1.2 Safety of Acalabrutinib Monotherapy in Chronic Lymphocytic Leukemia

As of 01 June 2016, over 1600 subjects have received acalabrutinib as monotherapy or in combination with other agents. Here we present an aggregated safety analysis of acalabrutinib monotherapy taken from the investigator's brochure. This pooled population represents 597 acalabrutinib-exposed subjects with median exposure of 8.4 months (range 0.03-27.4 months).

The most frequently reported Grade ≥3 AEs were neutropenia (6.4%), anemia (6.0%), pneumonia (4.9%), and thrombocytopenia (2.8%). The most frequently reported related Grade ≥3 AE was neutropenia (3.9%). A total of 15 (2.5%) subjects experienced Grade 5 events. Of the 15 subjects with a reported Grade 5 event, 10 subjects (1.7%) had an infection event, including 5 subjects (0.8%) who had a reported event of pneumonia, which was the most frequently reported fatal event. Serious AEs occurred in a total of 183 (30.7%) subjects. The most frequently reported SAEs were pneumonia (4.5%) and pyrexia (2.3%). Of the 183 subjects with a reported SAE, 76 (12.7%) subjects had a reported infection event, in which pneumonia (27 [4.5%] subjects) and sepsis (7 [1.2%] subjects) were the most commonly reported. A total of 30 (5.0%) subjects experienced AEs that led to study drug discontinuation, the most frequent of which included diarrhea, pneumonia, glioblastoma multiforme, and dyspnea (2 [0.3%] subjects for each).

Across this pooled population, atrial fibrillation of any grade has been reported in 8 subjects (1.3%) with Grade ≥3 in 4 subjects (0.7%). There were no reported Grade 4 or Grade 5 events of atrial fibrillation and there were no reported events of atrial flutter. Additionally, major hemorrhage events have been reported in 9 subjects (1.5%). The most frequently reported specific preferred terms representing major hemorrhage events were epistaxis and gastric hemorrhage, reported in 4 subjects (0.7%) and 2 subjects (0.3%), respectively. The majority of these subjects had confounding factors, which included concomitant use of NSAIDs/oral anticoagulants, and/or a medical history of events such as thrombocytopenia, stomach ulcer, and epistaxis. All major hemorrhage events were Grade 1-3. The incidence rates of atrial fibrillation and major hemorrhage (after a median time on therapy of 8.4 months) are less than what would be expected after a similar follow-up time on ibrutinib.

Long term follow-up of a phase I/II multicenter single arm study of acalabrutinib in 134 patients

with relapsed/refractory CLL has been recently published (Byrd et al., 2020). At time of data analysis, patients had received acalabrutinib for a median of 41 months. The most common adverse events (all/grade≥3) were diarrhea (52%/5.2%), headache (50.7%/0%) and upper respiratory tract infection (37.3%/0.7%). Major (grade ≥3) bleeding events occurred in 5% of patients, and atrial fibrillation occurred in 7% of patients, for a rate of 2.9 per 100 patient-years.

2.2.1.3 Safety of Acalabrutinib in Combination with Obinutuzumab

A phase Ib, multicenter, single-arm, open-label study of acalabrutinib in combination with obinutuzumab enrolled subjects with R/R or untreated CLL/SLL/PLL (ACE-CL-003). Safety data from this study is of particular relevance to the AVO combination investigated here. Safety and efficacy data from 45 subjects were recently published (Woyach et al., 2020). After a median follow-up of 39 months, the safety profile was favorable. The most common adverse event was upper respiratory tract infections (73.3% any grade, 0% grade ≥3). A list of the most frequent non-hematologic adverse events is reproduced in Table 1.

Table 1: Selected Adverse Events in a Phase I Trial of Acalabrutinib and Obinutuzumab

	Relapsed/Refractory n=26		**Treatment-Naïve n=19**		**Total N=45**	
Adverse event, n (%)	**All Grade**	**Grade 3/4**	**All Grade**	**Grade 3/4**	**All Grade**	**Grade 3/4**
Any adverse event	26 (100)	20 (76.9)	19 (100)	12 (63.2)	45 (100)	32 (71.1)
Upper respiratory tract infection	19 (73.1)	0	14 (73.7)	0	33 (73.3)	0
Weight increased	18 (69.2)	2 (7.7)	14 (73.7)	2 (10.5)	32 (71.1)	4 (8.9)
Rash, maculopapular	18 (69.2)	1 (3.8)	12 (63.2)	0	30 (66.7)	1 (2.2)
Cough	16 (61.5)	0	13 (68.4)	1 (5.3)	29 (64.4)	1 (2.2)
Diarrhea	19 (73.1)	2 (7.7)	10 (52.6)	0	29 (64.4)	2 (4.4)
Headache	17 (65.4)	1 (3.8)	8 (42.1)	0	25 (55.6)	1 (2.2)
Nausea	16 (61.5)	0	8 (42.1)	0	24 (53.3)	0
Arthralgia	13 (50.0)	1 (3.8)	8 (42.1)	0	21 (46.7)	1 (2.2)
Dizziness	11 (42.3)	0	10 (52.6)	0	21 (46.7)	0
Constipation	13 (50.0)	0	7 (36.8)	0	20 (44.4)	0
Contusion	11 (42.3)	0	8 (42.1)	0	19 (42.2)	0

2.2.1.4 Efficacy of Acalabrutinib in Relapsed/Refractory Chronic Lymphocytic Leukemia

As noted above, published experience with acalabrutinib consists of a phase I/II multicenter open-label, nonrandomized, sequential group, dose escalation single arm study (ACE-CL-001) of the drug in 61 patients with relapsed/refractory CLL (Byrd et al., 2016; Byrd et al., 2020). Subjects enrolled in the relapsed/refractory CLL cohort received acalabrutinib either QD or BID. The QD dosing cohorts comprised of 100 mg (*n*=9), 175 mg (*n*=8), 200 mg (*n*=33), 250 mg (*n*=7), and 400 mg (*n*=6). The twice daily dosing cohorts included a dose of 100 mg (*n*=65) and 200 mg (*n*=6) BID. There were no dose-limiting toxicities in any cohort. The acalabrutinib dose selected for dose expansion cohorts to support Phase 2 and 3 programs was 100 mg BID, and eventually all subjects in this trial were transitioned to the 100 mg BID dose.

The median age reported for the subject population was 66 years. A total of 132 subjects were diagnosed with CLL and 2 subjects were diagnosed with SLL. The median

time from initial diagnosis was 8.1 years (range 0.2 to 25.5 years). A total of 38.1% of subjects had baseline bulky disease of ≥ 5 cm, with 7.5% having ≥10 cm. Baseline Rai stage was 0 (low risk) in 0% of subjects, I-II (intermediate risk) in 28% and III-IV (high risk) in 49% (this data was missing for 23% of subjects). Subjects had received a median of 2 (range, 1 to 13) prior systemic therapies. Baseline ECOG status was 0 in 35.8%; 1 in 60.4% and 2 in 3% of subjects. With regard to chromosomal abnormalities, 23.9% of subjects had 17p13.1 deletion; 18.6% had 11q22.3 deletion without 17p13.1; 23% had 11q22.3 deletion; and 73.6 % had unmutated *IGHV* genes. Median time on study for this population was 15.5 months.

A total of 75 out of 134 subjects remained on study treatment as of the 04 Jan 2019 cutoff date, when the median length of follow-up was 41 months (Byrd et al., 2020). Discontinuation reasons include progressive disease (21%), adverse events (11%), death (4.5%), physician decision (4.5%), and withdrawal by subject (2.2%). The best ORR, including partial responses and partial responses with lymphocytosis (PR-L) was 94%. For subjects who experienced a response of PR-L or better, 63% had a sustained response for 45 months. The rate of progression-free survival at 12 months and 18 months was 96.1 and 88.6%, respectively. Response data for this subject population is provided in the table below.

Table 2: Best Response and Overall Response Rate by Investigator Assessment in Subjects with Relapsed/Refractory CLL (ACE-CL-001)

Best Response	**All R/R Cohorts (N=134)**[a]
CR	4%
PR	84%
PRL	6%
SD	1%
PD	1%
ORR (CR+CRi+PR+PRL) (95% CI)	94% (89-97%)

Abbreviations: CR = complete response; CRi = complete response with incomplete marrow recovery; PD = progressive disease; ORR = overall response rate; PR= partial response; PRL = partial response with lymphocytosis; SD = stable disease

[a] Efficacy evaluable population includes subjects with ≥ 1 response assessment after the first dose of study drug.

2.2.1.5 Efficacy of Acalabrutinib in Treatment-Naïve Chronic Lymphocytic Leukemia

Preliminary efficacy data are available for a total of 99 subjects with treatment-naive CLL enrolled in the treatment-naive cohort of study ACE-CL-001. A subset of subjects enrolled in this cohort initially received acalabrutinib at a dose level of 200 mg QD. The subjects in this dose cohort were subsequently transitioned to the planned expansion dose of 100 mg BID. The median age reported for this subject population is 64 years. The median time from initial diagnosis is 3.4 years (range 0.1 to 16.5 years). A total of 46.5% of subjects had baseline bulky disease of ≥ 5cm, with 6.1% of subjects having ≥ 10 cm. Baseline Rai was I-II (intermediate risk) in 48.5% and III-IV (high risk) in 50.5%, with no low risk subject enrolled. Baseline ECOG status was 0 in 34.3% and 1 in 65.7% of subjects. With regard to chromosomal abnormalities, 9.9% of subjects had 17p13.1 deletion, 19.8% had 11q22.3 deletion without 17p13.1, 22% had 11q22.3 deletion, and 62% had unmutated *IGHV* genes.

Median time on study for this subject population was 14.8 months. A total of 95 out of 99 subjects remain on study treatment as of the 01 June 2016 cutoff date. Currently 2 (2%) subjects have discontinued treatment due an AE; 1 (1%) subject has discontinued treatment due to pregnancy; and 1 (1%) subject withdrew consent. The best ORR, including PR and PRL was 97.9%. For subjects who experienced a response of PRL or better, 100% had a sustained response for 12 months. The rate of progression-free survival at 12 months and 18 months was 99% each. Response data for this subject population is provided in the table below.

Table 3: Best Response and Overall Response Rate by Investigator Assessment in Treatment-Naïve Subjects with CLL (ACE-CL-001)

Best Response	**Total (N=96)[a]**
CR	0
CRi	0
PR	88 (91.7)
PRL	6 (6.3)
SD	2 (2.1)
PD	0
ORR (CR+CRi+PR+PRL) 95% CI	94 (97.9) (92.7, 99.8)

Abbreviations: CR = complete response; CRi = complete response with incomplete marrow recovery; PD = progressive disease; ORR = overall response rate; PR= partial response; PRL = partial response with lymphocytosis; SD = stable disease

[a] Efficacy evaluable population includes treated subjects who have ≥ 1 response assessment after the first dose of study drug.

Preliminary efficacy data have also been presented in abstract form for two phase 3 trials of acalabrutinib. Most relevant to this study, results from ELEVATE-TN, a phase 3 trial comparing acalabrutinib vs. acalabrutinib plus obinutuzumab vs. chlorambucil plus obinutuzumab, were presented at ASH in 2019 (Sharman et al., 2019). In this trial, 535 patients with treatment naïve CLL (and either age ≥ 65 years or <65 years with coexisting conditions) were randomized among the three arms. The study met its primary endpoint, demonstrating an improvement in PFS in the acalabrutinib plus obinutuzumab arm (not reached) versus the chlorambucil plus obinutumab arm (22.6 months). Toxicities in the acalabrutinib plus obinutuzumab arm included headache (40% any grade, 1% grade ≥3), diarrhea (39% any grade, 4% grade ≥3), and neutropenia (31% any grade, 30% grade ≥3). The rate of infusion related reactions with obinutuzumab was less when the antibody was combined with acalabrutinib (13% any grade, 2% grade ≥3) than when the antibody was combined with chlorambucil (40% any grade, 5% grade ≥3). Preliminary results from a phase 3 trial of acalabrutinib monotherapy versus investigator's choice (either bendamustine/rituximab or idelalisib/rituximab) in relapsed/refractory CLL have been presented in abstract form (Ghia et al., 2019; Hebart et al., 2020). This study also met its primary endpoint, demonstrating a superior PFS in the acalabrutinib monotherapy arm (not reached in the acalabrutinib arm versus 16.5 months in the investigator's choice arm), with no new safety signals seen.

2.2.1.6 Acalabrutinib FDA Approval

Based on a phase 2 study in mantle cell lymphoma, acalabrutinib was granted accelerated approval in October 2017 for the treatment of patients with mantle cell lymphoma who have received at least one prior line of therapy. Based on the aforementioned phase 3 ELEVATE-TN and ASCEND trials, acalabrutinib (either with or without obinutuzumab) was granted FDA approval in November 2019 for the treatment of adult patients with CLL at any line of therapy.

Obinutuzumab

2.2.1.7 Structure and Mechanism of Action of Obinutuzumab

Obinutuzumab (GA101, RO5072759, Gazyva®, Gazyvaro®) is a glycoengineered type II anti-CD20 monoclonal antibody characterized by high-affinity binding to a CD20 epitope that is different from the epitope targeted by rituximab (which is currently the most widely used anti-CD20 monoclonal antibody). Obinutuzumab was derived by humanization of the parental B Ly1 mouse antibody and subsequent glycoengineering leading to the following characteristics: high-affinity binding to the CD20 antigen, high antibody-dependent cellular cytotoxicity (ADCC), and antibody-dependent cellular phagocytosis (ADCP); low complement-dependent cytotoxicity (CDC) activity; and high direct cell death induction. Preclinical studies with obinutuzumab in comparison with rituximab showed significantly greater ADCC and ADCP, increased direct cell-death induction, and lower CDC. Superior efficacy to rituximab has also been demonstrated in various human lymphoma xenograft models.

2.2.1.8 Tolerability and Efficacy of Obinutuzumab in Chronic Lymphocytic Leukemia

Study BO20999 (GAUGUIN; NCT00517530) (Phase I)
BO20999 was an open-label, multicenter, Phase I/II study to explore obinutuzumab safety and activity in relapsed/refractory NHL and CLL. Thirteen CLL patients received obinutuzumab at doses with a range of 400–2000 mg (given as a flat dose) across four cohorts (Cartron et al., 2014). There were no dose-limiting toxicities (DLTs) and no requirement for dose reductions. Infusion-related reactions (IRRs) occurred in all CLL patients and were characterized predominantly by National Cancer Institute Common Terminology Criteria (NCI-CTC) Grade 1–2 toxicities: chills, nausea, vomiting, fever, pyrexia, hypertension, hypotension, dyspnea, and dizziness. Ten patients experienced at least one grade ≥3 adverse event, IRRs and cytopenias being most common.

Although the safety profile appeared otherwise similar between NHL and CLL, there was an increase in NCI-CTC v3.0 Grade 3/4 neutropenia noted in CLL patients, which were observed in 7 patients across the four dose levels administered. Five patients experienced Grade 4 neutropenia and 2 patients experienced Grade 3 neutropenia as the maximum severity. All received G-CSF per institutional practice, and these patients responded quickly to G-CSF support. It is important to note that these neutropenia events did not appear to be accompanied by a higher incidence of infections, as only one patient experienced febrile neutropenia and there were no deaths on study. As assessed by 2008 International Workshop on CLL (IWCLL) criteria, the end-of-treatment response rate with obinutuzumab monotherapy was 62%, with all 8 responders having a PR.

Study BO21004 (CLL11; NCT01010061) (Phase III)
This was an open-label, multicenter, three-arm randomized, Phase III study comparing the efficacy and safety of obinutuzumab + chlorambucil (GClb), rituximab + GClb (RClb), or Clb alone in previously untreated CLL patients with comorbidities (Goede et al., 2014). BO21004 enrolled 781 patients and an additional 6 patients during a safety run-in period before randomization. None of the pre-defined stopping criteria were met during this safety run-in period, and Study BO21004 opened to randomization in April 2010.

Study BO21004 included two separate stages evaluating efficacy and the primary endpoint of progression-free survival (PFS). Stage 1 evaluated obinutuzumab + chlorambucil compared with chlorambucil alone. Compared with chlorambucil, both obinutuzumab + chlorambucil and rituximab + chlorambucil demonstrated statistically significant PFS benefit compared with chlorambucil alone. Median PFS of chlorambucil vs. obinutuzumab + chlorambucil was 11.1 vs. 26.7 months, respectively, (hazard ratio [HR], 0.18 [0.13 – 0.24]; $p< 0.001$) as assessed by independent review. Additionally, obinutuzumab + chlorambucil significantly improved overall survival compared to chlorambucil monotherapy (HR 0.41 [0.23-0.74]; $p<0.002$).

In the Stage 2 analysis of this study, obinutuzumab + chlorambucil was compared with rituximab + chlorambucil. At the pre-planned interim analysis, the primary endpoint was met early and the results were released by the independent data monitoring board. Median PFS was 26.7 months vs. 15.2 months for obinutuzumab + chlorambucil vs. rituximab + chlorambucil, respectively. The ORR was 78% vs. 65%, and CR was 21% vs. 7%, respectively. There was a non-significant trend towards improved overall survival in the obinutuzumab + chlorambucil group. Notably, the rate of MRD-negativity in the bone marrow was also significant higher in the obinutuzumab + chlorambucil group versus the rituximab + chlorambucil group (19.5% vs. 2.6%, respectively) (Goede et al., 2014).

For more information related to safety and efficacy in the CLL indication, please refer to the Obinutuzumab US prescribing information (USPI).

2.2.1.9 Additional Safety Overview of Obinutuzumab

Obinutuzumab has been administered in clinical trials to greater than 1,310 patients with CD20-positive malignancies. Both in patients with NHL and with CLL, IRRs were the most common AEs in clinical trials conducted to date. Other notable adverse events include neutropenia, hepatitis B reactivation, progressive multifocal leukoencephalopathy, tumor lysis syndrome, and infections. These events appeared to be more common in patients with CLL compared to NHL.

In trials investigating the combination of obinutuzumab and CHOP, FC, chlorambucil or bendamustine, the incidence of AEs in the treatment arms with combined use was consistent with the known safety profiles of the individual study drugs. So far, no maximum tolerated dose, no dose-limiting toxicities, and no clear dose-related trends in the incidence of AEs have been determined.

A pooled analysis of safety data for obinutuzumab collected during the monotherapy studies

BO20999 and BO21003 was conducted. These two studies included a total of 205 patients with NHL (49 aggressive NHL and 156 indolent NHL patients) and 38 patients with CLL who received obinutuzumab monotherapy. In the group of 38 CLL patients treated with obinutuzumab monotherapy, the majority of patients (25 [66%]) were treated for ≥4 weeks to <6 months. Eleven patients (29%) were exposed for 6-12 months, and two patients (5%) were exposed for 12 months or longer. Eight of 38 patients (21%) with CLL were withdrawn during the treatment phase; 4 patients (11%) were withdrawn due to AEs, which indicates that AEs were mostly manageable. Almost all patients (37/38 [97%]) experienced an IRR. The number of patients with Grade 3/4 IRRs was 11/38 (29%). Infusion related reactions were predominantly associated with the first infusion, generally occurring early during the infusion, shortly after, or in some cases up to 24 hours after the completion of the infusion with obinutuzumab. The incidence and intensity of IRRs decreased with subsequent infusions of obinutuzumab. In a few patients, concurrent signs of tumor lysis syndrome (TLS) were observed.

As is typical for patients with CLL, blood and lymphatic system disorders were among the most frequently reported AEs, in particular neutropenia (13/38 patients [34%]), febrile neutropenia (5/38 patients [13%]), and thrombocytopenia (7/38 patients [18%]).

Infections and infestations were common AEs, occurring in 21/38 patients (55%). Infections reported in more than one patient were nasopharyngitis (6 patients), bronchitis and sinusitis (4 patients each), influenza and lung infection (3 patients each), and herpes zoster and oral herpes (2 patients each).

Patients with CLL appeared to be at a higher risk of experiencing an AE of special interest than patients with NHL. The largest difference in the incidences was seen for neutropenia (occurring in 47% of patients with CLL [18/38] vs. 8% of patients with aNHL [4/49] and 8% of patients with iNHL [13/156]) and treatment-related AEs associated with the infusion (100% [38/38] vs. 80% (39/49) and 83% [129/156]).

In the Phase III Study BO21004 (CLL11), comparison of obinutuzumab + chlorambucil to chlorambucil alone showed that the most common AEs (all grades, Grades 3 – 4), respectively, were IRRs (69% vs. 0, 21% vs. 0), neutropenia (40% vs. 18%, 34% vs. 16%), thrombocytopenia (15% vs. 7%, 11% vs. 3%), anemia (12% vs. 10%, 4% vs. 5%), leukopenia (7% vs. 0, 5% vs. 0), pyrexia (10% vs. 7%, < 1 vs. 0), and cough (10% vs. 7%, 0 vs. < 1%).

The incidence of IRRs was 69% with the first infusion of obinutuzumab. The incidence of Grade 3 or 4 IRRs was 21%, with 8% of patients discontinuing therapy. The incidence of reactions with subsequent infusions was 3% with the second 1000-mg dose and <1% thereafter. No Grade 3 or 4 IRRs were reported beyond the first 1000-mg infusion. Of the first 53 patients receiving obinutuzumab in the trial, 47 (89%) experienced an IRR. After this occurrence, study protocol modifications were made to require pre-medication with a corticosteroid, antihistamine, and acetaminophen. The first dose was also divided into two infusions (100 mg on Day 1 and 900 mg on Day 2). Of the 45 patients for whom these mitigation measures were implemented, 21 patients (47%) experienced a reaction with the first 1000-mg dose and <2% thereafter.

The incidence of neutropenia reported as an AE was 40% in the obinutuzumab-treated arm and

18% in the chlorambucil-alone arm, with the incidence of SAEs being 1% and 0%, respectively. Cases of late-onset n-eutropenia (occurring 28 days after completion of treatment or later) were 16% in the obinutuzumab-treated arm and 12% in the chlorambucil-alone arm. The incidence of infections was similar between arms. Thirty-eight percent of patients in the obinutuzumab-treated arm experienced an infection, 9% were Grade 3 – 4, and none were fatal. The incidence of Grade 3 or 4 tumor lysis syndrome was 2% in the obinutuzumab-treated arm vs. 0% in the chlorambucil-alone arm. AEs related to musculoskeletal disorders, including pain (System Organ Class), have been reported with obinutuzumab with higher incidence than with the comparator (17% vs. 13%, respectively).

2.2.1.10 Obinutuzumab FDA Approval

Based on the data above including the positive results of the randomized phase III BO21004 (CLL11) study, obinutuzumab received FDA approval in late 2013 for use in combination with chlorambucil in previously untreated patients with CLL. In 2019, obinutuzumab was also approved in combination with ibrutinib for the front-line treatment of CLL, as well as in combination with venetoclax for the front-line treatment of CLL.

Venetoclax

2.2.1.11 Structure and Mechanism of Action of Venetoclax

Venetoclax (ABT-199, GDC-0199, Venclexta^{TM}) is a novel, orally bioavailable, small-molecule Bcl-2 family inhibitor in the biarylacylsulfonamide chemical class. Venetoclax binds with high affinity (K_i < 0.010 nM) to the antiapoptotic protein Bcl-2, and with far lower affinity to other antiapoptotic Bcl-2 family proteins, like Bcl-X_L and Bcl-w (> 4,000-fold and > 2,000- to > 20,000-fold lower affinity than to Bcl-2, respectively). Venetoclax was designed to have a different safety profile than dual Bcl-2/Bcl-X_L inhibitors such as its predecessor molecule navitoclax. Platelets depend on Bcl-X_L for survival, and thrombocytopenia is therefore a DLT caused by inhibition of Bcl-X_L in the clinic. Venetoclax has an improved therapeutic index, and thereby maintains efficacy against tumor cells while avoiding dose-limiting thrombocytopenia.

2.2.1.12 Safety of Venetoclax in CLL Patients

The safety of venetoclax was evaluated in study M12-175 (NCT01328626), a phase I, open-label, multicenter study evaluating the safety and pharmacokinetic profile of venetoclax in a one daily dosing schedule (Roberts et al., 2016). The CLL/SLL arm consisted of 116 patients with relapsed/refractory disease in need of therapy (56 patients in the dose escalation cohort and 60 in the expansion cohort). Patients were assigned sequentially to dose-escalation groups of three patients or more according to a 3+3 design. Patients received daily venetoclax until disease progression or unacceptable toxicity.

The most important toxic effect in the dose-escalation cohort was TLS, which occurred in all three patients that received a single initial dose of 200mg or 100mg, as well as in two subsequent patients (including one who required urgent hemodialysis and one with a fatal event presumed due to TLS). Subsequently, the protocol was revised to start at a lower dose of 50 mg, and

eventually further revised to a starting dose of 20mg with an extended stepwise ramp-up. Venetoclax dosing increases weekly as follows: 20mg, 50mg, 100mg, 200mg, and finally 400mg daily. Subjects also received TLS prophylaxis according to their level of TLS risk. In the expansion cohort that followed this dosing schedule, only one subject had laboratory evidence of TLS and no subjects had clinical TLS. The most common adverse event was diarrhea in 52% of patients (grade ≥3 in 15%). The most common grade ≥3 adverse event was neutropenia (41% of subjects), although the rate of febrile neutropenia was low (6%). The neutropenia responded to growth factor administration.

M13-365 (NCT01682616) was a phase Ib study evaluating venetoclax in combination with rituximab. The most common adverse events with this combination were low-grade self-limited gastrointestinal events (diarrhea in 55% of patients and nausea in 51% of patients). The most common grade ≥3 adverse event was neutropenia in 53% of patients, developing after a median time of 51 days which was managed with G-CSF support. Five patients had TLS, including one fatality. After the fatality, the protocol was amended to include a lower venetoclax starting dose of 20mg and modified TLS prophylaxis. Among 32 patients subsequently treated, no clinical TLS was observed (Seymour et al., 2017).

In study M13-982 (NCT01889186), a phase 2, open label multicenter study evaluating the efficacy and safety of venetoclax in patients with relapsed/refractory CLL harboring a 17p13 (*TP53*) deletion, the most common treatment-emergent adverse events were neutropenia (grade ≥3 in 40% of patients) and gastrointestinal disorders (63% of patients, with grade ≥3 in 7%). All neutropenia-related events were manageable with dose interruption, dose reduction, or G-CSF support; no subject had to permanently discontinue therapy due to neutropenia. Due to the TLS events observed in the phase I studies, a stepwise ramp-up strategy was used, starting at 20mg every week the dose was increased (20mg, 50mg, 100mg, 200mg, 400mg) until subjects received the final 400mg daily dose. With this protocol in place, laboratory evidence of TLS was only reported in five patients during the ramp-up period, and in all cases resolved without clinical sequelae (Stilgenbauer et al., 2016).

2.2.1.13 Efficacy of Venetoclax in CLL Patients

The studies cited above have also demonstrated the efficacy of venetoclax in CLL patients. In the phase I study of venetoclax monotherapy (study M12-175, NCT01328626), the ORR in all patients, including both the dose-escalation cohort and the expansion cohort, was 79%. In the subgroup of patients with a 17p deletion, the ORR remained high at 71%, and in the subgroup of patients refractory to fludarabine, the ORR was 79%. Complete responses (CRs) by IW-CLL criteria were seen in 20% of patients (with comparable CR rates in the high-risk subgroups), and 5% of patients achieved minimal residual disease (MRD) negativity. A report with longer follow-up on the cohort of patients who received the recommended phase II dose of 400 mg/day was notable for an ORR of 81% (CR rate of 16%) and also highlighted the durability of responses, with an estimated 24-month PFS of 62% (M. S. Davids et al., 2016; Roberts et al., 2016).

Venetoclax was also found to be effective in the phase 2 study specifically limited to patients with relapsed 17p deleted CLL (study M13-982, NCT01889186). One hundred seven subjects

were enrolled in the main cohort. After a median duration of follow-up of 12.1 months, the overall response rate (independent review committee assessment) was 79%, with 8% of subjects having a CR/CRi. Responses appeared to be durable, although follow-up was short, as the median PFS and OS had not been reached. The estimated 12-month PFS was 72% in this difficult-to-treat population (Stilgenbauer et al., 2016). Based upon this study, venetoclax was FDA-approved as monotherapy for the treatment of patients with CLL with 17p-deletion who have received at least one prior therapy.

2.2.1.14 Combination of Venetoclax with Obinutuzumab is Safe and Effective

The combination of venetoclax and obinutuzumab has been demonstrated to be safe in the phase 1 and phase 3 settings. Preliminary results from the phase I study (GP28331) identified a venetoclax dose of 400mg and an obinutuzumab dose of 1000mg as safe in combination (Flinn et al., 2014). These doses are equivalent to the recommended doses of each agent when used as monotherapy.

Practice-changing results from a randomized phase 3 trial of venetoclax and obinutuzumab versus obinutuzumab and chlorambucil in 432 patients with previously-untreated CLL have been published (CLL14) (K. Fischer et al., 2019; Kirsten Fischer et al., 2019). Subjects in the venetoclax-obinutuzumab arm received a lead-in with three doses of weekly obinutuzumab, followed by daily dosing of venetoclax beginning on day 22 in combination with obinutuzumab. This regimen last a fixed duration of time, with obinutuzumab administration ceasing after 6 months and venetoclax administration ceasing after 12 months. The combination was found to be both safe and effective. The study met its primary endpoint, with a superior PFS in the venetoclax-obinutuzmab arm compared to the chlorambucil-obinutuzumab arm (HR 0.35, 95% CI 0.23-0.53). More patients had a complete response in the venetoclax-obinutuzumab arm (49.5%) versus the chlorambucil-obinutuzumab arm (23.1%). There were no unexpected safety signals, with the most common grade ≥3 adverse events in the venetoclax-obintuzumab arm including neutropenia (52.8% grade ≥3) and infections (17.5% grade ≥3). Based on results of this study, venetoclax and obinutuzumab were approved as a fixed-duration regimen for the front-line treatment of CLL in 2019.

2.2.1.15 Triple Combination Therapy with Venetoclax, Obinutuzumab and Ibrutinib

The combination of ibrutinib, venetoclax, and obinutuzumab is being studied in a single center phase Ib/II open label study that has completed enrollment of 63 patients, OSU-14266 (NCT02427451) (Jones et al., 2016). There are two arms to the study, one enrolling patients with relapsed/refractory disease, and one enrolling treatment naïve patients. The dosing schedule in this trial begins with obinutuzumab on cycle 1 day 1 for up to 8 cycles. Beginning in cycle 2, subjects receive daily ibrutinib. Beginning in cycle 3, subjects initiate the ramp-up dosing with venetoclax. Treatment with ibrutinib and venetoclax continues until disease progression and toxicity. Preliminary results after enrollment of 12 patients in the relapsed/refractory arm have been presented in abstract form (Jones et al., 2016). Dose-limiting toxicities were not observed at any venetoclax dosing level, establishing venetoclax 400mg daily as safe in combination with standard doses of obinutuzumab and ibrutinib. The most common grade ≥3 adverse event was neutropenia (50%); the most common grade 1/2 adverse events were bruising (83%) and IRRs

(75%). Of the six patients who were evaluable for a response, there was one patient with a CR in the marrow who was also MRD-negative; two additional patients with partial responses were also MRD-negative.

2.2.1.16 Discontinuation of Venetoclax Is Feasible

In the phase Ib, open-label, multicenter study evaluated the safety and tolerability of venetoclax in combination with rituximab in 49 patients (Seymour et al., 2017), thirteen of the 49 patients in this trial who had a deep response (CR/CRi or PR with negative minimal residual disease) ceased venetoclax therapy after a median treatment duration of 10 months. Of the ten patients still being followed at the time of study publication, all eight who were MRD-negative at the time of therapy discontinuation remained in ongoing remission after a median of 9.7 months. The two patients who ceased venetoclax therapy while MRD-positive developed clinically asymptomatic disease recurrence (rising ALC) two years later. Venetoclax was re-initiated in both patients and they both achieved a partial response on venetoclax alone.

In the CLL14 trial, venetoclax was administered for a fixed duration of 12 months in combination with six cycles of obinutuzumab. All patients then discontinued therapy regardless of depth of response. At 24 months after initiation of therapy, 82% of patients in the venetoclax/obinutuzumab arm had still not progressed, demonstrating that fixed-duration venetoclax-based therapy is a feasible (and now FDA approved) approach. Multiple other trials have now incorporated venetoclax discontinuation as part of the trial design.

2.2.1.17 Venetoclax FDA Approval

Based on the aforementioned studies, venetoclax was granted accelerated approval in April 2016 for the treatment of patients with CLL with 17p deletion who have received at least one prior line of therapy, and in June 2018 received full approval for the treatment of patients with CLL who have received at least one prior line of therapy with or without 17p deletion. In May 2019, the combination of venetoclax and obinutuzumab was approved for the frontline treatment of patients with CLL.

2.3 Rationale

Current Therapy and Its Limitations

Current FDA-approved treatment options for CLL generally fall into one of two categories: combination chemoimmunotherapy or targeted therapy with novel agents. Each of these categories has their own benefits and drawbacks. For example, combination chemotherapy with FCR has demonstrated durable (>10 years) remissions in a subset of young, fit patients with lower risk CLL. However, due to cytopenias and an increased risk of severe infections, many older patients are not candidates for such therapy. Additionally, there are long-term complications of chemoimmunotherapy, including prolonged immune dysfunction and resultant late infections, pancytopenia, and therapy-related MDS/AML.

Novel targeted therapies for CLL have revolutionized the treatment of the disease due to their

efficacy and good tolerability in a broad patient population. However, long term novel agent monotherapy rarely results in CRs or MRD negativity. This leads to the need for continuous therapy, which has several drawbacks. First, consistent exposure to one mechanism of action leads to the development of resistance mutations such as the C481S *BTK* mutation in patients treated with ibrutinib. Second, patients continue to have toxicities while on active therapy, even with these relatively well-tolerated agents. Furthermore, the challenge of long term adherence as well as the financial expense of continuous therapy is burdensome both for patients and society, leading some patients to discontinue therapy early, which can have negative effects on their outcome.

With these novel targeted therapies, we are now in the same position that we were with chemotherapy many years ago – multiple agents with demonstrated efficacy as monotherapy (or in combination with anti-CD20 antibodies), but with the anticipation that combination therapy may hold the key to more durable responses. The prospect of developing a novel-agent-only approach to time-limited therapy in CLL is now on the horizon. Such an approach may achieve the prolonged, MRD-negative remissions seen in some patients with chemoimmunotherapy, but may be more tolerable than chemoimmunotherapy, given the non-overlapping toxicity profiles of these novel agents. This may open up the approach of highly effective time-limited therapies to the majority of older, frailer CLL patients who are not good candidates for chemoimmunotherapy.

Rationale for Combining Novel Agents

The reasons for combining novel agents can be summarized as follows:
(1) <u>The combination of novel agents may lead to deeper responses, and achievement of deeper responses may lead to longer remissions</u>. Combination chemotherapy leads to deeper responses in CLL compared to single chemotherapy agents, and current data suggest that the combination of novel agents also achieves deeper remissions. For example, the combination of venetoclax and rituximab achieves higher CR/CR rates than those typically seen with venetoclax monotherapy in a similar patient population (Seymour et al., 2017). Furthermore, at least for patients treated with chemoimmunotherapy, deeper responses lead to longer remissions. Patients who achieve MRD-negative status have a longer median PFS than their MRD-positive counterparts (Kovacs et al., 2016). Thus, MRD status has prognostic importance, and achieving and sustaining MRD-negativity is the first step toward a possible cure.

(2) <u>The combination of novel agents leads to deeper responses, and achievement of deeper responses may allow for discontinuation of therapy, lessening toxicities, improving adherence, and decreasing the financial burden on patients</u>. MRD-negativity has correlated historically with a longer time until disease recurrence. Moreover, MRD-negativity may even identify some patients likely to achieve prolonged disease-free survival. This assumption is supported by the long term results of FCR therapy, which show a survival plateau in patients with *IGHV*-mutated disease who reached an MRD-negative state in the peripheral blood (Thompson et al., 2016). Based on these data, it is possible that a significant fraction of patients who achieve MRD-negativity are cured of their disease. Therapy discontinuation would be a reasonable approach in this subgroup for multiple reasons. This would allow patients to avoid the toxicities and side effects that occur with these agents (e.g. neutropenia, gastrointestinal symptoms, etc.).

Furthermore, in the event of relapse, recurrent disease that developed while off drug may still be sensitive to the drug that induced remission in the first place.

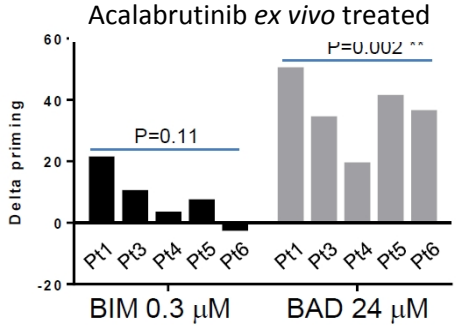
(3) The combination of novel agents may reduce toxicities compared to each agent given as monotherapy. The effects of one of the agents in the AVO triplet may decrease toxicities seen with the other agents. For example, the tumor debulking that will occur with three months of acalabrutinib and obinutuzumab lead-in therapy may reduce the risk of TLS with venetoclax. Additionally, there is preliminary data from a trial of ibrutinib and obinutuzumab in CLL to suggest that co-administration of ibrutinib with obinutuzumab decreases the rate of infusion reactions. No patients suffered infusion reactions when ibrutinib was given prior to, or simultaneously with, obinutuzumab; while 63% of patients had infusion reactions when obinutuzumab was given prior to ibrutinib (M. Davids et al., 2017). Thus, the administration of a BTK inhibitor prior to administration of obinutuzumab may decrease the high rate of infusion related reactions seen with obinutuzumab.

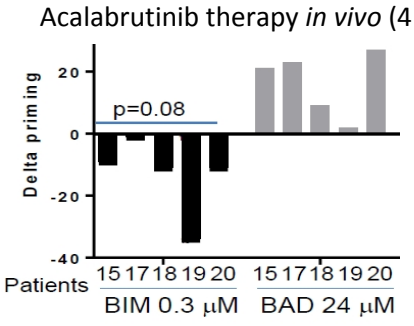
Rationale for Combining These Particular Novel Agents

Given the rationale above for exploring combination therapy of multiple novel agents, the exact agents to combine must next be considered. In this trial, the combination of acalabrutinib, venetoclax, and obinutuzumab has been chosen for investigation.

(1) Acalabrutinib: Acalabrutinib is a next-generation BTK inhibitor that is more specific for BTK than ibrutinib. This increased specificity may explain why atrial fibrillation and major hemorrhage, two potentially serious toxicities observed with ibrutinib, have not been observed as frequently with acalabrutinib monotherapy (Byrd et al., 2016). Furthermore, non-specific targeting of other kinases by ibrutinib may antagonize the action of obinutuzumab. Due to effects on ITK, ibrutinib has been shown *in vitro* to limit the efficacy of NK-cell mediated antibody dependent toxicity from rituximab (Kohrt et al., 2014). Therefore, a regimen including ibrutinib and CD20 antibodies may not be the optimal strategy for patients moving forward. Building an optimal chemotherapy-free regimen for CLL may require a more selective and better tolerated BTK inhibitor such as acalabrutinib.

(2) Venetoclax: Preclinical data suggest that the non-overlapping mechanisms of action of acalabrutinib and venetoclax enhance the effectiveness of either agent alone. We recently studied the effects of acalabrutinib on mitochondria in CLL cells using our dynamic BH3 profiling assay, a functional test which can identify the key anti-apoptotic proteins a cell depends on for its survival (see section 2.4.1). We found that acalabrutinib causes a significant delta priming in response to BAD BH3 peptide and no significant change in response to BIM BH3 peptide, suggesting that acalabrutinib selectively enhances the dependence of CLL cells on BCL-2. Importantly, we saw these same effects both in CLL cells treated *ex vivo* with acalabrutinib (left panel, below) and in CLL cells from patients treated *in vivo* with acalabrutinib monotherapy for 4 weeks on the phase I/II trial (right panel, below).





Based on these data, altho-ugh acalabrutinib can induce a modest amount of apoptosis on its own, an agent such as venetoclax that directly targets BCL-2 would be predicted to enhance the induction of apoptosis by acalabrutinib in CLL cells. Conversely, acalabrutinib's ability to release CLL cells from protective stromal microenvironments may enhance the access of venetoclax to CLL cells, suggesting that this combination may be synergistic.

(3) Obinutuzumab: Of the anti-CD20 antibodies currently available, obinutuzumab has demonstrated superior efficacy to rituximab in a phase III trial in combination with chlorambucil (Goede et al., 2014). When choosing which anti-CD20 antibody to incorporate into a chemotherapy-free regimen, we have thus chosen the antibody which has demonstrated superior efficacy.

Multiple studies are currently investigating novel agent combinations in CLL, including the OSU-14266 study investigating a similar combination of ibrutinib, obinutuzumab, and venetoclax. However, the study outlined here has some important differences from this other trial which should be highlighted to emphasize the unique contributions that our trial will make to the CLL field.

First, this study uses the BTK inhibitor acalabrutinib rather than ibrutinib. As discussed previously, acalabrutinib is more selective for BTK than ibrutinib and appears to have a more tolerable side effect profile, (see section 2.2.1), which is particularly important in combination regimens. Additionally, ibrutinib – but not acalabrutinib – may inhibit antibody-dependent cellular cytotoxicity (ADCC). Thus, acalabrutinib is the preferred BTK inhibitor to pair with obinutuzumab (which acts in large part through ADCC). Moreover, if the first generation BTK inhibitor ibrutinib is replaced over time by the second generation molecule acalabrutinib, we believe that our results could provide important preliminary data to support the further development of acalabrutinib as the preferred BTK inhibitor for use in combination therapy.

Second, in this study, patients receive lead-in therapy with the BTK inhibitor, after which obinutuzumab is introduced. This contrasts with the OSU study, where subjects receive a lead-in period with obinutuzumab followed by addition of ibrutinib. The OSU study had a relatively high rate of IRRs at 75%. Multiple lines of evidence suggest that a BTK inhibitor followed by administration of obinutuzumab is the preferred order to reduce infusion reactions. For example, data from a phase Ib/II combination trial of ibrutinib monotherapy followed by its combination with obinutuzumab (without venetoclax) had a very low rate of IRRs, with only one out of nine patients suffering a grade 1 IRR, with no grade ≥3 IRRs (Amaya-Chanaga et al., 2016). Therefore, in this trial, we will begin with a cycle of acalabrutinib prior to obinutuzumab initiation, with the hypothesis that this order may mitigate infusion-related adverse events.

Summary Statement
Acalabrutinib, venetoclax, and obinutuzumab (AVO) are three of the most active new agents in CLL. They have complementary mechanisms of action with non-overlapping toxicity profiles and preclinical data supporting their combination. Our phase 2 study will provide an initial assessment of the efficacy and safety of this combination. Our study could represent the first step

in a development plan that would eventually allow the AVO regimen to become a new standard of care for the treatment of CLL with a novel-agent-only approach.

2.4 Rationale for Study Expansion

Preliminary Safety Data
As of 1/1/2019, 25 patients have enrolled, with no screen failures. Twelve patients have reached the portion of the study where they take all three drugs in combination, and no unanticipated safety signals have been seen. The most frequent adverse events reported to date have been hematologic in nature, as is expected in the CLL patient population. We have seen a lower than expected rate of hospital admissions for TLS monitoring in patients initiating venetoclax, with only 1 patient requiring hospital admission due to persistent disease bulk at time of venetoclax initiation. This demonstrates that acalabrutinib and obinutuzumab therapy can successfully debulk the disease and decrease the risk of TLS with venetoclax initiation. One patient did experience laboratory TLS during cycle 2 with initial obinuzutumab administration, but this was prior to venetoclax initiation and resolved without clincial sequelae. No patients have discontinued study therapy due to toxicity.

Preliminary Efficacy Data
Twelve participants have reached cycle 4 day 1, when the initial disease re-staging occurs with CT scans and an optional bone marrow biopsy. Of the 12 patients who have reached this time point, 10 (83%) have already achieved a partial response by CT criteria; the remaining 2 patients had a 36% and 49% reduction in nodal burden, just missing the required 50% cut-off to achieve a partial response and thus being classified as stable disease. No patients have disease progression to date.

TP53-*Aberrant Disease Is an Unmet Need*
Long-term follow-up data from both chemoimmunotherapy trials and also early trials of novel agent monotherapy in CLL have demonstrated that patients with *TP53*-aberrant disease remain an unmet need. *TP53* aberrations include deletions of chromosome 17p (detected by either karyotype or FISH) or mutations in *TP53* itself. It has long been known that patients with *TP53* aberrations have lower response rates and shorter responses to standard chemoimmunotherapy. For example, in the CLL8 trial that compared FC to FCR for the up-front treatment of CLL, the group of patient with either *TP53* mutations or del(17p) had a PFS of approximately 12 months and an OS of approximately 40 months, shorter than any other genetically defined subgroup (Stilgenbauer et al., 2014). Regarding novel agents, the longest follow-up available is with ibrutinib (primarily in relapsed/refractory patients), and again this data suggests that while the overall response rates among those with and without del(17p) are similar, the presence of del(17p) clearly defines a subgroup of patients with a shorter duration of response to ibrutinib. After 5 years of follow-up on patients treated with ibrutinib monotherapy, a median PFS of only 26 months was seen in patients with del(17p), compared to a median PFS of 51 months for the overall group of relapsed/refractory patients. Furthermore, a multivariate analysis in this same population demonstrated that del(17p) was an independent risk factor for shorter PFS (S. O'Brien et al., 2018). In a study with shorter follow-up that examined the use of ibrutinib in the front-line setting outside of a clinical trial, patients with del(17p) again had an inferior PFS compared to those without del(17p) (Mato et al., 2018). These data support the conclusion that novel agent

monotherapy will be unable to reliably achieve durable responses in patients with *TP53*-aberrant disease and provide rationale for expanding this study to include more patients from this high-risk population. In addition to the unmet need, further justification for focusing our expansion cohort on these high risk patients is that the quality of response in our initial 12 patients treated on study to date (including 3 patients with *TP53*-aberrant disease) did not differ based on *TP53* status. This provides preliminary evidence that the "AVO" triplet regimen may potentially be able to overcome the inherent resistance of this high risk form of CLL.

Summary
Given the favorable safety profile, preliminary demonstration of efficacy, and rapid accrual of the study so far, we are expanding the study to provide additional power to more confidently report the primary outcome for the subgroup with *TP53*-abberant disease, the group most likely to benefit from this combination novel agent approach. Greater patient numbers will also allow for more robust analysis of the secondary endpoints including prespecified subgroup analyses, and to increase the number of samples available to perform the exploratory and correlative objectives.

Additionally, we plan to turn this into a multicenter study to allow our Dana-Farber Cancer Care Collaborative sites to participate. These are community sites closely tied to Dana-Farber but a different patient population from our main campus. The logistics of opening and monitoring these sites are more straightforward than for external sites, and by including these patients, we can demonstrate the feasibility of administering the AVO regimen outside of major academic medical centers.

2.5 Rationale for Modifying Treatment Discontinuation Parameters at Cycle 24

This trial was initially designed with optional treatment discontinuation at cycle 24 for subjects who achieve a complete response by IWCLL criteria as well as undetectable MRD as assessed by four color flow cytometry. We now update the trial to allow for treatment discontinuation at cycle 24 if the subject achieves undetectable MRD in the blood and marrow, regardless of IWCLL response. The primary driver for this change is recent data from CLL14 suggesting that the presence or absence of minimal residual disease, rather than the IWCLL response, is a more accurate predictor of a durable remission when a patient receives fixed-duration therapy with novel agents (Kirsten Fischer et al., 2019). For example, in the venetoclax-obinutuzumab arm of CLL14, there was no significant difference in the PFS between those who achieved undetectable MRD and a CR/CRi versus those who achieved undetectable MRD and a PR. This finding was also seen in the MURANO trial of venetoclax plus rituximab in relapsed/refractory CLL. Again, there was no significant difference in progression free survival between the group that achieved PR with undetectable MRD and the group that achieved a CR with undetectable MRD (Kater et al., 2019). Thus we now modify the treatment discontinuation parameters at cycle 24 to allow for cessation of acalabrutinib and venetoclax if the patient achieves undetectable MRD, regardless of IWCLL response. MRD will still be monitored in the peripheral blood every three months, with acalabrutinib and venetoclax resumed if it is detected. This modification to the study will allow some patients to enjoy treatment free periods, while staying within the confines of what is reasonable and safe according to currently available evidence. This change will also keep the study up-to-date with rapidly evolving standard-of-care approaches for the front-line treatment of

CLL.

2.6 Correlative Studies Background

BH3 Profiling

This study will incorporate a laboratory technique known as BH3 profiling, which is a functional assay we previously developed that detects the proximity of malignant cells to the threshold of apoptosis (what we call 'priming') through physiologic interrogation of BCL-2 family members (Ryan, Brunelle, & Letai, 2010). To perform a BH3 profile, we add individual BH3-only peptides to gently permeabilized primary CLL cells and use fluorescence activated cell sorting (FACS) to determine the amount of mitochondrial depolarization induced by each peptide, as measured by cytochrome c release. We previously found that in a small, heterogeneously treated cohort of CLL patients, increased priming was associated with improved clinical response (M. S. Davids et al., 2012). Building on these initial studies, we will incorporate BH3 profiling into this clinical trial to determine whether priming predicts degree of clinical response in this larger, homogeneously treated patient population.

Genomic Markers

We will perform whole exome sequencing on CLL cells and normal tissue from patients at baseline to evaluate for somatic mutations that may confer drug sensitivity and resistance. Our group and others have recently identified recurrent somatic mutations in the CLL genome which appear to associate with prognosis; these include *NOTCH1* and *SF3B1* (Wang et al., 2011). At present, whether other recurrent mutations associate with prognosis is less clear. Our group has also recently found that the presence of subclonal driver mutations was associated in a retrospective analysis with time to next treatment (Landau et al., 2013). In this trial, we will assess all of these recently described mutations as well as the presence of a subclonal driver mutation as potential predictors of response and progression-free survival. We will also bank samples at time of relapse for repeat analysis by whole exome sequencing to assess for the acquisition of resistance mutations. We will also evaluate established CLL prognostic markers such as cytogenetics by FISH, *TP53* mutation, IGHV status, and ZAP-70 status, and will determine whether these factors are associated with response to AVO.

Single Cell Sequencing and Cell Free DNA Sequencing Studies

Next generation sequencing (NGS) techniques have the potential for revolutionizing the treatment of CLL by allowing the detection of minimal residual disease or resistance mutations at levels below the sensitivity of current assays. We plan on using single cell DNA sequencing and RNA sequencing to identify the presence of oncogenic mutations, resistance mutations, and transcriptomic profiles of circulating CLL cells at various time points during the course of therapy. We will also retain plasma samples collected at the same time for cell-free DNA analysis, making use of an orthogonal assay to verify or expand upon results obtained from single cell sequencing.

ClonoSEQ Next Generation Sequencing Studies

Although conventional minimal residual disease (MRD) testing in CLL provides reasonable sensitivity for residual disease detection, novel methods of MRD-detection may allow even more sensitive detection with the potential for better prognostic power. Adaptive, Inc. has an assay known as ClonoSEQ, in which a baseline sample of genomic DNA is extracted from tumor cells, amplified using locus-specific primer sets for *IGH* and *IGK* rearrangements, and sequenced. Once a patient achieves a MRD-negative state, a peripheral blood sample is examined to determine whether tumor DNA is still detectable. This will be one of the first novel-agent only based studies to utilize ClonoSEQ MRD testing in parallel with conventional MRD testing for CLL patients who achieve a CR.

3. PARTICIPANT SELECTION

3.1 Eligibility Criteria

Unless otherwise specified, laboratory tests required for eligibility must be completed within 2 weeks prior to start of protocol therapy. Baseline tumor measurements by CT or PET/CT scan must be performed within approximately 30 days of starting study treatment. Bone marrow biopsy within approximately 6 months prior to study treatment will be acceptable if there has not been significant anti-cancer therapy given since the test was performed.

Inclusion Criteria

Participants must meet the following criteria on screening examination to be eligible to participate in the study:

- Subjects must meet IWCLL 2018 guidelines for the diagnosis of CLL or SLL, or WHO 2017 guidelines for the diagnosis of B-prolymphocytic leukemia (B-PLL)

- In cohort 2, subjects must have *TP53*-aberrant disease defined as:
    - Del(17p) detected by karyotype, FISH, or next-generation sequencing; OR
    - *TP53* mutation

- Participants must have measurable disease (lymphocytosis > 5,000 / µl, or palpable or CT measurable lymphadenopathy ≥ 1.5 cm, or bone marrow involvement ≥30%).

- Subjects must not have received any prior systemic therapy for CLL or SLL due to previously meeting IWCLL 2018 guidelines and must currently have an indication for treatment as defined by the IWCLL 2018 guidelines, or must not have received therapy for B-PLL and must currently have an indication for treatment:
    - Massive or progressive or symptomatic splenomegaly; OR
    - Massive lymph nodes, nodal clusters, or progressive lymphadenopathy; OR
    - Significant fatigue (i.e. ECOG PS 2 or worse; cannot work or unable to perform usual activities); OR
    - Fever ≥ 100.5°F for 2 or more weeks without evidence of infection; OR
    - Night sweats for ≥ 1 months without evidence of infection; OR
    - Presence of weight loss ≥ 10% over the preceding 6 months; OR
    - Progressive lymphocytosis with an increase of ≥ 50% over a 2-month period or lymphocyte doubling time of less than 6 months; OR
    - Evidence of progressive marrow failure as manifested by the development of or worsening of anemia and/or thrombocytopenia.
    - Autoimmune anemia and/or thrombocytopenia that is poorly responsive to corticosteroids and another standard therapy such as rituximab
    - Symptomatic or functional extranodal involvement; OR

- Age greater than or equal to 18 years. Because CLL is extremely rare in persons <18 years of age, children are excluded from this study.

- ECOG performance status ≤2 (see Appendix A)

- Participants must have adequate organ and marrow function as defined below:
    - total bilirubin ≤1.5 times upper limit of normal, unless there is disease involvement of the liver, hemolysis, or a known history of Gilbert's disease, in which case direct bilirubin must be ≤3 times the upper limit of normal
    - AST and ALT ≤ 2.5 times the upper limit of normal. If there is hemolysis or documented disease involvement of the liver, then patients with any AST or ALT abnormalities remain eligible.
    - creatinine clearance (CrCl) ≥ 50 mL/min using 24-hour urine collection for creatinine clearance or calculated CrCl OR calculated glomerular filtration rate (GFR) ≥ 50 mL/min/1.73m^2 using the CKD-EPI equation
    - PT/INR ≤2 times the upper limit of normal and PTT ≤2 times the upper limit of normal
    - Absolute neutrophil count ≥750 cells/mm^3 (0.75 x 10^9/L) or ≥500 cells/mm^3 in subjects with documented bone marrow involvement
    - Platelet count without transfusional support must be ≥50,000 cells/mm^3 or ≥ 30,000 cells/mm^3 in subjects with documented bone marrow involvement

- The effects of the study drugs on the developing human fetus are unknown. Women of child-bearing potential must agree to remain abstinent or use highly effective contraception (defined as contraceptive measures that result in a failure rate of <1% per year) during the treatment period and for at least 2 days after the last dose of acalabrutinib, 30 days after the last dose of venetoclax or 18 months after the last dose of obinutuzumab, whichever is longer. Men with female sexual partners of childbearing potential should agree to remain abstinent or use contraceptive measures which include a condom plus an additional contraceptive method that together result in a failure rate of< 1% per year during the treatment period and for at least 30 days after the last dose of venetoclax or 6 months after the last dose of obinutuzumab, whichever is longer. Men should refrain from donating sperm during the same period. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately.

- Ability to understand and the willingness to sign a written informed consent document.

Exclusion Criteria

Participants who exhibit any of the following conditions at screening will not be eligible for admission into the study:

- Participants who have a history of other malignancies except:
    - Malignancy treated with curative intent and with no known active disease present and felt to be at low risk for recurrence by treating physician. Current adjuvant hormonal therapy for disease treated with curative intent is permissible.
    - Adequately treated non-melanoma skin cancer or lentigo maligna without evidence of disease.

  - Adequately treated carcinoma in situ without evidence of disease.
  - Low-risk prostate cancer on active surveillance

- Participants who are receiving any other investigational agents.

- History of severe allergic reactions attributed to compounds of similar chemical or biologic composition to obinutuzumab, venetoclax, or acalabrutinib. Patients with reactions to other CD20 monoclonal antibodies (e.g. rituximab, ofatumumab) are not excluded.

- Unable to receive prophylactic treatment for pneumocystis, herpes simplex virus (HSV), and herpes zoster (VZV) at start of treatment

- Known or suspected Richter's transformation or known CNS involvement

- Concurrent systemic immunosuppressant therapy (e.g., cyclosporine A, tacrolimus, etc., within 28 days of the first dose of study drug or chronic administration of >20 mg/day of prednisone within 7 days of the first dose)

- Vaccinated with live, attenuated vaccines within 4 weeks of first dose of study drug.

- Ongoing or recent infection requiring intravenous antimicrobials at time of screening. Patients with ongoing use of prophylactic antibiotics are eligible as long as there is no evidence of active infection and the antibiotic is not included on the list of prohibited medications.

- Known bleeding disorders (e.g. von Willebrand's disease) or hemophilia.

- Presence of a gastrointestinal ulcer diagnosed by endoscopy within 3 months before screening.

- History of stroke or intracranial hemorrhage within 6 months prior to enrollment.

- Major surgery within 4 weeks of first dose of study drug. If a subject had major surgery greater than 4 weeks prior to the first dose, they must have recovered adequately from any toxicity and/or complications from the intervention before the first dose of study drug.

- Currently active, clinically significant cardiovascular disease, such as uncontrolled arrhythmia or Class 3 or 4 congestive heart failure as defined by the New York Heart Association Functional Classification; or a history of myocardial infarction, unstable angina, or acute coronary syndrome within 6 months prior to randomization. Patients with atrial fibrillation are allowed as long as they are adequately rate controlled.

- Baseline QTcF >480 ms. NOTE: This criterion does not apply to patients with a left bundle branch block.

- Patients who require warfarin or other vitamin K antagonists for anticoagulation (other anticoagulants are allowed).

- Patients who require treatment with proton pump inhibitors (see Appendix F). Subjects receiving proton pump inhibitors who switch to H2-receptor antagonists or antacids are eligible for enrollment on this study.

- Patients who require concurrent treatment with strong CYP3A inhibitors or strong CYP3A inducers are excluded from the study. If patients are receiving strong CYP3A inhibitors/inducers at time of screening but do not require continuous administration of these agents, these patients are eligible if there is a 3-day washout period between discontinuation of the strong CYP3A inhibitor/inducer and initiation of the first study drug, acalabrutinib.

- Patients who require concurrent treatment with P-gp inhibitors or narrow therapeutic index P-gp substrates are excluded from the study. If patients are receiving P-gp inhibitors or narrow therapeutic index P-gp substrates at time of screening but do not require continuous administration of these agents or can be switched to alternative agents, these patients are eligible if there is a 3-day washout period between discontinuation of the P-gp inhibitor and initiation of venetoclax.

- Unable to swallow tablets or malabsorption syndrome, disease significantly affecting gastrointestinal function, or resection of the stomach or small bowel if thought by the investigator to compromise systemic absorption, active, symptomatic inflammatory bowel disease or ulcerative colitis, or partial or complete bowel obstruction.

- Lactating or pregnant women are excluded from this study because venetoclax has been shown to decrease implantation, litter size, live fetuses, and fetal body weight in animal models. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with venetoclax*,* breastfeeding should be discontinued if the mother is treated with venetoclax. These potential risks may also apply to other agents used in this study.

- Patients with human immunodeficiency virus (HIV), human T cell leukemia virus (HTLV), active hepatitis C virus (HCV), or active hepatitis B virus (HBV) infection. HIV-positive participants are ineligible because of the potential for pharmacokinetic interactions between certain components of anti-retroviral therapy and venetoclax. Patients who are positive for hepatitis B core antibody or hepatitis B surface antigen must have a negative polymerase chain reaction (PCR) result before enrollment. Those who are PCR positive will be excluded. Those who are positive for either hepatitis B surface antigen and/or hepatitis B core antibody but negative for HBV DNA will be managed as detailed in section 5.4. Patients who are positive for HCV antibody must be negative for HCV by polymerase chain reaction (PCR) to be eligible for study participation.

- Unwilling or unable to participate in all required study evaluations and procedures.

- Unable to understand the purpose and risks of the study and to provide a signed and dated informed consent form (ICF) and authorization to use protected health information (in accordance with national and local subject privacy regulations)

- Significant co-morbid condition or disease which in the judgment of the Principal Investigator would place the patient at undue risk or interfere with the study

- Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements, compromise the subject's safety, or put the study outcomes at undue risk.

3.2 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups are eligible for this trial. The eligibility criteria will not have a negative impact on enrollment of women, particularly since most women with CLL are postmenopausal. We will make every effort to enroll minorities on this study.

4. REGISTRATION PROCEDURES

4.1 Informed Consent

The subject must read, understand and sign the consent form approved by the institutional review board, confirming his or her willingness to participate in this study before initiating any screening activity that is not standard of care. Subjects must also grant permission to use protected health information, if required by local regulations.

4.2 Medical History

Collect and record the subject's complete history through review of medical records and by interview. Concurrent medical signs and symptoms must be documented to establish baseline severities. A disease history, including the date of initial diagnosis and list of all prior anticancer treatments, and responses and duration of response to these treatments, also will be recorded.

4.3 Confirmation of Eligibility

Subject eligibility for enrollment will be assessed per Section 3.1. All screening procedures, unless otherwise indicated, should be completed within the timeframe as detailed in section 10.

4.4 General Guidelines for DF/HCC Institutions

Institutions will register eligible participants in the Clinical Trials Management System (CTMS) OnCore. Registrations must occur prior to the initiation of protocol therapy. Any participant not registered to the protocol before protocol therapy begins will be considered ineligible and

registration will be denied.

An investigator will confirm eligibility criteria and a member of the study team will complete the protocol-specific eligibility checklist.

Following registration, participants may begin protocol therapy. Issues that would cause treatment delays should be discussed with the Overall Principal Investigator (PI). If a participant does not receive protocol therapy following registration, the participant's registration on the study must be canceled. Registration cancellations must be made in the CTMS (OnCore) as soon as possible, with an appropriate date and reason entered.

4.5 Registration Process for DF/HCC Institutions

DF/HCC Standard Operating Procedure for Human Subject Research Titled *Subject Protocol Registration* (SOP #: REGIST-101) must be followed.

4.6 General Guidelines for Other Investigative Sites

Eligible participants will be entered on study centrally at Dana-Farber Cancer Institute by the lead site project manager. All sites should call the project manager to verify dose level and or slot availabilities.

Following registration, participants should begin protocol therapy as soon as feasible. Issues that would cause treatment delays should be discussed with the Overall PI. If the subject does not receive protocol therapy following registration, the subject must be taken off-study in the CTMS (OnCore) with an appropriate date and reason entered.

4.7 Registration Process for Other Investigative Sites

To register a participant, the following documents should be completed by the research nurse or data manager and faxed or e-mailed to the lead site project manager.

- Copy of relevant medical records that support the items on the eligibility checklist (with PMI redacted)
- Participant consent form signed by both the consenting physician investigator, and the participant
- HIPAA authorization form (if applicable)
- Completed eligibility checklist

The participating site will e-mail the lead site project manager to verify eligibility. The project manager will follow DF/HCC policy (REGIST-101) and register the participant on the protocol. The lead site project manager will email the participant study number, and if applicable the dose treatment level, to the participating site. The lead site may also contact the participating site and verbally confirm registration.

5. TREATMENT PLAN

5.1 Treatment Regimen

Treatment will generally be administered on an outpatient basis, except in the case that a patient meets the high risk criteria for TLS syndrome at the time of venetoclax initiation (see section 6.1.2 below). The length of each treatment cycle is 28 days. Expected toxicities and potential risks as well as dose modifications for venetoclax, obinutuzumab, and acalabrutinib are described in Section 6 (Expected Toxicities and Dosing Delays/Dose Modification). No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the participant's malignancy.

Acalabrutinib Lead-In: Cycle 1

All eligible subjects will begin by receiving one cycle (days 1-28) of acalabrutinib monotherapy. Acalabrutinib will be given continuously at the recommended phase 2 dose of 100mg orally twice a day (Byrd et al., 2016).

Regimen Description					
Agent	**Premedications; Precautions**	**Dose**	**Route**	**Schedule**	**Cycle Length**
Acalabrutinib	None	100mg	oral	bid	28 days (4 weeks)

Acalabrutinib and Obintuzumab Combination Therapy: Cycles 2-3

For the next two 28-day cycles, subjects will receive acalabrutinib and obinutuzumab combination therapy. Acalabrutinib will be continued at the dose of 100mg orally twice daily. Obintuzumab will be given as per the route, timing, and dose of the FDA-approved label. On cycle 2 day 1, after the morning dose of acalabrutinib is taken, obinutuzumab will begin with the 100mg intravenous dose. On cycle 2 day 2, after the morning dose of acalabrutinib is taken, the subject will receive 900mg of obinutuzumab. On cycle 2 day 8 and again on cycle 2 day 15, the subject will receive 1000mg of obinutuzumab. Finally, on cycle 3 day 1, the subject will receive 1000mg of obinutuzumab in addition to acalabrutinib.

Regimen Description					
Agent	**Premedications; Precautions**	**Dose**	**Route**	**Schedule**	**Cycle Length**
Acalabrutinib	None	100mg	oral	bid	28 days (4 weeks)
Obinutuzumab	Premedication before each infusion, as described in section 5.3.2	100mg	iv	Cycle 2 Day 1	
		900mg	iv	Cycle 2 Day 2	
		1000mg	iv	Cycle 2 Day 8	
		1000mg	iv	Cycle 2 Day 15	
		1000mg	iv	Cycle 3 Day 1	

Acalabrutinib, Obintuzumab, and Venetoclax Combination Therapy: Cycles 4-7

For the next four 28-day cycles, subjects will receive acalabrutinib, obinutuzumab, and venetoclax combination therapy. Acalabrutinib will be continued at the dose of 100mg orally twice daily. Obintuzumab will be given as four monthly 1000mg intravenous doses. Venetoclax will be given in a ramp-up dose-escalation fashion. Patients will be restaged after cycle 3 and this will be used to determine tumor lysis syndrome risk.

On days where multiple drugs are taken, unless specified otherwise in this document, there is no recommended order in which to take the drugs. The order in which the drugs can be taken is at the discretion of the treating investigator. On days when Obintuzumab is administered, the oral agents are to be dosed prior to the initiation of Obintuzumab.

Regimen Description					
Agent	**Premedications; Precautions**	**Dose**	**Route**	**Schedule**	**Cycle Length**
Acalabrutinib	None	100mg	oral	bid	28 days (4 weeks)
Obinutuzumab	Premedication before each infusion, as described in section 5.3.2	1000mg	iv	Cycle 4 Day 1	
		1000mg	iv	Cycle 5 Day 1	
		1000mg	iv	Cycle 6 Day 1	
		1000mg	iv	Cycle 7 Day 1	
Venetoclax	TLS monitoring labs, IV fluids, allopurinol prior to first dose (or rasburicase if deemed necessary)	20mg	oral	Cycle 4 Day 1	
		50mg	oral	Cycle 4 Days 2-7	
		100mg	oral	Cycle 4 Days 8-14	
		200mg	oral	Cycle 4 Days 15-21	
		400mg	oral	Cycle 4 Days 22-28	
		400mg	oral	daily, Cycles 5-7	

Acalabrutinib and Venetoclax Combination Therapy: Cycles 8-15

For the next eight 28-day cycles (cycles 8-15), patients will receive acalabrutinib and venetoclax combination therapy. Acalabrutinib will be continued at the dose of 100mg orally twice daily. Venetoclax will be continued at the dose of 400mg orally once daily.

Regimen Description					
Agent	**Premedications; Precautions**	**Dose**	**Route**	**Schedule**	**Cycle Length**
Acalabrutinib	None	100mg	oral	bid	28 days (4 weeks)
Venetoclax	None	400mg	oral	daily	

Cycles 15-24 and beyond

A bone marrow biopsy and response assessment will occur at the end of cycle 15. In patients who are negative for minimal residual disease in the bone marrow and in the peripheral blood at that time (as assessed by flow cytometry with a sensitivity of 10^{-4}) and who are also in complete

remission or complete remission with incomplete marrow recovery (all as defined in section 11.3 and Appendix B), acalabrutinib and venetoclax will have the option to discontinue therapy at the conclusion of cycle 15. Patients will receive peripheral blood monitoring for minimal residual disease at cycles 18, 20, 22, 25, and then every three cycles for two additional years. If recurrence is detected, the patient will undergo restaging with a CT scan (if not done within the prior three months) and a bone marrow biopsy and rapid heme panel (RHP) (if not done within the prior six months). Additionally, if recurrence is detected, the patient may resume acalabrutinib 100mg orally twice daily and venetoclax 400mg orally daily, continuing until disease progression or unacceptable toxicity.

In patients who are positive for minimal residual disease in the bone marrow or blood at the end of cycle 15 (in the setting of CR/CRi), or in patients who are in partial remission at the completion of cycle 15 (irrespective of MRD status), acalabrutinib 100mg orally twice daily and venetoclax 400mg orally daily will be continued through cycle 24. At cycle 24, patients will receive another bone marrow biopsy and disease assessment. If this demonstrates undetectable MRD in both the blood and bone marrow, patients will have the option to discontinue therapy at the conclusion of cycle 24. If the cycle 24 bone marrow biopsy or peripheral blood is positive for MRD, he/she will continue acalabrutinib 100mg orally twice daily and venetoclax 400mg orally daily; both drugs will be continued (acalabrutinib at 100mg bid and venetoclax at 400mg daily) until disease progression or unacceptable toxicity. Patients will have the option to discontinue therapy at any point after cycle 25 if they achieve two consecutive negative MRD assessments that are 3 months apart.

Patients who have discontinued therapy will receive peripheral blood monitoring for minimal residual disease (as assessed by flow cytometry with a sensitivity of 10^{-4}) every three cycles for 2 years after completion of cycle 24. Recurrent disease is defined as the appearance of progressive disease as defined by 2018 IWCLL criteria (see Appendix B); in addition, the reappearance of a clonal population on peripheral blood MRD testing after achievement of undetectable MRD will also be considered recurrent disease. If recurrence is detected, the patient will undergo restaging with a CT scan (if not done within the prior three months) and a bone marrow biopsy (if not done within the prior six months). Additionally, the patient may resume acalabrutinib and venetoclax. Acalabrutinib should be resumed at the dose level the patient was receiving when the drug was discontinued. For venetoclax, subjects may resume the drug at their prior dose only if disease recurrence is initially detected with peripheral blood flow cytometry MRD monitoring only. In that case when dosing is resumed patients should have TLS labs checked pre-dose, 6-8 hours post-dose, and 24 hours post-dose for close monitoring of TLS. If disease recurrence is detected through alternative means (e.g. physical exam, radiographic imaging, or complete blood count), then the subject should be assessed for risk of tumor lysis syndrome as detailed in section 6.1.2 and venetoclax should be reinitiated following the ramp-up schedule outlined in section 5.3.2. Subjects will then receive acalabrutinib and venetoclax continuously until experiencing progression or intolerable toxicity.

In patients who have stable disease or progressive disease at cycle 15 as compared to pre-treatment baseline scans, investigational therapy will be discontinued and patients will come off treatment. Patients deriving clinical benefit from study treatment may continue on therapy at

the discretion of the treating investigator to bridge to next treatment even if there is evidence of progressive disease.

5.2 Pre-Treatment Criteria

Cycle 1, Day 1

Participants must meet all of the inclusion criteria, have none of the exclusion criteria, and must be registered to the protocol prior to initiating therapy.

C1D1 laboratory results need not meet eligibility parameters again provided that they met criteria at screening and in the opinion of the treating investigator do not pose any undue risk to the participant.

Cycle 4 Days 1, 2, 8, 15, 22

These are the days of venetoclax initiation or dose escalation. Tumor lysis labs (potassium, uric acid, phosphorous, calcium, and creatinine) must be checked prior to the patient receiving their dose of venetoclax, and must not meet Cairo Bishop criteria (Appendix D) for TLS. If criteria for TLS are met, the venetoclax dose should be delayed until electrolytes return to baseline.

All cycles subsequent to cycle 1

If labs are checked prior to cycle initiation, then the following criteria must be met:

- ANC must be $\geq 500/mm^3$
- Platelet count must be $\geq$ 20k/μL (may be 10-20k/μL if transfusion support is also given)
- All non-hematologic toxicities must have resolved to $\leq$ Grade 2, or to the patient's baseline condition

5.3 Agent Administration

The participant will be requested to maintain a medication diary of each dose of medication. The medication diary will be returned to clinic staff at study visits. The date, time, and quantity of drug taken will be recorded in a drug self-administration diary. Patients should record any deviation from taking the full daily dose (e.g. vomited doses, missed doses, doses reduced due to missing or lost tablets). Missed doses outside the windows defined above or vomited doses should not be taken or repeated. At each visit, a sufficient number of doses will be dispensed so that the patient will have the required number of doses until the next visit, taking into account the window for the next visit. In extenuating circumstances study drug can be shipped directly from the study site pharmacy to the patient.

On days where multiple drugs are taken, unless specified otherwise in this document, there is no recommended order in which to take the drugs. The order in which the drugs can be taken is at the discretion of the treating investigator. On days when obintuzumab is administered, the oral agents are to be dosed prior to the initiation of obintuzumab.

Acalabrutinib

Acalabrutinib 100 mg will be administered orally twice daily and given in continuous 28-day cycles. The first dose of acalabrutinib 100 mg (one 100 mg tablet) will be self-administered by patients on C1D1. Acalabrutinib should be administered with 8 ounces (approximately 240 mL) of water. The tablets should be swallowed intact and patients should not attempt to break or crush tablets, or dissolve them in water. Doses of acalabrutinib should be taken approximately 12 hours apart, and each dose should be taken at approximately the same time each day. Acalabrutinib may be administered without regard to meals. If a dose is not taken at the scheduled time, it can be taken as soon as possible up to 3 hours later on the same day, with a return to the normal schedule the following day. If a dose is missed by more than 3 hours, it should be skipped and the next dose should be taken at its regularly scheduled time. The patient should not take extra tablets to make up the missed dose. The total length of acalabrutinib therapy will be 15 cycles for patients who have achieved MRD-negative CR/CRi at the conclusion of cycle 15 and choose to discontinue therapy. Patients who have not achieved MRD-negative CR/CRi at cycle 15 will continue acalabrutinib (with venetoclax) until cycle 24, at which point there will be another response assessment of the bone marrow. Patients who have achieved an MRD-negative CR/CRi at this time will have the option to cease therapy and begin surveillance; patients who are MRD positive will continue acalabrutinib (with venetoclax) until disease progression.

Obinutuzumab

Obinutuzumab will be administered by intravenous infusion over six 28-day cycles, beginning on cycle 2. Obinutuzumab will be given as the following doses: 100mg on D1, 900mg on D2, 1000mg on D8 and D15 in cycle 2, then five 28-day cycles in cycles 3-7. The schedule and details for obinutuzumab dosing are as follows. The cycle numbers are specific to this trial:

Day of treatment cycle		**Dose of Obinutuzumab**	**Rate of infusion** (in the absence of infusion reactions/hypersensitivity during previous infusions)
Cycle 2 (loading doses)	Day 1	100 mg	Administer at 25 mg/hr over 4 hours. Do not increase the infusion rate.
	Day 2	900 mg	Administer at 50 mg/hr. The rate of the infusion can be escalated in increments of 50 mg/hr every 30 minutes to a maximum rate of 400 mg/hr.
	Day 8	1000 mg	If no infusion reaction occurred during the previous infusion and the final infusion rate was 100 mg/hr or faster, infusions can be started at a rate of 100 mg/hr and increased by 100 mg/hr increments every 30 minutes to a maximum of 400 mg/hr.
	Day 15	1000 mg	
Cycles 3-7	Day 1	1000mg	

Obinutuzumab must be administered in a clinical setting (inpatient or outpatient). Full emergency resuscitation facilities should be immediately available, and patients should be under close supervision by a clinician at all times. Obinutuzumab should be given as a slow IV infusion through a dedicated line. IV infusion pumps should be used to control the infusion rate of obinutuzumab. Do not administer as an IV push or bolus. After the end of the first infusion, the IV line should remain in place for at least 2 hours in order to be able to administer IV drugs if necessary. If no AEs occur after 2 hours, the IV line may be removed. For subsequent infusions, the IV line should remain in place for at least 1 hour from the end of infusion; if no AEs occur after 1 hour, the IV line may be removed. There will be 1 hour observation following Obinutuzumab infusion in Cycle 2, and observation may be waived if participant has tolerated previous infusions without issue beginning with Cycle 3. If less than 100mg is delivered on the cycle 2 day 1 dose, the patient may still receive the 900mg dose on cycle 2 day 2, as per standard of care.

Patients who experience a significant obinutuzumab infusion reaction may be treated with an obinutuzumab desensitization protocol as per institutional standards, provided that this is approved by the PI and that the desensitization is done in collaboration with an allergist.

Premedication Requirements

Infusion-Related Reactions

Since some patients may develop hypersensitivity or other infusion-related reactions (IRRs) to obinutuzumab, pre-medication is recommended to reduce the risk of infusion reactions. Premedication will be given as outlined below. Cycle numbers are specific to this trial:

Day of treatment cycle	Patients Requiring Pre-medication	Premedication	Administration
Cycle 2, days 1 and 2	All patients	Intravenous glucocorticoid: methylprednisolone 80mg[1]	Completed at least one hour prior to obinutuzumab infusion.
		acetaminophen 1000mg orally	Taken at least 30 minutes before obinutuzumab infusion.
		diphenhydramine 50mg intravenously or orally	
All subsequent infusions	All patients	acetaminophen 1000mg orally	Taken at least 30 minutes before obinutuzumab infusion.
	Patients with an IRR (Grade 1-2) with the previous infusion	acetaminophen 1000mg orally	Taken at least 30minutes before obintuzumab infusion.
		diphenhydramine 50mg intravenously or orally	
	Patients with a Grade 3 IRR with the previous infusion OR with a lymphocyte count > $25x10^9$ / L prior to next treatment	intravenous glucocorticoid: methylprednisolone 80mg[1]	Completed at least one hour prior to obinutuzumab infusion.
		acetaminophen 1000mg orally	Taken at least 30 minutes before obinutuzumab infusion.
		diphenhydramine 50mg intravenously or orally	

[1] Hydrocortisone is not recommended as it has not been effective in reducing the rate of infusion reactions.

If a patient experiences any grade infusion reaction during infusion, adjust the infusion as outlined below:

- Grade 4 (life threatening): Stop infusion and discontinue obinutuzumab.
- Grade 3 (severe): Temporarily interrupt infusion and treat symptoms. Upon resolution of symptoms, restart infusion at no more than half the previous rate (the rate being used at the time that the infusion reaction occurred) and, if patient does not experience any infusion reaction symptoms, infusion rate escalation may resume at the increments and intervals as appropriate for the treatment dose.
- Grade 1-2 (mild to moderate): Reduce infusion rate and treat symptoms. Upon resolution of symptoms, continue infusion and, if patient does not experience any infusion reaction symptoms, infusion rate escalation may resume at the increments and intervals as appropriate for the treatment dose.

Hypotension may be expected to occur during obinutuzumab infusions. Withholding of antihypertensive treatments can be considered for 12 hours prior to and throughout each obinutuzumab infusion and for the first hour after administration. Patients at acute risk of hypertensive crisis should be evaluated for the benefits and risks of withholding their

hypertensive medication.

Venetoclax

Venetoclax will be administered orally once daily and given in continuous 28-day cycles. Venetoclax administration will begin on cycle 4 day 1. If patients are in an MRD-negative CR/CRi at the end of cycle 15, then patients will have the option to discontinue venetoclax at this time. If patients are not in an MRD-negative CR/CRi at the end of cycle 15, then venetoclax will be continued (along with acalabrutinib) through cycle 24, at which point another response assessment will occur. If patients are not in an MRD-negative state at this time, venetoclax (with acalabrutinib) will continue indefinitely until progression or toxicity. If patients are in an MRD-negative state, they will have the option to discontinue venetoclax and the patient will be monitored for disease recurrence.

Venetoclax is initiated with a dose ramp-up to reduce the risk of tumor lysis syndrome. In this trial, we will use a slightly modified dose escalation strategy since the patients will have already had significant debulking of their disease with three prior cycles of acalabrutinib and two prior cycles of obinutuzumab. This dosing regimen of escalating from 20 mg to 50 mg on day 2 is modeled after the experience from the M13-982 venetoclax trial, which showed that this is a safe approach (Stilgenbauer et al., 2016). The dosing schedule for venetoclax in this trial is as follows. Note that this ramp-up schedule cannot be shortened, but may be lengthened if TLS is observed.

Day of treatment cycle	Venetoclax Daily Dose
Cycle 4, day 1	20mg
Cycle 4, days 2-7	50mg
Cycle 4, days 8-14	100mg
Cycle 4, days 15-21	200mg
Cycle 4, days 22-28	400mg
Cycles 5 and beyond	400mg

- For patients at low or medium risk of tumor lysis syndrome (as defined in section 6.1.2), venetoclax will be given as a 20mg oral dose on cycle 4 day 1. Patients will receive intravenous hydration of 150-200ml/hr while in the outpatient infusion area. Tumor lysis labs (including potassium, uric acid, phosphorous, calcium, and creatinine) must be checked on cycle 4 day 1 prior to this dose, approximately 6-8 hours after dosing, and again the following day, approximately 24 hours after the initial dose (prior to the second dose of venetoclax on cycle 4 day 2). If Cairo-Bishop criteria (Appendix D) for laboratory or clinical tumor lysis syndrome (TLS) are met on these follow-up labs, TLS management will occur as per Appendix E, and the patient will either have drug held or continue at 20mg daily (depending on severity) until the changes resolve, at which point the patient will escalate to 50mg daily. If Cairo-Bishop criteria for TLS are not met, the patient will dose escalate to 50mg on day 2. TLS labs should be checked again on cycle 4 day 2 approximately 6-8 hours after dosing and approximately 24 hours post the first 50mg dose.

- Patients at high risk of TLS (as defined in section 6.1.2) will be admitted to the hospital for venetoclax initiation. Patients will receive intravenous hydration of 150-200ml/hr, prior to receiving their first dose of venetoclax 20mg. Tumor lysis labs will be checked on cycle 4 day 1 prior to this initial dose and then at approximately 4, 8, 12, and 24 hours later. If Cairo-Bishop criteria for tumor lysis syndrome are not met (see Appendix D), the patient will be escalated to 50mg daily on cycle 4 day 2. Tumor lysis labs will then be checked approximately 4, 8, 12, and 24 hours after this second dose.

Patients will continue on 50mg daily for cycle 4 days 2-7, then 100mg daily on cycle 4 days 8-14, then 200mg daily on cycle 4 days 15-21, and finally 400mg daily on cycle 4 days 22-28. For the initial doses of 100mg, 200mg, and 400mg, TLS labs should be checked prior to initiating the higher dose, approximately 6-8 hours post dose, and approximately 24 hours post dose, with dose escalation occurring only if criteria for TLS are not met.

For the remaining length of therapy, venetoclax will be given at a dose of 400mg daily.

Patients should take venetoclax within approximately 30 minutes after the completion of breakfast or the subject's first meal of the day. Patients may not consume: Grapefruit or grapefruit products, Seville oranges (including marmalade containing Seville oranges) or starfruit within the 3-day period prior to the first study drug administration and until the last day of treatment is completed due to possible CYP3A mediated metabolic interaction.

Other Modality(ies) or Procedures

N/A

5.4 General Concomitant Medication and Supportive Care Guidelines

General Supportive Care

Patients should receive full supportive care, including transfusions of blood and blood products, antibiotics, hematopoietic growth factors, analgesics, and antiemetics when appropriate.

Tumor Lysis Syndrome Prophylaxis

Tumor lysis syndrome (TLS), characterized by hyperkalemia, hyperuricemia, and hyperphosphatemia resulting from the rapid release of potassium, uric acid, and phosphate, has been reported in patients with CLL initiating venetoclax. There is a modest risk of TLS in patients treated with obinutuzumab, and as such it is recommended that patients receive allopurinol prior to starting on obinutuzumab. The risk of TLS on this study is highest during the first cycle of venetoclax combination therapy. It is strongly recommended that allopurinol 300mg PO daily begin 3 days before the initiation of venetoclax on cycle 4 day 1, and allopurinol should continue through the end of the first cycle of triple combination therapy. Patients with an allopurinol allergy should receive alternative TLS prophylaxis (e.g. febuxostat) if possible. In addition to pharmacologic TLS prophylaxis, all patients should be instructed to maintain adequate hydration and maintain urinary output as an additional measure to reduce the risk of TLS. Based on clinical and laboratory parameters, TLS prophylaxis may be continued or restarted as needed at the investigator’s discretion. All patients meeting criteria of laboratory TLS or clinical TLS according to the Cairo-Bishop definition of TLS (Appendix D) should receive vigorous intravenous hydration and should be considered for rasburicase therapy as needed to reduce hyperuricemia, until correction of electrolyte abnormalities. See section 6.1, dose modification table, for additional instructions.

Hepatitis B Monitoring and Prophylaxis

Testing for hepatitis B surface antigen (HBsAg) and hepatitis B core antibody (HBcAb) is required for all subjects at the time of enrollment. Subjects who are positive for either HBsAg and/or HBcAb must also be tested for serum HBV DNA. Subjects who are positive for HBV DNA are excluded from the study. Subjects who are negative for HBV DNA but positive for

either HBsAg or HBcAb must receive pharmacologic prophylaxis to prevent HBV reactivation. Entecavir is the preferred agent for pharmacologic prophylaxis. Use of an agent other than entecavir must be discussed with the principal investigator. Prophylaxis should be administered for as long as the subject is receiving a study medication or for up to 12 months after the last dose of obinutuzumab, whichever is longer. HBV DNA should be monitored every three months while a patient is receiving pharmacologic prophylaxis. If the HBV DNA level becomes detectable, all study medications should be discontinued and infectious disease consultation should be obtained. If a subject who meets criteria for prophylaxis is not willing or able to receive prophylaxis, then he/she will be excluded from the study.

Growth Factor Support

White blood cell growth factor support with Neulasta, Neupogen, or tbo-filgrastim (Granix) will be administered at the discretion of the treating investigators with dosing as per standard of care. Neulasta, Neupogen, and Granix will be from commercial supply.

Prophylactic Antibiotics

Prophylaxis for *Pneumocystis jiroveci* pneumonia (PCP) with trimethoprim/sulfamethoxazole (typically given as one single-strength tablet daily) or equivalent is mandatory for all patients. Anti-herpetic viral prophylaxis with acyclovir (typically given as 400mg po tid) or equivalent is mandatory for all patients.

Intravenous Immunoglobulin

Immunoglobulin replacement with intravenous immunoglobulin (IvIg) therapy is permitted for patients who, at the discretion of the investigator, require IvIg for primary prophylactic or secondary prevention of infection.

Moderate or Strong CYP3A Modulators

Minor CYP3A4 inducers or inhibitors (e.g. amlodipine) are allowed without any dose changes to the study drugs. Patients who require treatment with strong CYP3A inhibitors or inducers (partial list in Appendix F) are excluded from enrollment in the study. However, if treatment with strong CYP3A inhibitors becomes necessary while subjects are on study treatment, dosing of venetoclax and acalabrutinib should be managed as detailed in Table 4 below. Of note, venetoclax should held if treatment with strong CYP3A inhibitors or inducers is required during the ramp-up phase of drug administration. Dosing of acalabrutinib should be interrupted while the subject is on the strong CYP3A inhibitor, as detailed in Table 4 below. Three days after discontinuation of the strong CYP3A inhibitor, resume acalabrutinib and resume or increase venetoclax to the dose that was used prior to initiating the CYP3A inhibitor.

If treatment with moderate CYP3A inhibitors (see partial list in Appendix F) is required while subjects are on study, dosing of venetoclax and acalabrutinib should be reduced as detailed in Table 4 below. Three days after discontinuation of the moderate CYP3A inhibitor, increase acalabrutinib and venetoclax to the doses that were used prior to initiating the CYP3A inhibitor.

If treatment with strong or moderate CYP3A inducers is required while the subject is on study, doses of venetoclax and acalabrutinib should be modified as detailed in Table 4. Three days after discontinuation of the strong CYP3A inducer, reduce acalabrutinib and resume venetoclax at the dose that was used prior to initiating the CYP3A inhibitor.

Dosing modifications of acalabrutinib and venetoclax do not need to be made if patients are receiving inhibitors or inducers of other CYP450 enzymes.

Table 4: Dosing Modifications in the Setting of CYP3A Modulators and P-gp Inhibitors

Medication Class	**Initiation and Ramp-Up Phase of Venetoclax**	**Steady Daily Dose of Venetoclax (after ramp up phase)**	**Steady Daily Dose of Acalabrutinib**
Strong CYP3A Inhibitor	Contraindicated, hold venetoclax	Reduce venetoclax dose by 75% (e.g. 100mg daily)	Interrupt acalabrutinib dosing
Moderate CYP3A inhibitor	Reduce venetoclax dose by 50%	Reduce venetoclax dose by 50% (e.g. 200mg daily)	Reduce acalabrutinib dose by 50% (e.g. 100mg once daily)
Strong CYP3A Inducer	Contraindicated, hold venetoclax	Contraindicated, hold venetoclax	Double acalabrutinib dose (e.g. 200mg twice daily)
Moderate CYP3A Inducer	Contraindicated, hold venetoclax	Contraindicated, hold venetoclax	Maintain acalabrutinib dose (e.g. 100mg twice daily)
P-gp Inhibitors or P-gp Substrates of Narrow Therapeutic Index	Contraindicated, hold venetoclax	Reduce venetoclax dose by 50% (e.g. 200mg daily). Take inhibitor at least 6 hours before venetoclax.	No change to dose

P-gp Inhibitors or P-gp Substrates of Narrow Therapeutic Index

Patients who require treatment with P-gp inhibitors or P-gp substrates of narrow therapeutic index (partial list in Appendix F) are excluded from enrollment in the study. However, if treatment with these drugs becomes necessary while subjects are on study treatment, dosing of venetoclax should be managed as detailed in Table 4 above. If a narrow therapeutic index P-gp substrate must be used, it should be taken at least 6 hours before venetoclax. Resume the venetoclax dose that was used prior to initiating the P-gp inhibitor 3 days after discontinuation of the inhibitor.

Drug-Drug Interactions

Because there is a potential for interaction of acalabrutinib and venetoclax with other concomitantly administered drugs through the cytochrome P450 system, the case report form

must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. If there are known drug-drug interactions between the study drug(s) and other medications that the patient is taking, dose reductions of the study drug(s) may occur at the discretion of the treating investigator.

5.5 Criteria for Taking a Participant Off Protocol Therapy

Duration of therapy will depend on individual response, evidence of disease progression and tolerance. In the absence of treatment delays due to adverse event(s), treatment may continue until one of the following criteria applies:

- Disease progression
- Intercurrent illness that prevents further administration of treatment
- Pregnancy
- Unacceptable adverse event(s)
- Participant demonstrates an inability or unwillingness to comply with the oral medication regimen and/or documentation requirements
- Participant decides to withdraw from the protocol therapy
- General or specific changes in the participant's condition render the participant unacceptable for further treatment in the judgment of the treating investigator
- EXCEPTION: patients deriving clinical benefit from study treatment may continue on therapy at the discretion of the treating investigator to bridge to next treatment even if there is evidence of progressive disease

Participants will be removed from the protocol therapy when any of these criteria apply. The reason for removal from protocol therapy, and the date the participant was removed, must be documented in the case report form (CRF). Alternative care options will be discussed with the participant.
An ODQ Treatment Ended/Off Study Form will be filled out when a participant is removed from protocol therapy. This form can be found on the ODQ website or obtained from the ODQ registration staff.

In the event of unusual or life-threatening complications, treating investigators must notify the Overall PI, Matthew S. Davids, MD, as soon as possible, at 617-632-6331, DFCI pager 57215.

5.6 Duration of Follow Up

Once participants have completed study treatment they will be followed until initiation of new therapy, or until death, whichever occurs first. After cycle 24, subjects remaining on investigational therapy will be seen every 3 months in clinic for as long as they remain on study treatment. Subjects who discontinue acalabrutinib/venetoclax because they are in CR/CRi with MRD-negativity will be followed as per the study calendars in section 10. In these subjects, if no relapse is detected after 48 cycles of therapy, they may return to follow-up with a local

hematologist but will be followed by telephone for updates on disease progress and overall survival. Participants removed from treatment for unacceptable adverse events will be followed until resolution or stabilization of the adverse event, and/or until initiation of new therapy for their disease, whichever is longer.

5.7 Criteria for Taking a Participant Off Study

Participants will be removed from study when any of the following criteria apply:

- Lost to follow-up
- Withdrawal of consent for data submission
- Start of new CLL-directed therapy
- Disease Progression
- Death

Participants will be removed from the protocol therapy when any of these criteria apply. The reason for removal from protocol therapy, and the date the participant was removed, must be documented in the case report form (CRF). Alternative care options will be discussed with the participant.

When a participant is removed from protocol therapy and/or is off of the study, the relevant Off-Treatment/Off-Study information will be updated in OnCore. This form can be found on the ODQ website or obtained from the ODQ registration staff.

6. DOSING DELAYS / DOSE MODIFICATIONS

Dose delays and modifications will be made as indicated in the following table(s). Hematologic AEs will be graded as per the IWCLL Grading Scale for Hematologic Toxicities in CLL Studies (Appendix C). Nonhematologic AEs will be described and graded as per the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP website http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

Given that multiple drugs in this study can cause similar side effects, treating clinician discretion in consultation with the Principal Investigator can be used to decide which agent to dose reduce when patients experience a side effect that could be attributable to multiple drugs.

6.1 Venetoclax

Dose Reduction Table

The dose for venetoclax in this study is 400 mg QD (after intra-patient dose ramp-up). If known venetoclax toxicities such as tumor lysis syndrome, gastrointestinal adverse events, or neutropenia (as detailed in sections 6.1.2 and 6.1.3) are observed, dose reductions to the following dose levels for venetoclax may be utilized:

Table 5: Venetoclax Dose Reduction Schema

Dose at Interruption, mg	**Restart Dose, mg[a]**
400	300
300	200
200	100
100	50
50	20
20	10
[a]Continue the reduced dose for 1 week before increasing the dose.	

Prophylaxis and Management of Tumor Lysis Syndrome

Upon initial treatment of CLL with venetoclax, there is a potential for TLS, especially in those with elevated lymphocyte count, bulky lymphadenopathy, and renal dysfunction. To mitigate the risk for TLS, all patients on this study will be restaged with CT scan prior to initiation of venetoclax on cycle 4 day 1 to assess for risk of TLS. Patients will be managed according to Table 6.

Table 6: Prophylaxis and Monitoring For TLS Syndrome in Patients Receiving Venetoclax

Tumor Burden		**Prophylaxis**		**Blood Chemistry Monitoring**[c,d]
		Hydration[a]	**Antihyperuricemics**	**Setting and Frequency of Assessments**
Low	All LN <5 cm AND ALC <25 x10^9/L	Oral (1.5-2 L) and consider additional intravenous	Allopurinol[b]	Outpatient • Pre-dose, 6-8 hours post-dose, and 24 hours post-dose, on days where patient receives first dose of 20, 50, 100, 200, or 400mg • Consider hospitalization for patients with CrCl <80ml/min at first dose of 20 mg and 50 mg; see below for monitoring in hospital
Medium	Any LN 5 cm to <10 cm OR ALC ≥25 x10^9/L			
High	Any LN ≥10 cm OR ALC ≥25 x10^9/L AND any LN ≥5 cm	Oral (1.5-2 L) and intravenous[e]	Allopurinol; consider rasburicase if baseline uric acid is elevated[f]	Inpatient: • Pre-dose, 4, 8,12 and 24 hours post doses of 20 and 50mg Outpatient: • At subsequent ramp-up doses as outlined above for low and medium risk patients

ALC = absolute lymphocyte count; LN = lymph node.

[a] Administer intravenous hydration for any patient who cannot tolerate oral hydration.
[b] If possible start allopurinol or xanthine oxidase inhibitor about 3 days prior to initiation of venetoclax.
[c] Evaluate blood chemistries (potassium, uric acid, phosphorus, calcium, and creatinine); review in real time. During the dose ramp-up period, the subsequent dose should not be administered until the 24 hours post-dose laboratory values are reviewed by the investigator.
[d] All same-day laboratory assessments may be taken within ±2 hour window, and 24 hour laboratory assessments within a ±6 hour window, if necessary
[e] Intravenous fluids should be given at 150-200 mL/hr starting the night before the first dose of 20 mg, with a target of approximately 1.5 – 2 L/day (or as clinically appropriate), and continued for at least 24 hours after completing the 50mg dose.
[f] Rasburicase (given at a flat dose of 6 mg IV x 1, or per institutional standards) should be administered per institutional guidelines for patients with elevated uric acid level at baseline (> ULN) as prophylaxis prior to the initial dose of venetoclax. For patients with a contraindication to rasburicase (i.e., glucose-6-phosphate dehydrogenase [G6PD] deficiency), the TLS risk-mitigation plan must be reviewed with the Overall PI.

If any significant laboratory changes are observed within the first 24 hours after initiation of dosing, see Appendix E (Recommendations for Initial Management of Electrolyte Abnormalities and Prevention of Tumor Lysis Syndrome [TLS]), for additional laboratory assessments and management guidelines. If aggressive correction of electrolytes abnormalities was performed, the subsequent dose of venetoclax can only be given when electrolytes have been stable without any further treatment for approximately 24 hours.

Management of Other Toxicities

The following tables provide recommended guidelines for venetoclax dose reduction based on specific toxicities. Any subject who discontinues venetoclax due to toxicity may continue on the other study medications. Management not consistent with these guidelines should be discussed with the Overall PI:

Table 7: Dose Reduction Strategy for Patients Experiencing Non-Hematologic Toxicities on Venetoclax

Non-hematologic Toxicity Grade	**Recommended Management/Next Dose for Venetoclax**
≤ Grade 1 or 2	No change in dose
Grade 3	Hold* until ≤ Grade 1. At first occurrence, resume at same dose, no dose modification is required. At second and subsequent occurrences,** hold venetoclax and follow dose reduction guidelines in Table 5 when resuming treatment with venetoclax after resolution.
Grade 4	Permanently cease venetoclax therapy unless toxicity can be attributed to an alternative cause
*Participants requiring a hold of >4 weeks of venetoclax due to a toxicity thought to be related to venetoclax should go off venetoclax therapy unless approved by the overall PI to remain on therapy. **Participants requiring >3 dose reductions of venetoclax should go off venetoclax unless approved by the overall PI to remain on therapy.	

With regard to specific nonhematologic toxicities, the recommended management for venetoclax-associated diarrhea is loperamide, dosed at 4 mg at first onset, followed by 2 mg with each loose bowel movement until diarrhea-free for 12 hours (maximum dosage: 16 mg/24 hours). Adjunct anti-diarrheal therapy is permitted and should be recorded when used.

Table 8: Dose Reduction Strategy for Patients Experiencing Neutropenia on Venetoclax

Neutropenia in patients on venetoclax	**Recommended Management/Next Dose for Venetoclax**
≤ Grade 1 or Grade 2	No change in dose
Grade 3 (ANC 500-999 cells/mm^3)	Hold* until ≤ Grade 2 or continue at current dose as long as growth factor is administered. (If venetoclax is continued, repeat ANC within 3 days and if still grade ≥3, hold venetoclax until ≤ Grade 2.) If not previously growth factor responsive, resume at one dose level lower, if indicated.**
Grade 4 (ANC <500 cells/mm^3)	Hold* until ≤ Grade 3, then resume therapy at the same dose but with G-CSF support. On second and subsequent occurrences, follow dose reduction guidelines in Table 5 when resuming treatment.**
*Participants requiring a hold of >4 weeks of venetoclax due to a venetoclax related therapy should go off protocol therapy unless approved by the overall PI to remain on therapy. **Participants requiring >3 dose reductions of venetoclax should go off venetoclax unless with approval by the overall PI to remain on therapy.	

Neutropenia in patients on venetoclax	**Recommended Management/Next Dose for Venetoclax**
At any time during the study, if the patient presents with febrile neutropenia, it is recommended that venetoclax be interrupted until resolution of the fever or infection. At the first occurrence, venetoclax may then be re-initiated at the same dose. At a second or later occurrence, consideration should be given to venetoclax dose reduction.	
Growth factor support with filgrastim or pegfilgrastim may be utilized as necessary throughout the study and does not require holding venetoclax.	

Table 9: Dose Reduction Strategy for Patients Experiencing Venetoclax-Related Hematologic Toxicities (excluding Neutropenia)

Venetoclax-related Hematologic Toxicities	**Management/Next Dose for Venetoclax**
≤ Grade 1, 2, or 3	No change in dose
Grade 4	Hold* until ≤ Grade 3, unless the toxicity is thought to be due to a cause other than venetoclax. On first occurrence, venetoclax may be resumed at the same dose. On second and subsequent occurrences, follow the dose reduction guidelines in Table 5.**
*Participants requiring a hold of >4 weeks of venetoclax due to a venetoclax related therapy should go off protocol therapy unless approved by the overall PI to remain on therapy. **Participants requiring >3 dose reductions of venetoclax should go off venetoclax unless with approval by the overall PI to remain on therapy.	

6.2 Obinutuzumab

Dose Reduction Table

The dose of obinutuzumab is not modified but doses can be held or delayed as needed. Dose delays of obinutuzumab may be made as per the tables below if a toxicity is believed to be at least likely related to obinutuzumab. If a subject must permanently discontinue obinuzutumab prior to completion of the full six month course, acalabrutinib and venetoclax will continue to be administered as per the study calendar detailed in section 10. For the adverse event profile, see section 7.1.1 and the obinutuzumab IB.

Table 10: Dose Modifications for Non-Infusion-Related-Reaction, Non-Hematologic Toxicities

Worst CTCAE Grade Toxicity	Recommended Action to be Taken
Grade 1 or 2	None
Grade 3 CTCAE	Hold dosing of obinutuzumab until toxicity Grade ≤2
If toxicity remains grade 3 toxicity for longer than 2 weeks	Consider permanent discontinuation of obinutuzumab
Recurrence of Grade 3 toxicity	Consider permanent discontinuation of obinutuzumab
Grade 4 CTCAE	Hold dosing of obinutuzumab until toxicity resolves to Grade ≤2
Recurrence of Grade 4 toxicity	Consider permanent discontinuation of obinutuzumab

Infusion Related Reactions

If a patient experiences any grade infusion reaction during infusion, adjust the infusion as outlined below:

- Grade 4 (life-threatening): Stop infusion and permanently discontinue therapy.
- Grade 3 (severe): Temporarily interrupt infusion and treat symptoms. Upon resolution of symptoms, restart infusion at no more than half the previous rate (the rate being used at the time that the infusion reaction occurred) and, if patient does not experience any infusion reaction symptoms, infusion rate escalation may resume at the increments and intervals as appropriate for the treatment dose. The Day 1 infusion rate may be increased back up to 25 mg/hr after 1 hour but not increased further. Consideration can be given to following a desensitization protocol for future administration of the antibody.
- Grade 1-2 (mild to moderate): Reduce infusion rate and treat symptoms. Upon resolution of symptoms, continue infusion and, if patient does not experience any infusion reaction symptoms, infusion rate escalation may resume at the increments and intervals as appropriate for the treatment dose. The Day 1 infusion rate may be increased back up to 25 mg/hr after 1 hour but not increased further.

Hypotension may be expected to occur during obinutuzumab infusions. Withholding of antihypertensive treatments should be considered for 12 hours prior to and throughout each obinutuzumab infusion and for the first hour after administration. Patients at acute risk of hypertensive crisis should be evaluated for the benefits and risks of withholding their hypertensive medication.

Tumor Lysis Syndrome

Management of tumor lysis syndrome related to obinutuzumab should be managed as per standard of care. Subjects should be monitored for TLS after the first infusion of obinutuzumab on cycle 2 day 1 as outlined in the study calendar in section 10.

6.3 Acalabrutinib

Treatment with acalabrutinib should be held for any unmanageable, potentially study drug-

related toxicity that is Grade ≥ 3 in severity. Study drug may be held for a maximum of 28 consecutive days from expected dose due to toxicity. Acalabrutinib should be permanently discontinued in the event of a toxicity thought at least possibly due to acalabrutinib lasting > 28 days despite holding drug, unless reviewed and approved by the Overall PI. Any subject who permanently discontinues acalabrutinib prior to cycle 4 will no longer be eligible for any of the study drugs and will come off study treatment but continue to be followed as detailed in section 5.6. Any subject who discontinues acalabrutinib at cycle 4 or later may continue on the other study medications.

Dose Reduction Table

The actions in Table 11 below should be taken for the following toxicities if thought to be at least possibly related to acalabrutinib:

- Grade 4 ANC (< 500/μL) for > 7 days (Neutrophil growth factors are permitted and use must be recorded on the case report form [CRF]).
- Grade 3 or higher platelet decreases in presence of significant bleeding.
- Grade 4 platelet decreases.
- Grade 3 or 4 nausea, vomiting, or diarrhea, if persistent for >7 days despite optimal antiemetic and/or anti-diarrheal therapy.
- Any other Grade 4 toxicity or unmanageable Grade 3 toxicity.

Table 11: Dose Modifications for Acalabrutinib

Occurrence	Action
1st - 2nd	Hold acalabrutinib until recovery to Grade ≤ 1 or baseline; may restart at original dose level
3rd	Hold acalabrutinib until recovery to Grade ≤ 1 or baseline; restart at one dose level lower (100 mg once daily)
4th	Discontinue acalabrutinib

Any changes to the dosing regimen must be recorded in the Dosage Administration CRF.

If acalabrutinib is reduced for apparent treatment-related toxicity, the dose need not be re-escalated, even if there is minimal or no toxicity with the reduced dose. However, if the subject tolerates a reduced dose of acalabrutinib for ≥ 4 weeks then the dose may be increased to the next higher dose level, at the discretion of the investigator. Such re-escalation may be particularly warranted if further evaluation reveals that the AE that led to the dose reduction was not treatment-related.

6.4 Integrated Toxicity Management Guidelines When the Causative Agent is Unclear

The study drugs have some overlapping toxicities (e.g. diarrhea with venetoclax and acalabrutinib, neutropenia with all three agents), and in certain instances it may be unclear which agent is responsible for an adverse event that a patient is experiencing. If the treating clinician can identify the specific drug causing an adverse event, then the guidelines in sections 6.1-6.3 can be used to guide dose reductions of that individual drug. If, however, the treating clinician is

uncertain which agent is responsible for a particular adverse event, then the guidelines below should be used for management of study drug dosing in these circumstances:

For grade 1 or grade 2 toxicities where the causative drug is uncertain, no changes to study drug dosing need to be made.

For grade 3 or 4 hematologic and non-hematologic toxicities where the causative drug is uncertain, all suspected drugs should be held until the toxicity improves. One drug (at treating clinician discretion) should then be introduced (at the same dose as when the medication was discontinued). Of note, if one of the suspected causes is obinutuzumab, the other suspected agent(s) (with a shorter half-life) should be introduced first. If the toxicity recurs after reintroduction of the first drug, then the adverse effect is deemed attributable to this drug and management should proceed according to the guidelines in sections 6.1-6.3. The other suspected agents may then be reintroduced after the toxicity has improved. If, on the other hand, the toxicity does not occur after one week of therapy with the first drug, a second study drug should be reintroduced. If the toxicity occurs, then the adverse effect is deemed attributable to this second drug and management should proceed according to guidelines in sections 6.1-6.3. If a third study drug is also suspected, then it may be introduced one week later if no toxicity has been observed after introduction of the first two.

During cycle 8 and beyond, patients who have to hold either acalabrutinib or venetoclax due to drug-specific toxicity for longer than 4 weeks may stay on study and continue on monotherapy with the other agent that is not believed to be responsible for that toxicity.

7. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

The following list of reported and/or potential AEs and the characteristics of an observed AE will determine whether the event requires expedited reporting **in addition** to routine reporting.

7.1 Expected Toxicities

Adverse Events Lists

7.1.1.1 Adverse Event List for Obinutuzumab

Important risks identified in clinical investigations with obinutuzumab were: IRRs (infusion-related reactions), TLS, thrombocytopenia (including acute thrombocytopenia), neutropenia (including prolonged and late onset neutropenia), prolonged B-cell depletion, infections (including hepatitis B reactivation and PML), worsening of pre-existing cardiac conditions and GI perforation.

Hepatitis B Reactivation
HBV reactivation, in some cases resulting in fulminant hepatitis, hepatic failure, and death, can occur in patients treated with anti-CD20 antibodies such as obinutuzumab. HBV reactivation has been reported in patients who are HBsAg positive and also in patients who are HBsAg negative

but are anti-HBc positive. Reactivation has also occurred in patients who appear to have resolved hepatitis B infection (i.e., HBsAg negative, anti-HBc positive, and hepatitis B surface antibody [anti-HBs] positive). HBV reactivation is defined as an abrupt increase in HBV replication manifesting as a rapid increase in serum HBV DNA level or detection of HBsAg in a person who was previously HBsAg negative and anti-HBc positive. Reactivation of HBV replication is often followed by hepatitis, i.e., increase in transaminase levels and, in severe cases, increase in bilirubin levels, liver failure, and death. Patients in this study who are positive for either HBsAg and/or anti-HBc Ab but negative for HBV DNA should be managed as outlined in section 5.4.

Progressive Multifocal Leukoencephalopathy
JC virus infection resulting in progressive multifocal leukoencephalopathy (PML), which can be fatal, was observed in patients treated with obinutuzumab. The diagnosis of PML should be considered in any patient presenting with new onset or changes to preexisting neurologic manifestations. Evaluation of PML includes, but is not limited to, consultation with a neurologist, brain magnetic resonance imaging, and lumbar puncture. Discontinue obinutuzumab therapy and consider discontinuation of other agents in patients who develop PML.

Infusion Reactions
Obinutuzumab can cause severe and life-threatening IRRs; 65% of patients with CLL experienced a reaction to the first 1000 mg infused of obinutuzumab, and 38% of iNHL patients experienced a reaction on Day 1 of obinutuzumab infusion. IRRs within 24 hours of receiving obinutuzumab have occurred. IRRs can also occur with subsequent infusions. Symptoms may include hypotension, tachycardia, dyspnea, and respiratory symptoms (e.g., bronchospasm, larynx and throat irritation, wheezing, and laryngeal edema). Most frequently reported symptoms include nausea, fatigue, dizziness, vomiting, diarrhea, hypertension, flushing, headache, pyrexia, and chills. Management of IRRs is discussed in section 5.3.1.

Tumor Lysis Syndrome
TLS, including fatal cases, has been reported in patients receiving obinutuzumab. Patients with high tumor burden, high circulating lymphocyte count ($> 25 \times 10^9$/L) or renal impairment are at greater risk for TLS and should receive appropriate tumor lysis prophylaxis with anti-hyperuricemics (e.g., allopurinol or rasburicase) and hydration prior to the infusion of obinutuzumab. Continue prophylaxis prior to each subsequent obinutuzumab infusion, as needed. The laboratory parameters (including electrolytes and renal function) of patients considered at risk for TLS can be monitored as needed for patients at greater risk of TLS.

Infections
Serious bacterial, fungal, and new or reactivated viral infections can occur during and following the completion of obinutuzumab therapy. Fatal infections have been reported. Obinutuzumab should not be administered to patients with an active infection. Patients with a history of recurring or chronic infections may be at increased risk of infection.

Neutropenia
Severe and life-threatening neutropenia, including febrile neutropenia, has been reported during

treatment with obinutuzumab. Patients with Grade 3 – 4 neutropenia should be monitored frequently with regular laboratory tests until resolution. Anticipate, evaluate, and treat any symptoms or signs of developing infection. Consider administration of granulocyte colony-stimulating factors (G-CSF) in patients with Grade 3 or 4 neutropenia. Neutropenia can also be of late onset (occurring more than 28 days after completion of treatment) and/or prolonged (lasting longer than 28 days). Consider dose delays in the case of Grade 3 or 4 neutropenia. Patients with severe and long lasting (> 1 week) neutropenia are strongly recommended to receive antimicrobial prophylaxis until resolution of neutropenia to Grade 1 or 2. Antiviral and antifungal prophylaxis should be considered.

Thrombocytopenia
Severe and life-threatening thrombocytopenia has been reported during treatment with obinutuzumab in combination with chlorambucil or bendamustine. Fatal hemorrhagic events during Cycle 1 have also been reported in patients with CLL treated with obinutuzumab. Patients will be monitored frequently for thrombocytopenia and hemorrhagic events. Management of thrombocytopenia is discussed in section 6.2.1.

Immunization
The safety and efficacy of immunization with live or attenuated viral vaccines during or following obinutuzumab therapy have not been studied. Immunization with live-virus vaccines is not recommended during treatment and until B-cell recovery.

Adverse Event List for Venetoclax

Tumor Lysis Syndrome
Tumor lysis syndrome, including fatal events and renal failure requiring dialysis, has occurred in previously treated CLL patients with high tumor burden when treated with venetoclax.

Venetoclax can cause rapid reduction in tumor and thus poses a risk for TLS in the initial ramp-up phase. Changes in blood chemistries consistent with TLS that require prompt management can occur as early as 6 to 8 hours following the first dose of venetoclax and at each dose increase.

The risk of TLS is a continuum based on multiple factors, including tumor burden and comorbidities. Reduced renal function (CrCl <80 mL/min) further increases the risk. Patients should be assessed for risk and should receive appropriate prophylaxis for TLS, including hydration and anti-hyperuricemics, as discussed in section 6.1.2.

Concomitant use of venetoclax with strong or moderate CYP3A inhibitors and P-gp inhibitors increases venetoclax exposure, may increase the risk of TLS at initiation and during ramp-up phase and may require venetoclax dose adjustment.

Neutropenia
Neutropenia is an important identified risk for venetoclax. Grade 3 or 4 neutropenia occurs in 40-50% of patients treated with venetoclax. Clinical data from the oncology studies suggest that the neutropenia adverse events are observed among patients who receive venetoclax as a single

agent or in combination with other therapeutic agents, with slightly higher frequency observed in some combination studies. Serious adverse events of neutropenia or neutropenia events that lead to discontinuations are few across the entire venetoclax oncology program. Monitor complete blood counts throughout the treatment period. Neutropenia management guidelines are provided in section 6.1.3.

Infections
Infections have been reported in the oncology clinical studies; however, these events are confounded by the underlying disease, comorbidities, and other immunosuppressive medications. To date, no clear relationship has been noted between serious infectious events and neutropenia. The types of infectious events observed generally have been consistent with those anticipated in the elderly population of heavily pretreated patients with hematologic malignancies and are similar across all indications. Infections should be closely monitored in this study. Serious infections have been reported in patients treated with venetoclax in combination with other agents, including obinutuzumab.

Immunization
Do not administer live attenuated vaccines prior to, during, or after treatment with venetoclax until B-cell recovery occurs. The safety and efficacy of immunization with live attenuated vaccines during or following venetoclax therapy have not been studied. Advise patients that vaccinations may be less effective.

Reproductive System Effects and Embryo-Fetal Toxicity
Based on nonclinical studies, there is a potential for decreased spermatogenesis. Non-reversible depletion of testicular germ cells has been observed in dogs at all doses tested after 4 weeks of dosing. Male patients considering preservation of fertility should bank their sperm before treatment with venetoclax. Based on its mechanism of action and findings in animals, venetoclax may cause embryo-fetal harm when administered to a pregnant woman. In an embryo-fetal study conducted in mice, administration of venetoclax to pregnant animals at exposures equivalent to that observed in patients at the recommended dose of 400 mg daily resulted in post-implantation loss and decreased fetal weight. There are no adequate and well-controlled studies in pregnant women using venetoclax. Advise females of reproductive potential to avoid pregnancy during treatment. If venetoclax is used during pregnancy or if the patient becomes pregnant while taking venetoclax, the patient should be apprised of the potential hazard to the fetus and discontinued from study treatment. Venetoclax use is not advised in lactation either.

Treatment-Emergent Malignancies (Second Primary Malignancies)
Events of second primary malignancies have been reported across the oncology program. No pattern has been was observed. As venetoclax is being evaluated in subjects with relapsed/refractory disease who had previously been treated with various cytotoxic agents, second primary malignancies are closely monitored.

<u>Adverse Event List for Acalabrutinib</u>

Hemorrhage
Cases of hemorrhagic events, including central nervous system, respiratory, and gastrointestinal

hemorrhage, have been reported in clinical trials with acalabrutinib; some of these bleeding events resulted in fatal outcomes. The mechanism for hemorrhage is not well understood. Patients receiving antiplatelet or anticoagulant therapies may be at increased risk of hemorrhage and should be monitored for signs of bleeding. Consider the benefit-risk of withholding acalabrutinib for at least 3 days pre- and post-surgery. Subjects with hemorrhage should be managed per institutional guidelines with supportive care and diagnostic evaluations as clinically indicated.

Infection
Serious infections, including fatal events, have been reported in clinical studies with acalabrutinib. The most frequently reported Grade 3 or 4 infection was pneumonia. Across the acalabrutinib clinical development program (including subjects treated with acalabrutinib in combination with other drugs), cases of hepatitis B virus (HBV) reactivation (resulting in liver failure and death in 1 case) and cases of progressive multifocal leukoencephalopathy have occurred in subjects with hematologic malignancies. Subjects should be monitored for signs and symptoms of infection and treated as medically appropriate. Subjects with infection events should be managed according to institutional guidelines with maximal supportive care and diagnostic evaluations as clinically indicated.

Cytopenias
Treatment-emergent Grade 3 or 4 cytopenias including neutropenia, anemia, and thrombocytopenia have occurred in clinical studies with acalabrutinib. Monitor blood counts as medically appropriate. Subjects with cytopenias should be managed according to institutional guidelines with maximal supportive care and diagnostic evaluations as clinically indicated. Subjects should be closely monitored as appropriate.

Second Primary Malignancies
Second primary malignancies, including non-skin carcinomas, have been reported in clinical studies with acalabrutinib. The most frequent second primary malignancy was skin cancer. Subjects with a second primary malignancy should be managed according to institutional guidelines with maximal supportive and therapeutic care and diagnostic evaluations as clinically indicated.

Atrial Fibrillation
Atrial fibrillation/flutter have been reported in in clinical studies with acalabrutinib, particularly in subjects with cardiac risk factors, hypertension, diabetes mellitus, acute infections, and a previous history of atrial fibrillation. The mechanism for atrial fibrillation is not well understood. Subjects with atrial fibrillation should be managed per institutional guidelines with supportive care and diagnostic evaluations as clinically indicated.

7.2 Safety Parameters and Definitions

Safety assessments will consist of monitoring and reporting adverse events and serious adverse events per protocol. This includes all events of death and any study-specific issue of concern.

Adverse Events

Any untoward medical occurrence in a patient or clinical trial subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (e.g., an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. This includes the following:

- Adverse events not previously observed in the subject that emerge during the protocol-specified adverse event reporting period, including signs or symptoms associated with CLL that were not present prior to the adverse event reporting period.
- Complications that occur as a result of protocol-mandated interventions (e.g., invasive procedures such as cardiac catheterizations).
- If applicable, adverse events that occur prior to assignment of study treatment associated with medication washout, no treatment run-in, or other protocol-mandated intervention.

Preexisting medical conditions (other than the condition being studied) judged by the investigator to have worsened in severity or frequency or changed in character during the protocol-specified adverse event reporting period.

Serious Adverse Events

An adverse event should be classified as a serious adverse event if the following criteria are met:

- It results in death (i.e., the adverse event actually causes or leads to death)
- It is life threatening (i.e., the adverse event, in the view of the investigator, places the subject at immediate risk of death. It does not include an adverse event that, had it occurred in a more severe form, might have caused death.).
- It requires or prolongs inpatient hospitalization.
- It results in persistent or significant disability/incapacity (i.e., the adverse event results in substantial disruption of the subject's ability to conduct normal life functions).
- It results in a congenital anomaly/birth defect in a neonate/infant born to a mother exposed to the investigational medicinal product.
- It is considered a significant medical event by the investigator based on medical judgment (e.g., may jeopardize the subject or may require medical/surgical intervention to prevent one of the outcomes listed above).

Adverse Events of Special Interest

Adverse events of special interest which may be a focus of further analysis in this study include the following:

- Cases of potential drug-induced liver injury that include an elevated ALT or AST in combination with either an elevated bilirubin or clinical jaundice, as defined by Hy's law and based on the following observations:
    - Treatment-emergent ALT or AST > 3x baseline value in combination with total bilirubin > 2x ULN (of which > 35% is direct bilirubin)
    - Treatment-emergent ALT or AST > 3x baseline value in combination with clinical jaundice

- Cases of suspected transmission of an infectious agent by the study drug (STIAMP), as defined below:
    - Any organism, virus, or infectious particle (e.g., prion protein transmitting transmissible spongiform encephalopathy), pathogenic or non-pathogenic, is considered an infectious agent. A transmission of an infectious agent may be suspected from clinical symptoms or laboratory findings that indicate an infection in a patient exposed to a medicinal product. This term applies only when a contamination of the study drug is suspected.
- In addition, venetoclax events of special interest are:
    - TLS (irrespective of seriousness, causality, or severity)
- In addition, obinutuzumab events of special interest are:
    - TLS (irrespective of seriousness)
    - Second malignancies
- In addition, acalabrutinib events of special interest are
    - Any grade atrial fibrillation
    - Grade ≥3 bleeding events

Selected Adverse Events for Obinutuzumab

Selected events (in clinical trials, these are events for which additional data collection or analyses will be performed; no special case handling or follow-up is required) include the following:

- IRRs
- Infections (including PML and hepatitis B reactivation)
- Neutropenia (including late-onset neutropenia, defined as neutrophil count < 1000 cells/mm^3, occurring 28 days or more after obinutuzumab treatment has been completed or stopped; prolonged neutropenia, defined as neutrophil count < 1000 cells/mm^3, that does not resolve after 28 days (without obinutuzumab treatment)
- Thrombocytopenia (including acute thrombocytopenia occurring during and within 24 hours post obinutuzumab infusion)
- Cardiac events
- Second malignancies
- GI perforation

7.3 Methods and Timing for Assessing and Recording Safety Variables

The investigator is responsible for ensuring that all adverse events and serious adverse events that are observed or reported during the study, are collected and reported to the FDA, appropriate Institutional Review Boards (IRBs), AstraZeneca, and Genentech in accordance with instructions provided in this section and in Sections 7.4-7.6, as well as in accordance with CFR 312.32 (Investigational New Drug [IND] Safety Reports).

Adverse Event Reporting Period

The study period during which all adverse events and serious adverse events where the patient has been exposed to Genentech product must be reported. This reporting period begins after

informed consent is obtained. For AstraZeneca, the reporting period starts with first dose of AstraZenecaAstraZeneca's product (acalabrutinib) and for Genentech, the reporting period starts with the first dose of either venetoclax or obinutuzumab. The reporting period ends 30 days following the last administration of study treatment or study discontinuation/termination, whichever is earlier. After this period, investigators should only report serious adverse events that are attributed to prior study treatment.

Assessment of Adverse Events

All adverse events and serious adverse events whether volunteered by the subject, discovered by study personnel during questioning, or detected through physical examination, laboratory test, or other means will be reported appropriately. Each reported adverse event or serious adverse event will be described by its duration (i.e., start and end dates), regulatory seriousness criteria if applicable, suspected relationship to the study drugs (see following guidance), and actions taken. To ensure consistency of adverse event and serious adverse event causality assessments, investigators should apply the following general guideline:

Yes
There is a plausible temporal relationship between the onset of the adverse event and administration of acalabrutinib, venetoclax, or obinutuzumab, and the adverse event cannot be readily explained by the subject's clinical state, intercurrent illness, or concomitant therapies; and/or the adverse event follows a known pattern of response to acalabrutinib, venetoclax, or obinutuzumab; and/or the adverse event abates or resolves upon discontinuation of the study drugs or dose reduction and, if applicable, reappears upon re-challenge.

No
Evidence exists that the adverse event has an etiology other than the acalabrutinib, venetoclax, or obinutuzumab (e.g., preexisting medical condition, underlying disease, intercurrent illness, or concomitant medication); and/or the adverse event has no plausible temporal relationship to administration of acalabrutinib, venetoclax, or obinutuzumab (e.g., cancer diagnosed 2 days after first dose of study drug).

Expected adverse events are those adverse events that are listed or characterized in the USPI or current IB.

Unexpected adverse events are those not listed in the USPI or current IB or not identified. This includes adverse events for which the specificity or severity is not consistent with the description in the USPI or IB. For example, under this definition, hepatic necrosis would be unexpected if the USPI or IB only referred to elevated hepatic enzymes or hepatitis.

For patients receiving combination therapy, causality will be assessed individually for each protocol-mandated therapy.

7.3.1.1 Pregnancies

Pregnancies in Female Patients

If a female patient becomes pregnant while receiving the study drugs or within 18 months after the last dose of obinutuzumab or 30 days after the last dose of venetoclax or acalabrutinib, whichever is longer, a report should be completed and expeditiously submitted to AstraZeneca and Genentech. Follow-up to obtain the outcome of the pregnancy should also occur.

Pregnancies in Female Partners of Male Patients
If the female partner of male patients become pregnant while receiving the study drug or within 3 months after the last dose of obinutuzumab or 30 days after the last dose of venetoclax or acalabrutinib, whichever is longer, a report should be completed and expeditiously submitted to Genentech and AstraZeneca.

7.3.1.2 Congenital Anomalies / Birth Defects and Abortions

Pregnancy itself is not a serious adverse event, but it is a special situation that should always be reported within the same timelines as a serious adverse event. All pregnancies and outcomes should be reported to medical safety. Abortion, whether accidental, therapeutic, or spontaneous, should always be classified as serious, and expeditiously reported as an SAE. Similarly, any congenital anomaly / birth defect in a child born to a female subject exposed to the study drug should be reported as an SAE.

7.3.1.3 Post-Study Adverse Events

For studies involving collection of survival data and extended follow-up after the end of the adverse event reporting period (defined as 30 days after the last dose of study drug), the investigator should report all deaths (regardless of cause) and any serious adverse event including development of cancer or a congenital anomaly in a subsequently conceived offspring of a female subject who participated in the study (including pregnancy occurring in the partner of a male study subject) that is believed to be related to prior exposure to study drug.
Case Transmission Verification will be performed by both parties during this period to ensure successful transmission of single case reports.

7.3.1.4 Reconciliation

The Sponsor agrees to conduct reconciliation (case transmission verification) for the study drugs to ensure all single case reports have been adequately received by Genentech (for study drugs venetoclax and obinutuzumab) and AstraZeneca (for acalabrutinib). The Sponsor-Investigator will agree to the reconciliation periodicity and format, but agree at minimum to exchange quarterly line listings of single case reports sent to Genentech (for venetoclax and obinutuzumab) and AstraZeneca (for acalabrutinib) in the preceding time period.

The periodic line-listing will be exchanged within seven (7) calendar days of the end of the agreed time period. Confirmation of receipt should be received within the time period mutually agreed upon.

If discrepancies are identified, the Sponsor-Investigator and AstraZeneca (for acalabrutinib) and Genentech (for venetoclax and obinutuzumab) will cooperate in resolving the discrepancies. The

responsible individuals for each party shall handle the matter on a case-by-case basis until satisfactory resolution.

Following Case Transmission Verification, single case reports which have not been received by Genentech and/or AstraZeneca shall be forwarded by the Sponsor to Genentech and or AstraZeneca within five (5) calendar days from request by Genentech.

At the end of the study, a final cumulative Case Transmission Verification report will be sent to Genentech.

7.4 Adverse Event Reporting

The sponsor-investigator will be responsible for collecting all protocol-defined Adverse Events (AEs)/Serious Adverse Events (SAEs), AEs of Special Interest (AESIs), Special Situation Reports (including pregnancy reports) and Product Complaints (with or without an AE) originating from the Study for the Product.

The sponsor-investigator must report all serious adverse events to Genentech and AstraZeneca within the timelines described below. The completed MedWatch, CIOMS I form, or Genentech approved reporting form should be sent immediately upon completion to Genentech Drug Safety at:

Fax: (650) 238-6067
Email: usds_aereporting-d@gene.com

and to AstraZeneca at:

Email: AEMailboxClinicalTrialTCS@astrazeneca.com

Relevant follow-up information should be submitted to both Genentech Drug Safety and AstraZeneca Drug Safety as soon as it becomes available.

All product complaints (for Genentech products) without an AE should be sent to:
Email: kaiseraugst.global_impcomplaint_management@roche.com

–OR--

Serious adverse events, pregnancy reports (including pregnancy occurring in the partner of a male study subject), adverse events of special interest (AESIs), product complaints (with or without an AE) and special situation reports where the patient has been exposed to Genentech products (venetoclax and obinutuzumab) will be sent on a MedWatch or CIOMS I form to Genentech. Reports will be submitted to AstraZeneca using AstraZeneca SAE Reporting Form for entry into AstraZeneca's global safety database and cross-reporting to regulatory agencies if required. Transmission of these reports (initial and follow-up) will be either electronically or by fax and within the timelines specified below.

Serious Adverse Drug Reactions

The investigator-sponsor will notify AstraZeneca's Medical Safety department, in English, when

made aware of a Safety Report (Serious Adverse Event (SAE) or Special Situation Report) in a patient receiving AstraZeneca's investigational medicinal product (acalabrutinib). The investigator-sponsor will notify Genentech's Medical Safety department, in English, when made aware of a Safety Report (SAE) in a patient receiving obinutuzumab or venetoclax. Serious adverse event reports shall be transmitted to Genentech within fifteen (15) calendar days of the awareness date and to AstraZeneca within 7 calendar days for fatal or life-threatening events and 15 calendar days for all other SAEs, special situations, and adverse events of special interest.

Pregnancy Reports
While such reports are not serious adverse events per se, as defined herein, any reports of pregnancy (including pregnancy occurring in the partner of a male study subject), where the fetus may have been exposed to any of the study drugs, shall be transmitted to Genentech and/or AstraZeneca (depending on which drugs the subject was exposed to) within fifteen (15) calendar days of the awareness date. Pregnancies will be followed up until the outcome of the pregnancy is known, whenever possible, based upon due diligence taken to obtain the follow-up information.

Pregnancies in Female Partners of Male Patients
Male patients will be instructed through the Informed Consent Form to immediately inform the investigator if their partner becomes pregnant during the study or within 3 months after the last dose of obinutuzumab or 30 days after the last dose of venetoclax . A Clinical Trial Pregnancy Reporting Form should be completed and submitted to Genentech within thirty (30) calendar days of the awareness date.

Adverse Events of Special Interest
AESIs shall be forwarded to Genentech and AstraZeneca within fifteen (15) calendar days of the awareness date.

Special Situation Reports
In addition to all adverse events, pregnancy reports and AESIs, the following other Special Situations Reports (even in the absence of an Adverse Event) should be collected and transmitted to (depending on which drugs the subject was exposed to) Genentech within thirty (30) calendar days and AstraZeneca within fifteen (15) calendar days:

- Data related to study drug usage during **breastfeeding.**
- Data related to **overdose, abuse, misuse or medication error** (including potentially exposed or intercepted medication errors). Medication errors refers to any error in the prescribing, dispensing, or administration of a medicinal product while in the control of the healthcare professional, patient or consumer.
- **Occupational exposure** refers to the exposure to a medicinal product as a result of one's professional or nonprofessional occupation.
- Data related to suspected transmission of an infectious agent via a medicinal product
- Suspected Adverse Reaction related to quality defect or falsified medicinal products (whether suspected or confirmed)

In addition, reasonable attempts should made to obtain and submit the age or age group of the patient, in order to be able to identify potential safety signals specific to a particular population.

Product Complaints and Reporting
A product complaint is any written or oral information received from a complainant that alleges deficiencies related to identity, quality, safety, strength, purity, reliability, durability, effectiveness or performance of a product after it has been released and distributed to the commercial market or clinical trial.

To File A Complaint:

For all Investigator Initiated Studies (interventional and non-interventional):

Product Complaints with an AE (adverse event) should be reported via email/fax to: usds_aereporting-d@gene.com OR 650-238-6067

Product Complaints without an AE (adverse event) should be reported via email to:

- For Interventional Investigator Initiated Studies:
  kaiseraugst.global_impcomplaint_management@roche.com
- For Non-Interventional Investigator Initiated Studies:
  us-acmo-d@gene.com

All complaints must be filed within one (1) business day for pre-approved products and fifteen (15) calendar days for approved products. Complaints can be reported using a Medwatch, CIOMS or any Genentech-approved reporting form (same as SAEs, AESI etc.).

Reporting Requirements for Adverse Events Originating from Patient Reported Outcomes
Although sites are not expected to review the patient reported outcome (PRO) data, if physician/study personnel become aware of a potential adverse event during site review of the PRO questionnaire data, he/she will determine whether the criteria for an adverse event have been met and, if so, these must be reported using the Adverse Event and Special Situation Reporting Form or MedWatch form.

Aggregate Reports
Investigators will also report events to their IRB as required.

The investigator/sponsor will forward a copy of the Publication to AstraZeneca and Genentech/Roche upon completion of the Study.

The Sponsor agrees to share a copy of their own DSUR with AstraZeneca and Genentech/Roche as soon as reasonably possible after completion. Genentech/Roche agrees to forward to the Sponsor an executive summary of the Genentech/Roche DSUR upon request. Furthermore, Genentech/Roche agrees that the Sponsor may cross-reference the executive summary of the Genentech/Roche DSUR, as applicable.

MedWatch 3500A Reporting Guidelines
In addition to completing appropriate patient demographic and suspect medication information, the report should include the following information within the event description (Section 5) of

the MedWatch 3500A form:

- Study Identifiers (Investigator-sponsor and AstraZeneca protocol number and title description)
- Patient Identifiers (unique patient number, age, or sex)
- Study drugs including start date, most recent dose date, dose, units, frequency, and route
- Description of Serious Adverse Event / Special Situation Narrative and clearly identified event term should both be provided including:
    - Onset date
    - Resolution date
    - Outcome
    - Seriousness criteria
    - Action taken with study drugs
- Reporter (Investigator name, email, fax, institution name)
- Date of Investigator-sponsor awareness of Serious Adverse Event / Special Situation Report
- Relevant medical history, concomitant medications, or medical test results
- Investigator-sponsor causality assessment of relationship to study drugs (related or unrelated) and causality, if not related to study drugs
- Investigator-sponsor IND/CTA number
- IND/CTA Holder (if not reporting Investigator-sponsor)

Follow-up Information

Additional information may be added to a previously submitted report by any of the following methods:

- Adding to the original MedWatch 3500A form and submitting it as follow-up
- Adding supplemental summary information and submitting it as follow-up with the original MedWatch 3500A form
- Summarizing new information and faxing it with a cover letter, including patient identifiers (i.e., date of birth, initial, patient number), protocol description and number (if assigned), brief adverse event description, and notation that additional or follow-up information is being submitted. (The patient identifiers are important so that the new information is added to the correct initial report.)

Occasionally, Genentech or AstraZeneca may contact the reporter for additional information, clarification, or current status of the patient for whom and adverse event was reported. For questions regarding serious adverse event reporting, you may contact the Genentech Drug Safety representative noted above or the AstraZeneca Medical Safety team (AEMailboxClinicalTrialTCS@astrazeneca.com) or the Medical Science Liaison assigned to the study. Relevant follow-up information should be submitted to Genentech Drug Safety and AstraZeneca Medical Safety as soon as it becomes available and/or upon request.

The MedWatch 3500A (mandatory reporting) form is available at:
https://www.fda.gov/media/69876/download

IND Annual Reports

All IND annual reports submitted to the FDA by the Sponsor-investigator should be copied to Genentech and AstraZeneca. Copies of such reports should be emailed to Genentech Drug Safety CTV mailbox at ctvist_drugsafety@gene.com and AstraZeneca Drug Safety atAEMailboxClinicalTrialTCS@astrazeneca.com.

DF/HCC Expedited Reporting Guidelines

External (Non-DF/HCC) sites will send their report to the lead site project manager and Overall PI for review and submission to the appropriate entities according to DFCI IRB policies and procedures in reporting adverse events.

Attribution	DF/HCC Reportable AEs				
	Gr. 2 & 3 AE Expected	**Gr. 2 & 3 AE Unexpected**	**Gr. 4 AE Expected**	**Gr. 4 AE Unexpected**	**Gr. 5 AE Expected or Unexpected**
Unrelated Unlikely	Not required	Not required	Not required	10 calendar days	1 business day*
Possible Probable Definite	Not required	10 calendar days	Not required	10 calendar days	1 business day*
* For participants enrolled and actively participating in the study or for AEs occurring within 30 days of the last intervention, the AE should be reported within 1 business day of learning of the event.					

7.5 Study Closeout

Any study report submitted to the FDA by the Sponsor-investigator should be copied to Genentech and AstraZeneca. This includes all IND annual reports and the Clinical Study Report (final study report). Additionally, any literature articles that are a result of the study should be sent to Genentech and AstraZeneca. Copies of such reports should be mailed to the assigned AstraZeneca Medical Affairs contact for the study and to Genentech Drug Safety CTV oversight mailbox: ctvist_drugsafety@gene.com.

7.6 Safety Queries

Queries related to the Study will be answered by the Investigator. However, responses to all safety queries from regulatory authorities or for publications will be discussed and coordinated between the investigator and the funding sponsors. The investigator and funding sponsors agree that Genentech and AstraZeneca shall have the final say and control over safety queries relating to their respective Product. The Sponsor-Investigator agrees that he shall not answer such queries from regulatory authorities and other sources relating to the Product independently but shall redirect such queries to Genentech or AstraZeneca.

Both the investigator and the funding sponsors will use all reasonable effort to ensure that deadlines for responses to urgent requests for information or review of data are met. They will clearly indicate on the request the reason for urgency and the date by which a response is required.

7.7 Safety Crisis Management

In case of a safety crisis, e.g., where safety issues have a potential impact on the indication(s), on the conduct of the Study, may lead to labeling changes or regulatory actions that limit or restrict the way in which their respective Product is used, or where there is media involvement, the investigator or the funding sponsor (whomever first identified the crisis) will contact the other as soon as possible.

Both the investigator and the funding sponsors agree that Genentech and/or AstraZeneca shall have the final say and control over safety crisis management issues relating to the Product. The Sponsor-Investigator agrees that he shall not answer such queries from media and other sources relating to the Product but shall redirect such queries to Genentech and/or AstraZeneca.

7.8 Adverse Event Characteristics

- **IWCLL term (AE descriptions) and grade**: The descriptions and grading scales found in the IWCLL Grading Scale for Hematologic Toxicities in CLL Studies (Appendix C) will be utilized for hematologic AE reporting.

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for nonhematologic AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **For expedited reporting purposes only:**
  - AEs for the agent(s) that are listed above should be reported only if the adverse event varies in nature, intensity or frequency from the expected toxicity information which is provided.
  - Other AEs for the protocol that do not require expedited reporting are outlined in the next section (Expedited Adverse Event Reporting) under the sub-heading of Protocol-Specific Expedited Adverse Event Reporting Exclusions.

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

7.9 Expedited Adverse Event Reporting

Investigators must report to the Overall PI any serious adverse event (SAE) that occurs after the initial dose of study treatment, during treatment, or within 30 days of the last dose of treatment on the local institutional SAE form.

Investigative sites within DF/HCC will report AEs directly to the DFCI Office for Human Research Studies (OHRS) per the DFCI IRB reporting policy.

For Multi-Center Trials where a DF/HCC investigator is serving as the Sponsor, each participating institution **must** abide by the reporting requirements set by the DF/HCC. Criteria for reporting to DFCI listed in section 7.4.1.

This applies to any medical event equivalent to an unexpected grade 2 or 3 with a possible, probable or definite attribution, all grade 4 toxicities, (unless specifically mentioned above as not requiring expedited reporting) and grade 5 (death) regardless of study phase or attribution.

External Site Adverse Event Reporting Guidelines

In addition to following the SAE reporting requirements for DFCI, AstraZeneca, and Genentech, other investigative sites will report SAEs to their respective IRB per the local IRB's policies and procedures.

7.10 Expedited Reporting to the Food and Drug Administration (FDA)

The Overall PI, as study sponsor, will be responsible for all communications with the FDA. The Overall PI will report to the FDA, regardless of the site of occurrence, any serious adverse event that meets the FDA's criteria for expedited reporting following the reporting requirements and timelines set by the FDA.

7.11 Expedited Reporting to Hospital Risk Management

Participating investigators will report to their local Risk Management office any participant safety reports or sentinel events that require reporting according to institutional policy.

7.12 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions to the Overall PI on the toxicity case report forms. **AEs reported through expedited processes (e.g., reported to the IRB, FDA, etc.) must also be reported in routine study data submissions.**

8. PHARMACEUTICAL INFORMATION

A list of the adverse events and potential risks associated with the investigational agents administered in this study can be found in Section 7.1.

8.1 Acalabrutinib

8.1.1 Description

The investigational product, acalabrutinib maleate tablet, is presented as an orange film-coated tablet containing 129 mg of acalabrutinib maleate (equivalent to 100 mg of acalabrutinib) drug substance. The acalabrutinib maleate tablets are oval, biconvex, measuring approximately 7.5 × 13 mm. The tablets are debossed with 'ACA 100' on one side and are plain on the reverse. Each tablet also contains the following compendial inactive ingredients: mannitol, microcrystalline cellulose, low-substituted hydroxypropyl cellulose and sodium stearyl fumarate.
The tablet coating contains: hypromellose, copovidone, titanium dioxide, polyethylene glycol, caprylic/capric triglyceride, yellow iron oxide and red iron oxide.

8.1.2 Form

N/A

Storage and Stability

Acalabrutinib maleate tablets are packed in white, high-density polyethylene (HDPE) bottles containing a silica gel desiccant and should be stored according to the storage conditions as indicated on the label. Store at 20°C-25°C (68°F-77°F); excursions permitted to 15°C-30°C (59°F-86°F).
The investigational products are for investigational use only and are to be used only within the context of this study. The study drug supplied for this study must be maintained under adequate security and stored under the conditions specified on the label until dispensed for subject use or returned to AstraZeneca. Labels must remain attached to the containers.

If a temperature excursion is experienced at any site beyond the stated storage parameters, the site must report the excursion and associated details to the lead site project manager as soon as feasible.

Compatibility

N/A

Handling

Qualified personnel, familiar with procedures that minimize undue exposure to themselves and the environment, should undertake the preparation, handling, and safe disposal of the agent in a self-contained and protective environment.

Availability

Acalabrutinib will be supplied by AstraZeneca.

Preparation

N/A

Administration

Acalabrutinib is intended to be administered orally twice daily with 8 ounces (approximately 240 mL) of water. The tablets should be swallowed intact and subjects should not attempt to break tablets or dissolve them in water. Acalabrutinib can be taken with or without meals. Patients who require treatment with H2-receptor antagonists should take acalabrutinib two hours before taking the H2-receptor antagonist. Patients who require treatment with antacids should separate the dosing of acalabrutinib by 2 hours.

If a dose is not taken at the scheduled time, it can be taken as soon as possible up to 3 hours later on the same day, with a return to the normal schedule the following day. If a dose is missed by more than 3 hours, it should be skipped and the next dose should be taken at its regularly scheduled time. The patient should not take extra tablets to make up the missed dose. The missed dose will not be made up and must be returned to the site at the next scheduled visit.

Ordering

Acalabrutinib will be ordered from McKesson who will ship the drug directly to the DF/HCC study sites.

External (Non-DF/HCC) investigative sites will be provided with a study specific drug order form by the lead site project manager once all necessary documents are collected, and the site is otherwise approved to enroll participants on the study.

The drug will be provided at no cost to the participant.

Accountability

The investigator, or a responsible party designated by the investigator, should maintain a careful record of the inventory and disposition of the agent using a drug accountability form.

Destruction and Return

If pre-arranged between AstraZeneca and the site, destruction of used and unused study drug may be performed at the site. Empty containers will be destroyed at the site.

The Drug Accountability Log will contain the date and amount of study drugs received and

dispensed. All used and partially used study drug will be destroyed by the site, in accordance with the site's standard operating procedures (SOPs). All expired drug and any unused drug remaining once all patients are off study treatment will be destroyed on site in accordance with the site's standard operating procedures (SOPs).

8.1 Venetoclax

Description

Venetoclax (ABT-199/GDC-0199) is a novel, orally bioavailable, small-molecule Bcl-2 family inhibitor in the biarylacylsulfonamide chemical class. Venetoclax binds with high affinity (K_i < 0.010 nM) to antiapoptotic protein Bcl-2 and with lower affinity to other antiapoptotic Bcl-2 family proteins, like Bcl-X_L and Bcl-w (> 4,800-fold and > 24,500-fold lower affinity than to Bcl-2, respectively).

In vitro studies have shown that venetoclax is metabolized primarily by CYP3A4. Interaction of venetoclax with co-administered CYP3A inhibitors or inducers may cause an increase or decrease in the exposure of venetoclax. *In vivo*, venetoclax was also demonstrated to be metabolized by CYP3A4. Venetoclax is also substrate for P-glycoprotein (P-gp) and breast cancer resistance protein (BCRP) transporters. Renal contribution to the elimination of venetoclax is negligible, with < 0.1% of the dose being recovered in urine. Based on in vitro results, venetoclax was a P-gp, BCRP, and OATP1B1 inhibitor. It was not a potent in vitro inhibitor of CYP3A4, CYP1A2, CYP2B6, or CYP2D6 (IC50 > 30 μM); and it did not induce CYP3A4 or CYP1A2 at concentrations up to 10 μM. Venetoclax is also not predicted to cause inhibition of CYP2C19, CYP2C8, CYP2C9, and UGT1A1 at clinically relevant concentrations. It is not an inhibitor of UGT1A4, UGT1A6, UGT1A9 and UGT2B7. Following co-administration of a single dose of venetoclax with warfarin, R- and S-warfarin C_{max} and AUC_∞ increased by approximately 18% to 28%. Relatively low variability was observed in warfarin pharmacokinetics.

The venetoclax formulation currently used in clinical studies is a tablet formulation with strengths of 10, 50, and 100 mg. The tablet formulation was orally administered after a low-fat meal. Food increased the bioavailability of venetoclax by 3- to 5-fold. Preliminary pharmacokinetic results indicated that the absorption of venetoclax after the oral dosing was relatively slow. Venetoclax plasma concentrations peaked at approximately 5 to 8 hours after dosing. The mean harmonic terminal phase half-life from 17 to 41 hours and the mean oral clearance was approximately 13 L/hr after a single dose. Preliminary data did not suggest apparent pharmacokinetics differences among subjects with CLL/SLL, NHL, Multiple Myeloma, Acute Myeloid Leukemia, or systemic lupus erythematosus. The combined data from subjects with CLL/SLL and NHL suggested that venetoclax exposure was approximately dose proportional across the 150 to 800 mg dose levels at steady state. Additional information about venetoclax is available in the package insert.

Form

Venetoclax tablets are film coated and are available in strengths of 10 mg, 50 mg, or 100 mg.

Venetoclax will be packaged in high density polyethylene (HDPE) plastic bottles. Each bottle will be labeled per local regulatory requirements. Labels must remain affixed to the bottle.

Storage and Stability

Venetoclax must be stored at 15° to 25°C (59° to 77°F). The investigational products are for investigational use only and are to be used only within the context of this study. The study drug supplied for this study must be maintained under adequate security and stored under the conditions specified on the label until dispensed for subject use or returned to Genentech. Labels must remain attached to the containers.

If a temperature excursion is experienced at any site beyond the stated storage parameters, the site must report the excursion and associated details to the lead site project manager as soon as feasible.

Compatibility

N/A

Handling

Qualified personnel, familiar with procedures that minimize undue exposure to themselves and the environment, should undertake the preparation, handling, and safe disposal of the chemotherapeutic agent in a self-contained and protective environment.

Availability

Venetoclax is provided by Genentech directly to the study site.

Preparation

N/A

Administration

Oral. Patients should take venetoclax tablets with a meal and water at approximately the same time each day. Venetoclax tablets should be swallowed whole and not chewed, crushed, or broken prior to swallowing.

If vomiting occurs within 15 minutes after taking venetoclax and all expelled tablets are still intact, another dose may be given. Otherwise, no replacement dose is to be given. In cases where a dose of venetoclax is missed or forgotten, the patient should take the dose as soon as possible and ensure that the minimal interval between the current dose and the next dose is at least 16 hours in order to avoid excessive drug accumulation after the next dose.

Any overdose or incorrect administration of study drug should be noted in the medical chart.

Adverse events associated with an overdose or incorrect administration of study drug should be reported.

Ordering

Venetoclax will be ordered directly from Genentech, who will ship drug directly to the DF/HCC study sites with an order form provided at the time of study activation.

External (Non-DF/HCC) investigative sites will be provided with a study specific drug order form by the lead site project manager once all necessary documents are collected, and the site is otherwise approved to enroll participants on the study.

The drug will be provided at no cost to the participant.

Accountability

The investigator, or a responsible party designated by the investigator, should maintain a careful record of the inventory and disposition of the agent using a drug accountability form.

Destruction and Return

If pre-arranged between Genentech and the site, destruction of used and unused study drug may be performed at the site. Empty containers will be destroyed at the site.

8.2 Obinutuzumab

Description

Obinutuzumab (GA101, RO5072759), is a glycoengineered, humanized, type II anti-CD20 monoclonal antibody (mAb). Obinutuzumab was derived by humanization of the parental B-Ly1 mouse antibody and subsequent glycoengineering.

Form

Obinutuzumab is provided as a single-use vial. Each vial contains a sterile liquid formulation in a 50-mL pharmaceutical-grade glass vial containing a nominal dose of 1000 mg of obinutuzumab (G3 material). The formulated drug product consists of 25 mg/mL drug substance formulated in histidine/histidine-HCl, trehalose, and poloxamer 188. The vial contains 41 mL (with 2.5% overfill).

Storage and Stability

The recommended storage conditions for the obinutuzumab drug product are between 2°C and 8°C, protected from light. Chemical and physical in-use stability for obinutuzumab dilutions in 0.9% sodium chloride (NaCl) at concentrations of 0.2 - 20 mg/mL have been demonstrated for 24 hours at 2°C - 8°C and an additional 24 hours at ambient temperature and ambient room lighting. The prepared diluted product should generally be used immediately. If not used

immediately, in-use storage times and conditions prior to use are the responsibility of the user and would normally not be longer than 24 hours at 2°C - 8°C unless reconstitution/dilution has taken place in controlled and validated aseptic conditions. Obinutuzumab should not be frozen or shaken. Mix gently. All transfer procedures require strict adherence to aseptic techniques. Do not use an additional in line filter because of potential adsorption.

If a temperature excursion is experienced at any site beyond the stated storage parameters, the site must report the excursion and associated details to the lead site project manager as soon as feasible.

Compatibility

N/A

Handling

Qualified personnel, familiar with procedures that minimize undue exposure to themselves and the environment, should undertake the preparation, handling, and safe disposal of the chemotherapeutic agent in a self-contained and protective environment.

Preparation

Obinutuzumab drug product intended for IV infusion is prepared by dilution of the drug product into an infusion bag containing 0.9% NaCl.

One vial may be used to prepare both the 100-mg dose (equals 4 mL) and 900-mg dose (equals 36 mL) following the directions below. If both bags are prepared at the same time, the reconstitution/dilution has to take place in a controlled and validated aseptic conditions. Subsequently store the 900-mg bag for a maximum of 24 hours at 2°C – 8°C and administer the next day.

To prepare a 100-mg dose: The final drug concentration of a 100-mg dose should be in the range of 0.4 mg/mL to 4.0 mg/mL. Using a 250-mL infusion bag containing 0.9% NaCl, withdraw and discard 4 mL of the sodium chloride. Withdraw 4 mL of obinutuzumab from a single glass vial and inject it into the infusion bag (discard any unused portion of obinutuzumab left in the vial unless reconstitution/dilution has taken place in controlled and validated aseptic conditions). Gently invert the infusion bag to mix the solution. Do not shake.

To prepare a 900-mg dose: The final drug concentration of a 900-mg dose should be in the range of 0.4 mg/mL to 4.0 mg/mL. Using a 250-mL infusion bag containing 0.9% NaCl, withdraw and discard 36 mL of the sodium chloride. Withdraw 36 mL of obinutuzumab from a single glass vial and inject it into the infusion bag (discard any unused portion of obinutuzumab left in the vial unless reconstitution/dilution has taken place in controlled and validated aseptic conditions). Gently invert the infusion bag to mix the solution. Do not shake.

To prepare a 1000-mg dose: The final drug concentration of a 1000-mg dose should be 4

mg/mL. Using a 250-mL infusion bag containing 0.9% NaCl, withdraw and discard 40 mL of the NaCl. Withdraw 40 mL of obinutuzumab from a single glass vial and inject it into the infusion bag (discard any unused portion of obinutuzumab left in the vial). Gently invert the infusion bag to mix the solution. Do not shake.

Administration sets with polyvinyl chloride, polyurethane, or polyethylene as product contact surface and IV bags with polyolefin, polypropylene, polyvinyl chloride, or polyethylene as product contact surface are compatible and may be used. Use of a port or peripherally inserted central catheter line is acceptable.

Do not use obinutuzumab beyond the expiration date stamped on the carton.

Administration

Obinutuzumab is to be administered by IV infusion for up to 6 total cycles (28-day cycles):

- On first cycle, Day 1, 100 mg obinutuzumab will be administered
- On first cycle, Day 2, 900 mg of obinutuzumab will be administered
- On first cycle, Days 8 and 15, 1000 mg of obinutuzumab will be administered.
- On cycles 2-6, Day 1, 1000 mg of obinutuzumab will be administered (see section 5.3.2)

Obinutuzumab must be administered in a clinical setting (inpatient or outpatient). Full emergency resuscitation facilities should be immediately available, and patients should be under close supervision by the investigator at all times. Obinutuzumab should be given as a slow IV infusion through a dedicated line. IV infusion pumps (such as Braun Infusomat Space) should be used to control the infusion rate of obinutuzumab. Do not administer as an IV push or bolus. After the end of the first infusion, the IV line should remain in place for at least 2 hours in order to be able to administer IV drugs if necessary. If no AEs occur after 2 hours, the IV line may be removed. For subsequent infusions, the IV line should remain in place for at least 1 hour from the end of infusion; if no AEs occur after 1 hour, the IV line may be removed.

Ordering

Each participating site will utilize a study specific order form from Genentech provided at the time of study activation to order obinutuzumab.

External (Non-DF/HCC) investigative sites will be provided with a study specific drug order form by the lead site project manager once all necessary documents are collected, and the site is otherwise approved to enroll participants on the study.

The drug will be provided at no cost to the participant.

Accountability

The investigator, or a responsible party designated by the investigator, should maintain a careful record of the inventory and disposition of the agent using the NCI Drug Accountability Record

Form (DARF) or another comparable drug accountability form.

Destruction and Return

Destruction of used and unused study drug may be performed at the site as per institutional policy.

9. BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

9.1 Laboratory Correlative Studies

9.1.1 BH3 Profiling

BH3 profiling is a functional assay we previously developed that detects the proximity of CLL cells to the threshold of apoptosis (what we call 'priming') through interrogation of BCL-2 family members. To perform a BH3 profile, we add individual BH3-only peptides to gently permeabilized malignant cells and use FACS to measure the amount of mitochondrial depolarization induced by each peptide, as measured by cytochrome c release.

We hypothesize that patients whose baseline CLL cells, prior to therapy, undergo significant depolarization to BIM BH3 peptide (highly primed) will have superior clinical response to AVO combination therapy compared to patients whose cells undergo minimal BIM BH3 depolarization (unprimed). These assessments will be made on circulating CLL cells from the peripheral blood drawn from patients at baseline. We will attempt to identify potential resistance mechanisms by looking at whether patients who do not achieve CR have different upfront BH3 profiles from those who do. Additionally, another sample of peripheral blood will be collected from subjects at the time of disease progression, and the BH3 profile of these samples will be compared to subjects' baseline samples to help identify whether a change in anti-apoptotic protein dependence is observed as a possible mechanism of resistance.

If we have bone marrow aspirates available, we may also perform BH3 profiling to see whether the level of priming in CLL cells from this compartment is a better predictor of response than peripheral blood CLL cells.

We hypothesize that CLL cells taken from patients who have received acalabrutinib and obinutuzumab combination therapy will be more Bcl-2 dependent than their matched CLL cells at baseline. This will be measured by dynamic BH3 profiling, a technique that allows us to measure "delta priming," defined as the difference in mitochondrial response to a BH3 peptide in a drug-treated sample versus an untreated control. Specifically, we wish to confirm if acalabrutinib increases sensitivity to the BAD BH3 peptide as opposed to the BIM BH3 peptide, as we have previously reported (Deng et al., 2017). We will also determine if the potentiation of venetoclax activity that we see with acalabrutinib treatment *in vitro* can predict depth of response to the acalabrutinib/venetoclax combination *in vivo*. After completion of cycle 1 (on cycle 2 day 1 prior to administration of obinutuzumab) and after completion of cycle 3 (on cycle 4 day 1 prior to administration of venetoclax) we will obtain a peripheral blood sample. We will

compare the dynamic BH3 profile of these samples to a baseline sample, which will allow us to assess *in vivo* the shortterm change in apoptotic priming induced by acalabrutinib and also by acalabrutinib and obinutuzumab, respectively. We will correlate this with clinical response to subsequent venetoclax therapy. We will also compare the dynamic BH3 profile of a peripheral blood sample collected at cycle 8 day 1 (after 4 months of venetoclax therapy) to the dynamic BH3 profiles at baseline and immediately prior to venetoclax therapy.

All peripheral blood samples will promptly be delivered to the laboratory of Dr. Jennifer Brown, where they will undergo Ficoll purification and then be viably frozen in FBS with 10% DMSO. The viably-frozen samples will be batched and stored, and the BH3 profiling assays will be performed in Dr. Davids' lab (for detailed methods see Ryan et al., 2010).

9.1.2 Genomic Analyses

We plan to perform whole exome sequencing on CLL cells and normal tissue from patients at baseline to evaluate for somatic mutations such as *TP53*, *NOTCH1* and *SF3B1* that may confer drug sensitivity and resistance. If the patient does not have significant circulating CLL cells, we will pursue sequencing of bone marrow aspirate samples in patients who require a new bone marrow biopsy for screening. In addition, saliva as a source of germline will be collected prior to study initiation and may be collected more than once if inadequate specimen is obtained. All samples will promptly be delivered to the laboratory of Dr. Jennifer Brown, where DNA will be extracted and then sent to the Broad Institute (Cambridge, MA) for whole exome sequencing. In an exploratory analysis, we will assess novel mutations as potential predictors of response and progression-free survival. We will also collect a sample from each patient at time of relapse or progression for repeat analysis by whole exome sequencing to assess for acquired resistance mutations.

9.1.3 Adaptive ClonoSEQ Assay

To assess whether the presence of minimal residual CLL cells in the peripheral blood as measured by a novel sequencing platform is predictive of progression free and overall survival, peripheral blood and/or bone marrow samples will be collected for analysis at the following time points: screening visit, cycle 4 day 1, cycle 8 day 1, cycle 13 day 1, cycle 16 day 1, and cycle 25 day 1. Samples will be sent to the Brown Lab at DFCI where they will be stored initially prior to batch shipping to Adaptive for analysis. Using Adaptive's ClonoSEQ platform, rearranged immunoreceptor loci from genomic DNA will be extracted, amplified, and sequenced using V and J segment primers for each immunoreceptor gene. Tumor-specific clonotypes will be identified for each patient based on their high prevalence in peripheral blood or bone marrow. Sequences will be analyzed using standardized algorithms for clonotype determination. Adaptive MRD levels will be quantified using spiked-in reference sequences.

9.1.4 Additional Prognostic Tests

Patients who have not had ZAP-70 and *IGHV* testing done locally will have baseline samples sent for testing to Genzyme/LabCorp. These samples will be shipped alongside the Integrated Oncology/LabCorp requisition form ambient priority overnight to:

Genzyme/LabCorp Specialty Testing Group
521 West 57th Street
New York, NY 10000

Table 12: Sample Collection Schedule

Sample Time Point	Container[1]	Sample Type[2]	Shipping Method	Recipient
Screening or Pre-dose Day 1	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profiling, whole exome sequencing, Adaptive Clonoseq)
	2 x 6 mL Green Top	Bone Marrow Aspirate		
	1 x Oragene Kit	Saliva[3]	Ambient overnight	
	2 x 6mL Green Top*	Peripheral blood	Fridge pack overnight	Genzyme/LabCorp[4]
Cycle 2, Day 1 (start of obinutuzumab)	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile)
Cycle 2, Day 8	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile)
Cycle 4, Day 1 (start of venetoclax)	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
	2 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Brown Laboratory, DFCI (Adaptive Clonoseq)
	1 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Mayo Clinic (MRD Assessment)[5]
Cycle 8, Day 1 (one cycle after completion of obinutuzumab)	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
	2 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Brown Laboratory, DFCI (Adaptive Clonoseq)
	1 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Mayo Clinic (MRD Assessment)[5]
Cycle 13, Day 1	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral Blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
Cycle 16, Day 1 (primary endpoint assessment of MRD)	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral Blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
	2 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Brown Laboratory, DFCI (Adaptive Clonoseq)
	1 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Mayo Clinic (MRD Assessment)[5]
Cycle 25, Day 1	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral Blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
	2 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Brown Laboratory, DFCI (Adaptive Clonoseq)

	1 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	Mayo Clinic (MRD Assessment)[5]
Treatment Discontinuation	6x 6 mL Green Top 1 x 6 mL Red Top	Peripheral Blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profile, Adaptive Clonoseq)
	2 x 6mL Green Top	Bone Marrow Aspirate	Fridge pack overnight	
Relapse/Disease Progression	1 x 6 mL Red Top	Peripheral Blood	Fridge pack overnight	Brown Laboratory, DFCI (BH3 profiling, whole exome sequencing, Adaptive Clonoseq)
	6 x 6 mL Green Top	Peripheral Blood	Fridge pack overnight	
	2 x 6 mL Green Top	Bone Marrow Aspirate		

[1] Green top= sodium heparin tube; Red top= no additive;
[2] For time points that mention blood and or aspirate: If patient has few circulating CLL cells in peripheral blood, send additional aspirate (1 x 6 mL green top tube) in addition to blood
[3] Oragene kits require approximately 2 mLs of saliva
[4] Genzyme/LabCorp only provides requisition form
[5] Bone marrow and peripheral blood samples sent to Mayo Clinic for MRD assessment are considered part of standard of care disease monitoring, not research.

* Samples required for all patients at baseline unless previously tested for ZAP-70, *IGHV*, *TP53*, and *NOTCH1 (DFCI only)*.

Correlative studies note: While the goal of the correlative studies is to provide supportive data for the clinical study, there may be circumstances when a decision is made to stop a collection, not perform, or discontinue an analysis due to either practical reasons (e.g., inadequate sample number, issues related to the quality of the sample or issues related to the assay that preclude analysis, impossibility to perform correlative analyses, etc). Therefore, depending on the results obtained during the study, sample collection/analysis may be omitted at the discretion of the PI. Additional correlative samples may be collected after the 24 cycle time point at the discretion of the investigators.

9.2 Correlative Lab Sample Collection and Shipping Instructions

DFCI patient samples will be hand-delivered to the lab of Dr. Jennifer Brown. Other DF/HCC sites may send samples via preferred courier service, or hand delivery to the lab.

External Investigative Site Sample Preparation and Shipment:
Once fresh samples (blood and bone marrow aspirate) are collected, the vacutainers will be refrigerated and stored per instructions below. There is no required processing for any of the samples by the participating site prior to shipment.

1. Package tubes at room temperature and wrap in a liberal amount of paper towel around the tubes to ensure adequate insulation of the specimen(s) and absorption in the event of a breakage.
2. Place wrapped specimen in a biohazard labeled Ziploc bag or sealable bag with a fridge pack and securely close.
3. Wrap bubble wrap around the bag and place in a cardboard box. If space remains in the box, stuff with extra paper towel to reduce shifting of samples.

4. Complete the shipping requisition form using the address listed below. Prepare the package for shipping, applying packing tape as needed.
5. Ship the package using FedEx or UPS next day or overnight delivery the same day the sample was collected.
6. Only ship Monday-Thursday, and avoid shipping before a holiday as the lab does not receive packages on Saturday or on holidays.
7. With each shipment, please include the following:
    a. A sample requisition form (Appendix I), including the patient number, timepoint, study number, must accompany each shipment. Please sign and date the form, and retain a copy for site record maintenance.
    b. An electronic copy (Word or PDF) of the sample requisition form must also be sent via email and include the tracking number of the package. The listing must also include a contact name, address and phone number of the person who is responsible for the shipment.
8. Please email the Lead Site Research Project Manager and Brown Lab contact to notify of an incoming shipment at least 48 hours prior to a shipment
9. Samples may be shipped via overnight air to:

Stacey Fernandes, Principal Research Technician
DFCI CLL Center, J. Brown lab
1 Jimmy Fund Way
SM 648
Boston, MA 02115, U.S.A.
617-632-5828 (phone)

10. STUDY CALENDAR

Screening imaging should be conducted 30 days or less prior to start of patient registration, unless otherwise indicated. PET scans paired with nondiagnostic CT scans may substitute for CT scans, either at screening or at any point during the trial, if deemed clinically necessary by the investigator. CT scans must include neck, chest, abdomen, and pelvis. Bone marrow aspirate and/or biopsy is required within approximately 6 months prior to registration. Baseline laboratory testing should be completed within 2 weeks of initiation of therapy, unless otherwise indicated. In the event that the participant's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy.

Assessments must be performed prior to administration of any study agent. Study assessments and agents should be administered within $\pm$ 7 days of the protocol-specified date during the cycles 1-15, and within $\pm$14 days for cycles 16-24 and beyond, unless otherwise noted.

Study Calendar for all patients (Cycles 1-16)

Cycle = 28 days	**Screen**	**Cycle 1**	**Cycle 2** (± 3 days)					**Cycle 3** (±3 days)		**Cycle 4**[11] (±7 days)										**Cycle 5** (±7 days)		**Cycles 6-7** (±7 days)	**Cycle 8** (±7 days)	**Cycles 9, 11** (±7 days)	**Cycle 13** (±7 days)	**Cycle 16**[12] (±14 days)
Procedure	Day -30 to 0	Day 1	Day 1	Day 2	Day 8	Day 15	Day 22	Day 1	Day 15	Day 1	Day 2	Day 3	Day 8	Day 9	Day 15	Day 16	Day 22	Day 23	Day 1	Day 15	Day 1	Day 1	Day 1	Day 1	Day 1
Acalabrutinib		X----------Daily dosing days 1-28----------X																							
Obinutuzumab			X	X	X	X		X		X									X		X				
Venetoclax										X----------Daily dosing days 1-28----------X															
Informed Consent	X																								
Medical history, Physical Exam, Vital Signs ((height, weight, resp rate, HR, BP, temp, O_2 sat), ECOG Performance Status[17]	X	X	X		X	X	X	X	X	X	X		X		X		X		X	X	X	X	X	X	X
Urinalysis, EKG	X																								
Serum Pregnancy Test[1]	X																								
Bone Marrow aspirate / biopsy / MRD assessment[2,3,4, 16]	X									X[13]												X			X
SPEP + Immunoglobulins	X									X												X			X
HIV, HCV, HBV, HTLV serologies[5]	X																								
β_2 microglobulin, PT, PTT, INR	X																								
CT scan	X									X												X		X	X
Hematology[6]	X	X	X	X	X	X	X	X	X	X			X		X		X		X	X	X	X	X	X	X
Serum Chemistry[7]	X	X	X[14]	X	X	X	X	X	X	X[15]	X[15]	X	X[15]	X	X[15]	X	X[15]	X	X	X	X	X	X	X	X
FISH analysis	X																					X			X
IGHV, TP53, ZAP-70, NOTCH1	X																								
Adverse Event Evaluation	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
TLS Prophylaxis[8]			X	X					X----------X																
Peripheral Blood MRD Analysis[9, 16]										X												X		X	X
Adaptive ClonoSEQ MRD Analysis										X												X		X	X
Other correlative studies[10]	X	X	X		X					X												X			X

[1] Only required in women of childbearing potential

[2] Unilateral bone marrow aspirate and/or biopsy required within 6 months of screening.

[3] MRD assessment should be sent on marrow samples taken after cycles 3, 7, 15, and 24.

[4] Marrow sample requested for correlative studies if bone marrow biopsy is done as standard of care, or as part of protocol schedule. Flow cytometry (lymphoma panel), karyotype, and FISH (CLL) should be performed on all bone marrow biopsies (FISH may be performed on either marrow or blood and is not required on both).

[5] These baseline serologies may be completed within 4 weeks of initiation of therapy. For those patients who are either hepatitis B core antibody and/or hepatitis B surface antigen positive but HBV DNA negative, pharmacologic prophylaxis and monitoring of HBV DNA is required as detailed in section 5.4. HTLV serologic testing is not required but is strongly suggested if the treating clinician has a high suspicion for HTLV infection.

[6] CBC with differential, hemoglobin, hematocrit, platelets

[7] Calcium, Sodium, Potassium, Phosphorus, Chloride, Glucose, Magnesium, Albumin, AST, ALT, Creatinine, BUN, Bicarbonate, Total protein, Total bilirubin (and direct bilirubin if total Bilirubin is abnormal), Uric acid, LDH, Alkaline phosphatase
[8] TLS prophylaxis will consist of allopurinol in subjects without a history of an adverse reaction to this drug, or an alternative anti-hyperuricemic agent.
[9] Peripheral blood MRD analysis will be done by four color flow cytometry through a reference laboratory with a detection limit of 10^{-4} cells per leukocyte.
[10] Baseline correlative studies may be obtained either at the screening visit or prior to study drug administration on C1D1.
[11] Only cycle days requiring outpatient visits or laboratory evaluations are listed. Provided that there is no evidence of TLS or adverse events, venetoclax dosing is continuous throughout this cycle at doses as specified in the treatment plan.
[12] Subjects will have a response evaluation on cycle 16 day 1 and will continue on therapy until information on disease status is obtained from the restaging studies, at which point they will follow one of the two calendars below.
[13] Bone marrow biopsy at cycle 4 day 1 is optional.
[14] Chemistries to be drawn prior to administration of obinutuzumab and approximately 6-8 hours after initiation of obinutuzumab administration on this day.
[15] Chemistries to be drawn prior to administration of venetoclax and approximately 6-8 hours after administration of venetoclax on these days.
[16] Bone marrow and peripheral blood samples sent to Mayo Clinic for MRD assessment are considered part of standard of care disease monitoring, not research.
[17] Height needs to be collected in screening only.

Off-study evaluation. Note: for IND/IDE trials, follow up visits or other contact are required in order to identify SAEs during the 30 days following the end of study treatment.

Study Calendar for patients who **ARE** in complete remission with MRD-negativity in the bone marrow and blood at cycle 16 evaluation and choose to discontinue therapy
(Cycles 17 and beyond)

Cycle = 28 days	**Cycle 19** (±14 days)	**Cycle 22** (±14 days)	**Cycle 25** (±14 days)	**Every 3 cycles**[7] (±14 days)	**Off Treatment**	**Off Study**[8]
Procedure	Day 1	Day 1	Day 1			
Acalabrutinib[1]						
Obinutuzumab						
Venetoclax[1]						
Medical history, physical exam, vital signs (weight, resp rate, HR, BP, temp, O_2 sat) and ECOG Performance Status	X	X	X	X	X	X
Bone Marrow aspirate / biopsy / MRD assessment[2,3,9]			X			
CT scan (+/- 7 days)			X		X	X
Hematology[4] and Serum Chemistry[5]	X	X	X	X	X	X
Adverse Event Evaluation	X	X	X	X	X	X
Peripheral Blood MRD Analysis[6,9]	X	X	X	X	X	X
Adaptive ClonoSEQ MRD Analysis			X		X	X
Other correlative studies			X		X	X

[1] If relapse is detected (by clinical, radiographic, or MRD testing), subjects will resume acalabrutinib and venetoclax until progression / unacceptable toxicity. Follow-up schedule will be modified to match the calendar on the following page.
[2] MRD assessment should be sent on marrow sample taken after cycle 24.
[3] Marrow sample requested for correlative studies if bone marrow biopsy is done as standard of care, or as part of protocol schedule. Flow cytometry (lymphoma panel), karyotype, and FISH (CLL) should be performed on marrow at all bone marrow biopsies (FISH may be performed on either marrow or blood and is not required on both).
[4] CBC with differential, hemoglobin, hematocrit, platelets
[5] Calcium, Sodium, Potassium, Phosphorus, Chloride, Glucose, Magnesium, Albumin, AST, ALT, Creatinine, BUN, Bicarbonate, Total protein, Total bilirubin (and direct bilirubin if total Bilirubin is abnormal), Uric acid, LDH, Alkaline phosphatase
[6] Peripheral blood MRD analysis will be done by four color flow cytometry through a reference laboratory with a detection limit of 10^{-4} cells per leukocyte.
[7] Subjects with CR/CRi and MRD-negativity at C16D1 who choose to discontinue therapy will be seen in clinic on cycles 19, 22, and 25 and then every three cycles for 2 years (i.e. through cycle 48). If relapse is detected by PB flow cytometry at any of these time points, subjects will be re-staged with CT scan (if not done in the three months prior) and bone marrow biopsy (if not done in the six months prior) and will resume acalabrutinib and venetoclax until progression / unacceptable toxicity per the follow-up schedule on the following page. If no relapse is detected by cycle 48, subjects who are no longer on study treatment may return to follow-up with a local hematologist but will be followed by telephone for updates on disease progress and overall survival.
[8] If at the time of coming off study the subject is ≤2 weeks from coming off of therapy, the evaluations done at the off-treatment visit will suffice and need not be repeated. Follow-up data will be collected every three cycles until death, up to 5 years after the initial dosing.
[9]Bone marrow and peripheral blood samples sent to Mayo Clinic for MRD assessment are considered part of standard of care disease monitoring, not research.

Study Calendar for patients who **ARE NOT** in complete remission with MRD-negativity in the bone marrow and blood at cycle 16 evaluation OR who are in complete remission yet choose to continue therapy
(Cycles 17 and beyond)

Cycle = 28 days	**Cycle 19** (±14 days)	**Cycle 22** (±14 days)	**Cycle 25**[6] (±14 days)	**Every 3 cycles until cycle 48**[7] (±14 days)	**Clearance of MRD from the blood**[8]	**Off Treatment**	**Off Study**[9]
Procedure		Day 1	Day 1				
Acalabrutinib	X----------Daily dosing days 1-28----------X				X		
Obinutuzumab							
Venetoclax	X----------Daily dosing days 1-28----------X				X		
Medical history, physical exam, vital signs (weight, resp rate, HR, BP, temp, O_2 sat), and ECOG Performance Status	X	X	X	X		X	X
Bone Marrow aspirate / biopsy / MRD assessment[1,2,10]			X		X		
CT scan (+/- 7 days)			X		X	X	X
Hematology[3] and Serum Chemistry[4]	X	X	X	X	X	X	X
Adverse Event Evaluation	X	X	X	X		X	X
Peripheral Blood MRD Analysis[5,10]	X	X	X	X		X	X
Adaptive ClonoSEQ MRD Analysis			X		X	X	X
Other correlative studies			X		X	X	X

[1] MRD assessment should be sent on marrow samples taken after cycle 24.
[2] Marrow sample requested for correlative studies if bone marrow biopsy is done as standard of care, or as part of protocol schedule. Flow cytometry (lymphoma panel), karyotype, and FISH (CLL) should be performed on marrow at all bone marrow biopsies (FISH may be performed on either marrow or blood and is not required on both).
[3] CBC with differential, hemoglobin, hematocrit, platelets
[4] Calcium, Sodium, Potassium, Phosphorus, Chloride, Glucose, Magnesium, Albumin, AST, ALT, Creatinine, BUN, Bicarbonate, Total protein, Total bilirubin (and direct bilirubin if total Bilirubin is abnormal), Uric acid, LDH, Alkaline phosphatase
[5] Peripheral blood MRD analysis will be done by four color flow cytometry through a reference laboratory with a detection limit of 10^{-4} cells per leukocyte.
[6] Acalabrutinib and venetoclax are continued until cycle 24. Subjects who are MRD-negative at cycle 24 restaging will stop acalabrutinib and venetoclax. These subjects receive peripheral blood monitoring for minimal residual disease every three cycles for up to 2 years after completion of cycle 24 (i.e. cycle 48). If there is detectable recurrence by peripheral blood flow cytometry during that time, subjects will be re-staged (with CT scan if not done in the three months prior and bone marrow biopsy and rapid heme panel (RHP) if not done in the six months prior) and acalabrutinib and venetoclax will be resumed until clinical progression or unacceptable toxicity. Subjects who are MRD-positive at cycle 24 restaging will continue acalabrutinib and venetoclax with every three month follow-up visits until disease progression or unacceptable toxicity.
[7] Subjects will continue to be seen every three cycles for monitoring, for up to two years after cycle 24 (i.e. through cycle 48). After these two years, subjects who

are no longer on study treatment may return to follow-up with a local hematologist but will be followed by telephone for updates on disease progress and overall survival. Subjects who remain on study therapy will continue to be seen every three months. CT scans will be obtained every six months in patients who are not in remission and who are still on study therapy.

[8] Subjects who eventually achieve MRD-negativity in the blood after cycle 24 should have complete disease re-staging with studies as outlined in the table. These re-staging studies should occur within approximately one month of achievement of MRD-negativity. Per protocol, these subjects should continue to remain on therapy until the results of the MRD assessment are obtained. Patients will have the option to discontinue therapy at any point after cycle 25 if they achieve two consecutive negative MRD assessments that are 3 months apart.

[9] If at the time of coming off study the subject is ≤4 weeks from coming off of therapy, the evaluations done at the off-treatment visit will suffice and need not be repeated. Follow-up data will be collected every three cycles until death, up to 5 years after the initial dosing.

[10] Bone marrow and peripheral blood samples sent to Mayo Clinic for MRD assessment are considered part of standard of care disease monitoring, not research.

Study Calendar for patients in whom recurrence is detected after ceasing study therapy (Retreatment Cycles 1 and beyond)

Cycle = 28 days	**Detection of disease recurrence**[5]	**Retreatment Cycle 1**[6]	**Retreatment Cycle 2** (±3 days)	**Retreatment Cycle 3** (±3 days)	**Every 3 cycles** (±14 days)	**Off Treatment**	**Off Study**[7]
Procedure		Day 1	Day 1	Day 1			
Acalabrutinib		X----------Daily dosing days 1-28----------X					
Obinutuzumab							
Venetoclax		X----------Daily dosing days 1-28----------X					
Medical history, physical exam, vital signs (weight, resp rate, HR, BP, temp, O_2 sat), and ECOG Performance Status	X	X	X	X	X	X	X
Bone Marrow aspirate / biopsy[1]	X						
CT scan (+/- 7 days)	X					X	X
Hematology[3] and Serum Chemistry[4]	X	X	X	X	X	X	X
Adverse Event Evaluation		X	X	X	X	X	X
Peripheral Blood MRD Analysis[8]						X	X
Adaptive ClonoSEQ MRD Analysis	X					X	X
Other correlative studies	X		X			X	X

[1] Marrow sample requested for correlative studies if bone marrow biopsy is done as standard of care, or as part of protocol schedule. Flow cytometry (lymphoma panel), karyotype, and FISH (CLL) should be performed on marrow at all bone marrow biopsies (FISH may be performed on either marrow or blood and is not required on both).
[3] CBC with differential, hemoglobin, hematocrit, platelets. RHP (rapid heme panel) should be done if not done within 6 months of disease recurrence.
[4] Calcium, Sodium, Potassium, Phosphorus, Chloride, Glucose, Magnesium, Albumin, AST, ALT, Creatinine, BUN, Bicarbonate, Total protein, Total bilirubin (and direct bilirubin if total Bilirubin is abnormal), Uric acid, LDH, Alkaline phosphatase
[5] Subjects who develop disease recurrence must be re-evaluated with the studies listed in this column. These re-staging studies must occur within one month of detection of disease recurrence.
[6] Venetoclax may be reinitiated on retreatment cycle 1 day 1 at the patient's previously tolerated maximum daily dose only if recurrence was detected using peripheral blood MRD testing and the subject otherwise still meets IWCLL complete response criteria (as based on CT scans, bone marrow biopsy, and peripheral blood sampling). If the subject has recurrent disease and no longer meets complete response criteria then the subject should be re-assessed for TLS risk as discussed in section 6.1.2 and venetoclax should be re-initiated following the ramp-up schedule used in cycle 4 and detailed in section 5.3.3.
[7] If at the time of coming off study the subject is ≤4 weeks from coming off of therapy, the evaluations done at the off-treatment visit will suffice and need not be repeated. Follow-up data will be collected every three cycles until death, up to 5 years after the initial dosing.
[8] Bone marrow and peripheral blood samples sent to Mayo Clinic for MRD assessment are considered part of standard of care disease monitoring, not research.

11. MEASUREMENT OF EFFECT

11.1 Definitions

Evaluable for toxicity: All participants who receive at least one dose of study treatment will be evaluable for toxicity from the time of their first treatment on cycle 1 / day 1.

Evaluable for objective response: All subjects who have received at least one dose of study drug are evaluable for response. These participants will have their response classified according to the definitions stated below.

11.2 Methods for Evaluation of Measurable Disease

All baseline evaluations (physical exam and radiographic evaluations) should be performed as closely as possible to the beginning of treatment and not more than 30 days before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination when both methods have been used to assess the anti-tumor effect of a treatment.

Clinical lesions. Clinical lesions will only be considered measurable when they are superficial (e.g., skin nodules and palpable lymph nodes). For the case of skin lesions, documentation by color photography, including a ruler to estimate the size of the lesion, is recommended. All lymph node measurements should be taken and recorded in metric notation, using a ruler, calipers, or digital measurement tool.

Conventional CT and MRI. These techniques should be performed with cuts of 10 mm or less in slice thickness contiguously. Spiral CT should be performed using a 5 mm contiguous reconstruction algorithm. This applies to tumors of the chest, abdomen, and pelvis.

11.3 Response Criteria

Response and progression will be evaluated in this study using the 2018 IWCLL criteria for CLL (Hallek et al., 2018). These criteria are detailed in Appendix B.

11.4 Response Review

Radiology will be centrally reviewed by the DF/HCC Tumor Imaging Metrics Core (TIMC).

12. DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

12.1 Data Reporting

12.1.1 Method

The Office of Data Quality (ODQ) will collect, manage, and perform quality checks on the data for this study.

12.1.2 Responsibility for Data Submission

Investigative sites within DF/HCC or DF/PCC are responsible for submitting data and/or data forms to the Office of Data Quality in accordance with DF/HCC SOPs.

12.2 Data Safety Monitoring

The DF/HCC Data and Safety Monitoring Committee (DSMC) will review and monitor toxicity and accrual data from this study. The committee is composed of clinical specialists with experience in oncology and who have no direct relationship with the study. Information that raises any questions about participant safety will be addressed with the Overall PI and study team.

The DSMC will review each protocol up to four times a year or more often if required to review toxicity and accrual data. Information to be provided to the committee may include: up-to-date participant accrual; current dose level information; DLT information; all grade 2 or higher unexpected adverse events that have been reported; summary of all deaths occurring with 30 days of intervention for Phase I or II protocols; for gene therapy protocols, summary of all deaths while being treated and during active follow-up; any response information; audit results, and a summary provided by the study team. Other information (e.g. scans, laboratory values) will be provided upon request.

12.3 Multicenter Guidelines

This protocol will adhere to the policies and requirements of the DF/HCC Multi-Center Data and Safety Monitoring Plan. The specific responsibilities of the Overall PI, Coordinating Center, and Participating Institutions and the procedures for auditing are presented in Appendix B.

- The Overall PI/Coordinating Center is responsible for distributing all IND Action Letters or Safety Reports to all participating institutions for submission to their individual IRBs for action as required.

- Mechanisms will be in place to ensure quality assurance, protocol compliance, and adverse event reporting at each site.

- Except in very unusual circumstances, each participating institution will order the study agent(s) directly from supplier. A participating site may order the agent(s) only after the initial IRB approval for the site has been forwarded to the Coordinating Center.

12.4 Collaborative Agreements Language

N/A

13. STATISTICAL CONSIDERATIONS

13.1 Study Design/Endpoints

This is an open-label, single-arm, phase 2 study to assess the rate of bone marrow MRD negative complete response after 15 cycles of therapy with acalabrutinib, venetoclax, and obinutuzumab (AVO) combination therapy in previously untreated CLL patients. Acalabrutinib will be given at 100mg orally twice daily as monotherapy for one month. This will be followed by acalabrutinib and obinutuzumab combination therapy for two months. Then, acalabrutinib, obinutuzumab, and venetoclax therapy will be given as a triplet for four months, followed by acalabrutinib and venetoclax combination therapy. Disease assessments will be performed at baseline and after cycles 3, 7, and 15, with the primary endpoint of MRD-negativity in the bone marrow assessed at the completion of cycle 15. Patients who are in complete remission with marrow and peripheral blood MRD-negativity at this time will have the option to discontinue acalabrutinib and venetoclax and will be monitored for recurrence. Patients who are in partial remission and/or are marrow or blood MRD-positive at cycle 15 will continue acalabrutinib and venetoclax therapy through cycle 24, at which point another disease assessment will take place. Patients who are MRD-negative in marrow and blood at the end of cycle 24 will have the option to discontinue acalabrutinib and venetoclax and will be monitored for recurrence as per the study calendar in section 10. Patients who are MRD-positive at will continue acalabrutinib and venetoclax indefinitely until disease progression or unacceptable toxicity.

Primary Endpoint: Rate of bone marrow minimal residual disease (MRD) negative complete responses (CRs/CRis) assessed after 15 cycles of therapy.

13.2 Sample Size, Accrual Rate and Study Duration

This is a one-stage study to determine the MRD-negative CR/CRi rate in the bone marrow after 15 cycles of AVO combination therapy. In a previous study of venetoclax plus rituximab in the relapsed/refractory population, the CR/CRi rate was 51%, and 80% of these responses were bone-marrow MRD-negative, for an MRD-negative CR/CRi rate of 41% (Seymour et al., 2017). Furthermore, 21 out of 25 CRs in this study occurred after 18 months of therapy, thus we will assume that ~80% of all marrow MRD-negative CRs will have occurred after 15 months of therapy here, leading to an expected marrow MRD-negative CR/CRi rate of 33%. Of note, this study was done in a relapsed/refractory population. Given that the depth of response is likely to be greater in the frontline setting, we are using an MRD-negative CR/CRi rate of 40% as the null hypothesis. With the addition of acalabrutinib, we hypothesize that the MRD-negative CR/CRi rate will be 60% or higher (alternative hypothesis). Initially, we planned to enroll 37 patients, and assuming approximately 1 screening failure this meant that 36 patients would be evaluable for the primary endpoint. This cohort has now completed enrollment and is referred to as cohort 1. We will now expand the study to include a second cohort of 35 patients with high-risk disease, defined as 17p deletion and/or *TP53* mutation (cohort 2). In cohort 2, we will have 80% power and one-sided type I error of 0.07 for an exact one-sample binomial test comparing an expected 55% response rate to the assumed 35% response rate for standard-of-care treatment in this high risk group. A patient will be considered unevaluable for response and replaced if the patient decides to withdraw from the study prior to the administration of the first dose of study drug. Based on the accrual rate of the study to date of 3 patients per month, we anticipate that accrual

of this study will complete within approximately 12 months from activation of cohort 2. Up to an additional 15 months of follow-up will be required on the last participant accrued to observe participant response after the 15th cycle of protocol therapy, for a total study duration of approximately 2.5 years. Evaluation of secondary endpoints, such as 3 year overall survival, will take up to three years after enrollment of the final subject.

Gender of subjects will not be used as a criterion for inclusion or exclusion in this study and there are no restrictions on the accrual of minorities. In the CLL patient population at DFCI, approximately 70% of patients were men and approximately 15% of patients were minorities. Based on this self-reported ethnicity and gender, the anticipated accrual in subgroups defined by gender and race is summarized in the table below.

Accrual Targets						
Ethnic Category	**Sex/Gender**					
	Females		**Males**		**Total**	
Hispanic or Latino	2	+	2	=	4	
Not Hispanic or Latino	19	+	48	=	67	
Ethnic Category: Total of all subjects	21 (A1)	+	50 (B1)	=	71	(C1)
Racial Category						
American Indian or Alaskan Native		+		=	0	
Asian	3	+	3	=	6	
Black or African American	3	+	4	=	7	
Native Hawaiian or other Pacific Islander		+		=		
White	15	+	43	=	58	
Racial Category: Total of all subjects	21 (A2)	+	50 (B2)	=	71	(C2)
	(A1 = A2)		(B1 = B2)		(C1 = C2)	

13.3 Stratification Factors

There will be no stratification of patients on this study.

13.4 Interim Monitoring Plan

Data safety and monitoring will be performed per DFCI guidelines. We first note that veneteoclax and obinutuzumab (K. Fischer et al., 2019; Fischer et al., 2017), acalabrutinib and obinutuzumab (Woyach, Awan, et al., 2017; Woyach et al., 2020), and venetoclax plus ibrutinib (a related BTK inhibitor) (Tam et al., 2018) combination therapy has been tested in patients without any concerning safety signals so far. If in the first 10 patients who receive at least one cycle of triple therapy with acalabrutinib, venetoclax, and obinutuzumab, 3 or more patients develop unexpected toxicity, this will trigger a consultation with the DSMC about whether to stop accrual. Unexpected toxicities would include the following events that occur during the first cycle of triple therapy, unless they are clearly due to extraneous causes: grade 4 or higher infusion reaction, grade 4 or higher infection, grade 4 or higher neutropenia, any cases of clinical

TLS, or other grade 3 or higher, clinically significant nonhematologic toxicity related to study treatment, except asymptomatic laboratory abnormalities or nausea/vomiting/diarrhea that improves with supportive care. With this design, the probability of triggering a consultation is 0.07 if the true but unknown rate of grade 3 or higher treatment related toxicity is 10%, 0.18 if the rate is 15%, 0.74 if the rate is 35%, and 0.83 if the rate is 40%.

13.5 Analysis of Primary Endpoints

Please refer to section 13.1.

13.6 Analysis of Secondary Endpoints

The analysis of secondary endpoints will be primarily descriptive, including rate of PR after 15 cycles of therapy, rate of CR/CRi after 15 cycles of therapy, rate of best overall response, best rate of CR/CRi, best rate of peripheral blood and bone marrow MRD, progression free survival (including 2-year and 3-year landmark rate), overall survival (including 2-year and 3-year landmark rate), rate of MRD negativity in the bone marrow at 8 and 24 cycles, rate of peripheral blood MRD-negativity at 8, 15, and 24 cycles, correlation between MRD-negativity in the peripheral blood and bone marrow, time to MRD-positive disease recurrence in the peripheral blood, time to clinical disease progression, rate of infusion related reactions, and rate of tumor lysis syndrome. A descriptive analysis of rates of therapy discontinuation after 15 cycles will be performed, with subjects grouped by reasons for discontinuation (e.g. achievement of MRD-negative CR, progressive disease, or intolerability). Rates of discontinuation of individual components of the regimen will also be included in this analysis. The Kaplan Meier method will be used to estimate the median progression-free survival time, progression-free survival, median overall survival time, and overall survival. Association between clinical response and established CLL prognostic factors (ZAP70, FISH cytogenetics, *IGHV* mutation status, *TP53* mutation status) will be tested using Fisher's exact test. A similar descriptive analysis will be performed for laboratory correlative studies of BH3 profiling and genomic markers such as *SF3B1, NOTCH1,* and *BCR/NFKB* pathway somatic mutations. If feasible, association between clinical response and correlative endpoints will be explored. A descriptive safety analysis with attention to adverse events, adverse events of special interests, and serious adverse events will also be performed. We also plan to perform the above efficacy and safety analyses on cohort 1, cohort 2 plus the patients with TP53 aberrant disease in cohort 1, and in the entire group of cohort 1 plus cohort 2

13.7 Reporting and Exclusions

Subjects who never start protocol therapy will be excluded from all analyses.

<u>Evaluation of Toxicity</u>

All participants who receive any amount of study therapy will be evaluable for toxicity from the time of their first treatment.

Evaluation of the Primary Efficacy Endpoint

All participants who receive at least one dose of at least one of the study drugs will be considered evaluable for the primary efficacy endpoint in an intent-to-treat analysis.

14. PUBLICATION PLAN

The results should be made public within approximately 24 months of reaching the end of the study. The end of the study is the time point at which the last data items are to be reported, or after the outcome data are sufficiently mature for analysis, as defined in the section on Sample Size, Accrual Rate and Study Duration. If a report is planned to be published in a peer-reviewed journal, then that initial release may be an abstract that meets the requirements of the International Committee of Medical Journal Editors. A full report of the outcomes should be made public no later than three (3) years after the end of the study.

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APPENDIX A: PERFORMANCE STATUS CRITERIA

ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
1	Symptoms, but ambulatory. 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).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active work.
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

APPENDIX B: CLL RESPONSE DEFINITION (HALLEK ET AL. 2018)

Complete Remission (CR):

CR requires all of the following criteria:

- Peripheral blood lymphocytes (evaluated by blood and differential count) below $4x10^9$/L (4000/μL).
- Absence of significant lymphadenopathy by physical examination and CT scan. Lymph nodes should be < 1.5cm in longest diameter.
- No hepatomegaly or splenomegaly by physical examination. A CT scan of the abdomen should be performed at response assessment and should show no evidence for lymphadenopathy and splenomegaly. A measurement of 13cm in cranio-caudal length has been accepted as a consensus definition for splenomegaly.
- Absence of disease-related constitutional symptoms.
- Blood counts above the following values: Neutrophils more than 1.5 x 10^9/L (1,500/μL) without need for exogenous growth factors, Platelets more than 100 x 10^9/L (100,000/μL) without need for exogenous growth factors, Hemoglobin more than 11.0 g/dL without red blood cell transfusion or need for exogenous erythropoietin.
- Bone marrow aspirate and biopsy must be at least normocellular for age, without evidence for typical CLL lymphocytes by morphological criteria. This evaluation is not based on a flow cytometry-based MRD assessment. When performing marrow biopsies in clinical trials, lymphoid nodules can be found that may reflect residual disease. These nodules may be recorded as "nodular PR". Immunohistochemistry may be performed to define whether the nodules are comprised primarily of T cells, of B cells other than CLL cells or of CLL cells. If nodules are not composed of CLL cells, a CR can be documented provided all other criteria are met.

Complete Remission with incomplete marrow recovery (CRi):

Some patients fulfill all the criteria for a CR, but have a persistent anemia, thrombocytopenia or neutropenia apparently unrelated to CLL, but related to drug toxicity. These patients should be considered as a different category of remission, CR with incomplete marrow recovery (CRi). For the definition of this category, CRi, the marrow evaluation should be performed with scrutiny and not show any clonal disease infiltrate.

Minimal Residual Disease

Patients who achieve CR or CRi will be assessed for minimal residual disease (MRD). Such patients who have fewer than 0.01% (10^{-4}) CLL cell percentage of leukocytes in the bone marrow, as assessed by four-color flow cytometry, will be considered to be MRD-negative.

Partial Remission

To define a partial remission, at least one parameter of group A and one parameter of group B need to improve, if previously abnormal (see Table 13 and sections 5.2.1-5). If only one parameter of both groups A and B was abnormal prior to therapy, only 1 needs to improve.

Constitutional symptoms persisting for more than 1 month should be recorded.

Group A
a. Decrease in the number of blood lymphocytes by 50% or more from the value before therapy.
b. Reduction in lymphadenopathy compared to baseline (by cross-sectional imaging scans in clinical trials or by palpation in general practice) as defined by:

- A decrease in lymph node size by 50% or more in the sum of the products of the same enlarged lymph nodes selected at baseline as assessed by imaging (up to 6 lymph nodes if possible) AND the sum of the longest diameters of the same enlarged lymph nodes selected at baseline as assessed by physical exam.
- No increase in any lymph node, and no new enlarged lymph node (diameter ≥ 1.5 cm). For small lymph nodes (longest diameter < 1.5 cm), an increase of < 25% is not considered to be significant.

c. A regression of ≥ 50% of the extent of enlargement of the spleen below the costal margin defined by palpation, or normalization in size. When assessed by CT, scan spleen size must have regressed by ≥ 50% in length beyond normal. A persistence of splenomegaly post therapy may have limited influence on outcome in CLL.
d. A regression of ≥ 50% of the extent of enlargement of the liver below the costal margin defined by palpation, or normalization in size. Given the impact of numerous medical conditions, liver size by physical examination or by CT scan is not a reliable measure of hepatic involvement by CLL and should only be counted if hepatomegaly is clearly attributable to lymphoid involvement.

Group B
a. Blood count should show one of the following:

- Platelet count >100 x 10^9/L (100 000/μL) or 50% improvement over baseline.
- Hemoglobin >110 g/L (11.0 g/dL), or 50% improvement over baseline without requiring red blood cell transfusions or exogenous erythropoietin.

<u>Progressive Disease</u>

Progressive disease during or after therapy is characterized by at least one of the following, when compared to nadir values:

- Lymphadenopathy. Progression of lymphadenopathy is often discovered by physical examination and should be recorded. For CT scans used to confirm progression or relapse of lymphadenopathy, progression is defined as:
    - Appearance of any new lesion such as enlarged lymph nodes (≥ 1.5 cm), splenomegaly, hepatomegaly or other organ infiltrates. Transient increases of lymph node size during treatment with novel inhibitors may occur and should not be counted as PD.
    - An increase by 50% or more in greatest determined diameter of any previous site (≥ 1.5 cm).
- An increase in the spleen size by 50% or more or the de novo appearance of splenomegaly. In the setting of splenomegaly, the splenic length must increase by ≥ 50%

of the extent of its prior increase beyond baseline (e.g., a 15-cm spleen must increase to ≥16 cm). If no prior splenomegaly was observed at baseline or if splenomegaly has resolved with treatment, the spleen must increase by at least 2 cm from baseline.

- An increase in the liver size of ≥ 50% of the extent enlargement of the liver below the costal margin defined by palpation, or the de novo appearance of hepatomegaly. Given the impact of numerous medical conditions, liver size by physical examination or by CT scan is not a reliable measure of hepatic involvement by CLL and should only be counted if hepatomegaly is clearly attributable to lymphoid involvement.
- An increase in the number of blood lymphocytes by 50% or more with at least 5.000 B lymphocytes per μL. (*Note:* because of the well-described lymphocyte redistribution phenomenon, any increase in lymphocyte count during acalabrutinib monotherapy or any increase during combination therapy in the setting of improvement of lymph nodes or cytopenias will not be considered disease progression)
- Transformation to a more aggressive histology (eg, Richter syndrome). This diagnosis must be established by lymph node biopsy.
- Occurrence of cytopenia (neutropenia, anemia, or thrombocytopenia) due to CLL.
    - During therapy, cytopenias cannot be used to define disease progression.
    - After therapy, the progression of any cytopenia (unrelated to autoimmune cytopenia), as documented by a decrease of Hb levels by more than 2 g/dL or to less than 10 g/dL, or by a decrease of platelet counts by ≥50% or to less than 100 000/μL, which occurs at least 3 months after treatment, defines disease progression, if the marrow biopsy is consistent with the cytopenia due to increased marrow infiltration of clonal CLL cells and is not considered a treatment related toxicity.

Stable Disease

Patients who have not achieved a CR or a PR, and who have not exhibited progressive disease, will be considered to have stable disease.

Treatment failure

Responses that should be considered clinically beneficial include CR and PR; all others (eg, stable disease, non-response, progressive disease, or death from any cause) should be rated as a treatment failure.

Time to Progression, progression-free survival, and overall survival

"Progression-free survival" (PFS) is defined as the interval between the first treatment day to the first sign of disease progression or death from any cause. Subjects without progression are censored at date of last disease evaluation. "Event-free survival" (EFS) is defined as the interval between the first treatment day to the first sign of disease progression or start of a new treatment or withdrawal from the trial due to toxicity or death (whichever occurs first). "Overall survival" (OS) is defined as the interval between the first treatment day to death. Living subjects are censored at date last known alive. "Time to next treatment" is defined as interval between the first treatment day until the patient starts an alternative therapy for progressive CLL.

Table 13: Response definition after treatment for CLL patients

GROUP	PARAMETER	CR	PR	PD	SD
A	Lymph nodes	None ≥ 1,5 cm	Decrease ≥ 50% (from baseline) [1)]	Increase ≥ 50% from baseline or from response	Change of -49% to +49%
	Liver and/or spleen size*	Spleen size < 13 cm; liver size normal	Decrease ≥ 50% (from baseline)	Increase ≥ 50% from baseline or from response	Change of -49% to +49%
	Constitutional symptoms	None	Any	Any	Any
	Circulating lymphocyte count	Normal	Decrease ≥ 50% from baseline	Increase ≥ 50% over baseline	Change of -49% to +49%
B	Platelet count	≥ 100.000/µl	≥ 100.000/µl or increase ≥ 50% over baseline	Decrease of ≥ 50% from baseline secondary to CLL	Change of -49 to +49%
	Hemoglobin	≥ 11,0 g/dl (untransfused and without erythropoietin)	≥ 11 g/dl or increase ≥ 50% over baseline	Decrease of ≥ 2 g/dl from baseline secondary to CLL	Increase < 11,0 g/dl or < 50% over baseline, or decrease < 2 g/dl
	Marrow	Normocellular, no CLL cells, no B-lymphoid nodules.	Presence of CLL cells, or of B-lymphoid nodules, or not done	Increase of CLL cells by ≥ 50% on successive biopsies	No change in marrow infiltrate

1) Sum of the products of 6 or less lymph nodes (as evaluated by CT scans and physical exam in clinical trials, or by physical exam in general practice).

CR, complete remission: all of the criteria have to be met; **PR, partial remission**: for a PR at least 1 of the parameters of group A and 1 parameter of group B need to improve if previously abnormal. If only one parameter of both groups A and B is abnormal prior to therapy, only 1 needs to improve. **PD, progressive disease**: at least one of the above criteria of group A or group B has to be met; **SD, stable disease**: all of the above criteria have to be met. Constitutional symptoms alone do not define PD.

*Spleen size is considered normal if < 13 cm. There is not firmly established, international consensus of the size of a normal liver; therefore, liver size should be evaluated by imaging and manual palpation in clinical trials and be recorded according to the definition used in a study protocol.

APPENDIX C: IW-CLL GRADING SCALE FOR HEMATOLOGIC TOXICITY IN CLL STUDIES

Grade[#]	Decrease in Platelets[*] or Hb[˚] (nadir) from Baseline Value, %	Absolute neutrophil count/μL (nadir)[§]
0	No change to 10%	≥2000
1	11% - 24%	≥1500 and <2000
2	25% - 49%	≥1000 and <1500
3	50% - 74%	≥ 500 and <1000
4	≥75%	<500

Grades: 1, mild; 2, moderate; 3 severe; 4 life-threatening; 5 fatal. Death occurring as a result of toxicity at any level of decrease from baseline will be recorded as grade 5.

* - Platelet counts must be below normal levels for grades 1-4. If, at any level of decrease the platelet count is <20,000/μL, this will be considered grade 4 toxicity, unless a severe or life-threatening decrease in the initial platelet count (e.g. 20,000/μL) was present at baseline, in which case the patient is not evaluable for toxicity referable to platelet counts.

˚ - Hb levels must be below normal levels for grades 1 to 4. Baseline and subsequent Hb determinations must be performed before any given transfusions. The use of erythropoietin is irrelevant for the grading of toxicity but should be documented.

§ - If the absolute neutrophil count (ANC) reaches < 1000/μL, it should be judged to be grade 3 toxicity. Other decreases in the white blood cell count, or in circulating granulocytes, are not to be considered, since a decrease in the white blood cell count is a desired therapeutic endpoint. A gradual decrease in granulocytes is not a reliable index in CLL for stepwise grading of toxicity. If the ANC was < 1000/μL prior to therapy, the patient is not evaluable for toxicity referable to the ANC. The use of growth factors such as G-CSF is not relevant to the grading of toxicity, but should be documented.

APPENDIX D: CAIRO-BISHOP DEFINITIONS OF LABORATORY AND CLINICAL TUMOR LYSIS SYNDROME (TLS)

Table 1. Cairo-Bishop Definition of Laboratory Tumor Lysis Syndrome‡

Element	Value	Change from Baseline
Uric Acid	≥ 476 µmol/L or 8 mg/dL	25% increase
Potassium	≥ 6.0 mmol/L or 6 mg/L	25% increase
Inorganic Phosphorus	≥1.45 mmol/L or 5mg/dL	25% increase
Calcium	≤ 1.75 mmol/Lor 7mg/dL	25% decrease

Table 2. Cairo-Bishop Clinical Tumor Lysis Syndrome Definition and Grading†

	Grade					
Complication	**0**	**1**	**2**	**3**	**4**	**5**
Creatinine*	≤ 1.5 × ULN	1.5 × ULN	1.5 – 3.0 × ULN	> 3.0 – 6.0 × ULN	> 6.0 × ULN	Death
Cardiac Arrhythmia*	None	Intervention not indicated	Non-urgent medical intervention indicated	Symptomatic and incompletely controlled medically or controlled with device (e.g., defibrillator)	Life-threatening (e.g., arrhythmia associated with CHF, hypotension, syncope, shock)	Death
Seizure*	None	–	One brief, generalized seizure; seizure(s) well controlled by anticonvulsants or infrequent focal motor seizures not interfering with ADL	Seizure in which consciousness is altered; poorly controlled seizure disorder; with breakthrough generalized seizures despite medical intervention	Seizure of any kind which are prolonged, repetitive or difficult to control (e.g., status epilepticus, intractable epilepsy)	Death

ULN = upper limit of normal; CHF = congestive heart failure; ADL = activities of daily living

‡ Two or more values from Table 1 must have the prespecified change to meet criteria for laboratory TLS. Either the absolute value or the percent change for each element can count towards one criterion.

* Not directly or probably attributable to therapeutic agent.

† Clinical tumor lysis syndrome is defined as laboratory tumor lysis syndrome and at least one clinical complication.

APPENDIX E: RECOMMENDATIONS FOR INITIAL MANAGEMENT OF ELECTROLYTE ABNORMALITIES AND PREVENTION OF TUMOR LYSIS SYNDROME

1. FIRST DOSE OF VENETOCLAX OR DOSE INCREASE

- Within the first 24 hours after either the first dose or dose increase, if any laboratory criteria below are met, the patient should be hospitalized for monitoring and the investigator notified. No additional venetoclax doses should be administered until resolution. A rapidly rising serum potassium level is a medical emergency.
- Nephrology (or acute dialysis service) must be consulted/contacted on admission (per institutional standards to ensure emergency dialysis is available).
- IV fluids (e.g., D5 1/2 normal saline) should be initiated at a rate of at least 1 mL/kg/h rounded to the nearest 10 mL (target 150 to 200 mL/h; not < 50 mL/h). Modification of fluid rate should also be considered for individuals with specific medical needs.
- Monitor for symptoms or signs of TLS (e.g., fever, chills, tachycardia, nausea, vomiting, diarrhea, diaphoresis, hypotension, muscle aches, weakness, paresthesias, mental status changes, confusion, and seizures). If any clinical features are observed, recheck potassium, phosphorus, uric acid, calcium, and creatinine within 1 hour.
- Vital signs should be taken at time of all blood draws or any intervention.
- The management recommendations below focus on the minimum initial responses required. If a diagnosis of TLS is established, ongoing intensive monitoring and multi-disciplinary management will be per institutional protocols.

Recommendations for Initial Management of Electrolyte Abnormalities and Prevention of Tumor Lysis Syndrome (TLS)

Abnormality	Management Recommendations
Hyperkalemia (including rapidly rising potassium)	
Potassium ≥ 0.5 mmol/L increase from prior value (even if potassium within normal limits [WNL])	• Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 hour. If further ≥ 0.2 mmol/L increase in potassium, but still < upper limit of normal (ULN), manage per potassium ≥ ULN. Otherwise recheck in 1 hour. • Resume per protocol testing if change in potassium is < 0.2 mmol/L, and potassium < ULN, and no other evidence of tumor lysis. • At discretion of investigator, may recheck prior to hospitalization. If stable or decreased, and still WNL, hospitalization is at the discretion of the investigator. Potassium, phosphorus, uric acid, calcium, and creatinine must be rechecked within 24 hours.
Potassium > upper limit of normal	• Perform immediate ECG and commence telemetry. • Nephrology notification with consideration of initiating dialysis • Administer Kayexalate 60 g (or Resonium A 60 g). • Administer furosemide 20 mg IV × 1. • Administer calcium gluconate 100 to 200 mg/kg IV slowly if there is ECG/telemetry evidence of life-threatening arrhythmias. • Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 hour. • If potassium < ULN 1 hour later, repeat potassium, phosphorus, uric acid, calcium, and creatinine 1, 2, and 4 hours later, if no other evidence of tumor lysis.
Potassium ≥ 6.0 mmol/L (6.0 mEq/L) and/or symptomatic (e.g., muscle cramps, weakness, paresthesias, nausea, vomiting, diarrhea)	• Perform immediate ECG and commence telemetry. • Nephrology assessment with consideration of initiating dialysis • Administer Kayexalate 60 g (or Resonium A 60 g). • Administer furosemide 20 mg IV × 1. • Administer insulin 0.1 U/kg IV + D25 2 mL/kg IV. • Administer sodium bicarbonate 1 to 2 mEq/kg IV push. – If sodium bicarbonate is used, rasburicase should not be used as this may exacerbate calcium phosphate precipitation. • Administer calcium gluconate 100 to 200 mg/kg IV slowly if there is ECG/telemetry evidence of life-threatening arrhythmias. Do not administer in same IV line as sodium bicarbonate. • Recheck potassium, phosphorus, uric acid, calcium, and creatinine every hour.

Abnormality	Management Recommendations
Hyperuricemia	
Uric acid ≥ 8.0 mg/dL (476 μmol/L)	• Consider rasburicase (dose per institutional guidelines). – If rasburicase is used, sodium bicarbonate should not be used as this may exacerbate calcium phosphate precipitation. • Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 hour.
Uric acid ≥ 10 mg/dL (595 μmol/L) OR Uric acid ≥ 8.0 mg/dL (476 μmol/L) with 25% increase and creatinine increase ≥ 0.3 mg/dL (≥ 0.027 mmol/L) from predose level	• Administer rasburicase (dose per institutional guidelines). – If rasburicase is used, sodium bicarbonate should not be used as this may exacerbate calcium phosphate precipitation. • Consult nephrology. • Recheck potassium, phosphorus, uric acid, calcium and creatinine in 1 hour. • If uric acid < 8.0 mg/dL 1 hour later, repeat potassium, phosphorus, uric acid, calcium, and creatinine 2 and 4 hours later, if no other evidence of tumor lysis.
Hypocalcemia	
Corrected calcium ≤ 7.0 mg/dL (1.75 mmol/L) AND Patient symptomatic (e.g., muscle cramps, hypotension, tetany, cardiac arrhythmias) in the presence of hypocalcemia	• Administer calcium gluconate 50 to 100 mg/kg IV slowly with ECG monitoring. • Telemetry. • Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 hour. • If calcium normalized 1 hour later, repeat potassium, phosphorus, uric acid, calcium, and creatinine 2 and 4 hours later, if no other evidence of tumor lysis. • Calculate corrected calcium and ionized calcium if albumin is low.
Hyperphosphatemia	
Phosphorus ≥ 5.0 mg/dL (1.615 mmol/L) with ≥ 0.5 mg/dL (0.16 mmol/L) increase	• Administer a phosphate binder (e.g., aluminum hydroxide, calcium carbonate, sevelamer hydroxide, or lanthanum carbonate). • Nephrology notification (dialysis required for phosphorus > 10 mg/dL) • Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 hour. • If phosphorus < 5.0 mg/dL 1 hour later, repeat potassium, phosphorus, uric acid, calcium, and creatinine 2 and 4 hours later, if no other evidence of tumor lysis.
Creatinine	
Increase ≥ 25% from baseline	• Start or increase rate of IV fluids. • Recheck potassium, phosphorus, uric acid, calcium, and creatinine in 1 to 2 hours.

IV = intravenous; ULN = upper limit of normal; WNL = within normal limits.

2. ONGOING DOSING OF VENETOCLAX

Management of electrolyte changes from last value at intervals > 24 hours after either the first dose or dose increase (e.g., 48 or 72 hours) are as below. Note: If the patient is hospitalized, no additional venetoclax doses should be administered until resolution. If the patient meets criteria for laboratory TLS (i.e. two or more element abnormalities), no additional venetoclax doses should be administered until resolution.

- For potassium
  - For any increase ≥ 1.0 mmol/L (1.0 mEq/L) OR for any potassium > upper limit of normal, admit the patient to the hospital if the patient is not already admitted. Refer to the table above, "Recommendations for Initial Management of Electrolyte Abnormalities and Prevention of Tumor Lysis Syndrome" for guidance regarding interventions.
  - If a smaller potassium increase is observed that does not meet the criteria for admission above, recheck potassium, phosphorus, uric acid, calcium, and creatinine in 24 hours and confirm no evidence of TLS prior to further venetoclax dosing.
- For uric acid, calcium, phosphorus, and creatinine changes listed in Appendix D Table 1, refer to the management guidelines in the table above "Recommendations for Initial Management of Electrolyte Abnormalities and Prevention of Tumor Lysis Syndrome)." These recommendations are mandatory if the patient meets criteria for laboratory TLS (2 or more element abnormalities) in Appendix D Table 1 above.

APPENDIX F: SAMPLE LIST OF EXCLUDED AND CAUTIONARY MEDICATIONS
(this is not an exhaustive list)

Excluded
warfarin
biologic agents
anticancer therapy (excluding adjuvant hormonal therapy)
other investigational agents
proton pump inhibitors
steroid therapy with anti-neoplastic intent

Strong CYP3A Inhibitors (Excluded)
atazanavir
cerivastatin
chloroquine
clarithromycin
conivaptan
eltrombopag
fluvoxamine
indinavir
itraconazole
ketoconazole
lopinavir
lovastatin
nefazodone
nelfinavir
pioglitazone
posaconazole
repaglinide
ritonavir
rosiglitazone
saquinavir
telaprevir
telithromycin
voriconazole

Strong CYP3A Inducers (Excluded)
carbamazepine
mitotane
phenobarbital
phenytoin
rifabutin
rifampin
St. John's wort

P-gp Inhibitors (Excluded)
amiodarone
azithromycin
captopril
carvedilol
cyclosporine
felodipine
quercetin
quinidine
ranolazine
ticagrelor

Narrow Therapeutic Index P-gp Substrates (Excluded)
digoxin
everolimus
sirolimus

Moderate CYP3A Inducers (Cautionary)
bosentan
efavirenz
etravirine
modafinil
nafcillin
oxcarbazepine
rifapentine

Moderate CYP3A Inhibitors (Cautionary)
ciprofloxacin
diltiazem
dronedarone
erythromycin
fluconazole
verapamil

Proton Pump Inhibitors
dexlansoprazole
esomeprazole
lansoprazole
omeprazole
pantoprazole
rabeprazole

H2-receptor anatagonists
cimetidine
famotidine
nizatidine
ranitidine

Antacids
aluminum hydroxide
magnesium hydroxide
calcium carbonate

APPENDIX G: ACALABRUTINIB DRUG DIARY

Study Participant Self-Administration Instructions

The study staff will explain how to take acalabrutinib, but these are points to remember:

1. Take the drug twice daily approximately 12 hours apart.
2. Take the drug with about 8 ounces of water. Acalabrutinib can be taken with or without meals.
3. Do not open, break, chew, crush, dissolve, or cut the tablets.
4. You may **NOT** consume: Grapefruit or grapefruit products, Seville oranges (including marmalade containing Seville oranges) or starfruit within the 3-day period prior to the first study drug administration and until the last day of treatment is completed due to possible CYP3A mediated metabolic interaction.
5. If you miss taking your dose at the scheduled time, it can be taken as soon as possible up to 3 hours after the scheduled time. If a dose is missed by more than 3 hours, it should be skipped and the next dose should be taken at its regularly scheduled time. Please make sure to record the reason for missing the dose. If you vomit a dose, do not take it again unless you can see the tablet.
6. Please remember to bring your study drug supply and drug diary with you to your clinic appointment. The clinic or research staff will instruct you when to take your study drug on those days.
7. The drug can be stored at room temperature.
8. Keep this drug out of the reach of children.
9. If you are taking this drug in combination with venetoclax, you may take them in any order.

Please call your doctor or research nurse before taking any new prescription or over-the-counter medications/supplements other than the study drugs or if any other questions arise.

Study Participant Self-Administration Diary for Acalabrutinib

Please record how many tablets you take and the time you take them and bring the completed diary as well as your study drug supply, including empty bottles, to every study visit. This will help us keep track of your study drug and how well you are tolerating it.

Participant Identifier: _____ Cycle Number: _____
Protocol #: _____
Study Doctor: _____
Study Nurse: _____
Contact Number: _____

You will take the following number of tablets each time (per dose) as listed in the table below:

Study Drug Name	# of tablets to take per dose	# of times each day
Acalabrutinib		

FOR STUDY TEAM USE ONLY	
Staff initials:	
Date dispensed:	Date returned:
# pills/caps/tabs dispensed:	# pills/caps/tabs returned:
# pills/caps/tabs that should have been taken:	
Discrepancy Notes:	

Day	Date	Number of Acalabrutinib Tablets	Time of First Dose each day	Number of Acalabrutinib Tablets	Time of Second Dose each day
1			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
2			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
3			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
4			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
5			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
6			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
7			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
8			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
9			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
10			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
11			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
12			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
13			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
14			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
15			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
16			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
17			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____

Day	Date	Number of Acalabrutinib Tablets	Time of First Dose each day	Number of Acalabrutinib Tablets	Time of Second Dose each day
18			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
19			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
20			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
21			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
22			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
23			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
24			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
25			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
26			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
27			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
28			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____		_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____

Participant/Caregiver Signature: _____ Date: _____

APPENDIX H: VENETOCLAX DRUG DIARY

Study Participant Self-Administration Instructions

The study staff will explain how to take venetoclax, but these are points to remember:

1. Take the drug once daily at the same time each day.
2. Take the drug with a meal and water. Tablets should be swallowed whole and not chewed or crushed.
3. You may **NOT** consume: Grapefruit or grapefruit products, Seville oranges (including marmalade containing Seville oranges) or starfruit within the 3-day period prior to the first study drug administration and until the last day of treatment is completed due to possible CYP3A mediated metabolic interaction.
4. If you miss taking your dose at the scheduled time, it can be taken as soon as possible up to 8 hours after the scheduled time. If a dose is missed by more than 8 hours, it should be skipped and the next dose should be taken at its regularly scheduled time. Please make sure to record the reason for missing the dose. If you vomit a dose, do not take it again unless you can see the capsule.
5. Please remember to bring your study drug supply and drug diary with you to your clinic appointment. The clinic or research staff will instruct you when to take your study drug on those days.
6. The drug can be stored at room temperature.
7. Keep this drug out of the reach of children.
8. If you are taking this drug in combination with acalabrutinib, you may take them in any order.

Please call your doctor or research nurse before taking any new prescription or over-the-counter medications/supplements other than the study drugs or if any other questions arise.

Study Participant Self-Administration Diary for Venetoclax

Please record how many tablets you take and the time you take them and bring the completed Diary as well as your study drug supply, including empty bottles, to every study visit. This will help us keep track of your study drug and how well you are tolerating it.

Participant Identifier: _____
Protocol #: _____
Study Doctor: _____
Study Nurse: _____
Contact Number: _____

Cycle Number: _____
Venetoclax Dose: _____ mg

FOR STUDY TEAM USE ONLY	
Staff initials:	
Date dispensed:	Date returned:
# pills/caps/tabs dispensed:	# pills/caps/tabs returned:
# pills/caps/tabs that should have been taken:	
Discrepancy Notes:	

Day	Date	Number of Venetoclax Tablets	Time of Dose each day
1			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
2			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
3			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
4			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
5			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
6			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
7			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
8			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
9			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
10			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
11			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
12			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
13			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
14			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
15			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
16			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
17			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
18			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
19			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
20			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
21			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
22			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
23			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____

Day	Date	Number of venetoclax Tablets	Time of Dose each day
24			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
25			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
26			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
27			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____
28			_____:_____ □ AM / □ PM □ Dose Not Taken Why:_____

Participant/Caregiver Signature: _____ Date: _____

APPENDIX I: SAMPLE REQUISITION FORM

18-226: A Phase 2 Study of Acalabrutinib, Venetoclax, and Obinutuzumab (AVO) for Initial Therapy of Chronic Lymphocytic Leukemia

PI: Matthew Davids, MD Sponsor: Dana-Farber Cancer Institute

Brown Lab Contact: Stacey Fernandes

Davids Lab Contact: Mary Collins

Stacey Fernandes
Principal Research Technician
DFCI CLL Center, J. Brown lab
1 Jimmy Fund WaySM 648
Boston, MA 02115, U.S.A.
Stacey_Fernandes@dfci.harvard.edu
617-632-5828 (phone)

Instructions: Complete one form for each time point collected. Send one copy with shipment and save another copy with subject documentation. Please email the lab contact and the DFCI Project Manager with the completed requisition form and FedEx tracking number.

NOTE: Do NOT ship samples on Fridays and day prior to a holiday.
Please give 72-hour notice of draw.

Institution:		**Subject ID:**	
Samples sent by:		**Study Timepoint:**	
E-mail:		**Subject Initials:**	
Phone:		**Subject MRN:**	

Sample type	Number of tubes	Date Drawn	Time Drawn:
WBC			
ALC			

LAB USE ONLY
Time & Date Received: _____
Received by: _____
Date Entered into Database: _____
Version 2.0 Active October 2014

APPENDIX J: DANA-FARBER/HARVARD CANCER CARE DATA SAFETY MONITORING PLAN

INTRODUCTION

The Dana-Farber/Harvard Cancer Center Multi-Center Data and Safety Monitoring Plan (DF/HCC DSMP) outlines the procedures for conducting a DF/HCC Multi-Center research protocol. The DF/HCC DSMP serves as a reference for any sites external to DF/HCC that are participating in a DF/HCC clinical trial.

Purpose

To establish standards that will ensure that a Dana-Farber/Harvard Cancer Center Multi-Center protocol will comply with Federal Regulations, Health Insurance Portability and Accountability Act (HIPAA) requirements and applicable DF/HCC Standard Operating Procedures..

Multi-Center Data and Safety Monitoring Plan Definitions

DF/HCC Multi-Center Protocol: A research protocol in which one or more outside institutions are collaborating with Dana-Farber/Harvard Cancer Center where a DF/HCC investigator is the sponsor. DF/HCC includes Dana-Farber/Partners Cancer Care (DF/PCC) Network Clinical Trial Affiliates.

Lead Institution: One of the Dana-Farber/Harvard Cancer Center consortium members (Dana-Farber Cancer Institute (DFCI), Massachusetts General Hospital (MGH), Beth Israel Deaconess Medical Center (BIDMC), Boston Children's Hospital (BCH), Brigham and Women's Hospital (BWH) responsible for the coordination, development, submission, and approval of a protocol as well as its subsequent amendments per the DFCI IRB and applicable regulatory guidelines (CTEP, Food and Drug Administration (FDA), Office of Biotechnology Activities (OBA) etc.). The Lead Institution is typically the home of the DF/HCC Sponsor. The Lead Institution also typically serves as the Coordinating Center for the DF/HCC Multi-Center Protocol.

DF/HCC Sponsor: The person sponsoring the submitted Multi-Center protocol who takes responsibility for initiation, management and conduct of the protocol at all research locations. In applicable protocols, the DF/HCC Sponsor will serve as the single liaison with any regulatory agencies. The DF/HCC Sponsor has ultimate authority over the protocol and is responsible for the conduct of the study at DF/HCC and all Participating Institutions. In most cases the DF/HCC Sponsor is the same person as the DF/HCC Overall Principal Investigator; however, both roles can be filled by two different people.

Participating Institution: An institution that is outside the DF/HCC and DF/PCC consortium that is collaborating with DF/HCC on a protocol where the sponsor is a DF/HCC Investigator. The Participating Institution acknowledges the DF/HCC Sponsor as having

the ultimate authority and responsibility for the overall conduct of the study.

Coordinating Center: The entity (i.e. Lead Institution, Medical Monitor, Contract Research Organization (CRO), etc) that provides administrative support to the DF/HCC Sponsor in order that he/she may fulfill the responsibilities outlined in the protocol document and DSMP, and as specified in applicable regulatory guidelines (i.e. CTEP Multi-Center Guidelines). In general, the Lead Institution is the Coordinating Center for the DF/HCC Multi-Center Protocol.

DF/HCC Office of Data Quality (ODQ): A group within DF/HCC responsible ensuring high-quality standards are used for data collection and the ongoing management of clinical trials, auditing, and data and safety monitoring. ODQ also coordinates quality assurance efforts related to multi-center clinical research.

DF/HCC Research Informatics for Operations (RIO): A group within DF/HCC responsible for providing a comprehensive data management platform for managing clinical trial data.

GENERAL ROLES AND RESPONSIBILITIES

For DF/HCC Multi-Center Protocols, the DF/HCC Sponsor, the Coordinating Center, and the Participating Institutions are expected to adhere to the following general responsibilities:

DF/HCC Sponsor

The DF/HCC Sponsor, Dr. Matthew Davids, will accept responsibility for all aspects of conducting a DF/HCC Multi-Center protocol which includes but is not limited to:

- Oversee the coordination, development, submission, and approval of the protocol as well as subsequent amendments.
- Ensure that the investigators, study team members, and Participating Institutions are qualified and appropriately resourced to conduct the protocol.
- Include the Multi-Center Data and Safety Monitoring Plan as an appendix to the protocol.
- Ensure all Participating Institutions are using the correct version of the protocol.
- Ensure that each participating investigator and study team member receives adequate protocol training (and/or a Site Initiation Visit prior to enrolling participants) and throughout trial's conduct as needed.
- Ensure the protocol will be provided to each participating site in a language understandable to all applicable site personnel when English is not the primary language.
- Monitor progress and overall conduct of the study at all Participating Institutions.

- Ensure all DFCI Institutional Review Board (IRB), DF/HCC and other applicable (i.e. FDA) reporting requirements are met.
- Review data and maintain timely submission of data for study analysis.
- Act as the single liaison with FDA (investigator-held IND trials).
- Ensure compliance with all requirements as set forth in the Code of Federal Regulations, applicable DF/HCC requirements, HIPAA requirements, and the approved protocol.
- Commit to the provision that the protocol will not be rewritten or modified by anyone other than the DF/HCC Sponsor.
- Identify and qualify Participating Institutions and obtain accrual commitments prior to extending the protocol to that site.
- Monitor accrual and address Participating Institutions that are not meeting their accrual requirements.

Coordinating Center

The general responsibilities of the Coordinating Center may include but are not limited to:

- Assist in protocol development.
- Maintain FDA correspondence.
- Review registration materials for eligibility and register participants from Participating Institutions in the DF/HCC clinical trial management system (CTMS).
- Distribute protocol and informed consent document updates to Participating Institutions as needed.
- Oversee the data collection process from Participating Institutions.
- Maintain documentation of Serious Adverse Event (SAE) reports and deviations/violation submitted by Participating Institutions and provide to the DF/HCC Sponsor for timely review and submission to the DFCI IRB, as necessary.
- Distribute serious adverse events reported to the DF/HCC Sponsor that fall under the DFCI IRB Adverse Event Reporting pPolicy to all Participating Institutions.
- Provide Participating Institutions with information regarding DF/HCC requirements that they will be expected to comply with.

- Carry out plan to monitor Participating Institutions either by on-site or remote monitoring.
- Maintain Regulatory documents of all Participating Institutions which includes but is not limited to the following: local IRB approvals/notifications from all Participating Institutions, confirmation of Federalwide Assurances (FWAs) for all sites, all SAE submissions, Screening Logs for all sites, IRB approved consents for all sites
- Conduct regular communications with all Participating Institutions (conference calls, emails, etc) and maintain documentation all relevant communications.

Participating Institution

Each Participating Institution is expected to comply with all applicable federal regulations and DF/HCC requirements, the protocol and HIPAA requirements.

The general responsibilities for each Participating Institution may include but are not limited to:

- Document the delegation of research specific activities to study personnel.
- Commit to the accrual of participants to the protocol.
- Submit protocol and/or amendments to their local IRB of record.
- Maintain regulatory files as per sponsor requirements.
- Provide the Coordinating Center with regulatory documents or source documents as requested.
- Participate in protocol training prior to enrolling participants and throughout the trial as required (i.e. teleconferences).
- Update Coordinating Center with research staff changes on a timely basis.
- Register participants through the Coordinating Center prior to beginning research related activities.
- Submit Serious Adverse Event (SAE) reports to local IRB per institutional requirements and to the Coordinating Center, in accordance with DF/HCC requirements.
- Submit protocol deviations and violations to local IRB per institutional requirements and to the DF/HCC Sponsor in accordance with DF/HCC requirements.
- Order, store and dispense investigational agents and/or other protocol mandated drugs per federal guidelines and protocol requirements.

- Have office space, office equipment, and internet access that meet HIPAA standards.
- Participate in any quality assurance activities and meet with monitors or auditors at the conclusion of a visit to review findings.
- Promptly provide follow-up and/or corrective action plans for any monitoring queries or audit findings.

DF/HCC REQUIREMENTS FOR MULTI-CENTER PROTOCOLS

The following section will clarify DF/HCC Requirements and further detail the expectations for participating in a DF/HCC Multi-Center protocol.

Protocol Distribution

The Coordinating Center will distribute the final DFCI IRB approved protocol and any subsequent amended protocols to all Participating Institutions.

Protocol Revisions and Closures

The Participating Institutions will receive notification of protocol revisions and closures from the Coordinating Center. It is the individual Participating Institution's responsibility to notify its IRB of these revisions.

- **Non life-threatening revisions:** Participating Institutions will receive written notification of protocol revisions regarding non life-threatening events from the Coordinating Center. Non-life-threatening protocol revisions must be IRB approved and implemented within 90 days from receipt of the notification.

- **Revisions for life-threatening causes:** Participating Institutions will receive immediate notification from the Coordinating Center concerning protocol revisions required to protect lives with follow-up by fax, mail, e-mail, etc. Life-threatening protocol revisions will be implemented immediately followed by IRB request for approval.

- **Protocol closures and temporary holds:** Participating Institutions will receive notification of protocol closures and temporary holds from the Coordinating Center. Closures and holds will be effective immediately. In addition, the Coordinating Center, will update the Participating Institutions on an ongoing basis about protocol accrual data so that they will be aware of imminent protocol closures.

Informed Consent Requirements

The DF/HCC approved informed consent document will serve as a template for the informed consent for Participating Institutions. The Participating Institution consent form must follow the consent template as closely as possible and should adhere to specifications outlined in the DF/HCC Guidance Document on Model Consent Language for Investigator-Sponsored Multi-Center Trials. This document will be provided separately to each Participating Institution upon request.

Participating Institutions are to send their version of the informed consent document and HIPAA authorization, if a separate document, to the Coordinating Center for review and approval prior to submission to their local IRB. The approved consent form must also be submitted to the Coordinating Center after approval by the local IRB for all consent versions.

The Principal Investigator (PI) at each Participating Institution will identify the physician members of the study team who will be obtaining consent and signing the consent form for therapeutic protocols. Participating institutions must follow the DF/HCC requirement that for all interventional drug, biologic, or device research, only attending physicians may obtain initial informed consent and any re-consent that requires a full revised consent form.

IRB Documentation

The following must be on file with the Coordinating Center:

- Initial approval letter of the Participating Institution's IRB.
- Copy of the Informed Consent Form(s) approved by the Participating Institution's IRB.
- Participating Institution's IRB approval for all amendments.
- Annual approval letters by the Participating Institution's IRB.

IRB Re-Approval

Verification of IRB re-approval from the Participating Institutions is required in order to continue research activities. There is no grace period for continuing approvals.

The Coordinating Center will not register participants if a re-approval letter is not received from the Participating Institution on or before the anniversary of the previous approval date.

Participant Confidentiality and Authorization Statement

In 1996, congress passed the first federal law covering the privacy of health information known as the Health Insurance Portability and Accountability Act (HIPPA). Any information, related to the physical or mental health of an individual is called Protected Health Information (PHI). HIPAA outlines how and under what circumstances PHI can be used or disclosed.

In order for covered entities to use or disclose protected health information during the course of a study, the study participant must sign an authorization statement. This authorization statement may or may not be separate from the informed consent document. The Coordinating Center, with the approval from the DFCI IRB, will provide a consent template, with information regarding authorization for the disclosure of protected health information.

The DF/HCC Sponsor will use all efforts to limit its use of protected health information in its trials. However, because of the nature of these trials, certain protected health information must be collected. DF/HCC has chosen to use authorizations, signed by the participant in the trial, rather than limited data sets with data use agreements.

DF/HCC Multi-Center Protocol Confidentiality

All documents, investigative reports, or information relating to the participant are strictly confidential. Whenever reasonably feasible, any participant specific reports (i.e. Pathology Reports, MRI Reports, Operative Reports, etc.) submitted to the Coordinating Center should be de-identified. It is recommended that the assigned protocol case number (as described below) be used for all participant specific documents. Participant initials may be included or retained for cross verification of identification.

DF/HCC Multi-Center Protocol Registration Policy

Participant Registration and Randomization

To register a participant, the following documents should be completed by the Participating Institution and faxed or e-mailed to the Coordinating Center to the attention of Samantha Pazienza: Samantha_Pazienza@dfci.harvard.edu

- Copy of required laboratory tests
- Signed informed consent document
- HIPAA authorization form (if separate from the informed consent document)
- Completed Eligibility Checklist

The Coordinating Center will review the submitted documents in order to verify eligibility and consent. To complete the registration process, the Coordinating Center will:

- Register the participant on the study with the DF/HCC Clinical Trial Management System (CTMS).
- Upon receiving confirmation of registration, the Coordinating Center will inform the Participating Institution and provide the study specific participant case number, and, if applicable, assigned treatment and/or dose level.

Treatment or other protocol-specific interventions may not begin without confirmation from the Coordinating Center that the participant has been registered.

Initiation of Therapy

Participants must be registered with the DF/HCC CTMS before the initiation of treatment or other protocol-specific interventions. Treatment and other protocol-specific interventions may not be initiated until the Participating Institution receives confirmation of the participant's registration from the Coordinating Center. The DF/HCC Sponsor and DFCI IRB must be notified of any violations to this policy.

Eligibility Exceptions

No exceptions to the eligibility requirements for a protocol without DFCI IRB approval will be permitted. All Participating Institutions are required to fully comply with this requirement. The process for requesting an eligibility exception is defined below.

DF/HCC Protocol Case Number

At the time of registration, the following identifiers are required for all subjects: initials, date of birth, gender, race and ethnicity. Once eligibility has been established and the participant successfully registered, the participant is assigned a unique protocol case number. Participating Institutions should submit all de-identified subsequent communication and documents to the Coordinating Center, using this case number to identify the subject.

Protocol Deviations, Exceptions and Violations

Federal Regulations require an IRB to review proposed changes in a research activity to ensure that researchers do not initiate changes in approved research without IRB review and approval, except when necessary to eliminate apparent immediate hazards to the participant. DF/HCC requires all departures from the defined procedures set forth in the IRB approved protocol to be reported to the DF/HCC Sponsor, who in turn is responsible for reporting to the DFCI IRB.

For reporting purposes, DF/HCC uses the terms "violation", "deviation" and "exception" to describe departures from a protocol. All Participating Institutions must adhere to these requirements for reporting to the DF/HCC Sponsor and will follow their institutional policy for reporting to their local IRB.

Definitions

Protocol Deviation: Any departure from the defined procedures set forth in the IRB-approved protocol which is *prospectively approved* prior to its implementation.

Protocol Exception: Any protocol deviation that relates to the eligibility criteria, e.g. enrollment of a participant who does not meet all inclusion/exclusion criteria.

Protocol Violation: Any protocol departure that was not *prospectively approved* by the IRB prior to its initiation or implementation.

Reporting Procedures

DF/HCC Sponsor: is responsible for ensuring that clear documentation is available in the medical record and/or regulatory documents to describe all protocol exceptions, deviations and violations. The DF/HCC Sponsor will also be responsible for ensuring that all protocol violations/deviations are promptly reported per DFCI IRB guidelines.

Participating Institutions: Protocol deviations require prospective approval from the DFCI IRB. The Participating Institution must submit the deviation request to the Coordinating Center who will then submit the deviation request to the DFCI IRB. Upon DFCI IRB approval the deviation is submitted to the Participating Institution IRB, per institutional policy. A copy of the Participating Institution's IRB report and determination will be forwarded to the Coordinating Center within 10 business days after the original submission. The deviation may not be implemented without all required approvals.

All protocol violations must be sent to the Coordinating Center in a timely manner. The Coordinating Center will provide training for the requirements for the reporting of violations.

Coordinating Center: Upon receipt of the violation/deviation report from the Participating Institution, the Coordinating Center will submit the report to the DF/HCC Sponsor for review. Subsequently, the Participating Institution's IRB violation/deviation report will be submitted to the DFCI IRB for review per DFCI IRB reporting guidelines.

Safety Assessments and Toxicity Monitoring

The study teams at all participating institutions are responsible for protecting the safety, rights and well-being of study participants. Recording and reporting of adverse events that occur during the course of a study help ensure the continuing safety of study participants.

All participants receiving investigational agents and/or other protocol mandated therapy will be evaluated for safety. The safety parameters include all laboratory tests and hematological abnormalities, physical examination findings, and spontaneous reports of adverse events reported by participants. All toxicities encountered during the study will be evaluated according to the NCI criteria specified in the protocol. Life-threatening toxicities must be reported immediately to the DF/HCC Sponsor via the Coordinating Center.

Additional safety assessments and toxicity monitoring will be outlined in the protocol.

Guidelines for Reporting Serious Adverse Events

Guidelines for reporting Adverse Events (AEs) and Serious Adverse Events (SAEs) are detailed in protocol section 7.4.

Participating Institutions must report the SAEs to the DF/HCC Sponsor and the Coordinating Center following the DFCI IRB Adverse Event Reporting Policy IRB of record's Adverse Event Reporting Policy.

The Coordinating Center will maintain documentation of all Participating Institution Adverse Event reports and be responsible for communicating to all participating investigators, any observations reportable under the DFCI IRB Reporting Requirements. Participating Institutions will review and submit to their IRB according to their institutional policies and procedures

Guidelines for Processing IND Safety Reports

The DF/HCC Sponsor will review all IND Safety Reports and ensure that all IND Safety Reports are distributed to the Participating Institutions. Participating Institutions will review/submit to the IRB according to their institutional policies and procedures.

Data Management

DF/HCC RIO develops case report forms (CRF/eCRFs), for use with the protocol. These forms are designed to collect data for each study. DF/HCC RIO provides a web based training for all eCRF users.

Data Forms Review

Data submissions are monitored for timeliness and completeness of submission. If study forms are received with missing or questionable data, the submitting institution will receive a written or electronic query from the DF/HCC Office of Data Quality, Coordinating Center, or designee.

Responses to all queries should be completed and submitted within about 14 calendar days.

Responses may be returned on the written query or on an amended paper case report form, or in the case of electronic queries, within the electronic data capture (eDC) system. In the case of a written query for data submitted on a paper case report form, the query must be attached to the specific data being re-submitted in response.

If study forms are not submitted on schedule, the Participating Institution will periodically receive a Missing Form Report from the Coordinating Center noting the missing forms.

REQUISITIONING INVESTIGATIONAL DRUG

The ordering of investigational agent is specified in the protocol section 8.
Participating Institutions should order their own agent regardless of the supplier.

MONITORING: QUALITY CONTROL

The quality control process for a clinical trial requires verification of protocol compliance and data accuracy. The Coordinating Center, with the aid of the DF/HCC Office of Data Quality, provides quality control oversight for the protocol.

Ongoing Monitoring of Protocol Compliance

The Participating Institutions may be required to submit participant source documents to the Coordinating Center for monitoring. Participating Institution may also be subject to on-site monitoring conducted by the Coordinating Center.

The Coordinating Center will implement ongoing monitoring activities to ensure that Participating Institutions are complying with regulatory and protocol requirements, data quality, and participant safety. Monitoring practices may include but are not limited to source data verification, and review and analysis of eligibility requirements, informed consent procedures, adverse events and all associated documentation, review of study drug administration/treatment, regulatory files, protocol departures reporting, pharmacy records, response assessments, and data management.

Participating institutions will be required to participate in monthly Coordinating Center initiated teleconferences. Emails highlighting overall protocol progress and important announcements will be distributed as needed.

Remote Monitoring: Participating Institutions will be required to forward de-identified copies of participants' medical record and source documents to the Coordinating Center to aid in source data verification.

and

On-Site Monitoring: Source documentation verification (SDV) may be conducted by having access to participants' complete medical record and source documents.

Monitoring Reports

The DF/HCC Sponsor will review all monitoring reports to ensure protocol compliance. The DF/HCC Sponsor may increase the monitoring activities at Participating Institutions that are unable to comply with the protocol, DF/HCC Sponsor requirements or federal and local regulations.

Accrual Monitoring

Prior to extending a protocol to an external site, the DF/HCC Sponsor will establish accrual requirements for each participating institution. Accrual will be monitored for each participating institution by the DF/HCC Sponsor or designee. Sites that are not meeting their accrual expectations may be subject to termination. Participating institutions are required to accrue 3 patients per site per year.

AUDITING: QUALITY ASSURANCE

Auditing is a method of Quality Assurance and involves the systematic and independent examination of all trial related activities and documents. Audits determine if evaluated activities were appropriately conducted and whether data was generated, recorded and analyzed, and accurately reported per the protocol, applicable Policies, and the Code of Federal Regulations (CFR).

DF/HCC Internal Audits

All Participating Institutions are subject to audit by the DF/HCC Office of Data Quality (ODQ). Typically, approximately 3-4 participants would be audited at the site over a 2-day period. If violations which impact participant safety or the integrity of the study are found, more participant records may be audited.

Audit Notifications

It is the Participating Institution's responsibility to notify the Coordinating Center of all external audits or inspections (e.g., FDA, EMA, NCI) that involve this protocol. All institutions will forward a copy of final audit and/or re-audit reports and corrective action plans (if applicable) to the Coordinating Center, within 12 weeks after the audit date.

Audit Reports

The DF/HCC Sponsor will review all final audit reports and corrective action plans, if applicable. The Coordinating Center, must forward any reports to the DF/HCC ODQ per DF/HCC policy for review by the DF/HCC Audit Committee. For unacceptable audits, the DF/HCC Audit Committee would forward the final audit report and corrective action plan to the DFCI IRB as applicable.

Participating Institution Performance

The DF/HCC Sponsor and the IRB of record are charged with considering the totality of an institution's performance in considering institutional participation in the protocol.

Participating Institutions that fail to meet the performance goals of accrual, submission of timely and accurate data, adherence to protocol requirements, and compliance with state and federal regulations, may be recommended for a six-month probation period. Such institutions must respond with a corrective action plan and must demonstrate during the probation period that deficiencies have been corrected, as evidenced by the improved

performance measures. Participating Institutions that fail to demonstrate significant improvement will be considered by the DF/HCC Sponsor for revocation of participation. A DF/HCC Sponsor and/or the DFCI IRB may terminate a site's participation if it is determined that a site is not fulfilling its responsibilities as described above.