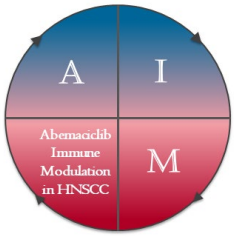

Immune Modulation by Abemaciclib in HPV-Negative Head and Neck Squamous Cell Carcinoma (HNSCC): A Phase II Window Trial

Short Title: Abemaciclib Immune Modulation (AIM) Trial

UA IRB Protocol #: 1906702171
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Phase: II
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Study Drug: Abemaciclib
National Clinical Trial (NCT): NCT04169074

COORDINATING INVESTIGATOR: Ricklie Julian, MD
University of Arizona Cancer Center
1515 N. Campbell Ave.
Tucson, AZ 85724
Phone: 520-694-2873
Email: rickliej@arizona.edu

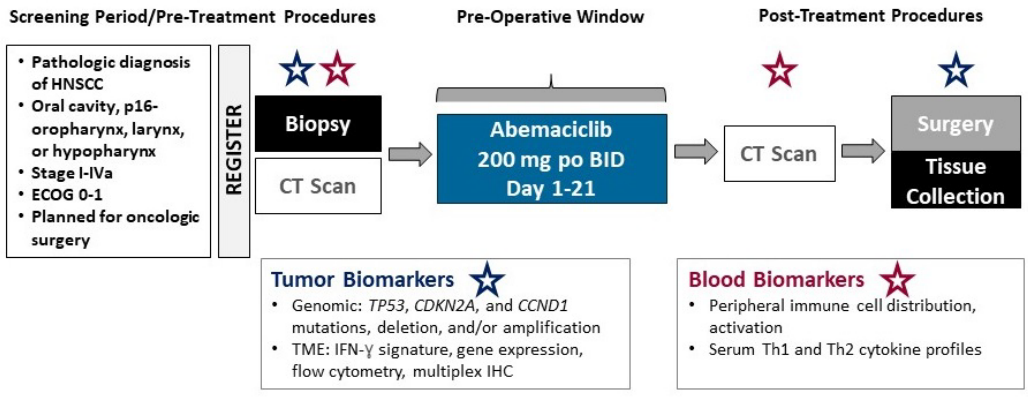
Site Investigator : Julie Bauman, MD, MPH
George Washington Cancer Center
2150 Pennsylvania Ave, NW Washington, DC 20037

Source of Support: Eli Lilly and Company

Study Monitoring Entity: University of Arizona Cancer Center Data and Safety Monitoring Board

STUDY OVERVIEW	
Title	Immune Modulation by Abemaciclib in HPV-Negative Head and Neck Squamous Cell Carcinoma (HNSCC): A Phase II Window Trial
Patient population	Patients with de novo HPV-negative HNSCC who are planned for oncologic surgery.
Rationale for Study	Current guidelines for the treatment of HNSCC incorporate HPV as a major stratification marker. Patients with HPV-negative tumors represent approximately 80% of HNSCC diagnoses worldwide, and they carry a significantly worse overall prognosis compared to their HPV-positive counterparts. New therapeutic strategies for HPV-negative HNSCC represent a major unmet clinical need. Genetic and molecular studies have revealed distinct genetic and phenotypic differences between HPV-negative and HPV-positive HNSCC tumors. HPV-negative tumors harbor a high mutational burden in genes associated with cellular growth, proliferation, and survival, such as *CCND1, PIK3CA*, *PTEN*, and *RAS* and tumor suppressor genes such as *TP53* and *CDKN2A.* In particular, loss of CDKN2A (p16) function leads to unbridled activity of the cyclin dependent kinase 4/6 (CDK4/6)-cyclin D1 pathway, driving oncogenic cellular proliferation. The tumor-intrinsic rationale for the development of CDK 4/6 inhibitors in HPV-negative HNSCC is strong. Moreover, increasing evidence points to unbridled CDK 4/6 signaling as a source of immune suppression in the tumor microenvironment (TME). CDK 4/6 inhibition may reverse immunosuppression, resulting in a T cell-inflamed phenotype.
Primary Objective	To evaluate the clinical activity of abemaciclib in patients with operable, HPV-negative HNSCC as measured by quantitative change in tumor size (ΔT)[1] following 10-21 (+7) days of neoadjuvant exposure.
Secondary Objective	• To describe the safety and tolerability of neoadjuvant exposure to abemaciclib in accordance with NCI Common Terminology Criteria for Adverse Events (CTCAE) v5.0.

Exploratory Objectives	• To evaluate tumor-intrinsic genetic biomarkers associated with ΔT, including somatic genetic or epigenetic alterations in *CCND1, CDKN2A,* and *TP53*
	• To evaluate baseline and pharmacodynamic biomarkers within the TME associated with ΔT, including:
	  ○ IFN-ɣ gene expression signature in baseline and post-treatment tumor biopsies. ***The primary biomarker hypothesis is that abemaciclib will increase the proportion of tumors that are T-cell inflamed.***
	  ○ The distribution of tumor-infiltrating immune cell subtypes and their activation status, including lymphocytes and myeloid-derived stem cells, as measured by gene expression, flow cytometry, and multiplex immunohistochemistry (IHC).
	• To evaluate the distribution of peripheral immune cell subtypes and their activation status, and how this is altered by abemaciclib.

	• To evaluate serum Th1 and Th2 cytokine profiles, and how they are altered by abemaciclib. • To evaluate tumor-intrinsic molecular mediators of response and resistance to abemaciclib in baseline and post-treatment tumor biopsies, including expression of CDKN2A (p16), CCND1 (cyclin D1), and pRB (retinoblastoma). • To evaluate the anti-proliferative activity of abemaciclib as measured by change in the proliferative index (ΔKi67) in pre- and post-treatment tumor specimens. •
Study Design	This is a Phase II window trial to assess the clinical activity and immune biomarker modulation of 10-21 days of neoadjuvant abemaciclib in patients with HPV-negative HNSCC who are planned for oncologic surgery, with pre-clinically informed genetic and immune biomarker correlatives. In the window-of-opportunity clinical trial, patients planned for oncologic surgery are briefly exposed to a novel cancer agent in the window between diagnostic biopsy and definitive surgery. Paired, pre- and post-surgical tumor specimens permit *ex vivo* analysis of target modulation in both tumor and the tumor microenvironment (TME) – providing insight into mechanism of action and paving the way for rigorous companion biomarker development. Clinical activity is assessed by quantitative change in tumor size (ΔT), which is correlated to hypothesis-driven genomic and immune biomarkers of interest. We hypothesize that abemaciclib will significantly reduce tumor burden as measured by ΔT. Further, we will test the primary biomarker hypothesis that the clinical activity of abemaciclib is associated with an increased proportion of tumors that are T-cell inflamed. We will evaluate tumor-intrinsic and TME hypotheses in specific genetic contexts, including tumors with specific classes of *TP53* mutations, p16 loss, and/or *CCND1* amplification. Dr. Julie E. Bauman's group has successfully conducted and published three window trials in HNSCC [1, 61, 72].
Study Schema	
Number of patients	Projected accrual is 20 patients, for a total of 14 biomarker-evaluable patients.
Study Drug	Abemaciclib is an orally available CDK inhibitor that targets the CDK4 (cyclin D1) and CDK6 (cyclin D3) cell cycle pathway. Abemaciclib specifically inhibits

	CDK4 and 6, thereby inhibiting retinoblastoma (Rb) protein phosphorylation in early G1. Inhibition of Rb phosphorylation prevents CDK-mediated G1-S phase transition, thereby arresting the cell cycle in the G1 phase, suppressing DNA synthesis and inhibiting cancer cell growth.
Duration of Therapy	Treatment will consist of a single neoadjuvant cycle of 10-21 days (+7 days). Abemaciclib will be administered from days 1-21. Abemaciclib may be continued for an additional 7 days, or up to 28 days, for delays in planned surgery.
Duration of Follow up	4-weeks post-operation, or as deemed clinically necessary.
Duration of study	The study will accrue over 24 months. Clinical and biomarker analyses will be completed 36 months after the study opens to accrual. *Note: Accrual to this study has been significantly affected by the COVID-19 pandemic. Thus, the accrual time will be extended to 48 months from the time the study opened and will close 60 months after the study opens.*
Study Drug Dose and Route	Abemaciclib 200 mg PO twice daily on Days 1-21 (+7 days).
Safety Assessments	The safety of this window intervention will be reported descriptively, including tabulation of toxicities according to NCI CTCAE v5.0.
Efficacy Assessments	The primary endpoint is quantitative change in tumor size (ΔT). ΔT will be treated as a continuous variable, based upon the percent change in RECIST-determined target lesions pre- and post-treatment as previously described by Dr. Julie E. Bauman's group[1].

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1 Introduction

1.1 Background and Rationale

1.1.1 HPV-Negative Head and Neck Squamous Cell Carcinoma

Head and neck squamous cell carcinoma (HNSCC) is the sixth leading incident cancer worldwide [2]. In the United States, the disease burden for 2016 was estimated at 62,000 new cases and 12,500 deaths [3]. Among the 700,000 cases expected globally in 2017, the majority (80%) is caused by environmental carcinogens: specifically tobacco, alcohol, and the areca nut. In the United States, Western Europe, and Australia, an increasing proportion of HNSCC is caused by high risk human papillomavirus (HPV), an etiology associated with a much improved prognosis due to enhanced sensitivity to standard therapeutic modalities. For HPV-negative HNSCC, the 5-year overall survival (OS) after curative-intent treatment remains 30-50% despite advances in multimodal therapy over the past three decades. Poor outcomes persist in spite of therapeutic intensification with altered radiotherapy fractionation, multi-drug induction chemotherapy, or integration of EGFR-targeted monoclonal antibodies (mAb). New therapeutic strategies for HPV-negative HNSCC represent a major unmet clinical need.

The Genetic Landscape of HPV-Negative HNSCC. The transformation of oral epithelial cells by environmental carcinogens is based upon the principle of multistep carcinogenesis, where the accumulation of structural and functional DNA damage drives stepwise clonal evolution. Loss of heterozygosity (LOH), also known as allelic imbalance, has been quantified across the histopathologic spectrum of hyperplasia, dysplasia and carcinoma in situ in order to model malignant transformation. In essence, environmentally-induced HNSCC is characterized by loss of function of tumor suppressor genes. A central, early molecular event is LOH at 9p21, where the gene *CDKN2A* encodes p16, a tumor suppressor regulating cell cycle traversal from G1 to S-phase. Mutation of *TP53* is a later molecular event. The p53 tumor suppressor is considered the guardian of the genome, as it triggers cell cycle arrest and apoptosis upon sensing DNA damage. Mutations of *CDKN2A* and *TP53* are the most common genetic alterations in HPV-negative HNSCC, observed in 57% and 84% respectively. Moreover, p16 expression is lost due to epigenetic silencing in an additional 30%, leading to the histopathologic classification of HPV-negative HNSCC as “p16-negative.” [4-7].

Most *TP53* mutations found in human cancers are missense mutations that encode mutant forms of p53. Approximately 30% of HPV-negative HNSCC harbors a mutant p53 that can transform cells as a result of acquired gain-of-function (GOF) activities [8]. Patients with HPV-negative HNSCC harboring GOF *TP53* mutations demonstrate increased extranodal extension, decreased OS, and resistance to platinum chemotherapy [9-11]. In line with these clinical observations, we generated mouse models that demonstrated that the p53 GOF mutation $p53^{R172H}$ (equivalent to the human hot spot $p53^{R175H}$) promotes SCC development, metastasis and resistance to platinum chemotherapy and immune checkpoint inhibitors [12-14]. These findings suggest that GOF p53 mutations serve not only as oncogenes driving tumor progression and resistance to chemotherapy, but also induce an immunosuppressed tumor microenvironment resistant to immunotherapy.

In vitro studies have shown that HPV-negative squamous cell carcinoma lines behave differently than identical cell lines that express HPV. Nagel *et al* found that HPV-positive and negative cells are equally sensitive to both radiation and cetuximab, but that, paradoxically, HPV-negative cells are more sensitive to cisplatin [15]. Busch *et al* observed similar cisplatin sensitivities in HPV negative and positive cells but a reduced ability of HPV-negative cells to clear cisplatin-induced DNA adducts, in particular in the most cisplatin-sensitive cell lines [16]. Arenz *et al* and Ziemman *et al* showed that HPV-negative cell lines are more resistant to radiation than HPV-positive cell lines [17, 18]. These studies reveal that squamous cell carcinoma cells *in vitro* respond to clinically utilized chemotherapies and radiotherapies in a manner that is dependent on HPV infection.

Whereas the molecular pathogenesis of HPV-positive HNSCC cancers is well characterized, specifically, involving the oncogenic effects of HPV E6 and E7 proteins [19], the molecular drivers

of HPV-negative HNSCC are relatively poorly understood. As the biology of HPV-negative HNSCC is better understood, investigators have the opportunity to design trials focused on molecular targets unique to HPV-negative tumors.

1.1.2 Genetic Alterations of *TP53*, *CDKN2A*, and *CCND1* and Resistance to Immunotherapy in HPV-negative HNSCC: Preclinical Data

TP53 Mutations and the Immune Microenvironment. Although high-risk *TP53* mutations have been associated with resistance to conventional therapies, the role of p53 mutations in response to immunotherapy in HNSCC has not been explored. Using mouse models that combine the genetic activation of p53 mutations with carcinogen-induced oral tumor development, our preliminary studies suggest that p53 GOF mutations promote resistance to PD1 inhibitors, a current FDA-approved immunotherapy in recurrent/metastatic HNSCC (Fig. 1).

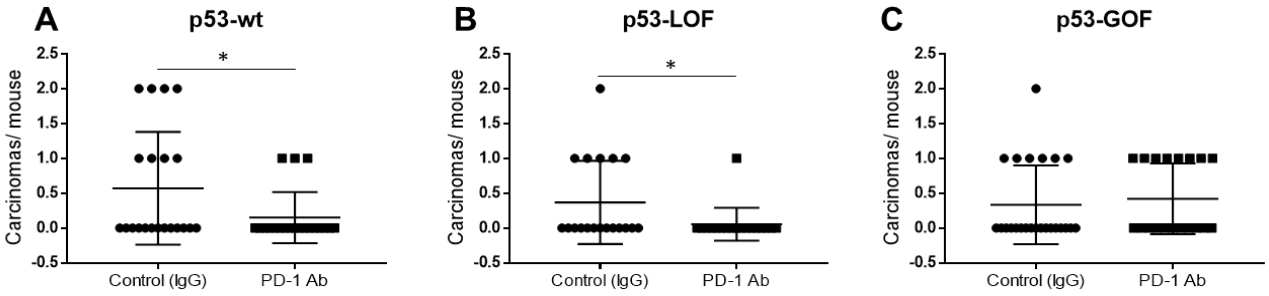


Fig. 1. Anti PD1 treatment prevents progression of oral premalignant lesions induced by 4NQO in mice with wild type p53 (p53-wt) or a p53 LOF mutation (deletion of p53) (p53-LOF), but not in mice expressing the $p53^{R172H}$ GOF mutation (p53-GOF). Mice in which oral tumors were induced upon exposure to 4NQO were treated with an anti-PD-1 antibody or a control IgG, for 4 weeks, twice a week. Six weeks after completion of the treatment the tongues were harvested and assessed for the presence of carcinomas. The graphs show the number of carcinomas per mouse for each mouse in the study.

Preliminary analysis of the immune component of the tumors using nanostring technology, suggest that the tumor microenvironment of $p53^{R172H}$ tumors is depleted of exhausted CD8 cells, and may contain lower levels of negative regulatory T cells (Tregs), compared to tumors with p53LOF mutations or those with wild type p53 (Fig. 2). These studies suggest that mutant p53 expressed in the epithelial tumor cells may promote the generation of a tumor microenvironment that lacks immune cell populations that are reactivated by PD-1 inhibitors, leading to resistance to PD1 blockade in those tumors. Understanding mechanisms that promote a “cold microenvironment” in tumors with p53 mutations is critical to design strategies that result in T cell inflammation, with the potential to overcome resistance to immune checkpoint inhibitors. We hypothesize that the oncogenic function of p53 GOF mutation contributes to remodeling the tumor microenvironment and therefore targeting this oncogenic function of p53 mutations could sensitize tumors to immunotherapy. Interestingly, we observed that the most aggressive SCCs that develop in mice upon activation of $p53^{R172H}$ express high levels of cyclin D1 (Fig. 3), which contributes to over activation of CDK4/6 (Fig. 4). In addition, we found that combining the $p53^{R172H}$ mutation with deletion of *CDKN2A* results in the formation of metastatic tumors [12]. Overall, these studies suggest that either cyclin D1 overexpression or *CDKN2A* deletion contribute to promote malignancy in tumors with p53 GOF mutations, which may also lead to a cold TME and resistance to immunotherapy.

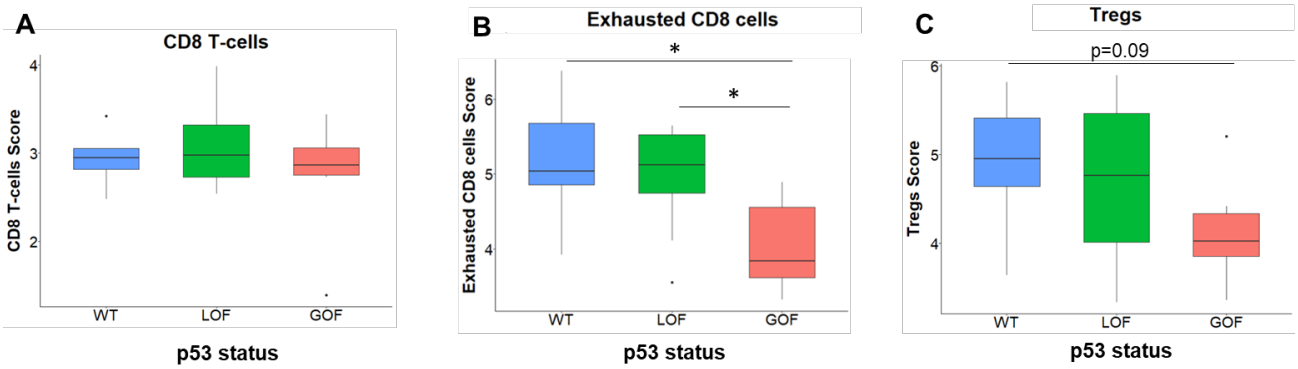


Fig. 2. Immune cell type analysis using nanostring platform. Scores for CD8 T cells (A), exhausted CB8 cells (B) and Tregs (C) are shown in oral SCCs induced by 4NQO in genetically engineered mice in which a p53 gain-of-function mutation $p53^{R172H}$ (GOF), or p53 deletion (LOF) was induced in oral epithelial cells. Note that exhausted CD8 cells are depleted in SCCs expressing the p53 GOF mutation, compared to both p53 LOF or mice with wt p53 (WT).

1.1.3 CDK 4/6 in HPV-negative HNSCC: Tumor Intrinsic Signaling

The rationale for targeting CDK4/6 in HNSCC is supported by genetic [6, 39, 40], *in vitro* [15, 16], epidemiological [41-43], and now clinical studies [44]. Oral SCCs, more than 95% of which are HPV-negative[45], demonstrate overexpression of CDK4 [45, 46], CDK6 [46] and/or cyclin D1 [47]. One study found that 56% of oral squamous cell carcinomas harbored mutations in the p16 gene [48]. As shown in Figure 3, HPV-negativity strongly correlates with genetic alterations of *CCND1* and *CDKN2A*, the genes that code for cyclin D1 and p16, respectively. Specifically, whole-exome sequencing studies have revealed that HPV-negative tumors demonstrate frequent amplifications of CCND1 and deletions of CDKN2A [4, 6, 49, 50].

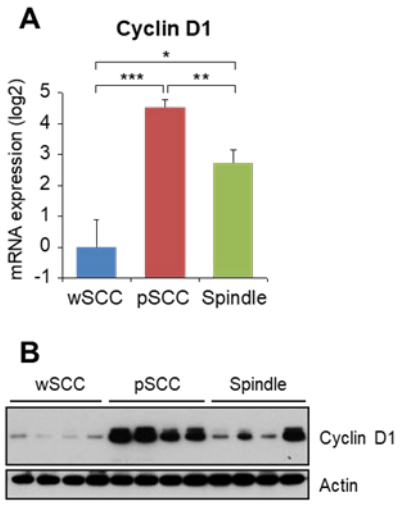


Fig. 3. Cyclin D1 is overexpressed in poorly differentiated SCC (pSCC), compared to well-differentiated SCCs (wSCCs) and spindle tumors induced by activation of p53 mutations in mice. (A) Cyclin D1 RNA expression in the tumors . (B) Western blot for Cyclin D1 protein expression. Actin was used a loading control. Of note, our studies (Li et, J Pathol, 2016) showed that pSCC are the most aggressive tumors that developed in these mice, and the only ones that

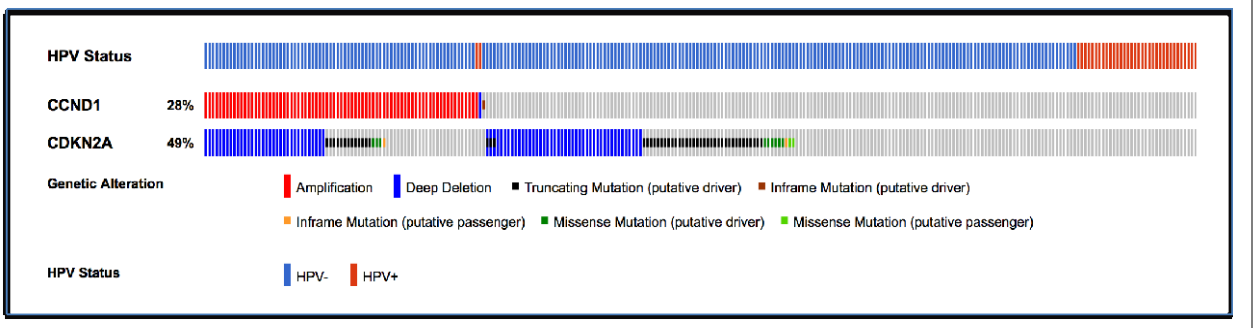


Fig. 4. Expression of CCND1 and CDKN2A genes, encoding for cyclin D1 and p16 proteins, respectively, as a function of HPV status. Data taken from TCGA; courtesy of E. Knudsen, PhD.

These well-documented alterations in the CDK4/6-pRb pathway provide insight into the molecular pathogenesis of HPV-negative HNSCC. Because p16 binds to CDK4/6 and inhibits its phosphorylation of pRb, loss of p16 via deletion of CDKN2A leads to constitutive activation of CDK4/6, phosphorylation of pRb, dissociation of hyperphosphoyrlated pRb from E2F, and

subsequent expression of E2F target genes that facilitate progression into the S phase of the cell cycle. Similarly, amplification of CCND1 causes overexpression of cyclin D1, which also promotes CDK4/6-mediated pRb phosphorylation and unchecked cellular proliferation.

These genetic features indicate a reliance on CDK4/6 for malignant transformation and tumor proliferation in HPV-negative HNSCC. Indeed, numerous studies have compared CDK4/6 inhibition in HPV-negative vs HPV-positive HNSCC *in vitro.* Bo *et al* found that CDK4/6 inhibition blocked cell growth and proliferation in HPV-negative oral cavity (OSC-19[51] and YD-10B[52]) and hypopharyngeal (FaDu)[51] cell lines and also reduced tumor growth in xenograft models using these HPV-negative cells.[53] Beck *et al* directly compared HPV positive and negative cells and found that CDK4/6 inhibition reduced proliferation and pRb phosphorylation at 2, 24, and 48 hours significantly more in HPV-negative hypopharyngeal (FaDu) and oral cavity (SCC61)[51] cell lines compared to HPV-positive cell lines.[54] Clinical trials are currently underway to investigate the efficacy of combined CDK4/6 and EGFR inhibition in HPV-negative HNSCC (NCT02101034, NCT02499120). Phase I clinical data investigating CDK4/6 inhibition in combination with cetuximab in HPV-negative HNSCC have shown promise, both in treatment naïve and cetuximab and platinum-resistant patients combination with cetuximab.[44] D. Adkins and colleagues presented a phase II trial evaluating the combination of palbociclib and cetuximab at the annual meeting of the American Society of Clinical Oncology in 2018, demonstrating a promising overall response rate of 35%, median PFS of 6.4 months, and median OS of 12.1 months [55]. These oncologic outcomes compare favorably to historical data for cetuximab monotherapy, where ORR is 13% and median PFS is 2 months, suggesting that targeting the deregulation of the CDK/cyclin D regulatory complex is a rational molecular strategy that may translate well to the clinic in HPV-negative HNSCC.

1.1.4 CDK 4/6 and the Tumor Immune Microenvironment

The tumor-intrinsic rationale for the development of CDK 4/6 inhibitors in HPV-negative HNSCC is strong, due to the genomic alterations that lead to aberrantly active CDK/cyclin D in the vast majority of cases. Moreover, increasing evidence points to unbridled CDK 4/6 signaling as a source of immune suppression in the tumor microenvironment (TME). CDK 4/6 inhibition may reverse immunosuppression, resulting in the T cell-inflamed phenotype more responsive to immunotherapy, including PD-1 inhibition.

In the complex setting of a solid tumor and its microenvironment, efficacious and durable treatment requires a systemic approach encompassing more than inhibition of cellular proliferation. As has become clear with the success of novel immunotherapies, mounting a successful anti-tumor immune response plays a crucial role in overall treatment efficacy. Indeed, how an anti-proliferative or cytotoxic chemotherapy affects the host immune response intimately impacts the overall anti-tumor efficacy. Inhibition of the CDK4/6-pRb pathway has been shown to impact host immunity on a variety of levels, ranging from the TME to the overall host immune system.

In addition to blocking pRb phosphorylation and cellular proliferation, Goel *et al* recently showed that inhibition of CDK4/6 in mice harboring mammary-gland tumors caused upregulation of genes involved in antigen processing and presentation, including genes encoding for mouse major histocompatibility complex (MHC) class I molecules (H2d1, H2k1, and B2m) and antigen processing proteins (Erap1, Tap1, Tap2, and Tapbp).[56] Similarly, in human breast cancer cells, CDK4/6 inhibition led to upregulation of analogous HLA-1, -B, and -C genes, as well as genes that mediate interferon signaling (eg STAT1/STAT2). Further, CDK4/6 inhibition increased recruitment of CD4+ and CD8+ cells and decreased T regulatory cells within tumors. Altogether, these findings demonstrate that inhibition of CDK4/6 leads to expression of genes that promote a robust immune response within the tumor microenvironment, as shown in Figure 5.

CDK4/6 inhibition impacts the host immune response outside of the tumor microenvironment as well, specifically via protective effects on the hematopoietic system [57]. Murine and canine *in vivo* experiments revealed that CDK4/6 inhibition causes transient and dose-dependent cell cycle arrest in hematopoietic stem cells, protecting them from apoptosis induced by cytotoxic agents like etoposide and 5-FU [58]. Similar studies in tumor-bearing mice demonstrated that CDK4/6 inhibition caused myeloprotection from concurrent treatment with carboplatin [59] and cisplatin [60]. By preserving the integrity of the host immune system, CDK4/6 inhibitors not only prevent myelosuppression-related morbidity but also facilitate a more robust anti-tumor immune response. This is anticipated to promote an immune response within TME, ultimately leading to a robust anti-tumor immune response, as outlined in Figure 5.

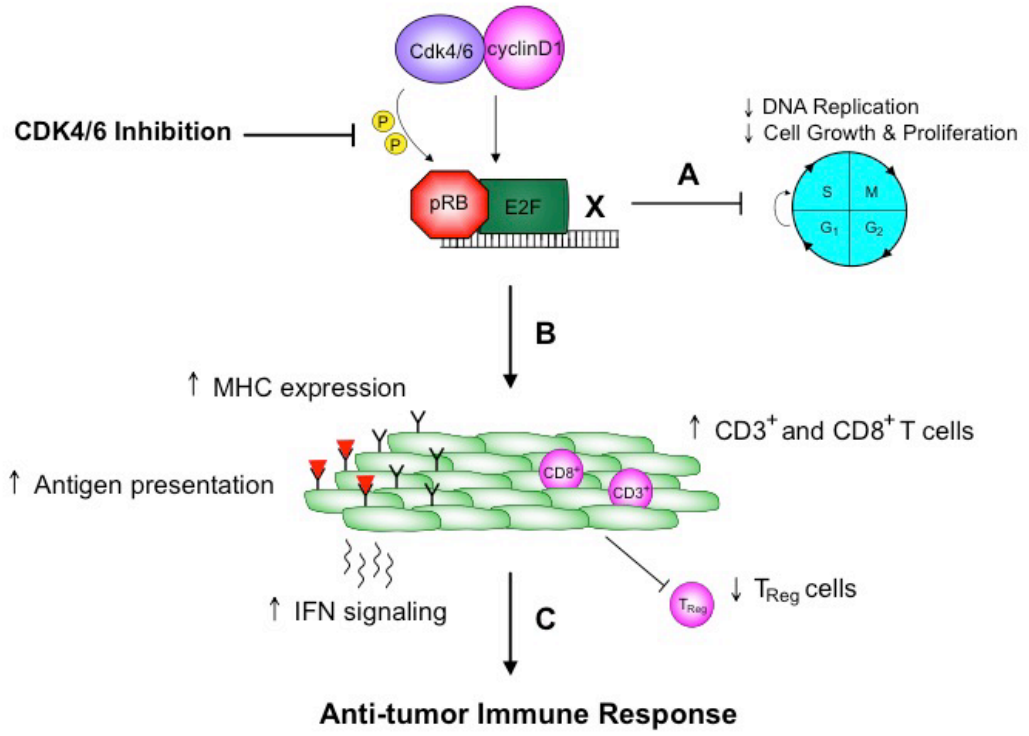


Fig. 5. CDK4/6 inhibition promotes an immune response within the tumor microenvironment. In addition to the well-known cell-cycle blockade (**A**), CDK4/6 inhibition has been shown to induce expression of genes that stimulate an immune response within the tumor microenvironment (**B**). Up-regulation of MHC genes, increased interferon signaling and antigen presentation, and the recruitment of CD3+ and CD8+ T cells together facilitate a robust anti-tumor immune response (**C**).

2 Background on Abemaciclib (LY2835219)

For detailed information on abemaciclib, please see the Investigator's Brochure as well as the FDA Package Insert:

https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/208716s000lbl.pdf

https://uspl.lilly.com/verzenio/verzenio.html#ppi

Abemaciclib is an orally-administered pyridopyrimidine-based compound that inhibits the kinase activity cyclin-dependent kinases 4 and 6 (CDK4/6).[62] Inhibition of CDK4/6 blocks phosphorylation of the retinoblastoma tumor suppressor protein (pRb), the product of the RB1 tumor suppressor gene and the best characterized cellular target of CDK4/6 activity. Abemaciclib binds to CDK4/6-cyclin D complexes selectively, potently, and reversibly, and it exhibits essentially no activity

against other cellular CDKs or tyrosine/serine/threonine kinases. Abemaciclib thus blocks cellular proliferation via highly selective and potent inhibition of pRb phosphorylation.

Current drug discovery and development in oncology has shifted towards molecularly targeted therapies. A key focus has been on identifying inhibitors against components of pathways that drive tumor cell proliferation, survival, and metastasis, such as the pRb pathway, including its upstream regulators, CDK4/6.

Targeted inhibition of CDK4/6 exhibits cytostatic activity against breast[62, 63] and other malignant cells in vitro,[64, 65] and phase II trials demonstrated anti-tumor activity in breast cancer, lymphoma, and sarcoma patients.[66] A series of landmark phase III trials has led to three FDA indications for abemaciclib (Verzenio) in breast cancer as follows:

1) As monotherapy for the treatment of adult patients with HR-positive, HER2-negative advanced or metastatic breast cancer with disease progression following endocrine therapy and prior chemotherapy in the metastatic setting (MONARCH 1[67]);
2) In combination with fulvestrant for the treatment of women with HR-positive, HER2-negative advanced or metastatic breast cancer with disease progression following endocrine therapy (MONARCH 2[68]);
3) In combination with an aromatase inhibitor as initial endocrine-based therapy for the treatment of postmenopausal women with hormone receptor (HR)-positive, human epidermal growth factor receptor 2 (HER2)-negative advanced or metastatic breast cancer (MONARCH 3[69]).

2.1 Preclinical studies and pharmacodynamics

In biochemical assays, abemaciclib exhibits potent and selective inhibition of CDK4/6-cyclin D complexes. As outlined below, reduction in CDK4/6 activity directly correlates to pRb phosphorylation and in turn cell cycle arrest. *In vitro* studies of abemaciclib in colon cancer cell lines demonstrated inhibition of Rb phosphorylation with IC_{50} values of < 120 nM. Kinase panel screens have shown that abemaciclib demonstrates inhibition (IC_{50} <0.3 μM) of several other human protein kinases (hCDK9, hPIM1, hPIM2, hHIPK2, hDYRK2, GSK3β, hCDK5/P35, and CK2); however, the overwhelming and reversible G1 arrest observed after abemaciclib treatment indicates CDK4/6 inhibition predominates over these other activities. Further, three of the major abemaciclib *in vivo* metabolites (LSN3106726, LSN2839567, LSN3106729) demonstrated CDK4/6 inhibition that was nearly identical to parent abemaciclib, with IC50s between 1 and 3 nM. Because these compounds have little effect on the other protein kinases screened, this indicates that selective CDK4/6 dependent cell cycle arrest is maintained even after abemaciclib is metabolized.

In animal models of xenograft tumors, numerous markers of selective G1 arrest, including phosphorylation of pRb at serine-780 (one of the Cdk4/6 target sites), TopoIIa, and histone H3, were used to evaluate abemaciclib efficacy. In Colo-205 murine xenografts, dose-dependent reductions of pRb, TopoIIα, and histone H3 were all observed, demonstrating abemaciclib-mediated cell cycle arrest via CDK4/6 inhibition. Further, abemaciclib treatment led to significant inhibition of tumor growth in multiple different xenograft models, including Colo-205 (colorectal cancer), NCI-H460 (NSCLC), U87 MG (glioblastoma), and JeKo-1 (MCL) xenografts. Each of these cell lines have intact and functional pRb expression, and xenograft growth inhibition was dose-dependent from 15 to 100 mg/kg following daily oral administration for 21 days. The tumor growth inhibition was concomitant with a sustained inhibition of pRb, TopoIIα, and histone H3. The inhibition of tumor growth corresponded to a statistically significant and dose-dependent improvement in survival a rat orthotropic brain tumor model, in which tumor cells were implanted intracerebrally after which the mice were treated daily for 21 days. At doses of 20, 40, and 80 mg/kg, a survival benefit was observed after treatment at 40 and 80 mg/kg.

Altogether, these studies demonstrate that abemaciclib mediates potent and selective inhibition of CDK4/6, that this CdDK/6 inhibition reduces pRb phosphorylation and inhibits cell cycle

progression, and that this cell cycle arrest which inhibits tumor growth *in vivo* and ultimately provides a survival benefit in murine xenograft models.

2.2 Preclinical Pharmacokinetics

Preclinical studies to evaluate the absorption, distribution, metabolism, and excretion of abemaciclib were conducted in rats and dogs. Radiolabeled abemaciclib administered as a single oral dose was found to be slowly absorbed with a time to maximum observed plasma concentration [Tmax] ranging from 6.7 to 8 hours. The oral bioavailability of abemaciclib was determined to be 83.5% in dogs. Repeated abemaciclib administration led to increased plasma concentrations when compared to single-dose administration. The elimination of abemaciclib in dogs was approximately 2x faster than the elimination of radioactivity, indicating that some abemaciclib metabolites (which have similar CDK4/6 inhibition efficacy as the parent compound) are cleared more slowly than parent compound. In rats, the elimination of abemaciclib was comparable to the elimination of radioactivity.

Tissue and organ distribution was similarly evaluated with radioactive [14C] abemaciclib in pigmented (Long–Evans) and non-pigmented (Sprague Dawley) rats. The tissue distribution appeared to be concentrated to melanin-containing tissues. Abemaciclib was found to penetrate the blood-brain barrier as measured by radioactivity in CNS tissues (cerebellum, cerebrum, medulla, and spinal cord) up to 24 hours post-dose. Abemaciclib exhibited very high plasma protein binding, approximately 95% to 99%, across species, with similar binding for its active metabolites.

In vivo metabolism of abemaciclib was evaluated in intact or biliary-cannulated male Sprague Dawley rats and male beagle dogs via administration of IV or PO [14C]abemaciclib. Unmodified abemaciclib was the major circulating entity in both rats and dogs, and N-desethyl-abemaciclib (LSN2839567) was the major metabolic pathway in both species as well as in humans. Liver metabolism revealed 4 oxidative metabolites, including the CDK4/6-active metabolites, LSN2839567 and LSN3106726.

Excretion studies in rats and dogs indicated that, following oral administration, the majority of the dose was excreted in the bile or feces. Following a single oral dose or single IV dose of [14C]abemaciclib, the majority of the dose was excreted in the bile (approximately 52% in bile-cannulated rats) or feces (approximately 91% in intact rats and 86% in dogs).

Regarding interaction with cellular transporters, *in vitro* metabolic studies indicated that abemaciclib is a substrate for both P-glycoprotein (P-gp) and breast cancer resistance protein (BCRP). This suggests that *in vivo* drug interactions may occur when abemaciclib is co-administered with P-gp substrate drugs, such as digoxin. Abemaciclib and its major metabolites, LSN2839567, and LSN3106726 did not significantly inhibit the hepatic uptake transporters OCT1, OATP1B1, and OATP1B3 or the renal transporters, OAT1 and OAT2, at clinically relevant concentrations. Therefore, in vivo drug interactions with substrates of these transporters are unlikely. Abemaciclib, LSN2839567, and LSN3106726 exhibited significant inhibition of the renal transporters OCT2, MATE1, and MATE2-K at clinically relevant concentrations, indicating that abemaciclib may reduce the renal clearance of substrate drugs of these transporters, such as metformin. Inhibition of these transporters by abemaciclib and its metabolites may also reduce renal clearance of creatinine, leading to a rise in serum creatinine levels. Indeed, the serum creatinine increases observed in abemaciclib clinical studies are likely due to the inhibition of creatinine tubular secretion via OCT2, MATE1, and MATE2-K, and do not necessarily represent a decline in renal function. *In vivo*, <10% of unchanged abemaciclib is recovered in the feces, confirming that abemaciclib is cleared primarily through metabolism and indicating that P-gp and BCRP do not prevent oral absorption or disposition of abemaciclib. Collectively, these data indicate that the likelihood of drug interactions is low when abemaciclib is co-administered with specific inhibitors or inducers of P-gp and BCRP.

2.3 Clinical Pharmacokinetics

The pharmacokinetic profile of abemaciclib was studied using data from 222 patients enrolled in Study I3Y-MC-JPBA (JPBA). Consistent with animal studies above, human abemaciclib absorption was found to be slow, with a Tmax ranging from 4 to 6 hours over the dose range of 50 to 275 mg. After repeated oral Q12H dosing of 200 mg, the mean minimum plasma concentration at steady state (Cmin,ss) and maximum observed plasma concentration at steady state (Cmax,ss) were 197 and 298 ng/mL, respectively. The mean absolute bioavailability of abemaciclib after a single oral dose of 200 mg was found to be 45%.

In a study to evaluate the effect of food on abemaciclib absorption (I3Y-MC-JPBG [JPBG]), abemaciclib was administered as a 50% w/w capsule to healthy subjects with a high-fat meal. This study revealed that the Cmax was increased by 24% while the AUC remained unchanged, when compared to the fasted state. A similar study (I3Y-MC-JPBU [JPBU]) found a similar in crease in abemaciclib absorption with high fat meals. Based on these data, abemaciclib can be taken without regard to co-administration of food, and future protocols (including this one) need not contain requirements regarding food restrictions.

Similar to animal studies, abemaciclib was highly bound to plasma proteins in humans (mean bound fraction was approximately 96-98%) and the binding was independent of concentration. Abemaciclib binds to both serum albumin and alpha-1-acid glycoprotein, and the active metabolites are also high protein-bound (approximately 89% to 94%). The geometric mean apparent volume of distribution at steady state for abemaciclib was found to be approximately 1300 L.

As outlined above in animal studies, abemaciclib is predominantly eliminated via metabolism, with the majority of metabolism being mediated by the liver. In human subjects, no significant changes in half-life were observed in patients with mild and moderate liver dysfunction. However, in subjects with severe hepatic impairment, the abemaciclib $t_{1/2}$ increased from 24 to 55 hours. Therefore, no dosage adjustment is required for patients with mild or moderate hepatic impairment, but changing the dosing frequency to once daily may be required for patients with severe hepatic impairment. No dose adjustment is necessary for patients with mild or moderate renal impairment.

3 Study Objectives

3.1 Primary Objective

To evaluate the clinical activity of abemaciclib in patients with operable, HPV-negative HNSCC as measured by quantitative change in tumor size (ΔT)[1] following 10-21 (+7) days of neoadjuvant exposure.

Hypothesis: Abemaciclib will significantly reduce tumor burden as measured by ΔT.

3.2 Secondary Objectives

- To describe the safety and tolerability of neoadjuvant exposure to abemaciclib in accordance with NCI Common Terminology Criteria for Adverse Events (CTCAE) v5.0.

3.3 Exploratory Objectives

- To evaluate tumor-intrinsic genetic biomarkers associated with ΔT, including somatic genetic or epigenetic alterations in *CCND1, CDKN2A,* and *TP53*
- To evaluate baseline and pharmacodynamic biomarkers within the TME associated with ΔT, including:

- IFN-ɣ gene expression signature in baseline and post-treatment tumor biopsies. ***The primary biomarker hypothesis is that abemaciclib will increase the proportion of tumors that are T-cell inflamed.***
- The distribution of tumor-infiltrating immune cell subtypes and their activation status, including lymphocytes and myeloid-derived stem cells, as measured by gene expression, flow cytometry, and multiplex immunohistochemistry (IHC).

- To evaluate the distribution of peripheral immune cell subtypes and their activation status, and how this is altered by abemaciclib.
- To evaluate serum Th1 and Th2 cytokine profiles, and how they are altered by abemaciclib.
- To evaluate tumor-intrinsic molecular mediators of response and resistance to abemaciclib in baseline and post-treatment tumor biopsies, including expression of CDKN2A (p16), CCND1 (cyclin D1), and pRB (retinoblastoma).
- To evaluate the anti-proliferative activity of abemaciclib, as measured by change in the proliferative index (ΔKi67) in pre- and post-treatment tumor specimens.

4 Study Design

4.1 Study Characteristics

This is a phase II window trial to assess the clinical activity and immune biomarker modulation of 10-21 (+7) days of neoadjuvant treatment with abemaciclib in patients planned for oncologic surgery for HPV-negative HNSCC. The study includes rigorous collection of tumor and peripheral blood pre- and post-treatment, in order to study immune modulation in specific genomic contexts.

4.2 Patient Population

Patients with de novo HPV-negative HNSCC who are planned for oncologic surgery.

4.3 Number of Subjects

Up to 20 patients will be accrued to achieve the target of 14 evaluable patients, as defined in Section 12.2.1.

4.4 Eligibility Criteria

The planned (standard of care) surgery will be the primary curative treatment for patients enrolled in this study. Patients must not require any standard induction treatment prior to surgery. Surgery must be the best treatment option as determined by the treating physician as to not be delaying chemoradiotherapy or other curative treatment.

The trial includes any stage of oral cavity, hypopharynx, larynx, or p16-negative oropharynx HNSCC that will be managed by primary surgery.

4.4.1 Inclusion Criteria

1. Males and females, ≥ 18 years of age.
2. New cytologically or histologically diagnosed squamous cell carcinoma of the oral cavity, hypopharynx, larynx, or p16-negative oropharynx.
    a. Patients with two simultaneous primary tumors or bilateral tumors are eligible.
    b. For eligibility, tumors of the oral cavity, hypopharynx, or larynx will be considered HPV-negative without specialized testing.

c. Tumors of the oropharynx must be HPV-negative as determined by p16 immunohistochemistry and/or HPV-DNA per local standard.

d. The study-qualifying HNSCC may be a second primary HNSCC, provided the following criteria are met:

i. The previously treated HNSCC was treated with curative intent.

ii. The HNSCC target lesion(s) are at least 1 cm from the previously treated HNSCC.

iii. At least 2 years have elapsed since curative treatment of the previous HNSCC without evidence for recurrence.

3. AJCC 8th edition clinical TNM staging classifications T1-T4a, any N, and M0.
4. Appropriate and planned for oncologic resection of the primary tumor and/or neck dissection.
5. Clinically or radiologically measurable disease of at least the primary tumor and/or cervical nodes.
    a. These must be measurable according to RECIST 1.1 (tumor diameter ≥ 1 cm; short-axis lymph node diameter ≥ 1.5 cm) OR by caliper/ruler measurement (tumor diameter ≥ 1 cm).
    b. Refer to study definition of Target Lesions.
6. Eastern Cooperative Oncology Group (ECOG) performance status 0-1. (See Appendix 1.)
7. Adequate hematologic function as defined by:
    a. Absolute neutrophil count (ANC) ≥ 1,500/μL
    b. Platelets ≥ 100,000/μL
    c. Hemoglobin ≥ 8 g/dL
        i. Erythrocyte transfusions to achieve this hemoglobin level are permitted.
8. Adequate liver function as defined by:
    a. Bilirubin ≤ 1.5 x institutional upper limit of normal (ULN). Patients with Gilbert's syndrome with a total bilirubin ≤ 2.0 x ULN and direct bilirubin within normal limits are permitted.
    b. AST (aspartate aminotransferase) ≤ 3 x ULN
    c. ALT (alanine aminotransferase) ≤ 3 x ULN
9. Adequate renal function, as defined by:
    a. Creatinine ≤ 1.5 x ULN.
10. Have signed written informed consent.
11. Consent to biomarker collection requirements, including mandatory baseline and intra-operative research biopsies of the study-qualifying HNSCC.

4.4.2 Exclusion Criteria

1. Prior treatment with any CDK 4/6 inhibitor.
2. Prior treatment for the study-qualifying HNSCC.
3. Current or prior use of immunosuppressive medication within 14 days before the first dose of study drug. The following are exceptions to this criterion unless otherwise indicated:

a. Intranasal, inhaled, or topical steroids, or local steroid injections (e.g., intra-articular injection)

b. Systemic corticosteroids at physiologic doses not to exceed 10 mg/day of prednisone or its equivalent

c. Steroids as premedication for hypersensitivity reactions (e.g., CT premedication) and/or as anti-emetics

4. Patient with personal history of any of the following cardiac conditions:

   a. Syncope of cardiovascular etiology

   b. Ventricular arrhythmia of pathologic origin (including, but limited to, ventricular tachycardia and ventricular fibrillation)

   c. Sudden cardiac arrest

   d. Documented history of New York Heart Association functional classification III-IV congestive heart failure

   e. Myocardial infarction ≤ 6 months prior to start of study treatment

   f. Current unstable angina pectoris

5. Personal history of any of the following pulmonary abnormalities:

   a. Interstitial lung disease

   b. Severe dyspnea at rest or requiring oxygen therapy

6. Impaired gastrointestinal (GI) function or GI disease that may significantly alter the absorption of oral abemaciclib (e.g., history of major surgical resection involving the stomach or small bowel, malabsorption syndrome, preexisting Crohn's disease or ulcerative colitis, preexisting chronic condition resulting in baseline Grade 2 or higher diarrhea).

7. Requires chronic administration of drugs that are strong and moderate **inducers** of CYP3A, and no acceptable substitute can be identified (See Appendix 2). Such drugs must be discontinued at least 7 days before the start of study treatment.

8. Requires chronic administration of drugs that are strong **inhibitors** of CYP3A, and no acceptable substitute can be identified (See Appendix 2). Such drugs must be discontinued at least 7 days before the start of study treatment.

9. Active systemic infections including:

   a. Active bacterial infection requiring intravenous antibiotics at time of initiating study treatment,

   b. Fungal infection, or

   c. Detectable viral infection (such as known human immunodeficiency virus positivity or known active hepatitis B or C [e.g. hepatitis B surface antigen positive or hepatitis C antibody positive with detectable viral load]). (Screening for HIV or hepatitis is not required for enrollment.)

      Note: Active topical infections (for example oral thrush, folliculitis, adult acne) do not exclude a subject even if treated with systemic antibiotics or systemic antifungals.

10. Any other condition that would, in the Investigator's judgment, preclude the patient's participation in the clinical study due to safety concerns or compliance with clinical study procedures, e.g. social/psychological complications.

11. Pregnant or nursing (lactating) women.

12. Does not agree to use highly effective contraception (defined below) during the study and through at least 3 weeks after the final dose of abemaciclib:

a. Sexually active males must agree to use a condom during intercourse while taking abemaciclib and for 3 weeks after the final dose of abemaciclib.

b. Males who are sexually active with a woman of child-bearing potential must agree to apply highly effective contraception during the study in order not to father a child in this period and through at least 3 weeks after the final dose of abemaciclib.

c. Women of child-bearing potential (WOCBP), defined as all women physiologically capable of becoming pregnant, must agree to use highly effective contraception during the study and through at least 3 weeks after the final dose of abemaciclib. Highly effective contraception is defined as:

    i. Total abstinence: When this is in line with the preferred and usual lifestyle of the subject. Periodic abstinence, e.g., calendar, ovulation, symptothermal, postovulation methods and withdrawal are not acceptable methods of contraception.

    ii. Female sterilization: have had surgical bilateral oophorectomy (with or without hysterectomy) or tubal ligation at least six weeks before taking study treatment.

    iii. Male partner sterilization (with the appropriate post-vasectomy documentation of the absence of sperm in the ejaculate). (For female study subjects, the vasectomized male partner should be the sole partner for that patient.)

    iv. Use a combination of the following (both 1+2):

        1. Placement of an intrauterine device (IUD) or intrauterine system (IUS)

        2. Barrier methods of contraception: Condom or Occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream/vaginal suppository.

    v. Note: Hormonal contraception methods (e.g. oral, injected, and implanted) are not considered effective as abemaciclib may decrease the effectiveness of hormonal contraceptives.

    vi. Note: Women are considered post-menopausal and not of child-bearing potential if they have had 12 months of natural (spontaneous) amenorrhea with an appropriate clinical profile (e.g. age appropriate, history of vasomotor symptoms) or have had surgical bilateral oophorectomy (with or without hysterectomy) at least six weeks ago.

13. Prisoners or subjects who are compulsorily detained (involuntarily incarcerated) for treatment of either a psychiatric or physical (e.g., infectious) illness.

4.5 Duration of Therapy

In the absence of treatment delays due to adverse events, treatment will consist of a single neoadjuvant cycle administered during the pre-operative window of 10-21 days or up to 28 days for delays in planned surgery or until:

- Progressive disease at any time
- Any clinical adverse event, laboratory abnormality or intercurrent illness which, in the opinion of the Investigator, indicates that continued treatment with study drug is not in the best interest of the subject
- Excessive toxicity
- Refusal of further treatment
- Withdrawal of informed consent (subject's decision to withdraw for any reason)

- Pregnancy
- Study closure or termination
- Subject noncompliance (in the opinion of the treating investigator)

Subjects who discontinue the study intervention will transition to the follow-up portion of the trial.

4.6 Duration of Follow Up

Patients will be followed through the end-of-study visit to be done 4 weeks (+/- 1 week). Patients removed from treatment for unacceptable treatment-related adverse event(s) will be followed weekly until resolution or stabilization of all treatment-related adverse events as described in Section 7.4.1.

4.7 Procedures for Subject Discontinuation from the Trial

Subjects who discontinue the trial during treatment or during follow up should, if possible, be seen and assessed by an Investigator. If a subject chooses to withdraw, the reason for withdrawal (if given) and the date of withdrawal must be documented. If the patient chooses not to disclose the reason for withdrawal, please document as unknown. If possible, any medication diaries and study drug/materials should be returned by the subject as soon as possible after study discontinuation.

5 Study Drug: Abemaciclib

NOTE: For current information, please see the FDA package insert and the Investigator's Brochure.

5.1 Potential Risks of Abemaciclib

Comprehensive information on the risks of abemaciclib are found in the Investigator's Brochure.

Per prior trials of abemaciclib use in combination with endocrine therapies and as a single agent, adverse events of special interests were determined to be events that were:

- Of clinical significance,
- Potentially associated with other agents that inhibit CDK4 and CDK6, or
- Observed in preclinical evaluation or earlier clinical studies of abemaciclib, although in some instances a causal association with abemaciclib could not be clearly established.

The following AEs are considered to be AESIs for abemaciclib:

- Neutropenia,
- Infections,
- Diarrhea,
- hepatic events, including increases in AST and ALT,
- venous thromboembolic events (VTEs), and
- ILD/pneumonitis.

The following are considered potential risks:

- Arterial thromboembolic events (ATEs)
- Ocular effects
- Carcinogenicity
- Reproductive safety

For purposes of reporting of Suspected Unexpected Serious Adverse Reactions (SUSARs) for Abemaciclib as a single agent, only neutropenia is considered ***expected****.*

5.1.1 Reproductive Safety

Abemaciclib has not been studied in pregnant or lactating women; therefore, no clinical data are available. Animal studies indicate that abemaciclib has the potential to cause fetal harm when administered to a pregnant woman.

Male animals given abemaciclib had injury to their testes; therefore, male patients should be advised about the possibility of infertility.

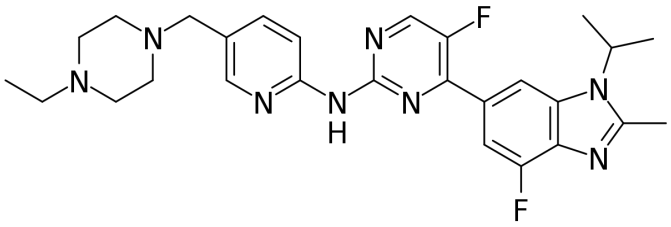
5.2 Description, Supply and Storage of Abemaciclib

5.2.1 Classification

Abemaciclib is an oral small molecule inhibitor that selectively targets the CDK4/6 enzymes.

5.2.2 Description

Abemaciclib is a powder with the empirical formula $C_{27}H_{32}F_2N_8$ and a molecular weight 506.59. The chemical name for abemaciclib is 2-Pyrimidinamine, *N*-[5-[(4-ethyl-1-piperazinyl)methyl]-2-pyridinyl]-5-fluoro-4-[4fluoro-2-methyl-1-(1-methylethyl)-1*H*-benzimidazol-6-yl]-. Abemaciclib has the following structure:



Abemaciclib tablets are provided as immediate-release tablets. Inactive ingredients are as follows: Excipients—microcrystalline cellulose 102, microcrystalline cellulose 101, lactose monohydrate, croscarmellose sodium, sodium stearyl fumarate, silicon dioxide. Color mixture ingredients—polyvinyl alcohol, titanium dioxide, polyethylene glycol, talc, iron oxide yellow, and iron oxide red.

5.2.3 Mechanism of Action

Abemaciclib is an inhibitor of cyclin-dependent kinases 4 and 6 (CDK4 and CDK6). These kinases are activated upon binding to D-cyclins. In estrogen receptor-positive (ER+) breast cancer cell lines, cyclin D1 and CDK4/6 promote phosphorylation of the retinoblastoma protein (Rb), cell cycle progression, and cell proliferation. In vitro, continuous exposure to abemaciclib inhibited Rb phosphorylation and blocked progression from G1 into S phase of the cell cycle, resulting in senescence and apoptosis. In breast cancer xenograft models, abemaciclib dosed daily without interruption as a single agent or in combination with antiestrogens resulted in reduction of tumor size.

5.2.4 Metabolism

Hepatic metabolism is the main route of clearance for abemaciclib. Abemaciclib is oxidatively metabolized to several metabolites primarily by cytochrome P450 (CYP) 3A4, with formation of N-desethylabemaciclib (M2) representing the major metabolism pathway.

5.2.5 Cardiac Electrophysiology

Based on evaluation of the QTc interval in patients and in a healthy volunteer study, abemaciclib did not cause large mean increases (i.e., 20 ms) in the QTc interval.

5.3 Supply, receipt and storage

Abemaciclib will be supplied in 50 mg tablets in bottles. The bottles of 50 mg tablets will have 60 tablets each. Abemaciclib is for oral administration only. It is not available as a parenteral product for intravenous administration.

Study drug must be received by a designated person at the study site, handled and stored safely and properly, and kept in a secured location to which only the investigator and designated assistants have access. Abemaciclib must be stored at room temperature. For specific storage instructions refer to the product label.

Study medication will be dispensed by an authorized person at the investigator's site.

Patients will be provided with a 21-day supply of abemaciclib for self-administration at home. In the event of logistical delays in surgery requiring longer treatment with abemaciclib, an additional 7-day supply will be dispensed.

5.4 Drug compliance and accountability

Clinical drug supply must be accounted for and patients will be asked to return all unused study drug and packaging on a regular basis, at the end of the study, or at the time of study drug discontinuation.

The University of Arizona Investigational Drug Service (IDS) pharmacy uses the National Cancer Institute (NCI) drug accountability logs for all studies at this institution. Control logs will be used for inventory and patient dispensing at the University of Arizona Cancer Center and George Washington Cancer Center.

The University of Arizona and George Washington Investigational Pharmacists will manage drug accountability records. Damaged supplies will be destroyed at the study site and adequate records of the damaged supplies will be kept at the site. Study investigators will be responsible for drug accountability.

5.5 Disposal and Destruction

The drug supply will be destroyed at the University of Arizona and George Washington Cancer Centers in accordance with local policies and Lilly guidelines (if applicable) for destruction.

5.6 Drug Ordering

Abemaciclib will be supplied by Lilly Pharmaceuticals.

5.7 Packaging and Labeling of the Study Drug

Study drug will be packaged and labeled in accordance with US Regulations.

6 Treatment Plan

6.1 Dosage and Administration

Abemaciclib Dosage

Treatment will be administered on an outpatient basis.

Treatment will be administered as a single neoadjuvant cycle.

The FDA approved dose for abemaciclib is 200 mg twice a day when administered as monotherapy.

Abemaciclib will be administered at 200 mg (four 50 mg tablets) by mouth twice a day for 10-21 days, with the option to extend the neoadjuvant cycle up to 28 days for logistical or scheduling purposes.

Study Drug	Dose	Route	Schedule	Cycle Length
Abemaciclib	200 mg	PO	Twice daily	10-21 days (+7 days)

Abemaciclib will be discontinued the day prior to planned surgery, after the evening dose. If surgery is delayed for logistical reasons, abemaciclib can be continued until surgery for a maximum of 28 days.

Abemaciclib Administration

Abemaciclib will be supplied as 50 mg tablets; each dose is 4 tablets. Subjects will be provided with three 60-count bottles.

The following general guidelines should be followed for abemaciclib administration:

- Abemaciclib should be taken at approximately the same time every day approximately 12 hours apart.
    - Preclinical results have also demonstrated that a chronic (or continuous) dosing strategy is important for achieving durable cell-cycle arrest
    - To encourage compliance, patients should be encouraged to take abemaciclib at the same time every morning and evening, approximately 12 hours apart (e.g., 7 am and 7 pm and will document each dose in the study diary.
- Abemaciclib may be taken with or without food.
- Abemaciclib should be swallowed whole.
- Broken, cracked, or otherwise not intact abemaciclib tablets should not be ingested (except as described in the bullet below).
- Administration for subjects unable to swallow:
    - If a patient cannot swallow the abemaciclib tablet whole, the tablet may be crushed, dispersed in at least 10 mL of water, and taken completely and immediately within 10 minutes of dispersion in water.
        - It is not expected that breaking or crushing of the tablet will alter its effectiveness. It must be noted that the tablet will not dissolve in the water and may taste bitter if swallowed, crushed, or broken.
    - Other than being crushed and dispersed in at least 10 mL of water:
        - Patients should not chew, crush, or split tablets before swallowing.
        - Patients are instructed not to ingest any tablet that is broken, cracked, or otherwise not intact except as described in the bullet above.
    - Lilly has not performed specific studies to evaluate the impact of administering the tablets through a feeding tube such as a G-tube or J-tube. However, it is not expected that breaking or crushing the tablet for administration through a gastric tube will impact the effectiveness of the tablet.
    - Crushing or breaking the abemaciclib tablet may lead to accidental exposure to the active ingredient inside the tablet to patients not on abemaciclib therapy. Repeated exposure to the active ingredient may result in impaired fertility or organ toxicity.
- Patients should avoid grapefruit products.
- Abemaciclib is primarily metabolized by CYP3A4.

- If co-administration with a CYP3A inhibitor is unavoidable, adjust abemaciclib dose. (Subjects requiring chronic administration of strong inhibitors of CYP3A are not eligible for this trial.)
- Avoid concomitant use of strong or moderate CYP3A inducers and consider alternative agents. (Subjects requiring chronic administration of strong and moderate inducers of CYP3A are not eligible for this trial.)

- If a patient vomits or misses a dose of abemaciclib, the patient should be instructed to take the next dose at its scheduled time.
- If the patient forgets to take study dose, it may be made up within 6 hours of the scheduled time. Once a dose is missed by 6 hours or more, it should be withheld. Abemaciclib will be restarted at the next scheduled administration. Missed doses should be recorded in the study diary.

6.2 Abemaciclib Dose Reductions/Dose Modifications

A continuous monitoring rule for safety will be instituted, to guard against excess toxicity. See Section 12.3.1.

Abemaciclib dose modification is recommended based on individual safety and tolerability. Management of specific adverse reactions (e.g., diarrhea, hematologic toxicities, or hepatotoxicity) require dose interruption and/or dose modification as shown in the Safety Monitoring and Toxicity Management Section.

All dose modifications must be based on the worst preceding toxicity as graded by the NCI CTCAE v5.0.

Dose level reductions should be made in 50 mg increments. For example, if the starting dose of abemaciclib for a study is 200 mg q12 hours, dose reduction 1 would be 150 mg q12 hours, dose reduction 2 would be 100 mg q12 hours. Refer to Table 1 below for the starting dose level and dose reduction levels.

Patients requiring > 2 dose reductions for abemaciclib should be permanently discontinued. Table 1 below describes the dose reduction steps for abemaciclib.

Table 1. Abemaciclib Dose Reduction Table

Starting dose level	200 mg twice daily
Dose level -1*	150 mg twice daily
Dose level -2*	100 mg twice daily

*Dose reduction should be based on the worst preceding toxicity.

6.3 Abemaciclib Treatment Interruption and Discontinuation

If the administration of abemaciclib is interrupted for reasons other than toxicity, then treatment with abemaciclib may be resumed at the same dose.

All abemaciclib dose interruptions or discontinuations due to toxicity must be based on the worst preceding toxicity as graded by the NCI CTCAE v5.0.

Grade 4 adverse events due to toxicity will lead to permanent discontinuation, irrespective of recovery time, **unless otherwise specified** in this protocol, e.g., in the Abemaciclib Safety Monitoring and Toxicity Management Section regarding dose modifications.

Patients who develop an AE considered at least possibly related to abemaciclib leading to treatment discontinuation will be followed per Section 7.4.1.

6.4 Abemaciclib Procedures in Case of Overdose

There is no known antidote for abemaciclib overdose. Patients experiencing toxicity upon overdosing must be treated at the discretion of the treating physician with adequate supportive care as indicated by the symptoms observed in the patient. Patients will have to be followed until full recovery or confirmed stabilization of the events considered related to an overdose.

6.5 General Guidance for Women of Childbearing Potential and/or Use of Contraceptive Methods

Based on findings in animals, abemaciclib can cause fetal harm when administered to a pregnant woman. In animal studies, abemaciclib was teratogenic and caused decreased fetal weight at maternal exposures that were similar to human clinical exposure based on the area under the plasma concentration versus time curve (AUC) at the recommended human dose. Therefore, teratogenicity is considered an important potential risk for abemaciclib. There are no available human data informing the drug-associated risk. Advise pregnant women of the potential risk to a fetus. Additionally, there are no available data on effects of breastfeeding. Advise a nursing woman to discontinue breastfeeding during treatment with abemaciclib.

- A female of childbearing potential, must have a negative serum pregnancy test within 3 days of the first dose of abemaciclib and agree to use a highly effective contraception method during the treatment period and for 3 weeks following the last dose of abemaciclib.
- Contraceptive methods may include an intrauterine device [IUD] or barrier method. If condoms are used as a barrier method, a spermicidal agent should be added as a double barrier protection.

6.6 Abemaciclib Procedures in Case of Pregnancy

Cases of pregnancy that occur during maternal exposures to abemaciclib must be reported. If a patient or spouse/partner is determined to be pregnant following abemaciclib initiation, she must discontinue treatment immediately. Data on fetal outcome and breast feeding are to be collected for regulatory reporting and drug safety evaluation.

Any pregnancy occurring in a patient or spouse/partner must be reported to Lilly.

6.7 Abemaciclib Safety Monitoring and Toxicity Management

The safety parameters include all laboratory tests and hematological abnormalities, physical findings, and spontaneous reports of adverse events reported to the investigator by patients.

Each patient will be assessed periodically for the development of any toxicity as outlined in Section 7 and Section 10. Toxicity will be assessed according to the NCI CTCAE v5.0. Dose adjustments will be made according to the system showing the greatest degree of toxicity.

Management guidelines for abemaciclib are provided below for the following anticipated adverse events.

6.7.1 Guidelines for Diarrhea Management

Clinical trial data indicates the majority of patients who receive abemaciclib will develop diarrhea. Our experience indicates early identification and intervention for the management of diarrhea has been helpful to patients.

At enrollment, patients should receive instructions on the prompt management of diarrhea. In the event of diarrhea, supportive care measures should be initiated as early as possible. These include the following:

- At the first sign of loose stools, the patient should initiate antidiarrheal therapy (e.g., loperamide) and notify the investigator for further instructions and appropriate follow-up.
- Patients should also be encouraged to drink fluids (e.g., 8 to 10 glasses of clear liquids per day).

- Site personnel should assess response within 24 hours.
- Refer to Table 2 for additional information for diarrhea management and dose modification.

Refer to Table 1 for the dose level reduction levels.

Table 2. Dose Modification and Management for Abemaciclib: Diarrhea

At the first sign of loose stools, start treatment with antidiarrheal agents, such as loperamide.	
CTCAE v5.0 Grade	**Abemaciclib Dose Modifications**
Grade 1	No dose modification is required.
Grade 2	If toxicity does not resolve within 24 hours to ≤ Grade 1, suspend dose until resolution. Dose reduction is not required.
Grade 2 that persists or recurs after resuming the same dose despite maximal supportive measures	Suspend dose until toxicity resolves to ≤ Grade 1. Resume at next lower dose.
Grade 3 or Grade 4 or requires hospitalization	

6.7.2 General Guidance for Interstitial Lung Disease (ILD)/Pneumonitis Events

Interstitial lung disease (ILD) /pneumonitis has been identified as an adverse drug reaction for abemaciclib. The majority of events observed in clinical trial were Grade 1 or Grade 2 with serious cases and fatal events reported. Additional information is available in the IB.

Monitor for clinical symptoms or radiological changes indicative of ILD/pneumonitis and ask your patients to report any new or worsening pulmonary symptoms such as dyspnea, cough, and fever, and investigate and treat as per your local clinical practice (including corticosteroids as appropriate). If ILD/pneumonitis is suspected, investigations may include imaging, such as high resolution computed tomography, broncheoalveolar lavage, and biopsy as clinically indicated. Discontinue abemaciclib in cases of severe (Grade 3 or 4) ILD/pneumonitis.

Table 3: Dose Modification and Management: Interstitial Lung Disease/Pneumonitis

CTCAE v5.0 Grade	Abemaciclib Dose Modifications
Grade 1 or Grade 2	No dose modification is required.
Persistent or recurrent Grade 2 toxicity that does not resolve with maximal supportive measures within 7 days to baseline or Grade 1	Suspend dose until toxicity resolves to baseline or ≤Grade 1. Resume at next lower dose.
Grade 3 or Grade 4	Discontinue abemaciclib.

6.7.3 General Guidance for Hepatic Monitoring

Patients with severe hepatic impairment will not be enrolled in this trial.

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) elevation are considered as ADR with the use of abemaciclib. Abemaciclib dose modification and management of increased ALT is in Table 4.

Table 4. Dose Modification and Management of Abemaciclib: Increased ALT/AST

Monitor ALT/AST prior to the start of abemaciclib therapy, every 2 weeks for the first 2 months, monthly for the next 2 months, and as clinically indicated.	
CTCAE v5.0 Grade	**Abemaciclib Dose Modifications**
Grade 1 (>ULN-3.0 x ULN) Grade 2 (>3.0-5.0 x ULN)	No dose modification is required.
Persistent or Recurrent Grade 2, or Grade 3 (>5.0-20.0 x ULN) WITHOUT increase in total bilirubin above 2 XULN	Suspend dose until toxicity resolves to baseline or Grade 1. Resume at next lower dose.
Elevation in AST and/or ALT>3 x ULN WITH total bilirubin >2 x ULN, in the absence of cholestasis	Discontinue abemaciclib.
Grade 4 (>20.0 x ULN)	Discontinue abemaciclib.

Close Hepatic Monitoring and Evaluation

Liver testing including ALT, AST, alkaline phosphatase (ALP), total bilirubin (TBL), direct bilirubin (D. Bil), gamma-glutamyltransferase (GGT), and creatine kinase (CK), should be repeated within 2 to 4 days to confirm the abnormality and to determine if it is increasing or decreasing, if one or more of these conditions occur:

If a participant with baseline results of...	**develops the following elevations:**
ALT or AST <1.5×ULN	ALT or AST ≥5×ULN or ALT or AST ≥3×ULN concurrent with TBL ≥2×ULN
ALT or AST ≥1.5×ULN	ALT or AST ≥3× baseline or ALT or AST ≥2× baseline concurrent with TBL ≥2×ULN

If the abnormality persists or worsens, clinical and laboratory monitoring and evaluation for possible causes of abnormal liver tests, should be initiated by the investigator. At a minimum, this evaluation should include physical examination and a thorough medical history, including symptoms, recent illnesses (for example, heart failure, systemic infection, hypotension, or seizures), history of concomitant medications (including over-the-counter, herbal and dietary supplements, history of alcohol drinking and other substance abuse). In addition, the evaluation should include a blood test for prothrombin time (PT-INR); serological tests for viral hepatitis A, B, C, E, autoimmune hepatitis; and an abdominal imaging study (for example, ultrasound or CT scan)

Based on the patient's history and initial evaluation results, further testing should be considered, including tests for hepatitis D virus (HDV), cytomegalovirus (CMV), Epstein-

Barr virus (EBV), acetaminophen levels, acetaminophen protein adducts, urine toxicology screen, Wilson's disease, blood alcohol levels, urinary ethyl glucuronide, and blood phosphatidylethanol. Based on the circumstances and the investigator's assessment of the participant's clinical condition, the investigator should consider referring the participant for a hepatologist or gastroenterologist consultation, magnetic resonance cholangiopancreatography (MRCP), endoscopic retrograde cholangiopancreatography (ERCP), cardiac echocardiogram, and/or a liver biopsy.

Additional Hepatic Safety Data Collection

Additional safety data (Table 5) should be collected via the CRF if 1 or more of the following conditions occur:

In participants enrolled with baseline ALT or AST <1.5×ULN

- Elevation of serum ALT or AST to ≥5×ULN on 2 or more consecutive blood tests
- The combination of elevated ALT or AST ≥3×ULN and elevated TBL ≥2×ULN

In participants enrolled with baseline ALT or AST ≥1.5×ULN

- Elevated ALT or AST ≥3× baseline on 2 or more consecutive tests
- The combination of elevated ALT or AST ≥2× baseline and elevated TBL ≥2×ULN

In all study participants

- Discontinuation from study intervention due to a hepatic event or abnormality of liver tests
- Occurrence of a hepatic event considered to be an SAE

Table 5. Hepatic Monitoring Tests for a Hepatic Treatment-Emergent Abnormality

Hematology	**Clinical Chemistry**
Hemoglobin	Total bilirubin
Hematocrit	Direct bilirubin
Erythrocytes (RBCs - red blood cells)	Alkaline phosphatase (ALP)
Leukocytes (WBCs - white blood cells)	Alanine aminotransferase (ALT)
Differential:	Aspartate aminotransferase (AST)
Neutrophils, segmented	Gamma-glutamyl transferase (GGT)
Lymphocytes	Creatine kinase (CK)
Monocytes	**Other Chemistry**
Basophils	Acetaminophen
Eosinophils	Acetaminophen protein adducts
Platelets	Alkaline phosphatase isoenzymes
Cell morphology (RBC and WBC)	Ceruloplasmin
Coagulation	Copper
Prothrombin time, INR (PT-INR)	Ethyl alcohol (EtOH)
Serology	Haptoglobin
Hepatitis A virus (HAV) testing:	Immunoglobulin IgA (quantitative)
HAV total antibody	Immunoglobulin IgG (quantitative)
HAV IgM antibody	Immunoglobulin IgM (quantitative)
Hepatitis B virus (HBV) testing:	Phosphatidylethanol (PEth)
Hepatitis B surface antigen (HBsAg)	**Urine Chemistry**
Hepatitis B surface antibody (anti-HBs)	Drug screen
Hepatitis B core total antibody (anti-HBc)	Ethyl glucuronide (EtG)
Hepatitis B core IgM antibody	**Other Serology**
Hepatitis B core IgG antibody	Anti-nuclear antibody (ANA)
HBV DNA [c]	Anti-smooth muscle antibody (ASMA) [a]
Hepatis C virus (HCV) testing:	Anti-actin antibody [b]
HCV antibody	Epstein-Barr virus (EBV) testing:
HCV RNA [c]	EBV antibody
Hepatitis D virus (HDV) testing:	EBV DNA [c]
HDV antibody	Cytomegalovirus (CMV) testing:
Hepatitis E virus (HEV) testing:	CMV antibody
HEV IgG antibody	CMV DNA [c]
HEV IgM antibody	Herpes simplex virus (HSV) testing:
HEV RNA [c]	HSV (Type 1 and 2) antibody
Microbiology	HSV (Type 1 and 2) DNA [c]
Culture:	Liver kidney microsomal type 1 (LKM-1) antibody
Blood	
Urine	

[a] Not required if anti-actin antibody is tested.

[b] Not required if anti-smooth muscle antibody (ASMA) is tested.

[c] Reflex/confirmation dependent on regulatory requirements, testing availability, or both.

6.7.4 General Guidance for Hematology Toxicity and Dose Modification

Hematologic toxicities including neutropenia, leukopenia, anemia, and thrombocytopenia have been observed in patients treated with abemaciclib, and causality has been established. Severe (Grade 3 and 4) neutropenia was observed in patients receiving abemaciclib. Patients should be monitored closely for signs of infection, anemia, and bleeding.

Abemaciclib dose modification and management of hematologic toxicity is in Table 6.

Table 6. Dose Modification and Management: Hematologic Toxicities

Monitor complete blood counts prior to the start of abemaciclib therapy, every 2 weeks for the first 2 months, monthly for the next 2 months, and as clinically indicated.	
CTCAE v5.0 Grade	**Abemaciclib Dose Modifications**
Grade 1 or Grade 2	No dose modification is required.
Grade 3	Suspend dose until toxicity resolves to ≤ Grade 2. Dose reduction is not required.
Grade 3, recurrent, or Grade 4	Suspend dose until toxicity resolves to ≤ Grade 2. Resume at next lower dose.
Patient requires administration of a blood cell growth factor	Suspend abemaciclib dose for at least 48 hours after the last dose of blood cell growth factor and until toxicity resolves to ≤ Grade 2. Resume abemaciclib at next lower dose unless the dose was already reduced for the toxicity that led to the use of the growth factor.

6.7.5 General Guidance for Monitoring Nonhematologic Toxicities Excluding diarrhea (see Table 2), ILD/Pneumonitis (see Table 3), and increased ALT (see Table 4)

Table 7. Dose Modification and Management: Nonhematologic Toxicities Excluding Diarrhea, ALT/AST Increased, and ILD/Pneumonitis

CTCAE v5.0 Grade	Abemaciclib Dose Modifications
Grade 1 or Grade 2	No dose modification is required.
Persistent or recurrent Grade 2 toxicity that does not resolve with maximal supportive measures within 7 days to baseline or Grade 1	Suspend dose until toxicity resolves to baseline or Grade 1. Resume at next lower dose.
Grade 3 or Grade 4	

6.7.6 General Guidance for Increases in Serum Creatinine and Assessment of Renal Insufficiency

Abemaciclib has been shown to increase serum creatinine due to inhibition of renal tubular transporters without affecting glomerular function (as measured by iohexol clearance). In clinical studies, increases in serum creatinine occurred within the first month of abemaciclib dosing, remained elevated but stable through the treatment period, were reversible upon treatment discontinuation, and were not accompanied by changes in markers of renal function, such as blood urea nitrogen (BUN), cystatin C, or calculated glomerular filtration rate based on cystatin C.

7 Study Procedures and Assessments

The study-specific assessments are detailed in this section, as well as in the study calendar in Appendix 3.

Any results not meeting the eligibility criteria may be repeated at the discretion of the Investigator.

Treatment or visit delays for public holidays or weather conditions do not constitute a protocol violation/deviation.

A written, signed, informed consent form (ICF) and a protected health information (PHI) use and disclosure authorization must be obtained before any study-specific assessments are initiated. A copy of the signed ICF will be given to the subject. The original will be kept on file with the study records.

All patients who sign an informed consent will be entered into the Clinical Trial Management System (CTMS).

All subjects who sign an informed consent will be assigned a sequential subject identifier. Each subject identifier will begin with AIM, the year the trial opened to accrual, a study-specific numerical identifier, a site number, and subject-specific, three-digit number. Example: AIM21-04-01-001. Subjects that are determined to be ineligible (screen failure) will have “-SF” added to the end of the subject ID. Subjects who withdraw prior to registration will have “-WD” added to the end of the subject ID.

7.1 Screening Period & Pre-treatment Procedures

7.1.1 Screening Evaluations

Screening Evaluations/Baseline procedures must occur within 4 weeks prior to study registration unless otherwise specified.

Required screening evaluations:

- Medical history assessment
- Concomitant medication assessment (medications, herbal medications, and food or vitamin supplements administered within 28 days prior to first administration of abemaciclib must be documented)
- Physical examination
- Surgical/medical evaluation, within 8 weeks prior to study registration, including a careful description of the location and extent of the primary lesion and nodal spread. (This may have been done previously per standard of care or may be done simultaneously with the pre-treatment tumor biopsy.)
    - A thorough description of the anatomical extent of the head and neck primary tumor must be detailed by the surgeon in the outpatient note or the EUA operative report.
    - Fiberoptic nasopharyngoscopy/laryngoscopy is recommended, unless fiberoptic exam is deemed unnecessary by the treating surgeon.
    - Examination under anesthesia (EUA) with triple endoscopy is strongly recommended, if not previously performed and/or if an appropriate standard of care for surgical staging. EUA may include direct laryngoscopy, bronchoscopy and esophagoscopy in order to fully evaluate the extent of primary tumor and rule out evidence of a second primary aerodigestive tract tumor.

  - Confirmatory biopsy of the primary tumor for diagnosis can also be performed during the outpatient visit or during the EUA according to standard pathologic procedures.
- Vital signs, weight, height within 8 weeks prior to study registration
- ECOG Performance Status (PS) within 8 weeks prior to study registration
- Clinical laboratory studies:
  - CBC with differential (CBCD; to include total white blood cell count, absolute neutrophil count, hemoglobin, hematocrit and platelets),
  - Comprehensive metabolic panel (CMP must include sodium, potassium, chloride, carbon dioxide, BUN, creatinine, glucose, and calcium, total bilirubin, AST, ALT, total protein, albumin and alkaline phosphatase)
- Negative **serum** pregnancy test for WOCBP
  - An additional negative pregnancy test (urine or serum) is required within 3 days prior to starting protocol treatment.
    - If the **serum** screening pregnancy test was performed within 3 days prior to Day 1, pregnancy test does not need to be repeated.
  - All WOCBP should be instructed to contact the Investigator immediately if they suspect they might be pregnant (e.g., missed or late menstrual period) at any time during study participation.
- Tumor Measurements - Clinically or radiologically measurable disease must be established within 8 weeks *prior to the first dose of study drug* (within 4 weeks is strongly preferred).
  - The minimum required cross-sectional evaluation is a diagnostic, contrasted computed tomography (CT) scan of the neck.
  - PET/CT is the preferred modality, provided it includes a diagnostic, contrasted CT scan of the neck – however is not mandatory.
  - For patients with severe allergy to iodinated contrast dye despite premedication, or in the case of physician preference, neck MRI with gadolidium contrast may be substituted.
  - NOTE: In the event that a patient lacks a measurable primary tumor or metastatic cervical lymph node on cross sectional imaging, measurable disease may be established at baseline by caliper measurement of an oral cavity or oropharyngeal tumor. In this case, the Investigator will measure the primary tumor by caliper or ruler and record the measurement in the patient's chart. A photograph with superimposed caliper or ruler is strongly preferred.
  - The pre- and post-treatment imaging modality should be the same. I.e., if MRI was used at study entry, it should be used at the post-treatment, pre-surgical assessment.
- For oropharyngeal primary tumors: confirmation of p16-negative status.

7.1.2 Registration

- Questions regarding the eligibility of subjects must be directed to Ricklie Julian, MD.
- After eligibility has been confirmed, registration will be submitted to the University of Arizona Cancer Center using the eligibility checklist. The following source documents must be submitted with the eligibility checklist.

- For oropharyngeal primary tumors: confirmation of p16-negative status
- Signed consent page with subject ID added, and PHI redacted

- "Registration" is defined as the day the treating investigator signs the Eligibility Checklist confirming that the subject is eligible to start the study drug.

7.1.3 Mandatory Baseline Biomarker Specimen Collection & Tobacco Assessment

These procedures must occur prior to the initial dose of study drug. They can occur any time after the subject signs the informed consent.

These Procedures are Mandatory.

- **Baseline, pre-treatment tumor biopsy/pre-treatment tissue**
  - Note that a research biopsy of the primary tumor or malignant lymph node for biomarker studies is a condition for enrollment unless sufficient tissue is available to be repurposed for use in this trial.
  - The biopsy will be performed per standard-of-care procedure in the surgeon's outpatient office, or during the standard of care examination under anesthesia (EUA), as preferred by the surgeon.
  - The tissue can be obtained during the standard-of-care surgical/medical evaluation of the extent of primary lesion and nodal spread.
  - **NOTE**: Patients who have had research tissue procured under an omnibus tissue consent, who are determined to have sufficient fresh-frozen and paraffin tissue for biomarker analysis according to the PI or the Translational Science Co-Chair, may substitute the archived tissue and do not need to undergo baseline research biopsy. Such tissue must have been obtained within 24 weeks prior to registration, provided no interval anti-neoplastic therapy was given.
  - Collection and preparation of tissue for biomarker analysis will be processed according to procedures described in in the Laboratory Manual.
- **Baseline research blood collection**
  - For logistical purposes, research labs may be drawn simultaneous with blood labs required for standard clinical management, including on Day 1 of protocol treatment provided the blood is collected **prior** to first dose of protocol treatment.
  - Blood for biomarker analysis will be collected and processed according to procedures described in the Laboratory Manual.
  -
- **Baseline Tobacco Assessment questionnaire (see Appendix 5).**
  - The patient must complete the Tobacco Assessment questionnaire.

7.2 Treatment Period

7.2.1 Day 1 Study Visit

- Physical examination
- Vitals, weight
- ECOG PS

- WOCBP - pregnancy test (serum or urine) within 3 days prior to start of treatment.
    - If the **serum** screening pregnancy test was performed within 3 days prior to Day 1, pregnancy test does not need to be repeated.
- Clinical blood draw: CBCD, CMP (If performed within 14 days prior to Day 1, screening labs may be substituted)
    - Research blood draw, if not already performed during the screening evaluations (can be obtained simultaneously with clinical blood draw)
- Loperamide (generic Imodium) will be dispensed and diarrhea management instructions will be provided to the subject.
- Abemaciclib will be dispensed (up to 3 bottles of 60 tablets each. Note, 3 bottles will last 22.5 days)
- Study Diary will be provided and explained
- Adverse event assessment (and reporting of SAEs) will begin upon the first dose of study drug.
    - Prior to the first dose, active diagnosis and conditions will be considered medical history
    - For all grade 2 adverse events, treating investigator's determination of tolerable or intolerable must be documented for determination of AEs to be entered into the AE CRF
- Concomitant medication assessment (medications, herbal medications, and food or vitamin supplements taken within 28 days prior to Day 1 must be documented)

7.2.2 Study Drug Treatment - Days 1-21 (window of +7 days)

NOTE: Abemaciclib treatment is planned for 21 days, however may be administered up to 28 days if required for logistical/scheduling purposes or due to delay of the planned surgery.

- Abemaciclib will be administered at 200 mg orally twice daily.
- The last dose of abemaciclib will be taken on the evening prior to surgical resection.

If surgery is delayed, abemaciclib may be continued until the day prior to surgery, for a maximum of 28 days.

Subjects will be asked to indicate on a drug diary (see Appendix 4) when doses of abemaciclib are taken in order to monitor for compliance. Subjects will also be encouraged to use the drug diary for notations on side effects and other treatment related events.

7.2.3 "Day 15" Study Visit (This visit can occur on Day 11 through Day 22, inclusive of surgery)

This visit may occur anywhere from Day 11 through Day 22 (inclusive of surgery), depending on logistics and scheduling.

A "Day 15" visit may not need to be scheduled depending on the planned surgery date, however all subjects must have a Pre-operative Assessments visit. If the "Day 15" visit procedures are planned to occur within the 5 days prior to surgery, only the Pre-operative Assessments visit will be performed. Refer to the Pre-operative Assessments, section 7.2.4, below. **Note** that tumor measurements **are required** for pre-operative assessments, but not for the "Day 15" visit.

- Physical examination

- Vital signs, weight
- ECOG PS
- Adverse event assessment with tolerable/intolerable assessment for grade 2 AEs
- Concomitant medication assessment
- Compliance assessment as evidenced by pill count and patient drug diary
- Clinical blood draw: CBCD, CMP
- Research blood draw (can be obtained simultaneously with clinical blood draw)
- Loperamide can be dispensed, if needed
- Abemaciclib will be dispensed, if needed (1 bottle of 60 tablets each which will last through day 28, if needed)

7.2.4 Pre-operative assessments - "Pre-op visit" - Day 11-22 (window of +7 days through Day 29, inclusive of day of surgery)

NOTE: all efforts should be made to schedule these evaluations on the day prior to or the day of planned surgery, however a 5-day pre-operative window is provided for logistical purposes. (For example, these assessments can occur on day 17 if surgery is planned for day 22, day 18 if surgery is planned for day 23, etc., including day 24 if surgery is planned for day 29.)

If the Pre-operative assessments are being scheduled **within the "Day 15" visit window AND within the 5 days prior** to the planned surgery, the Pre-operative assessments will substitute for the "Day 15" visit. **Note** that tumor measurements **are required** for pre-operative assessments, but not for "Day 15".

Refer to the "Day 15" Visit, section 7.2.3, to ensure protocol compliance.

Subjects will continue to take abemaciclib through the day prior to planned surgery; abemaciclib will be discontinued after the evening dose. The interval between the last dose of abemaciclib and surgery must be approximately 12 hours.

NOTE: If surgery is delayed, subjects will continue to take abemaciclib twice daily through the day prior to rescheduled surgery, not to exceed 28 days.

*If the Pre-operative assessments visit substitutes for the "Day 15" visit and the surgery is unexpectedly delayed, resulting in the Pre-op visit procedures having occurred more than 5 days prior to surgery, those procedures can be considered the "Day 15" visit and efforts should be made to perform a Pre-op visit. If the procedures cannot be performed, this will not be considered a deviation. The CT scan does **not** need to be repeated in this scenario.*

- Physical examination.
- Vital signs, weight
- ECOG PS
- Adverse event assessment with tolerable/intolerable assessment for grade 2 AEs.
- Concomitant medication assessment
- Compliance assessment as evidenced by pill count and patient drug diary
- Clinical blood draw: CBCD, CMP
- Research blood draw. Research blood may be drawn simultaneously with standard pre-operative labs.
- Loperamide can be dispensed, if needed

- Tumor Measurements. Clinical and/or radiologic measurements consistent with the method used for the baseline procedure. Every effort should be made to use the same modality (CT or MRI, or caliper/ruler) as used for baseline tumor measurements.

7.3 Surgery and Post-Treatment Tumor Tissue Collection

7.3.1 Days 11-22 (window of +7 days up to day 29, inclusive of day of surgery)

- Oncologic head and neck cancer surgery.
    - After 10-21 days of neoadjuvant treatment (+7 days if required for logistical and scheduling purposes), the patient will undergo the planned oncologic head and neck cancer surgery. The nature of complete resection of the primary head and neck tumor, levels of nodes to be dissected, and type of reconstruction will be determined by the treating surgeon.
    - Prior to surgery, dental evaluation is recommended, to allow for any necessary dental extractions to be planned in conjunction with surgery. Consultations with a nutritionist and speech and language pathologist are strongly recommended prior to surgery and as ongoing support post-operatively. Placement of a nasogastric (e.g., Dobhoff) or gastrostomy feeding tube is at the discretion of the subject and the study physicians.
- Intraoperative tumor tissue for biomarkers (post-treatment tumor tissue collection).
    - Part of the tumor specimen will be collected for the research for mandatory biomarker analyses. See the Laboratory Manual for tissue collection and processing instructions.
    - Ideally, the pre-treatment biopsy and the intraoperative sample will be obtained from the same site (when there are multiple lesions, e.g., primary tumor and malignant lymph nodes).
    - If surgery is unexpectedly cancelled, and primary tumor or malignant lymph node is accessible for in-office biopsy in the judgment of the surgeon-investigator, a tumor biopsy may be substituted for the intraoperative specimen.
    - In the rare case that neither the intraoperative specimen or in-office biopsy are available, tissue from another procedure may be requested only if there is no interval anti-neoplastic therapy given between the baseline and the procedure.

7.3.2 Post-operative therapy

- After surgery, subjects may undergo adjuvant radiation or radiation plus chemotherapy in accordance with appropriate standards, as determined by the subject's treating physicians.

7.4 End-of-Treatment/Post-Operative/Final Study Visit

7.4.1 End of treatment due to related AE

Subjects who end treatment due to an AE determined to be at least possibly related to the study must have weekly follow up to assess the AE until the AE is resolved, returns to baseline grade, or stabilizes, whichever occurs first. If the weekly AE follow up stops before the 4-week post treatment visit, these subjects will also have the 4-week post treatment/post-operative visit (+/- 1 week) as described in Section 7.4.3 below.

7.4.2 Procedures if Subject Withdraws During Treatment Period

Subjects withdraw during the treatment period will be asked to have the following procedures. Subjects should be asked to provide remaining study drug and their diary to the study team. Other procedures should be completed if possible, if clinically indicated, and only if not performed within the previous 2 weeks. These EOT evaluations should be performed as soon as possible.

- Physical examination
- Vital signs, weight
- ECOG PS
- Clinical blood tests (CBC, chemistries, liver function tests in accordance with investigator judgment)
- Adverse event and pregnancy assessment with tolerable/intolerable assessment for grade 2 AEs
- Concomitant medication assessment
- Compliance assessment as evidenced by pill count and patient drug diary

7.4.3 4-Week (+/- 1 week) Follow-up Assessment

All subjects who take at least 1 dose of the study drug will have a 4-week (+/- 1 week) follow-up assessment after the last dose of study drug. AE/SAE, pregnancy, and concomitant medication assessments are required. Drug compliance assessment must be done, if necessary. *Other procedures are to be done as clinically indicated.*

- Physical examination
- Vital signs, weight
- ECOG PS
- Clinical blood tests (CBC, chemistries, liver function tests in accordance with investigator judgment)
- Adverse event and pregnancy assessment with tolerable/intolerable assessment for grade 2 AEs
- Concomitant medication assessment (Note: medications administered specifically for the standard-of-care surgery are not required to be documented.)
- Compliance assessment as evidenced by pill count and patient drug diary (if not completed at the pre-operative visit.)

Additional non-protocol visits may occur during the post-operative period as deemed clinically necessary.

In addition, the following peri-operative data points will be recorded:

- grade 3 or higher post-operative complications;
- number of hospital days;
- number of ICU days.

8 Usage of Concurrent/Concomitant Medications

Please refer to the Investigator's Brochure for additional details.

CYP450 substrates

Abemaciclib was found to elicit significant interactions with only CYP3A.

Patients that require chronic administration of drugs that are strong and moderate inducers of CYP3A and/or strong inhibitors of CYP3A are not eligible for this trial. Such drugs must be discontinued at least 7 days before the start of study treatment.

8.1 General Guidance for Concomitant Therapy

In general, the use of any concomitant medication or therapies deemed necessary for the care of the patient is permitted, except as specifically prohibited.

Abemaciclib is predominantly cleared by oxidative metabolism via CYP3A4. Clinical drug interaction studies with a CYP3A inhibitor and CYP3A inducer significantly altered the PK of abemaciclib and its circulating major metabolites.

CYP3A inducers

Avoid concomitant use of CYP3A inducers and consider alternative agents.

CYP3A inhibitors

Avoid concomitant use of strong CYP3A inhibitors (for example, voriconazole) and use caution with coadministered moderate (for example, ciprofloxacin) or weak (for example, ranitidine) CYP3A inhibitors. If coadministration with a strong CYP3A inhibitor is unavoidable, reduce the abemaciclib dose to 100 mg twice daily or, in the case of ketoconazole, reduce the abemaciclib dose to 50 mg twice daily. In patients who have had a dose reduction to 100 mg twice daily due to adverse reactions, further reduce the abemaciclib dose to 50 mg twice daily. Avoid grapefruit or grapefruit juice. If a CYP3A inhibitor is discontinued, increase the abemaciclib dose (after 3-5 half-lives of the inhibitor) to the dose that was used before starting the inhibitor. For example, coadministration of itraconazole, a strong CYP3A inhibitor, increased the area under the plasma concentration-time curve (AUC) of abemaciclib in healthy subjects by 87%.

In terms of CYP3A inducers, coadministration of a strong CYP3A inducer (rifampin) decreased the AUC of abemaciclib in healthy subjects by 85%. Strong CYP3A inducers (e.g., phenytoin, rifampicin, carbamazepine and St John's Wort) will thus be avoided. Coadministration of moderate CYP3A inducers (e.g., bosentan, efavirenz, etravirine, modafinil, and nafcillin) may also decrease the plasma exposure of abemaciclib and will thus be avoided as well. In A5481004, abemaciclib was administered in combination with dexamethasone. Dexamethasone is a known inducer of CYP3A4. The exposure of abemaciclib observed from A5481004 was considerably lower compared to exposures observed in studies A5481001 (single agent) or A5481003 (in combination with letrozole) likely due to the induction effect of dexamethasone.

8.2 Guidance for Specific Concomitant Medications

Medications include not only physician prescribed medications, but also all over-the counter medications, herbal medications and food or vitamin supplements. The Investigator should instruct the patient to notify the investigational site about any new medications (s)he takes after start of the study drug.

Antiemetics

Use of anti-emetics is allowed as clinically necessary per the discretion of the Investigator.

Anticoagulation, including warfarin and other coumarin derivatives

Anticoagulants may be administered as clinically necessary per the discretion of the Investigator.

The major CYP450 enzymes inhibited by warfarin are CP2C8 and 2C9, on which abemaciclib has negligible impact. No contraindications to warfarin therapy are thus recommended while being treated with abemaciclib.

Contraceptives

Hormonal contraceptives may be affected by cytochrome P450 interactions, and are therefore not considered effective for this study. For allowed contraception methods, refer to Exclusion Criteria. Highly effective contraception should be maintained throughout the study and for 3 weeks after study treatment discontinuation.

Gastric protection agents

Gastric protection agents, including proton pump inhibitors (PPI) and H2 blockers may be administered as clinically necessary per the discretion of the Investigator.

Other anticancer therapy

Anti-cancer therapies other than the study agent, abemaciclib, are prohibited for the duration of the study, and for 4 weeks after the last abemaciclib dose.

Herbal medications

Any herbal preparations containing St. John's Wort must be avoided during the study. Patients should stop using St. John's Wort 7 days prior to first dose of study drug.

Growth factors

Growth factors may be administered as clinically necessary per the discretion of the Investigator.

9 Tumor Measurements

The primary objective of this study is to evaluate ΔTumor size, which will be measured using established RECIST v1.1 metrics for target lesions, however treated as a continuous variable as previously described by Dr. Julie E. Bauman's group [1]. Target lesions will be evaluated using the new international criteria proposed by the Response Evaluation Criteria in Solid Tumor (RECIST) Committee [71]. Changes in only the largest diameter (unidimensional measurement) of the tumor lesions and the shortest diameter of metastatic lymph nodes are used in the RECIST v1.1 criteria.

NOTE: Because this is a pre-operative window study with only brief exposure to study drug, and RECIST responses are not anticipated, target lesions will be measured bi-dimensionally and recorded quantitatively. Quantitative change in the RECIST-defined baseline sum diameter (longest diameter of tumor + short axis diameter of up to three pathologic lymph nodes), expressed as a percent, will be the primary endpoint and will be correlated with biomarker modulation [1].

All tumor measurements pre- and post-treatment will be performed independently by a single neuroradiologist.

9.1 Disease Parameter Definitions

Measurable disease

Primary Tumors. Measurable disease is defined as a primary tumor that can be accurately measured by one of the following techniques:

- Cross-sectional imaging (spiral CT or MRI): the primary tumor can be measured in at least one dimension (longest diameter to be recorded) and measures at least 10 mm, when using spiral CT or MRI with slice thickness no thicker than 5 mm.

- Caliper exam: the primary tumor measured at least 10mm in longest diameter by caliper measurement on clinical exam. Note: A photograph with superimposed caliper or ruler is strongly preferred for documentation of baseline and response.
- PET/CT

Lymph Nodes. Measurable disease is defined as a lymph node that can be accurately measured by cross-sectional imaging (spiral CT or MRI) and measures at least 15 mm in shortest diameter, when using spiral CT or MRI with slice thickness no thicker than 5 mm.

All tumor measurements will be recorded in millimeters or decimal fractions of centimeters.

Target lesions

Only the following will be used as target lesions in this HNSCC window trial:

- Primary tumor (long axis)
- Up to two metastatic cervical lymph nodes (short axis)

Non-target lesions

All other lesions (or sites of disease) including any measurable lesions over and above the target lesions may be identified as non-target lesions and may also be recorded at baseline.

Non-measurable disease (Tumor Markers)

Non-measurable disease is all other lesions (or sites of disease), including small lesions (longest diameter < 20 mm with conventional techniques or < 10 mm using spiral CT scan). Leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniques are all non-measurable. (e.g., PSA, CA-125, CA19-9, CEA)

9.2 Methods for Evaluation of Measurable Disease

All measurements will be taken and recorded in metric notation using a ruler or calipers.

The same method of assessment and the same technique will 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 antitumor effect of a treatment.

10 Documentation and Reporting of Adverse Events & Unanticipated Problems

10.1 Definitions of Adverse Events

10.1.1 Adverse Event

An adverse event (also known as an adverse experience) is defined as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. More specifically, an adverse event (can be any unfavorable and unintended sign (e.g., an abnormal laboratory finding), symptom, or disease temporally associated with the use of a drug, without any judgment about causality. An adverse event can arise from any use of the drug (e.g., off-label use, use in combination with another drug) and from any route of administration, formulation, or dose, including an overdose.

10.1.2 Adverse reaction

An adverse reaction is defined as any adverse event caused by the use of a drug. Adverse reactions are a subset of all suspected adverse reactions for which there is reason to conclude that the drug caused the event.

10.1.2.1 Suspected

A suspected adverse reaction is defined as any adverse event for which there is a reasonable possibility that the drug caused the adverse event. For the purposes of IND safety reporting, "reasonable possibility" indicates that there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than an adverse reaction.

10.1.2.2 Unexpected

An adverse event or suspected adverse reaction is considered *unexpected* if it is not listed in the investigator brochure or package insert(s), or is not listed at the specificity or severity that has been observed, or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application.

"Unexpected," as used in this definition, also refers to adverse events or suspected adverse reactions that are mentioned in the investigator brochure as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

Adverse events that would be anticipated to occur as part of the disease process are considered *unexpected* for the purposes of reporting because they would not be listed in the investigator brochure. For example, a certain number of non-acute deaths in a cancer trial would be anticipated as an outcome of the underlying disease, but such deaths would generally not be listed as a suspected adverse reaction in the investigator brochure.

Some adverse events are listed in the Investigator Brochure as occurring with the same class of drugs, or as anticipated from the pharmacological properties of the drug, even though they have not been observed with the drug under investigation. Such events would be considered *unexpected* until they have been observed with the drug under investigation. For example, although angioedema is anticipated to occur in some patients exposed to drugs in the ACE inhibitor class and angioedema would be described in the investigator brochure as a class effect, the first case of angioedema observed with the drug under investigation should be considered *unexpected* for reporting purposes.

10.1.3 Life-threatening

An adverse event or suspected adverse reaction is considered *life-threatening* if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.

10.1.4 Serious adverse events

Serious adverse event (SAE) is defined as one of the following:

- Pregnancy of subject or subject's partner
- Is fatal or life-threatening
    - *For this trial, death unequivocally due to disease progression is not considered an SAE*
- Results in persistent or significant disability/incapacity
- Constitutes a congenital anomaly/birth defect
- Is medically significant (important medical event), i.e., defined as an event that jeopardizes the patient or may require medical or surgical intervention to prevent one of the outcomes listed above

- Requires inpatient hospitalization or prolongation of existing hospitalization, *unless, per the determination of the investigator, the hospitalization is for*:
    - Routine treatment or monitoring of the patient's cancer indication, not associated with any deterioration in condition
    - Elective or pre-planned treatment for a pre-existing condition that is unrelated to the patient's cancer indication and has not worsened since the start of the treatment with alpelisib
    - Social reasons and respite care in the absence of any deterioration in the patient's general condition
    - Treatment on an emergency outpatient basis for an event not fulfilling any of the definitions of a SAE given above and not resulting in hospital admission
    - Transmission of infectious agent via medicinal product

10.2 Recording Adverse Events

Only AEs meeting any of the following criteria will be entered into the study case report forms. Data about these events and their severity will be recorded using the NCI CTCAE v5.0.

- Any AE that is Grade 3 or higher, regardless of relationship to the study drug
- Any **intolerable** Grade 2 AE at least possibly related to study drug, in the judgment of the Investigator (For all grade 2 adverse events, the treating investigator will make a determination of tolerable or intolerable, and only intolerable grade 2 AEs must be documented on the AE Log source document and entered into the AE CRF.)
- Any Grade 2 laboratory values at least possibly related to study drug and is deemed clinically significant by the treating Investigator
- Any Grade 2 vital sign values at least possibly related to study drug and is deemed clinically significant by the treating Investigator
- Any Grade AE resulting in holding or dose-reducing abemaciclib
- Any Grade AE at least possibly related to study drug that results in the delay of surgery to more than 42 days after start of protocol treatment
- Any Grade AE that results in reporting of an SAE
- Any Grade AE in the following categories of interest:
    - Diarrhea
    - Rash
    - Pneumonitis
    - AST or ALT elevation
    - Any AE determined to be immune-related
- In addition, the following peri-operative adverse events must be recorded:
    - Grade 3 or higher post-operative complications
- Pregnancy
    - Subjects must be followed for pregnancy or pregnancy of a female partner. Pregnancy must be documented and submitted as an SAE per Section 10.1.4, Serious adverse events.

For each AE, the following will be assessed and recorded:

- Grade

- Relationship to study drug
- Causality if other than study drug (disease related, concomitant medication related, intercurrent illness, other)
- Date of onset
- Date of resolution
- Frequency of event (single, intermittent, continuous)
- Event outcome (resolved, ongoing, death)
- Action taken (none, held, dose reduced, discontinued)

Relationship

The Investigator will assign attribution of the possible association of the event with use of the study drug classification system listed below:

Relationship	Attribution	Description
Unrelated to study drug/intervention	Unrelated	The AE *is clearly NOT related* to the intervention
	Unlikely	The AE *is doubtfully related* to the intervention
Related to study drug/intervention	Possible	The AE *may be related* to the intervention
	Probable	The AE *is likely related* to the intervention
	Definite	The AE *is clearly related* to the intervention

CTCAE v5.0 Grading

Signs or symptoms reported as AEs will be graded and recorded by the Investigator according to the CTCAE v5.0. The Investigator will grade AEs as none, mild, moderate or severe according to the following grades and definitions.

Grade 0:	No AE (or within normal limits)
Grade 1:	Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated
Grade 2:	Moderate; minimal, local, or noninvasive intervention (e.g., packing, cautery) indicated; limiting age-appropriate instrumental activities of daily living (ADL)
Grade 3:	Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL
Grade 4:	Life-threatening consequences; urgent intervention indicated
Grade 5:	Death related to AE

10.3 Follow up of Adverse Events

All AEs will be followed with appropriate medical management. Patients removed from study for unacceptable adverse events will be followed until resolution, return to baseline grade, or stabilization (whichever occurs first) of the adverse event. For selected adverse events for which administration of the study drug was stopped, a re-challenge of the subject with the study drug may be conducted if considered both safe and ethical by the Investigator.

Subjects who end treatment due to an AE determined to be at least possibly related to the study must have weekly follow up to assess the AE until the AE is resolved, returns to baseline grade, or stabilizes, whichever occurs first. Refer to Section 7.4.1.

10.4 Adverse Event Monitoring

The Investigator will assess all adverse events and determine reportability requirements to the University of Arizona Cancer Center Data and Safety Monitoring Board (DSMB) and University of Arizona Institutional Review Board, and, when the study is conducted under an Investigational New Drug Application (IND), to the Food and Drug Administration (FDA) if it meets the FDA reporting criteria.

10.5 Serious Adverse Event Reporting

In the event of a serious adverse event, the PI, the institutional review board (if applicable, per institutional reporting requirements), and Lilly will be notified using the Site specific SAE form.

SAEs must be reported within 24 hours of learning of its occurrence. Additional information must be provided in a follow-up SAE report and also reported within 24 hours after receiving the information.

ALL events meeting the definition of a serious adverse event must be reported using the current FDA MedWatch 3500 Form and submitted to:

- UACC DSMB
- Lilly

In addition to completing appropriate patient demographic and suspect medication information, the report should include as applicable the following information that is available at the time of report within the Event Description of the MedWatch 3500 form:

- CTCAE term(s) and grade(s)
- Current status of study drug
- All interventions to address the AE (testing and result, treatment and response)
- Hospitalization and/or discharge dates
- Event relationship to study drug

Follow-up SAE Reports

Additional information may be added to a previously submitted SAE report and submitted as follow-up SAE Report.

Reporting to the UACC Data and Safety Monitoring Board

To ensure patient safety, every SAE, regardless of suspected causality, occurring after the patient has taken the first dose of study drug and through the end of the study follow-up period must be reported to the DSMB Chair (or qualified alternate) within 24 hours of learning of its occurrence.

If a death occurs during the treatment phase of the study or during the study follow-up period and it is determined to be related to the study drug or to a study procedure, the Investigator or his/her designee must notify the DSMB Chair (or qualified alternate) within 1 business day of knowledge of the event. The contact may be by phone or e-mail.

Reporting Safety Information to Lilly

To ensure patient safety, every SAE, regardless of suspected causality, must be reported to the investigator and Lilly. The following are required:

- To comply with applicable laws, regulations and standards regarding Investigator's and Institution's obligations, as the sponsor of the Study, to collect and report adverse events to regulatory authorities, IRBs, Ethics Committees or other third parties. In addition to the obligations set forth below, Investigator and Institution agree to provide Lilly with a copy of all information Investigator and/or Institution submit to regulators related to any adverse events for the Study Drug that occur during the Study that Investigator and/or Institution have not otherwise provided Lilly;
- To notify Lilly, sub-investigators, and the IRB of any problems involving risk to Study patients and report new safety information to IRBs in accordance with applicable requirements;
- To notify Lilly within fifteen (15) business days after Investigator and/or Institution receives notification of any "serious" adverse event experienced by a patient participating in the Study and receiving Study Drug. For purposes of this requirement, "serious" means: (1) death; (2) in-patient hospitalization or prolonged hospitalization; (3) life-threatening; (4) persistent or significant disability or incapacity; (5) congenital anomaly or birth defect; or (6) other serious events that may jeopardize the patient and may require medical or surgical intervention to prevent one of the other five listed outcomes. Serious adverse events should be reported to Lilly using a CIOMS Form or other form acceptable to Lilly. Investigator and Institution further agree to make available promptly to Lilly such records as may be necessary and pertinent for Lilly to further investigate an adverse event in the Study that is possibly associated with the Study Drug.
- For Lilly all SAEs should be submitted within 15 days through the study portal at www.lillyinvestigatorresearch.com.

Recurrent episodes, complications, or progression of the initial SAE must be reported as follow-up to the original episode within 24 hours of the investigator receiving the follow-up information. An SAE occurring at a different time interval or otherwise considered completely unrelated to a previously reported one should be reported separately as a new event.

Information about all SAEs is collected and recorded on the MedWatch Form 3500; all applicable sections of the form must be completed in order to provide a clinically thorough report. The investigator must assess and record the relationship of each SAE to the study treatment.

Follow-up information is sent to the same contact(s) to whom the original SAE Report Form was sent, using a new SAE Report Form stating that this is a follow-up to the previously reported SAE and giving the date of the original report. Each re-occurrence, complication, or progression of the original event should be reported as a follow-up to that event regardless of when it occurs. The follow-up information should describe whether the event has resolved or continues, if and how it was treated, whether the blind was broken or not, and whether the patient continued or withdrew from study participation.

10.6 Reporting Unanticipated Problems

Adverse events, serious adverse events, and deviations may be considered "unanticipated problems." Unanticipated problems, as defined below, will be reported to oversight entities as required by regulation and/or policy.

Note: Treatment or visit delays for public holidays or weather conditions do not constitute a protocol violation/deviation.

The Office for Human Research Protections (OHRP) considers unanticipated problems involving risks to participants or others to include, in general, any incident, experience, or outcome that meets **all** of the following criteria:

- Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the Institutional Review Board (IRB)-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

11 Data and Safety Monitoring

11.1 Safety Monitoring

Dr. Julian at the University of Arizona Cancer Center (UACC) will assume the role of Coordinating site Investigator (C-I). The UACC Data and Safety Monitoring Board (DSMB) will provide ongoing oversight of safety monitoring for this trial. Based on the UACC DSMB Charter, this is a high-risk study.

The UACC DSMB will monitor the study in accordance with the UACC NCI-approved Data and Safety Monitoring Board Charter. The DSMB will routinely review all adverse events and suspected adverse reactions considered "serious". The DSMB may audit study-related activities to ensure that the study is conducted in accordance with the protocol, local standard operating procedures, FDA regulations, and Good Clinical Practice (GCP). Significant results of the DSMB audit will be communicated to the IRB and the appropriate regulatory authorities at the time of review, or in an expedited fashion, as applicable.

Routine study activity and safety information will be reported to the DSMB on a monthly basis, or more frequently if requested. These reports will include, as applicable:

- Study activity, cumulative and for the period under review;
- Safety (narrative description on non-serious and serious adverse events, protocol pre-determined early stopping rules for safety or treatment-emergent adverse events);
- Predetermined protocol early stopping rules for efficacy/futility;
- Status of study in relationship to stopping rules;
- Current dose level of study agent;
- Routine monitoring and protocol compliance (describe the monitoring process and identify the status of the monitoring);
- Comments;
- Attachments (AE data reviewed by the PI to compile the report, SAE letters and reports, results of any review(s), applicable correspondence with the IRB or other regulatory agencies.

DSMB Determinations will be submitted to the UA IRB at least annually.

The UACC DSMB will also perform the review described in the Delay of Surgery section of this protocol. That is, if the number of patients with an unacceptable delay in surgery ever exceeds 3 the study will be referred to the UACC DSMB for evaluation of unacceptable delays in surgery, and recommendations for study redesign or closure

The PI will immediately notify, in writing, the funding agency, if applicable, any action resulting in a temporary or permanent suspension of the study. A copy of this correspondence will also be forwarded to the DSMB and the SRC.

11.2 Data Monitoring

Routine monitoring will be provided by the UACC Compliance and Quality Assurance (CQA) Program. Monitoring is conducted to ensure that the rights and well-being of trial participants are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial is in compliance with the currently approved protocol/amendment(s), with International Conference on Harmonisation Good Clinical Practice (ICH GCP), and with applicable regulatory requirement(s).

This trial will also undergo real-time monitoring by the PI and study team, including documentation of real-time monitoring of any new or ongoing safety issues.

The investigational site will provide direct access to all trial related sites, source data/documents, and reports for the purpose of monitoring and auditing/inspection by local and regulatory authorities.

Routine monitoring of subject data will be conducted in compliance with the UACC Data and Safety Monitoring Board (DSMB) Charter. For affiliate sites UACC CQA will conduct remote monitoring in accordance with the UACC's DSMB charter.

Monitoring reports will be provided to the PI, UACC DSMB, and UA IRB. The PI is responsible to follow up on any action items and/or findings in a timely manner.

12 Statistical Considerations and Evaluation of Results

12.1 Study Endpoints

12.1.1 Primary Endpoint

The primary objective is to evaluate the clinical activity of neoadjuvant abemaciclib.

The primary endpoint is quantitative change in tumor size (ΔT). ΔT will be treated as a continuous variable, based upon the percent change in RECIST-determined target lesions pre- and post-treatment as previously described by Dr. Julie E. Bauman's group [1].

ΔT will be assessed using a paired *t* test.

12.1.2 Secondary Endpoint

The secondary objective of this study is to evaluate the safety and tolerability of neoadjuvant exposure to abemaciclib in accordance with NCI Common Terminology Criteria for Adverse Events (CTCAE) v5.0.

12.1.3 Exploratory Endpoints

The exploratory objectives of this study are to evaluate the baseline and pharmacodynamic tumor-intrinsic, tumor microenvironment (TME) and peripheral immune biomarkers that correlate with clinical activity. Exploratory endpoints include:

- Tumor-intrinsic genetic biomarkers associated with ΔT, including somatic genetic or epigenetic alterations in *CCND1, CDKN2A,* and *TP53*
- Baseline and pharmacodynamic biomarkers within the TME associated with ΔT, including:
    - IFN-ɣ gene expression signature in baseline and post-treatment tumor biopsies. ***The primary biomarker hypothesis is that abemaciclib will increase the proportion of tumors that are T-cell inflamed.***

- The distribution of tumor-infiltrating immune cell subtypes and their activation status, including lymphocytes and myeloid-derived stem cells, as measured by gene expression, flow cytometry, and multiplex immunohistochemistry (IHC).

- The distribution of peripheral immune cell subtypes and their activation status, and how this is altered by abemaciclib.
- Serum Th1 and Th2 cytokine profiles, and how they are altered by abemaciclib.
- Tumor-intrinsic molecular mediators of response and resistance to abemaciclib in baseline and post-treatment tumor biopsies, including expression of p16, CCND1, and pRB.
- Anti-proliferative activity of abemaciclib as measured by change in the proliferative index (∆Ki67) in pre- and post-treatment tumor specimens.

Change in ΔKi-67 will be assessed using a paired *t* test. Assessment of the associations between ΔT and the candidate biomarkers will be performed using Pearson correlation coefficients, with appropriate transformation of the biomarker measurements to ensure normality.

12.2 Determination of Sample Size and Accrual Rate

12.2.1 Sample Size and Power Estimate

The overall required sample size is 14 evaluable patients.

“Evaluable” is defined as a patient who demonstrates treatment compliance, with exposure to at least 10 days of abemaciclib with at least one dose, has undergone response assessment, and has provided paired pre- and post-tumor specimens sufficient in quantity and quality for both DNA and gene expression (RNA) analyses as determined by the Translational Science Co-Chair.

Based upon prior window trials, up to 20 patients will be enrolled to meet these criteria. To justify the sample size, the standard deviation of ΔT was estimated from the erlotinib arm of Dr. Julie E. Bauman’s previously completed 4-arm window study of erlotinib, dasatinib, or the combination versus placebo in HNSCC [1]. In that study, ∆T after 14 – 21 days of targeted therapy ranged from -30% to 55%. Assuming the same standard deviation for ∆T after abemaciclib treatment as observed for the erlotinib arm (sd = 8%), the study will have > 80% statistical power to detect a mean decrease in ∆T of 6.5%, assuming a two-sided alpha level of 0.05.

As a biomarker study, the statistical design is to accrue a sufficient number of patients and provide 80% power to test one or any of 4 key hypotheses with a shared alpha of 0.15: 1) relationship between ΔT and genetic alterations in *TP53*; 2) relationship between ΔT and genetic alterations in *CDKN2A*; 3) relationship between ΔT and change in IFN-γ gene expression signature; 4) the relationship between ΔT and change in the tumor immune infiltrate (CD8+/Treg ratio). An overall alpha of 0.15 will be shared equally among the four primary hypotheses.

Fourteen evaluable patients with complete biomarker data will provide 80% statistical power to detect a Pearson correlation coefficient of 0.69 or greater, assuming a two-sided alpha level of 0.0375.

12.2.2 Replacement Policy

Because this is a window trial designed to investigate mechanisms associated with the clinical activity of abemaciclib in patients with HPV-negative HNSCC, the sample size of 14 evaluable patients is strictly required.

Additional patients will be enrolled to replace patients who do not meet criteria for evaluability. It is planned that up to 20 patients will be eligible and registered in order to obtain 14 evaluable subjects.

12.3 Protocol Early Stopping Rules

12.3.1 Toxicity

Each patient who initiates protocol treatment, receiving at least one dose of abemaciclib will be evaluable for safety and toxicity. Safety evaluation will be reported descriptively, including tabulation of toxicities according to NCI CTCAE v5.0.

A continuous monitoring rule for safety will be instituted, to guard against excess toxicity from pre-operative treatment. After enrollment of the 6th patient the trial will be continuously monitored the number of patients who discontinue abemaciclib and/or come off study for toxicity. A qualifying toxicity must be at least possibly related to abemaciclib as judged by the treating investigator and fulfill one of the following definitions:

- Grade ≥ 3 non-hematologic toxicity, with the exceptions of asymptomatic electrolyte abnormalities manageable with repletion or grade 3 diarrhea with optimal medical management lasting < 3 days.
- Intolerable non-hematologic toxicities of any grade, which persist despite optimal medical management.
- Grade ≥ 4 thrombocytopenia or neutropenia.
- Any grade toxicity that results in a unacceptable delay of surgery, defined as more than 42 days from Day 1 of study drug treatment.

If the posterior probability of ≥ 33% toxicity rate exceeds 50% for patients having to discontinue study treatment due to qualifying toxicity, this would be considered unacceptable. The study would be placed on hold and would be referred to the UACC DSMB for evaluation of excess toxicity and recommendations for design change or study closure.

Table 8 below describes the number of toxicity events per number of treated patients required to trigger referral to the UACC DSMB. Table 8 also shows the posterior probability that the rate exceeds 33%, and the binomial probability associated with the decision for an assumed 33% discontinuation rate. The prior probability has a beta distribution with parameters a = 1 and b = 9, so assumes a 10% mean with an approximate 80% mid-range of .0 to .22.

Table 8. Boundaries for the Excess Toxicity Rule

Subjects	Treatment-Related Discontinuations	PP(π > 33%)*	Pr(X ≥ r|p = .33)
6	5	.629	.017
8	6	.685	.019
11	7	.673	.037
14	8	.664	.054
17	9	.657	.071
20	10	.650	.087

*π is the discontinuation rate. The minimum acceptable upper bound of a treatment-related discontinuation is 33%. PP (π > 33%) is the posterior probability that the discontinuation rate exceeds this 33% upper bound. This posterior probability of discontinuation is calculated from the prior distribution, the number of subjects treated and the observed number of treatment-related discontinuations.

12.3.2 Delay in Surgery

Because the protocol enrolls patients who will undergo potentially curative surgery, a conservative stopping rule is in place to guard against unacceptable delays in surgery. An unacceptable delay

in surgery is defined as surgery occurring more than 42 days after the start of protocol treatment, AND is due to a toxicity that is at least possibly related to abemaciclib as judged by the treating investigator. Among the first 14 patients, if the number of patients with an unacceptable delay in surgery ever exceeds 3, then enrollment will be stopped and the study will be referred to the UACC DSMB for evaluation of unacceptable delays in surgery, and recommendations for study redesign or closure. Table 9 below describes the probability of observing at least 3 of 14 patients with unacceptable delay in surgery for varying values of the underlying assumed probability. As shown below, if the probability of an unacceptable delay in surgery is 0.20 or greater, the probability of observing at least 3 out of 14 patients is > 0.50.

Table 9. Probability of Observing ≥ 3/14 Patients with Unacceptable Delay in Surgery

Probability of Unacceptable Delay	Probability of Observing ≥ 3/14 Patients with Unacceptable Delay
0.05	0.03
0.10	0.16
0.15	0.35
0.20	0.55
0.25	0.72
0.30	0.84

13 Study Management

13.1 Pre-study Documentation

This study will be conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki and consistent with GCP and all applicable regulatory requirements.

Before initiating this trial, the Investigator will have written and dated approval from the Institutional Review Board for the protocol, written informed consent form, subject recruitment materials, and any other written information to be provided to subjects before any protocol related procedures are performed on any subjects.

The Investigator must comply with the applicable regulations in Title 21 of the Code of Federal Regulations (21 CFR §50, §54, and §312), GCP/ICH guidelines, and all applicable regulatory requirements. The IRB must comply with the regulations in 21 CFR §56 and applicable regulatory requirements.

13.2 Institutional Review Board Approval

The protocol, the proposed informed consent form, and all forms of participant information related to the study (e.g., advertisements used to recruit participants) will be reviewed and approved by the IRB of Record. Prior to obtaining IRB approval, the protocol must be approved by the University of Arizona Cancer Center Scientific Review Committee and by the Protocol Review Committee (PRC). The initial protocol and all protocol amendments must be approved by the IRB prior to implementation.

13.3 Informed Consent

All participants must be provided a consent form describing the study with sufficient information for each participant to make an informed decision regarding their participation. Participants must sign the IRB -approved informed consent form prior to participation in any study specific procedure. The participant must receive a copy of the signed and dated consent document. The original signed copy of the consent document must be retained in the medical record or research file.

13.4 Changes in the Protocol

Once the protocol has been approved by the IRB, any changes to the protocol must be documented in the form of an amendment. The amendment must be signed by the Investigator and approved by the IRB prior to implementation.

If it becomes necessary to alter the protocol to eliminate an immediate hazard to patients, an amendment may be implemented prior to IRB approval. In this circumstance, however, the Investigator must then notify the IRB in writing within the timeframe required by the IRB of Record. The Study Chair and the UA study team will be responsible for updating any participating sites.

13.5 Deviations

A protocol deviation (noncompliance, unanticipated problem) may be either on the part of the participant, the investigator, or the study site staff. No changes from current IRB-approved research protocol are allowed except where a change may be necessary to eliminate an apparent immediate hazard to a human subject. Deviations may be identified by any study staff or monitoring/oversight entity. As a result of deviations, as applicable, corrective actions are to be developed by the site and implemented promptly.

Deviations will be documented in the CTMS and reported to oversight entities, as applicable.

13.6 Maintenance of Subject Specimens

Specimens collected as part of this study will be maintained for future use to learn about preventing or treating cancer. Subjects will be asked to consent to the future use during the informed consent process, and asked to positively indicate their consent to future use in writing within the informed consent document. Samples will be maintained in a secure location within the UACC.

13.7 Study Discontinuation and Closure

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to the PI, study participants, investigator, Lilly, the FDA Investigational New Drug (IND and other regulatory authorities as applicable. If the study is prematurely terminated or suspended, the Principal Investigator (PI) will promptly inform study participants, the Institutional Review Board (IRB), and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Demonstration of efficacy that would warrant stopping
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination that the primary endpoint has been met
- Determination of the study meeting futility and/or stopping rules

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the sponsor, IRB, and/or Food and Drug Administration (FDA).

13.8 Handling and Documentation of Clinical Supplies

The UA Principal Investigator and each participating site will maintain complete records showing the receipt, dispensation, return, or other disposition of the study drug. The date, quantity and batch or code number of the drug, and the identification of patients to whom study drug has been

dispensed by patient number and initials will be included. The sponsor-investigator will maintain written records of any disposition of the study drug.

The Principal Investigator shall not make the study drug available to any individuals other than to qualified study patients. Furthermore, the Principal Investigator will not allow the study drug to be used in any manner other than that specified in this protocol.

13.9 Case Report Forms (CRFs)

The Principal Investigator and/or his/her designee will prepare and maintain adequate and accurate participant case histories with observations and data pertinent to the study. Study specific Case Report Forms (CRFs) will document safety and treatment outcomes for safety monitoring and data analysis. All study data will be entered into standardized CRFs, using single data entry with a secure access account. The study staff will complete the CRFs as soon as possible upon completion of the study visit.

The information collected on CRFs shall be identical to that appearing in original source documents. Source documents will be found in the patient's medical records maintained by UA personnel or per the covered entity's electronic medical record (EMR) per institutional procedure, and separate Research Chart, if applicable.

In accordance with federal regulations, the Investigator is responsible for the accuracy and authenticity of all clinical and laboratory data entered onto CRFs.

All source documentation and CTMS data will be available for review/monitoring by the UACC DSMB and regulatory agencies.

The Principal Investigator will be responsible for ensuring the accurate capture of study data. At study completion, when the CRFs have been declared to be complete and accurate, the database will be locked. Any changes to the data entered into the CRFs after that time can only be made by joint written agreement among the Study Chair, the Trial Statistician, and the Protocol Project Manager.

13.10 Record Keeping and Record Retention

The Principal Investigator is required to maintain adequate records of the disposition of the drug, including dates, quantity, and use by subjects, as well as written records of the disposition of the drug when the study ends.

The Principal Investigator is required to prepare and maintain adequate and accurate case histories that record all observations and other data pertinent to the investigation on each individual administered the study drug or employed as a control in the investigation. Case histories include the case report forms and supporting data (e.g., signed and dated consent forms and medical records, such as progress notes of the physician, the individual's hospital chart(s), and the nurses' notes. The case history for each individual shall document that informed consent was obtained prior to participation in the study.

Study documentation includes all CRFs, data correction forms or queries, source documents, Sponsor-Investigator correspondence, monitoring logs/letters, and regulatory documents (e.g., protocol and amendments, IRB correspondence and approval, signed patient consent forms).

Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical research study.

In accordance with FDA regulations, the investigator shall retain records for a period of 2 years following the date a marketing application is approved for the drug for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 2 years after the investigation is discontinued and FDA is notified.

14 Protection of Human Subjects

14.1 Protection from Unnecessary Harm

Each clinical site is responsible for protecting all subjects involved in human experimentation. This is accomplished through the IRB mechanism and the process of informed consent. The IRB reviews all proposed studies involving human experimentation and ensures that the subject's rights and welfare are protected and that the potential benefits and/or the importance of the knowledge to be gained outweigh the risks to the individual. The IRB also reviews the informed consent document associated with each study in order to ensure that the consent document accurately and clearly communicates the nature of the research to be done and its associated risks and benefits.

14.2 Protection of Privacy & Confidentiality

Patients will be informed of the extent to which their protected health information may be collected, used, and/or disclosed for research purposes. Following this discussion, they will be asked to sign the PHI Authorization form (to authorize use and disclosure of PHI in compliance with HIPAA) and informed consent document. The original signed document will become part of the patient's medical records, and each patient will receive a copy of the signed document. The use and disclosure of protected health information will be limited to the individuals and entities described in the PHI authorization and informed consent document.

In the event that a subject revokes authorization to collect, use, and/or disclose PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e., that the subject is alive) at the end of their scheduled study period.

Only authorized individuals will have access to the identifiable study data. All records identifying subjects must be kept confidential per local and Federal policy. Minimum necessary standards must be followed. All subjects are assigned a study ID number. The study data to be used for purposes of analysis and reporting will be coded. Specimens will also be coded using the study ID number. Subject identities will not be used in any results that are published.

Study subjects and study data will be entered into the CTMS. The system is password protected and meets HIPAA requirements.

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Appendices

Appendix 1. Performance Status Criteria

ECOG Performance Status Scale	
Grade	Descriptions
0	Normal activity Fully active, able to carry on all pre-disease performance without restriction
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)
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
3	In bed > 50% of the time Capable of only limited self-care, confined to bed or chair more than 50% of waking hours
4	100% bedridden Completely disabled Cannot carry on any self-care Totally confined to bed or chair
5	Dead

Appendix 2. Inhibitors and Inducers of CYP3A4

Patients should be instructed to avoid grapefruit, grapefruit juice, or grapefruit products for the duration of the study due to the potential for CYP3A4 interactions.

Patients who require the chronic administration of drugs that are strong and moderate inducers of CYP3A and/or strong inhibitors of CYP3A, and no acceptable substitute can be identified, are not eligible for study. Such drugs should be discontinued at least 7 days before the start of study treatment.

If during the course of the study, the patient develops a new requirement for the chronic administration of drugs that are strong and moderate inducers of CYP3A and/or strong inhibitors of CYP3A, and no acceptable substitute can be identified, then abemaciclib dose adjustments are required. Such circumstances are expected to be very rare, and should be discussed with the PI.

Examples of acceptable dose adjustments follow:

- Patients who must take CYP3A inhibitors such as clarithromycin, diltiazem, or verapamil should reduce the abemaciclib dose to 100 mg twice daily.
- Patients who must take itraconazole should reduce the abemaciclib dose to 50 mg twice daily.
- Patients who must take ketoconazole should reduce the abemaciclib dose to 50 mg once daily.

Category	Drug Name
Strong CYP3A Inhibitors	Boceprevir, clarithromycin, cobicistat, conivaptan, elvitegravir, indinavir, itraconazole, ketoconazole, lopinavir, mibefradil, nefazodone, nelfinavir, posaconazole, ritonavir, saquinavir, telaprevir, telithromycin, tipranavir, troleandomycin, voriconazole
Strong CYP3A Inducers	Avasimibe, carbamazepine, mitotane, phenobarbital, phenytoin, rifabutin, rifampin (rifampicin), St. John's wort (hypericum perforatum)
ModerateCYP3A Inhibitors	Amprenavir, atazanavir, casopitant, cimetidine, ciprofloxacin, cyclosporine, darunavir, diltiazem, dronedarone, erythromycin, fluconazole, fosamprenavir, grapefruit juice (citrus paradisi fruit juice), imatinib, Schisandra sphenanthera, tofisopam, verapamil
Moderate CYP3A Inducers	Bosentan, efavirenz, etravirine, genistein, modafinil, nafcillin, ritonavir, talviraline, thioridazine, tipranavir

Strong CYP3A Inhibitors

Strong and moderate CYP3A4 inhibitors increased the exposure of abemaciclib plus its active metabolites to a clinically meaningful extent and may lead to increased toxicity.1 Strong CYP3A inhibitors include

- **Ketoconazole**: Predicted to increase the AUC of abemaciclib by up to 16-fold. Avoid concomitant use of ketoconazole,[1] and
- **Clarithromycin**: Coadministration of clarithromycin 500 mg twice daily with a single 50-mg dose of abemaciclib (0.3 times the approved recommended 150-mg dosage), increased the relative potency adjusted unbound $AUC_{0\text{-}INF}$ of abemaciclib plus its active metabolites by 2.5-fold relative to abemaciclib alone in cancer patients.[1]

In patients with recommended starting doses of 200 mg twice daily or 150 mg twice daily, reduce the abemaciclib dose to 100 mg twice daily with concomitant use of strong CYP3A inhibitors other than ketoconazole. In patients who have had a dose reduction to 100 mg twice daily due to adverse reactions, further reduce the abemaciclib dose to 50 mg twice daily with concomitant use of strong CYP3A inhibitors.[1]

If a patient taking abemaciclib discontinues a CYP3A inhibitor, increase the abemaciclib dose (after 3-5 half-lives of the inhibitor) to the dose that was used before starting the inhibitor.[1]

Avoid grapefruit or grapefruit products.[1]

Moderate CYP3A Inhibitors

With concomitant use of moderate CYP3A inhibitors, monitor for adverse reactions and consider reducing the abemaciclib dose in 50 mg decrements, if necessary.[1] Verapamil and diltiazem are moderate CYP3A inhibitors which are predicted to increase the relative potency adjusted unbound AUC of abemaciclib plus its active metabolites by approximately 1.6-fold and 2.4-fold, respectively.[1]

CYP3A Inducers

Coadministration of CYP3A inducers decreased the plasma concentrations of abemaciclib plus its active metabolites and may lead to reduced activity. Avoid concomitant use of CYP3A inducers and consider alternative agents.[1]

Loperamide

Coadministration of a single 8-mg dose of loperamide with a single 400-mg dose of abemaciclib in healthy subjects increased the relative potency adjusted unbound $AUC_{0\text{-}INF}$ of abemaciclib plus its active metabolites by 12%, which is not considered clinically relevant.[1]

Endocrine Therapies

In clinical studies in patients with breast cancer, there was no clinically relevant effect of fulvestrant, anastrozole, letrozole, or exemestane on the PK of abemaciclib.[1]

EFFECT OF ABEMACICLIB ON OTHER DRUGS

Loperamide

In a clinical drug interaction study in healthy subjects, coadministration of a single 8-mg dose of loperamide with a single 400-mg dose of abemaciclib in healthy subjects (2.7 times the approved recommended 150-mg dosage) increased the relative potency $AUC_{0\text{-}INF}$ of abemaciclib plus its active metabolites by 12%, and increased loperamide $AUC_{0\text{-}INF}$ by 9% and C_{max} by 35% relative to loperamide alone. These effects are not considered clinically relevant.[1]

Metformin

In a clinical drug interaction study in healthy subjects, coadministration of a single 1000-mg dose of metformin, a clinically relevant substrate of renal OCT2, and MATE1 and 2-K transporters, with a single 400-mg dose of abemaciclib (2.7 times the approved recommended 150-mg dosage) increased metformin $AUC_{0\text{-}INF}$ by 37% and C_{max} by 22% relative to metformin alone. Abemaciclib reduced the renal clearance and renal secretion of metformin by 45% and 62%, respectively, relative to metformin alone, without any effect on glomerular filtration rate as measured by iohexol clearance and serum cystatin C.[1]

Endocrine Therapies

In clinical studies in patients with breast cancer, there was no clinically relevant effect of abemaciclib on the PK of fulvestrant, anastrozole, letrozole, or exemestane.

Appendix 3. Study Calendar: Schedule of Procedures and Assessments

Study Procedures [1]	Baseline, Mandatory Procedures, & Registration [2]	Treatment Period Days 1 through 21 (+7 days)			Surgery & Biopsy [3]	Follow Up / Final Study Visit: 4 (+/-1) Weeks after last dose [4]
		Day 1 [2]	Day 15 [5]	Pre-op Visit [5] (-5 day window)		
			Day 11 through 29 inclusive of surgery [5]			
Informed consent	X					
Medical History	X					
Physical exam	X	X	X	X		X [7]
CBC with differential [6]	X	X	X	X		X [7]
Comprehensive metabolic panel [8]	X	X	X	X		X [7]
Pregnancy test	X [9]	X [9]				
Vital signs	X [10]	X	X	X		X [7]
Weight	X [10]	X	X	X		X [7]
Height	X [10]					
ECOG-PS	X [10]	X	X	X		X [7]
Surgical/Medical tumor evaluation [11]	X [10]					
Tumor measurements [12]	X [13]			X		
p16 IHC (OP tumors only)	X					
Concomitant medication assessment [14]	X	X	X	X		X
Adverse Event/pregnancy assessment [15]		X	X	X		X [16]
Registration	X					
Tobacco assessment questionnaire	X					
Research blood collection [17]	X		X	X		
Research tumor biopsy/tissue collection	X [18]				X [19]	
Provide Loperamide & Diarrhea Management Instructions		X	X [20]	X [20]		
Study Drug Dispensing		X	X [21]			
Provide Diary & Instructions		X				
Abemaciclib administration		X (Day 1-21 & up to 28) [22]				
Compliance assessment			X	X		X [23]
Record Peri-operative information						X [24]

[1] Any results obtained *during screening* that do not meet the eligibility criteria and/or study windows may be repeated at the discretion of the investigator.

[2] Baseline procedures are to be done within 4 weeks of registration unless otherwise specified. Screening procedures can be used as Day 1 procedures if they occur within the protocol-required window. Refer to Section 7.2.1 for the protocol-required windows. The baseline tumor tissue may be collected during the surgical/medical tumor evaluation.

[3] Prior to surgery, dental evaluation is recommended, to allow for any necessary dental extractions to be planned in conjunction with surgery. Consultations with a nutritionist and speech and language pathologist are strongly recommended prior to surgery and as ongoing support post-operatively. Placement of a nasogastric (e.g., Dobhoff) or gastrostomy feeding tube is at the discretion of the subject and the study physicians.

[4] All subjects who take at least 1 dose of study drug will have a Final Study Visit/4-week (+/- 1 week) follow up assessment after the last dose of study drug. AE/SAE, pregnancy, and concomitant medication assessments are required. Drug compliance assessment must be done, if necessary. *Other procedures are to be done as clinically indicated.* Refer to Section 7.4.1 and Section 7.4.3.

[5] These procedures may occur anywhere from Day 11 through Day 29 (inclusive of surgery). All efforts should be made to schedule them on the day prior or day of planned surgery. If the Pre-op assessments are being scheduled **within the "Day 15" visit window AND within the 5 days prior** to the planned surgery, the Pre-op assessments will substitute for the "Day 15" visit. **Note** that tumor measurements **are required** for pre-operative assessments, but not for "Day 15". Refer to the "Day 15" Visit & Pre-operative Assessments sections to ensure protocol compliance. (Also, see Section 7.2.4 regarding unexpected delays in surgery and potential to consider the "Pre-op" assessments as the "Day 15" Visit.)

[6] CBCD must include total white blood cell count, absolute neutrophil count, hemoglobin, hematocrit, and platelets.

[7] As clinically indicated, in accordance with investigator judgement.

[8] CMP must include sodium, potassium, chloride, carbon dioxide, BUN, creatinine, glucose, and calcium, total bilirubin, AST, ALT, total protein, albumin, and alkaline phosphatase.

[9] For WOCBP only. Screening pregnancy test is required to be serum. Pregnancy test can occur anytime during screening but a negative **serum** pregnancy test must be documented within 3 days prior to **first dose** of drug.

[10] Must be performed within 8 weeks of registration.

[11] This must include documentation of a careful description of the location and extent of the primary lesion and nodal spread. Refer to Section 7.1.1 for recommendations and requirements of the Surgical/Medical Evaluation.

[12] This includes clinical OR radiological documentation of measurable disease. Refer to Section 7.1.1 for details.

[13] Must be performed within 8 weeks of *the first dose of study drug* (within 4 weeks is strongly preferred).

[14] Medications taken within the 28 days prior to Day 1 must be documented; medications administered for the standard-of-care surgery are not required to be documented.

[15] Active diagnosis and conditions at baseline and prior to first dose are considered medical history. Refer to Section 10 of the protocol for requirements for AE and SAE monitoring and documentation.

[16] Subjects who end treatment due to an AE at least possibly related to the study must have weekly follow up to assess the AE until the AE is resolved, returns to baseline grade, or stabilizes, whichever occurs first. See Section 7.4.1.

[17] Whenever possible, research blood collection should occur simultaneously with clinical labs blood collection. Baseline research blood can occur on Day 1.

[18] The baseline tumor tissue may be collected during the surgical/medical tumor evaluation. Tissue procured under an omnibus tissue consent and determined to have sufficient fresh-frozen and paraffin tissue for biomarker analysis according to the PI or the Translational Science Co-Chair, may substitute for the baseline research biopsy. Such tissue must have been obtained within 24 weeks prior to registration, provided no interval anti-neoplastic therapy was given.

[19] Part of the tumor specimen from the planned surgery will be sent to the research laboratory for mandatory post-treatment biomarker analyses. If surgery is unexpectedly cancelled, tissue may be obtained via in-office biopsy if appropriate in the judgment of the surgeon-investigator.

[20] Dispense additional bottle of loperamide, *if necessary*.

[21] Dispense additional bottle of abemaciclib, *if necessary*.

[22] Abemaciclib treatment is planned for 21 days, however may be administered up to a maximum of 28 days if required for logistical/scheduling purposes or due to delay of the planned surgery. The last dose will be the night prior to the planned surgery. The interval between the last dose and surgery must be approximately 12 hours.

[23] If not completed at the pre-op visit.

[24] Documentation must include grade 3 or higher post-operative complications, number of hospital days, and number of ICU days.

Appendix 4. Study Medication Diary

Instructions for Taking Abemaciclib

Please complete the diary each day even if you miss your dose. Document missed doses on the diary. Also write in any side effects that you experience. *It is very important that you maintain this diary and bring it with you when you see your study doctor or nurse. Also bring all tablets and bottles with you.*

Study dosing:

- Take abemaciclib at the same time every morning and evening, approximately 12 hours apart (for example, at 7 am and 7 pm).
- You will take 4 abemaciclib tablets by mouth twice a day.
- The abemaciclib tablets are provided in 50 mg tablets.
- The study dose is 200 mg taken twice per day.

If you experience side effects, you may be given a reduced dose. If this occurs, the study doctor and/or nurse will discuss this with you and inform you of the number of tablets to take.

The following general guidelines should be followed when taking the medication:

- Abemaciclib can be taken with or without food.
  - You should avoid grapefruit products while you are taking this medication.
- The tablets should be swallowed whole.
  - Broken, cracked, or otherwise not intact abemaciclib tablets should not be ingested.
  - If you cannot swallow the tablets whole, refer to the instructions on the following page.
- If vomiting occurs do not make up the dose.
  - Take the medication at the next scheduled time.
  - Make note in this diary when vomiting occurs.
- If you forget to take study dose, it may be made up *within 6 hours* after the time it was supposed to be taken.
  - Once a dose is missed by 6 or more hours, do not make it up; just take the next regularly schedule dose.
  - Document the missed dose in the study diary.
- If you do not take a dose of the medication, please write the reason on the diary.

THE UNIVERSITY OF ARIZONA Cancer Center
The AIM Trial

- If you lose tablets, make note in the comment section on the last page. Specify the number of tablets lost.
- A pill count will be performed when you see the study doctor or nurse.
  - You will be provided 3 bottles with 60 tablets each.
  - It is expected that you will have tablets to return to the study staff since the supply may last longer than the time between when you start taking the tablets and when your surgery is scheduled.
- Bring all bottles and tablets with you each time you see the study doctor or nurse, even if you will continue taking them after the visit.
- There are medications you need to avoid while you are on the study medication. These are listed in the consent form.
  - Make sure you have told the study team about all medications you are currently taking.
  - Also, before you begin any new medications, talk to the study doctor.
- The medication must be stored at 20°C to 25°C (68°F to 77°F).
- Keep the medication out of reach of children.

If you cannot swallow the tablets whole, follow the procedures listed here.

- The tablet may be crushed and dispersed in at least a half ounce of water, and taken completely and immediately within 10 minutes of dispersion in water.
  - The tablet will not dissolve in the water and may taste bitter.
  - Avoid the crushed tablets from being handled by people other than yourself.
- Other than being crushed and dispersed in at least a half ounce water, do not chew, crush, or split tablets before swallowing.
- It is possible to have the tablets administered through a feeding tube such as a G-tube or J-tube. If you think this is necessary, discuss this with the study doctor.

If you have any questions or concerns, contact the study team.

Study Doctor:	Dr. Ricklie Julian	520-694-2873
Research Nurse:		
Study Coordinator:		

THE UNIVERSITY OF ARIZONA Cancer Center	Initials	Participant ID		Year of Birth	Protocol #	AIM Trial Patient Diary
					AIM 1906702170	

Day	Date	Morning dose time	Morning # of Tablets Taken OR Dose Missed	Evening dose time	Evening # of Tablets Taken OR Dose Missed	Comments/Side Effects/Vomiting/Reason Missed
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						

Other comments, including number of tablets lost, if any:

You will be asked to sign this document (both pages, if necessary) once complete to confirm that you or a caregiver completed the diary.

Subject or Caregiver Signature: _____ **Date:** _____

THE UNIVERSITY OF ARIZONA Cancer Center	Initials	Participant ID		Year of Birth	Protocol #	AIM Trial Patient Diary
					AIM 1906702170	

Day	Date	Morning dose time	Morning # of Tablets Taken OR Dose Missed	Evening dose time	Evening # of Tablets Taken OR Dose Missed	Comments/Side Effects/Vomiting/Reason Missed
15						
16						
17						
18						
19						
20						
21						
22						
23						
24						
25						
26						
27						
28						

Other comments, including number of tablets lost, if any:

You will be asked to sign this document (both pages, if necessary) once complete to confirm that you or a caregiver completed the diary.

Subject or Caregiver Signature: _____ **Date:** _____

Appendix 5. Tobacco Assessment Questionnaire

TOBACCO **ASSESSMENT – BASELINE**

REGISTERING INSTITUTION	PARTICIPANT ID	VISIT TYPE	VISIT DATE (MM/DD/YYYY)
_____	_____	_____	__ __/__ __/__ __ __ __

Instructions:
When a number is requested in the response, please enter a whole number (i.e. "4") and not a range or fraction of a number.

Section A. Basic Cigarette Use Information

1. Have you smoked at least 100 cigarettes (5 packs = 100 cigarettes) in your entire life?

☐ Yes
☐ No → **Skip to Section B**
☐ Don't know/Not sure → **Skip to Section B**

2. How old were you when you first smoked a cigarette (even one or two puffs)?

_____ Years old

3. How old were you when you first began smoking cigarettes regularly?

_____ Years old
☐ Check here if you have never smoked cigarettes regularly.

4. How many total years have you smoked (or did you smoke) cigarettes? Do not count any time you may have stayed off cigarettes.

_____ Years (If you smoked less than one year, write "1.")

5. On average when you have smoked, about how many cigarettes do you (or did you) smoke a day? (A pack usually has 20 cigarettes in it).

_____ Number of cigarettes per day

6. Do you NOW smoke cigarettes?

☐ Everyday
☐ Some days
☐ Not at all → **Skip to question 8**

7. How soon after you wake up do you smoke your first cigarette?

☐ Within 30 minutes
☐ After 30 minutes

8. How long has it been since you last smoked a cigarette (even one or two puffs)?

First check which one of the following choices applies to you. Then, if applicable, write a number on the line for how many days, weeks, months, or years it has been since your last cigarette.

☐ I smoked a cigarette today (at least one puff)
☐ 1-7 days → Number of days since last cigarette _____
☐ Less than 1 month → Number of weeks since last cigarette _____
☐ Less than 1 year → Number of months since last cigarette _____
☐ More than 1 year → Number of years since last cigarette _____
☐ Don't know/Don't remember

Section B. Use of Other Forms of Tobacco

9. Have you ever used other forms of tobacco, not including cigarettes?

☐ Yes
☐ No → **Skip to Section C**

10. How often do you/did you use other forms of tobacco?

☐ Every day → Number of times per day _____
☐ Some days → Number of days _____ per ☐ Week ☐ Month ☐ Year

11. Which of the following products have you ever used regularly?
Check all that apply

☐ Cigarettes
☐ E-cigarettes or other electronic nicotine delivery system
☐ Traditional cigars, cigarillos or filtered cigars
☐ Pipes
☐ Waterpipe
☐ Hookah
☐ Clove cigarettes or kreteks
☐ Bidis
☐ Smokeless tobacco, like dip, chew, or snuff
☐ Snus
☐ Paan with tobacco, gutka, zarda, khaini
☐ Other, Please specify:_____

12. If you do not currently use other forms of tobacco, but did in the past, how long has it been since you last used other forms of tobacco regularly?

☐ Within the past month (0 to 1 month ago)
☐ Between 1 and 3 months (1 to 3 months ago)
☐ Between 3 and 6 months (3 to 6 months ago)
☐ Between 6 and 12 months (6 to 12 months ago)
☐ Between 1 and 5 years (1 to 5 years ago)
☐ Between 5 and 15 years (5 to 15 years ago)
☐ More than 15 years ago
☐ Don't know/Not sure
☐ Never used other forms of tobacco regularly

Section C. Second-Hand Smoke Exposure

13. Are you currently living with a smoker?

☐ Yes
☐ No

14. In the past 30 days, have you lived in a place where other people smoked cigarettes indoors?

☐ Yes
☐ No

15. In the past 30 days, have you worked in a place where other people smoked cigarettes indoors?

☐ Yes
☐ No

16. Thinking of all your childhood and adult years, have you ever lived in a place where other people smoked cigarettes indoors?

☐ Yes In total, for about how many years?_____ If less than 1, write "1."
☐ No

17. Thinking of all the years you have worked, have you ever worked in a place where other people smoked cigarettes indoors?

☐ Yes → In total, for about how many years?_____ If less than 1, write "1."
☐ No

Investigator Signature _____ Date __ __/__ __/__ __ __ __ (MM/DD/YYYY)

Investigator Name (please print) _____

Appendix 6. Diarrhea Management Instructions

English Diarrhea Management Instructions

Diarrhea Management Instructions

The AIM Trial
Immune Modulation by Abemaciclib in HPV-Negative Head and Neck Squamous Cell Carcinoma (HNSCC): A Phase II Window Trial

Diarrhea Management Instructions

EXPECT
Diarrhea is a common side effect of abemaciclib and may sometimes be severe. Be on the lookout for cramping and frequent, loose, or watery stools.

PLAN
It is important to plan ahead of time. Have your loperamide (Imodium) on hand on the first day of dosing.

ACT

1. At the first loose stool, take 4 mg (2 capsules) of loperamide and notify physician/research nurse.
2. Once you've taken the first dose of loperamide, start taking 2 mg (1 capsule) of loperamide twice a day, every day, while taking abemaciclib.
3. Do not take more than 4 capsules within a 24-hour period.
4. Drink at least 8-10 glasses of clear fluids any day you have diarrhea.
5. Notify physician/research nurse if diarrhea is not controlled within 24 hours.
6. Contact the physician/research nurse right away if you see blood in your stool or develop a fever or abdominal bloating or swelling.

Note: Loperamide may cause tiredness, drowsiness, and dizziness, and therefore, be cautious when driving or operating machinery.

Study Doctor:	Dr. Ricklie Julian	520-694-2873
Research Nurse:		
Study Coordinator:		