

Duvelisib for Critically Ill Patients With Coronavirus Disease 2019: An Investigator-Initiated, Randomized, Placebo-Controlled, Double-Blind Pilot Trial

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Background. Despite improvements in prevention and treatment, severe coronavirus disease 2019 (COVID-19) is associated with high mortality. Phosphoinositide 3-kinase (PI3K) pathways contribute to cytokine and cell-mediated lung inflammation. We conducted a randomized, placebo-controlled, double-blind pilot trial to determine the feasibility, safety, and preliminary activity of duvelisib, a PI3K δ inhibitor, for the treatment of COVID-19 critical illness.

Methods. We enrolled adults aged ≥ 18 years with a primary diagnosis of COVID-19 with hypoxic respiratory failure, shock, and/or new cardiac disease, without improvement after at least 48 hours of corticosteroid. Participants received duvelisib (25 mg) or placebo for up to 10 days. Participants had daily semi-quantitative viral load measurements performed. Dose modifications were protocol driven due to adverse events (AEs) or logarithmic change in viral load. The primary endpoint was 28-day overall survival (OS). Secondary endpoints included hospital and intensive care unit length of stay, 60-day OS, and duration of critical care interventions. Safety endpoints included viral kinetics and AEs. Exploratory endpoints included serial cytokine measurements and cytometric analysis.

Results. Fifteen patients were treated in the duvelisib cohort, and 13 in the placebo cohort. OS at 28 days was 67% (95% confidence interval [CI], 38%–88%) compared to 62% (95% CI, 32%–86%) for placebo ($P = .544$). Sixty-day OS was 60% versus 46%, respectively (hazard ratio, 0.66 [95% CI, .22–1.96]; $P = .454$). Other secondary outcomes were comparable. Duvelisib was associated with lower inflammatory cytokines.

Conclusions. In this pilot study, duvelisib did not significantly improve 28-day OS compared to placebo for severe COVID-19. Duvelisib appeared safe in this critically ill population and was associated with reduction in cytokines implicated in COVID-19 and acute respiratory distress syndrome, supporting further investigation.

Clinical Trials Registration. NCT04372602.

Keywords. ARDS; COVID-19; cytokine storm; duvelisib; PI3K inhibition.

Over the course of the coronavirus disease 2019 (COVID-19) pandemic, treatment strategies have evolved significantly [1–4]. With the advent of highly effective vaccines, the incidence

of severe disease has dramatically improved [5]. Nevertheless, those who develop severe and critical illness as a result of poor response to vaccination or vaccine hesitancy still suffer from excessive morbidity and mortality [6–8].

Dexamethasone became the first intervention to yield a significant survival benefit in such patients, highlighting the role of immunosuppression in curbing disease and preventing death [9]. Acute inflammation was hypothesized and demonstrated to underlie acute respiratory distress syndrome (ARDS) and “cytokine storm” [10, 11]. As such, several other immunosuppressive interventions were examined in clinical trials, some of which have achieved regulatory approval based on metrics such as decreased duration of ventilation or hospitalization [12–15].

Duvelisib is a potent inhibitor of phosphoinositide 3-kinase delta and gamma (PI3K δ) and used for treatment of chronic

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lymphocytic leukemia, follicular lymphoma, and peripheral T-cell lymphomas [16, 17]. Disruption of many PI3K-dependent pathways, including those involved in cellular metabolism, cell cycling, and DNA repair, leads to both potent anti-neoplastic and anti-inflammatory effects. Preclinical research evaluating the anti-inflammatory effects of duvelisib demonstrated its ability to significantly downregulate expression of inflammatory cytokines and chemokines such as granulocyte and granulocyte/macrophage colony-stimulating factors, macrophage inflammatory protein-1 α and -1 β , and several chemotactic factors [18]. These same inflammatory molecules are upregulated in ARDS and COVID-19 [19–21]. Several murine models of acute lung inflammation and sepsis have demonstrated improved markers of inflammation and survival of mice treated with PI3K inhibitors [22]. The original severe acute respiratory syndrome coronavirus (SARS-CoV) had demonstrated the ability to hijack the PI3K signaling machinery to persist within human airway cells [23–26]. Additionally, in the context of neoplasia, duvelisib polarizes macrophages to the M1 phenotype, which have effector functions in contrast to the proinflammatory M2 phenotype [17]. Moreover, the PI3K cascade can be triggered by JAK-dependent and independent mechanisms, with interleukin (IL)-2 directly inducing a physical interaction between JAK2 and PI3K to mediate these cascades, with significant implications for mitigating inflammation given the role of JAK inhibitors and IL-2 disruption in COVID-19 and other hyperinflammatory states [27].

Based on these potential mechanisms of anti-inflammatory and antiviral activity, along with the continued excessive mortality for this critically ill population, we conducted an investigator-initiated, randomized, double-blind pilot trial of duvelisib versus placebo in patients with COVID-19–related critical illness in order to assess the impact of duvelisib on 28-day mortality and other critical illness outcomes.

METHODS

Patient Consent Statement

Between November 2020 and January 2022, we enrolled participants at 2 hospitals within the BJC HealthCare system. The protocol was approved by their respective institutional review boards and conducted in compliance with the principles of the Declaration of Helsinki. All participants or their legally authorized representatives provided written informed consent.

Trial Design and Patients

Eligible participants were 18 years or older who were hospitalized with a primary diagnosis of severe COVID-19, which required (1) a positive reverse-transcription polymerase chain reaction (RT-PCR) test for SARS-CoV-2 RNA collected from either the upper or lower respiratory tract; and (2) severe disease considered attributable to COVID-19 as manifested by

any of the following: (i) pulmonary infiltrates with $\geq 50\%$ lung involvement; (ii) respiratory failure requiring invasive mechanical ventilation, noninvasive mechanical ventilation, supplementary oxygen with ≥ 6 L/minute, or extracorporeal membrane oxygenation (ECMO); (iii) shock, defined as mean arterial pressure ≤ 65 mm Hg unresponsive to 25 mL/kg isotonic intravenous fluid resuscitation and/or requiring vasopressor support; and (iv) cardiac dysfunction defined by new global systolic dysfunction with ejection fraction $\leq 40\%$ or Takotsubo cardiomyopathy. Participants were required to have absolute neutrophil count $\geq 1000/\mu\text{L}$ and platelet count $\geq 50\,000/\mu\text{L}$ without growth factor or transfusion support for 7 days prior to screening, have a creatinine clearance ≥ 15 mL/minute or be receiving renal replacement therapy (RRT), and have aminotransferase levels < 3 times the upper limit of normal. Additionally, participants were required to have enteral access for study drug administration. Given the proposed anti-inflammatory mechanism of action of duvelisib and the potential for delayed or subacute inflammatory manifestations (eg, ARDS in chronically immunosuppressed patients), patients with critical illness definitively attributable to COVID-19 could be enrolled without limit as to the timing of their initial COVID-19 diagnosis. Patients were excluded if they had a known allergy or intolerance to duvelisib or another PI3K inhibitor, were pregnant or breastfeeding, or had a known or suspected active viral, bacterial, or fungal infection at the time of screening. Cytomegalovirus (CMV) seroconversion was assessed at screening and those who were seropositive required a negative plasma CMV PCR test in order to be eligible.

Procedures

Eligible patients underwent 1:1 randomization to receive either duvelisib or placebo. Investigators and participants were blinded to allocation. Participants were allocated to study cohort in blocks of 10. Computerized random sequence generation assigned participants to matching study drug kits, and the master list was held by third-party individuals uninvolved in the direct conduct of the study. Participants were not stratified by baseline characteristics. Verastem Oncology provided the duvelisib and placebo capsules and funding for the study. Starting on day 1, participants received either duvelisib 25 mg or placebo, orally or per tube.

Participants were monitored and evaluated at least daily for safety and toxicity according to Common Terminology Criteria for Adverse Events of the National Cancer Institute version 5.0. Adverse events (AEs) that were considered at least possibly related to the study drug resulted in protocol-driven dose holds and modifications. All participants received *Pneumocystis jirovecii* pneumonia (PJP) prophylaxis (trimethoprim-sulfamethoxazole or atovaquone) until post-treatment absolute CD4⁺ T-cell count was > 200 cells/ μL or 30 days after study drug discontinuation.

All participants received therapy that was deemed standard of care (SOC) at the time of their respective enrollment. Participants were required to have completed at least a 10-day course of corticosteroid (dexamethasone 6 mg or equivalent) or were required to start corticosteroids at screening and receive corticosteroids for at least 48 hours prior to first study drug administration, unless contraindicated. As the SOC evolved throughout the pandemic, we included patients who had SOC therapies prior to enrollment, and permitted the continuation of remdesivir and corticosteroids to complete their prescribed courses. We discouraged the initiation of other anti-inflammatory medications once subjects initiated the study treatment and required principal investigator approval. Prior investigational therapy was permitted but prohibited after study enrollment. Critical care practices were conducted per institutional guidelines.

Participants continued study therapy for up to 10 days. Those with rapid improvement allowing for hospital discharge could discontinue therapy early. Additionally, discontinuation could occur in the event of withdrawal of consent, AE, or change in participants' conditions that, in the judgment of the investigator or provider, prohibited further continuation. An unblinded, independent data and safety monitoring committee reviewed the patient outcomes data on a quarterly basis.

Viral Load Monitoring and Dose Modifications

Due to concerns the Washington University COVID-19 study oversight board had over the administration of an immunosuppressant to participants with a viral infection, the study protocol included daily nasopharyngeal or oropharyngeal sampling in order to quantitate SARS-CoV-2 viral load and make study drug dose modifications in response to viral load elevations. At study inception, only nasopharyngeal samples were utilized for viral kinetics, although oropharyngeal samples were permitted following internal validation of that sample site on the RT-PCR machinery. Lower respiratory tract sampling was not utilized for viral load monitoring due to the inability to repeatedly sample this site. We used the same sample site repeatedly for inpatient comparisons and trends. We permitted switching samples sites only in situations where the primary sample site was no longer feasible (eg, nasopharyngeal bleeding), although this occurred infrequently. Samples were run on US Food and Drug Administration–approved, Clinical Laboratory Improvement Amendments–certified RT-PCR equipment and, while different platforms were used throughout the study, individual participants' samples were run on the same platform to ensure comparability. In the rare situation in which a platform became unavailable, a participant's subsequent samples would be run on the alternate platform. Viral loads were compared only in an inpatient fashion to establish trends and were calculated based on the known cycle limit of the assay and assumed efficiency of the reaction.

All participants had 3 consecutive pretreatment samples at screening, day –1, and day 1, prior to the first-dose administration, in order to establish a baseline trajectory of viral kinetics. After subjects initiated study therapy, they had daily sampling performed. Viral load increases of >0.5 log on 2 consecutive days or >1 log increase in 1 day could potentially trigger dose holds, if deemed at least potentially attributable to study treatment. If viral load elevations were suspected to be at least potentially related to study treatment, treatment was held until it returned to baseline, and reduced 1 dose level when reinitiated. Principal investigator approval was required for reinitiation of study treatment. The starting dose of duvelisib/placebo was 25 mg twice daily. One dose reduction was to 15 mg twice daily, after which the drug would be discontinued in the event of a subsequent AE or viral load shift requiring dose modification. Viral load increases that occurred below the accurate level of detection (cycle threshold >34) were not acted upon. On occasion, viral load testing or reporting was delayed in which case participants could continue to receive treatment if deemed safe by the investigators. If 2 viral load values were reported simultaneously, the one most recently sampled guided treatment decisions.

Outcomes

The primary endpoint was 28-day overall survival (OS), defined as the proportion of participants alive on day 29. Secondary efficacy endpoints included hospital length of stay (LOS), intensive care unit LOS, duration on mechanical ventilation, duration on vasopressors, duration on RRT, and 60-day OS. Safety and tolerability of duvelisib in this population was the secondary objective, assessed through endpoints of viral kinetics and incidence of adverse events. Exploratory endpoints included serial cytokine measurements and flow cytometry–based measurements of immunocompetence and effector cell activation.

Statistical Analysis

The characteristics of both cohorts were described and bivariate analysis was performed comparing the outcomes using the Fisher exact test. At the time of trial design, outcome data of COVID-19 were still developing, but observational studies suggested that 28-day OS was poor, ranging from approximately 10% to 40% [4, 28]. Sample size considerations were largely based on feasibility and detecting hypothesis-generating, clinically meaningful effect. For duvelisib to have meaningful efficacy, we estimated that 28-day OS would need to be $>60\%$. Based on this assumption, a priori sample size calculations suggested that a sample of 28 participants, approximately 14 in each arm, would allow us to detect an improvement in 28-day OS from approximately 20% to 60% with $>80\%$ power, with a 1-sided P value $<.20$ being considered statistically significant for the primary endpoint. OS was compared using Fisher exact test

and Cox regression analysis. Analyses were conducted using IBM SPSS version 24 (Armonk, New York).

Correlative Studies

Serum cytokines were analyzed as we previously described (see [Supplementary Methods](#)) [29]. Flow cytometry of cryopreserved peripheral blood mononuclear cells (PBMCs) was performed as previously described ([Supplementary Methods](#)) [30]. The fluorochrome-labeled antibodies used for flow cytometry are listed in [Supplementary Table 1](#). Statistical analyses for correlative studies are described in the [Supplementary Methods](#).

RESULTS

Between November 2020 and January 2022, 42 patients provided informed consent, and 13 were ineligible. Twenty-nine participants were randomized to either duvelisib or placebo. One patient randomized to placebo withdrew consent prior to initiation of therapy. Due to this and blocked allocation, 15 participants received duvelisib whereas 13 received placebo. Baseline characteristics were well-balanced, although a numerically higher percentage of patients who received placebo had advanced renal dysfunction, which may have reflected a lower rate of remdesivir usage ([Table 1](#)). Two patients (1 on each arm) were receiving remdesivir at study enrollment and completed their courses shortly after starting study treatment. Fourteen patients (6 on duvelisib and 8 on placebo) had received an adequate course of corticosteroids prior to enrollment and did not restart corticosteroids. Fourteen patients (8 in the duvelisib arm and 6 in the placebo arm) received corticosteroids while on study treatment, with 10 continuing a course started prior to enrollment, 2 restarting a course due to the prior course being considered suboptimal, and 2 starting dexamethasone de novo 48 hours prior to study treatment initiation.

Efficacy

At day 29, 67% of those in the duvelisib arm and 62% of those in the placebo arm were alive ($P = .544$). The estimated 28-day survival when treated with duvelisib is 38%–88% compared to 32%–86% receiving placebo. There was a trend toward improved 60-day OS in the duvelisib arm ([Figure 1](#)); at day 60, 60% of those in the duvelisib arm and 46% of those in the placebo arm were alive ($P = .362$). In comparison to placebo, the 60-day hazard ratio for death of those on duvelisib was 0.66 (95% confidence interval [CI], .22–1.96; $P = .454$). Secondary efficacy endpoints were comparable between the 2 cohorts ([Table 2](#)).

Safety

Three patients withdrew from the study prematurely due to worsening clinical status. AEs occurred at similar rates between

Table 1. Baseline Demographic, Disease, and Treatment Characteristics of Patients Randomized to Duvelisib or Placebo

Characteristic	Duvelisib (n = 15)	Placebo (n = 13)
Age, y, median (range)	63 (26–79)	64 (45–74)
Race/Ethnicity		
Black	6 (40)	5 (38)
White	9 (60)	8 (62)
Sex		
Female	6 (40)	5 (38)
Male	9 (60)	8 (62)
Time from diagnosis to enrollment, d, median (range)	14 (2–60)	14 (2–36)
Predominant local circulating variant at enrollment		
No predominant variant (Nov 2020–Mar 2021)	6 (40)	6 (46)
Alpha (Apr–Jun 2021)	4 (27)	4 (31)
Delta (Jul–Nov 2021)	5 (33)	1 (8)
Omicron (Dec 2021 to Jan 2022)	0 (0)	2 (15)
Status at enrollment		
Mechanical ventilation	9 (60)	9 (69)
Vasopressors	3 (20)	5 (38)
Renal replacement therapy	1 (7)	4 (31)
Prior therapy		
Vaccination	6 (40)	3 (23)
Corticosteroid	15 (100)	11 (85)
Remdesivir	13 (87)	9 (69)
Tocilizumab	4 (27)	4 (31)
Baricitinib	1 (7)	1 (8)
Convalescent plasma	5 (33)	4 (31)
Extracorporeal membrane oxygenation	4 (27)	4 (31)

Data are presented as No. (%) unless otherwise indicated.

arms ([Supplementary Table 2](#)). Among AEs of special interest, 1 patient developed colitis deemed to be stercoral colitis and unrelated to treatment. All respiratory failure was secondary to COVID-19, no pneumonitis. No CMV reactivation or PJP occurred. Dose modifications occurred in response to increasing viral loads ($n = 7$), nasopharyngeal bleeding ($n = 1$), worsening respiratory status ($n = 1$), hyperkalemia ($n = 1$), alkaline phosphatase ($n = 1$), and hypotension ($n = 1$). Eighty percent of deaths were due to respiratory failure. In the placebo arm, 1 patient died of *Pseudomonas* pneumonia and 1 died of cholecystitis.

Immune Repertoire and Cytokine Analysis

Serum Cytokine Levels

A multiplexed bead assay was used to measure the serum concentrations of 30 cytokines or chemokines immediately before (day 0) and 2, 4, 8, 10, 15, and 28 days after initiation of placebo or duvelisib treatment. Five analytes exhibited a significant decrease at a single timepoint upon treatment with duvelisib; IL-5, IL-6, CCL3, and interferon- $\alpha 2$ were all decreased on day 4 whereas macrophage colony-stimulating factor was reduced on day 15 ([Figure 2A](#)). Furthermore, 5 analytes (CCL2, CXCL9, IL-1R α , IL-8, and tumor necrosis factor alpha [TNF- α]) were significantly decreased upon treatment with duvelisib in a groupwise comparison ([Figure 2B](#)). There were no

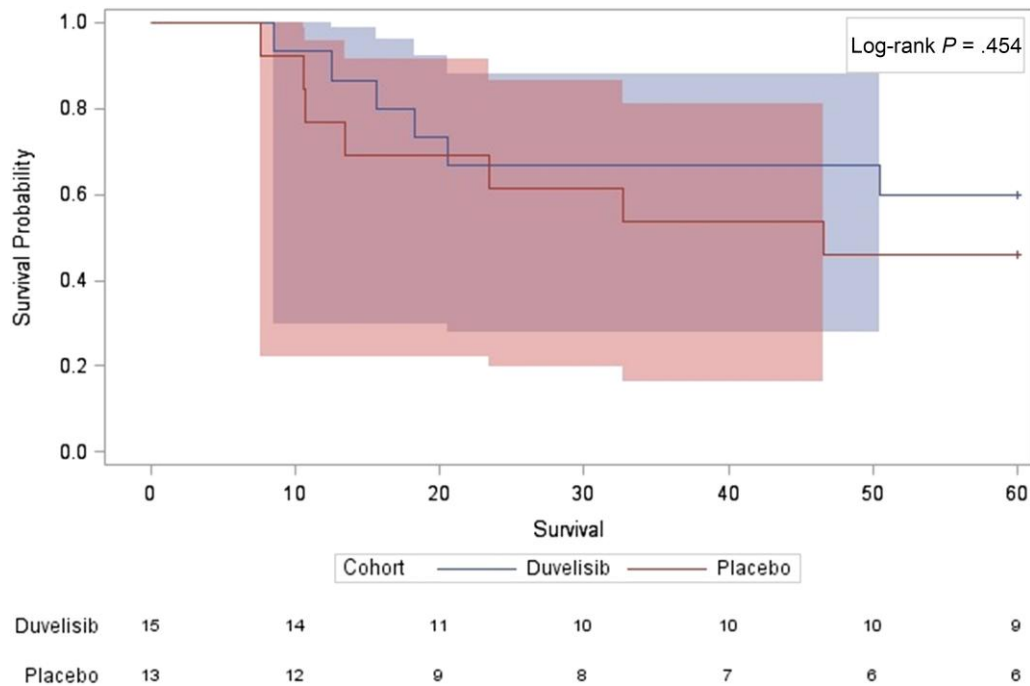


Figure 1. Kaplan-Meier curve comparing overall survival between the duvelisib and placebo cohorts (with 95% confidence intervals).

Table 2. Primary and Secondary Efficacy Endpoints at Day 29

Endpoint	Duvelisib (n = 15)	Placebo (n = 13)	P Value (1-Sided Exact)
Alive	10/15 (67)	7/13 (62)	.554
Weaned from ventilator	5/9 (56)	5/9 (56)	.362
Median time on ventilator	25 d	20 d	.919
Weaned from vasopressors	1/3 (33)	2/5 (40)	.681
Median time on vasopressors	8 d	18 d	.910
Weaned from RRT	0/1 (0)	1/4 (25)	.714
Median time on RRT	28 d	19 d	UTQ
Transferred from ICU	5/15 (33)	4/12 (33)	1.00
Median time in ICU	20 d	15 d	.894
Discharged from hospital	5/15 (33)	4/13 (31)	.885
Median time in hospital	18 d	23 d	.834

Data are presented as no./No. (%) unless otherwise indicated.

Abbreviations: ICU, intensive care unit; RRT, renal replacement therapy; UTQ, unable to quantify.

significant differences between placebo versus duvelisib treatment groups with the remaining 20 analytes (Supplementary Figure 1).

PBMC Subsets After Duvelisib Treatment

To explore whether duvelisib treatment was associated with modulation of a particular cell subset or phenotype, flow cytometry was performed on peripheral blood samples collected at baseline (day 0) and 2, 4, 8, 10, 15, and 28 days after initiation of placebo or duvelisib treatment. Although similar percentages

of circulating monocytes, basophils, dendritic, natural killer (NK), natural killer T (NKT), and T cells were observed, participants treated with duvelisib exhibited significantly reduced percentages of B cells compared to the placebo-treated controls (Figure 3A). This decreased percentage of B cells in the duvelisib-treated cohort was primarily within the IgD⁺CD27⁻ naive B-cell subset (Figure 3B), existed at baseline (day 0, pre-duvelisib), and remained throughout the 28-day observation period (Figure 3A and 3B). However, no significant differences in the absolute numbers of circulating B cells, monocytes, basophils, dendritic, NK, NKT, and T cells were observed upon treatment with duvelisib (Supplementary Figure 2). Extended immunophenotyping of the CD4 and CD8 T-cell subsets revealed no significant differences in the relative percentages or absolute numbers of naive (T_n), central memory (T_{cm}), effector memory (T_{em}), and effector memory CD45RA⁺ (T_{emra}) T cells (Supplementary Figure 3) nor CD4 regulatory T-cell or helper T-cell subsets after duvelisib treatment (Supplementary Figure 4). Likewise, although the relative percentage of CD141⁺CX3CR1⁻ classical monocytes was decreased 4 and 8 days after starting duvelisib administration (Supplementary Figure 5A), the absolute numbers of this circulating subset, as well as other classical and intermediate monocyte subsets, were not significantly altered (Supplementary Figure 5B).

T-Cell Phenotype After Duvelisib Treatment

We also included antibodies in our flow cytometric analyses to evaluate T-cell activation (HLA-DR, CD38, CD69, CD25, KI67,

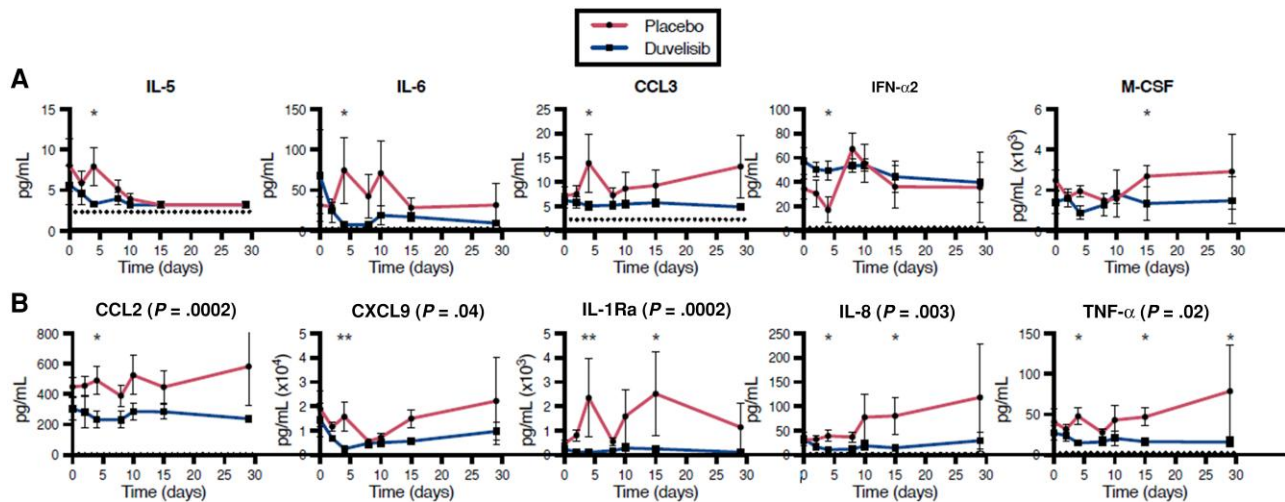


Figure 2. Multiplex bead assay on patient serum at baseline and several timepoints after treatment with either duvelisib or placebo measuring the concentration of 30 cytokines or chemokines. *A*, Analytes with a significant decrease at a single timepoint from duvelisib. *B*, Analytes that were significantly decreased upon duvelisib treatment in a groupwise comparison. [Supplementary Figure 1](#) displays the remaining 20 analytes with no significant differences. * $P < .05$, ** $P < .001$. Abbreviations: IFN, interferon; IL, interleukin; M-CSF, macrophage colony-stimulating factor; TNF, tumor necrosis factor.

perforin, granzyme A and B), exhaustion (PD1, Tim3, Lag3, TIGIT, CTLA-4), and/or senescence (KLRG1, CD57) in all of the major T-cell subsets. No significant differences were detected in the percentage of CD4 ([Supplementary Figure 6](#)) or CD8 ([Supplementary Figure 7](#)) T cells expressing the 15 different activation, exhaustion, and/or senescence markers. We next evaluated these phenotypic markers on the Tn, Tcm, Tem, and Temra CD4 ([Supplementary Figures 8–11](#)) and CD8 ([Supplementary Figures 12–15](#)) T-cell subsets. CD4 Tem and Temra cells exhibited slightly decreased expression of Tim3 and KI67, respectively, at baseline and during the first 10 days of duvelisib treatment ([Supplementary Figures 10–11](#)). Likewise, fewer activated (HLA-DR⁺CD38⁺) and Lag3-expressing CD8 Tem were observed in the duvelisib-treated cohort ([Supplementary Figure 14](#)). Although they represent <1% of the total CD45⁺ circulating cells ([Figure 3A](#)), the greatest phenotypic differences were observed within the T-cell receptor (TCR)- $\gamma\delta$ T-cell subset. Specifically, the percentage of activated TCR- $\gamma\delta$ T cells, as determined by KI67 or HLA-DR/CD38 coexpression, decreased from >40% in the placebo cohort to <1% in the duvelisib-treated participants at 15 days after initiation of drug treatment ([Supplementary Figure 16](#)). No significant phenotypic changes were observed within the CD4 or CD8 NKT subsets ([Supplementary Figures 17–18](#)).

Viral Kinetics

Monitoring viral load kinetics proved feasible and actionable. Dose modifications occurred in 7 participants in response to uptrending viral load. However, there were no trends or significant differences in the trajectory between treatment arms and

no observed influence of duvelisib on viral load kinetics ([Supplementary Figure 19](#)).

DISCUSSION

In this investigator-initiated, randomized, double-blind pilot trial in patients with COVID-19–related critical illness, duvelisib was not associated with improved 28-day OS compared to placebo. This study has inherent limitations that are important for contextualization of any conclusions. As this was an investigator-initiated pilot study, the main objectives were to demonstrate the safety and feasibility of duvelisib therapy for severe COVID-19, and to perform robust correlatives and exploratory analysis on efficacy in order to potentially support larger studies. Sample size was calculated based on the only available data at study inception, which was survival rate, and influenced by feasibility and fiscal constraints. To maintain clinical equipoise, regulatory bodies required the trial be double-blinded and placebo-controlled, which further limited sample size as often such pilot studies are single-armed and uncontrolled. We acknowledge that the population size for both arms is too small to draw conclusions relating to efficacy. The population examined represented a highly refractory cohort of patients, many of whom were excluded from other ongoing clinical trials due to exceedingly high levels of support (eg, ECMO, vasopressors, continuous RRT). Additionally, several participants had an earlier diagnosis of COVID-19 with prolonged ARDS, which introduced confounders such as prior therapies and additional comorbidities. Based on the putative effects of PI3K inhibition on inflammatory cells and cytokines relating to ARDS more so than an antiviral effect, we thought it

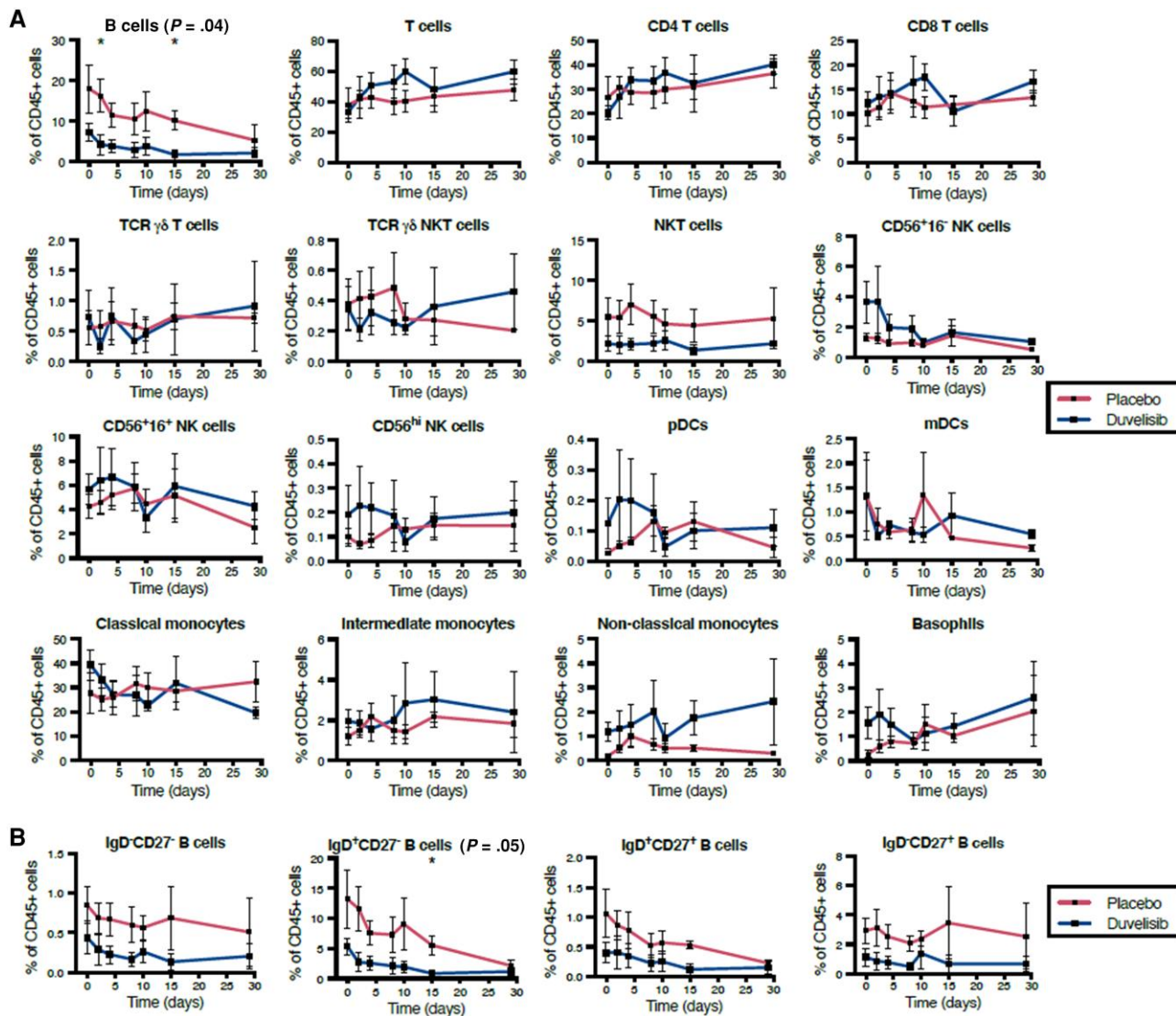


Figure 3. A, Flow cytometric analysis of peripheral blood mononuclear cells (PBMCs) demonstrated that B-cell populations were fewer among those treated with duvelisib but had preceded initiation of treatment. There were no significant differences in proportion of other immune effector cells. B, Differences in B cells were predominantly among the IgD⁺CD27⁻ naive B-cell subset. * $P < .05$. Abbreviations: mDC, myeloid dendritic cells; NK, natural killer cell; NKT, natural killer T cell; pDC, plasmacytoid dendritic cells; TCR, T-cell receptor.

necessary to include participants who had COVID-19–related ARDS that persisted even for an extended duration, and anecdotally saw radiographic and clinical improvement in several such participants who ended up receiving duvelisib (Supplementary Figure 20). However, the heterogeneity in prior therapies and disease specifics confounds the interpretation of correlative and survival outcomes, and we acknowledge that anecdotal evidence does not translate to actual drug activity.

Despite these limitations, we demonstrated, in a placebo-controlled, double-blind fashion, the feasibility and safety of duvelisib for the treatment of severe COVID-19. Adverse events occurred at a similar frequency and severity between

duvelisib and placebo without any AEs of special interest occurring with which duvelisib has been associated in the oncologic setting. We did not observe a positive or negative impact of duvelisib on 28-day or 60-day OS.

Duvelisib was not posited to directly contribute to decreased viral replication, although there was concern that immunosuppression could escalate viral replication. We demonstrated the feasibility of utilizing semi-quantitative viral kinetic monitoring with protocol-driven dose adjustments in response to potentially consequential changes. We encountered several challenges including supply rationing and hemorrhagic complications from naso- and oropharyngeal sampling in critically ill participants. Nevertheless, we carried this out with high

fidelity. Ultimately, the viral kinetics and dose modifications relating to up-trending viral loads were comparable between duvelisib and placebo, providing reassurance that our intervention did not significantly impair the immune response to viral replication.

Our exploratory correlatives suggest that duvelisib is associated with downregulation of these ARDS-associated cytokines in participants with SARS-CoV-2 acute lung injury, in addition to those classically associated with “cytokine storm” such as IL-6 and TNF- α , as seen in the oncologic setting. CXCL9 is detected in lavage samples from participants with ARDS, and Callahan et al demonstrated SARS-CoV-2-mediated transcriptional induction of this proinflammatory cytokine which is mitigated by PI3K/Akt inhibition [31, 32]. CCL2 and IL-8 have long been implicated in neutrophilic chemotaxis contributing to the hyperinflammatory pathogenesis of both COVID and non-COVID ARDS [33, 34]. A larger study examining duvelisib therapy at an earlier disease stage would provide purer results on its impact on inflammation, since there was heterogeneity in prior therapies and corticosteroid timing.

The impact of duvelisib on PBMCs within this study is less conclusive. Those treated with duvelisib had lower B-cell percentages including IgD⁺CD27⁻ B cells, although this included the pretreatment samples, which may be related to prior B-cell-depleting therapies for other diseases (eg, rituximab). While the difference was predominantly seen among naive B cells, future study of duvelisib in COVID-19 ARDS should examine the impact on memory B cells and antiviral titers among those previously vaccinated. Duvelisib appears to potentially modulate the activation and exhaustion states of CD8⁺ Tem, which have been implicated in the severity and resolution of COVID-19, although studies on bronchioalveolar lavage fluid could provide more insight [35, 36].

In conclusion, we conducted an investigator-initiated, COVID-19 clinical trial in critically ill participants with highly granular and informative correlative data encompassing viral kinetics and immune activity. We demonstrated the safety of duvelisib in comparison to placebo. While we did not observe any impact of duvelisib on survival, a larger clinical trial in participants with COVID-19 at an earlier stage may see such benefits, and the safety and correlative data from our trial would support such an investigation. Given that the mechanism of duvelisib-mediated attenuation of the inflammatory response is not virus-specific, and preclinical data support PI3K inhibition in models of sepsis and ARDS, duvelisib could also be investigated in realms of critical care medicine beyond COVID-19.

Supplementary Data

Supplementary materials are available at *Open Forum Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

Author contributions. S. R. G., F. C., M. F., Z. X., Z. I., N. M., E. B., B. C., B. P., E. R. D., and J. F. D. designed the study and contributed to the analysis. M. P. R., S. C., L. G., E. S., N. W., and J. R. conducted the correlative studies. F. G. conducted statistical analysis on the correlative studies. All authors contributed to, reviewed, and edited the manuscript.

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Potential conflicts of interest. J. P. reports employment with Verastem Oncology Inc. All other authors report no potential conflicts.

References

1. Murakami N, Hayden R, Hills T, et al. Therapeutic advances in COVID-19. *Nat Rev Nephrol* 2023; 19:38–52.
2. Murthy S, Gomersall CD, Fowler RA. Care for critically ill patients with COVID-19. *JAMA* 2020; 323:1499–500.
3. Bhimraj A, Morgan RL, Hirsch Shumaker A, et al. Infectious Diseases Society of America guidelines on the treatment and management of patients with COVID-19. Available at: www.idsociety.org/COVID19guidelines. Accessed 3 January 2021.
4. Richardson S, Hirsch JS, Narasimhan M, et al. Presenting characteristics, comorbidities, and outcomes among 5700 patients hospitalized with COVID-19 in the New York City area. *JAMA* 2020; 323:2052–9.
5. Scobie HM, Johnson AG, Suthar AB, et al. Monitoring incidence of COVID-19 cases, hospitalizations, and deaths, by vaccination status—13 U.S. jurisdictions, April 4–July 17, 2021. *MMWR Morb Mortal Wkly Rep* 2021; 70:1284–90.
6. Johnson AG, Amin AB, Ali AR, et al. COVID-19 incidence and death rates among unvaccinated and fully vaccinated adults with and without booster doses during periods of Delta and Omicron variant emergence—25 U.S. jurisdictions, April 4–December 25, 2021. *MMWR Morb Mortal Wkly Rep* 2022; 71:132–8.
7. Ludwig H, Sonneveld P, Facon T, et al. COVID-19 vaccination in patients with multiple myeloma: a consensus of the European Myeloma Network. *Lancet Haematol* 2021; 8:e934–46.
8. Boekel L, Stalman EW, Wieske L, et al. Breakthrough SARS-CoV-2 infections with the Delta (B.1.617.2) variant in vaccinated patients with immune-mediated inflammatory diseases using immunosuppressants: a substudy of two prospective cohort studies. *Lancet Rheumatol* 2022; 4:e417–29.
9. Dexamethasone in hospitalized patients with Covid-19. *N Engl J Med* 2021; 384:693–704.
10. Mehta P, McAuley DF, Brown M, Sanchez E, Tattersall RS, Manson JJ. COVID-19: consider cytokine storm syndromes and immunosuppression. *Lancet* 2020; 395:1033–4.
11. Stebbing J, Phelan A, Griffin I, et al. COVID-19: combining antiviral and anti-inflammatory treatments. *Lancet Infect Dis* 2020; 20:400–2.
12. Rosas IO, Bräu N, Waters M, et al. Tocilizumab in hospitalized patients with severe Covid-19 pneumonia. *N Engl J Med* 2021; 384:1503–16.
13. Kalil AC, Patterson TF, Mehta AK, et al. Baricitinib plus remdesivir for hospitalized adults with Covid-19. *N Engl J Med* 2021; 384:795–807.
14. Selvaraj V, Finn A, Lal A, Khan MS, Dapaah-Afiriye K, Carino GP. Baricitinib in hospitalised patients with COVID-19: a meta-analysis of randomised controlled trials. *EClinicalMedicine* 2022; 49:101489.
15. Roschewski M, Lionakis MS, Sharman JP, et al. Inhibition of Bruton tyrosine kinase in patients with severe COVID-19. *Sci Immunol* 2020; 5:eabd0110.
16. Vangapandu HV, Jain N, Gandhi V. Duvelisib: a phosphoinositide-3 kinase δ/γ inhibitor for chronic lymphocytic leukemia. *Expert Opin Investig Drugs* 2017; 26:625–32.
17. Horwitz SM, Koch R, Porcu P, et al. Activity of the PI3K- δ, γ inhibitor duvelisib in a phase 1 trial and preclinical models of T-cell lymphoma. *Blood* 2018; 131:888–98.
18. Xie S, Chen M, Yan B, He X, Chen X, Li D. Identification of a role for the PI3K/AKT/mTOR signaling pathway in innate immune cells. *PLoS One* 2014; 9:e94496.
19. Montazersaheb S, Hosseiniyan Khatibi SM, Hejazi MS, et al. COVID-19 infection: an overview on cytokine storm and related interventions. *Virol J* 2022; 19:92.
20. Han S, Mallampalli RK. The acute respiratory distress syndrome: from mechanism to translation. *J Immunol* 2015; 194:855–60.
21. Margaria JP, Moretta L, Alves-Filho JC, Hirsch E. PI3K signaling in mechanisms and treatments of pulmonary fibrosis following sepsis and acute lung injury. *Biomedicine* 2022; 10:756.
22. Winkler DG, Faia KL, DiNitto JP, et al. PI3K- δ and PI3K- γ inhibition by IPI-145 abrogates immune responses and suppresses activity in autoimmune and inflammatory disease models. *Chem Biol* 2013; 20:1364–74.

23. Dunn EF, Connor JH. Hijack: the PI3K/Akt pathway in virus replication and pathogenesis. *Prog Mol Biol Transl Sci* **2012**; 106:223–50.
24. Mizutani T, Fukushi S, Saijo M, Kurane I, Morikawa S. JNK And PI3k/Akt signaling pathways are required for establishing persistent SARS-CoV infection in Vero E6 cells. *Biochim Biophys Acta* **2005**; 1741(1–2):4–10.
25. Fujita A, Kan-O K, Tonai K, et al. Inhibition of PI3K δ enhances poly I:C-induced antiviral responses and inhibits replication of human metapneumovirus in murine lungs and human bronchial epithelial cells. *Front Immunol* **2020**; 11:432.
26. Kindrachuk J, Ork B, Hart BJ, et al. Antiviral potential of ERK/MAPK and PI3K/AKT/mTOR signaling modulation for Middle East respiratory syndrome coronavirus infection as identified by temporal kinome analysis. *Antimicrob Agents Chemother* **2015**; 59:1088–99.
27. Yamada O, Ozaki K, Akiyama M, Kawauchi K. JAK-STAT and JAK-PI3K-mTORC1 pathways regulate telomerase transcriptionally and post-translationally in ATL cells. *Mol Cancer Ther* **2012**; 11:1112–21.
28. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* **2020**; 395:497–506.
29. Abboud R, Keller J, Slade M, et al. Severe cytokine-release syndrome after T cell-replete peripheral blood haploidentical donor transplantation is associated with poor survival and anti-IL-6 therapy is safe and well tolerated. *Biol Blood Marrow Transplant* **2016**; 22:1851–60.
30. Huselton E, Rettig MP, Campbell K, et al. Combination of dociparstat sodium (DSTAT), a CXCL12/CXCR4 inhibitor, with azacitidine for the treatment of hypomethylating agent refractory AML and MDS. *Leuk Res* **2021**; 110:106713.
31. Callahan V, Hawks S, Crawford MA, et al. The pro-inflammatory chemokines CXCL9, CXCL10 and CXCL11 are upregulated following SARS-CoV-2 infection in an AKT-dependent manner. *Viruses* **2021**; 13:1062.
32. Wong CK, Lam CWK, Wu AKL, et al. Plasma inflammatory cytokines and chemokines in severe acute respiratory syndrome. *Clin Exp Immunol* **2004**; 136: 95–103.
33. Williams AE, José RJ, Mercer PF, et al. Evidence for chemokine synergy during neutrophil migration in ARDS. *Thorax* **2017**; 72:66–73.
34. Cesta MC, Zippoli M, Marsiglia C, et al. The role of interleukin-8 in lung inflammation and injury: implications for the management of COVID-19 and hyperinflammatory acute respiratory distress syndrome. *Front Pharmacol* **2022**; 12: 808797.
35. Kusnadi A, Ramírez-Suástegui C, Fajardo V, et al. Severely ill patients with COVID-19 display impaired exhaustion features in SARS-CoV-2-reactive CD8⁺ T cells. *Sci Immunol* **2021**; 6:eabe4782.
36. Rha MS, Shin EC. Activation or exhaustion of CD8⁺ T cells in patients with COVID-19. *Cell Mol Immunol* **2021**; 18:2325–33.