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Ceralasertib-Mediated ATR Inhibition Combined With Olaparib in Advanced Cancers Harboring DNA Damage Response and Repair Alterations (Olaparib Combinations)

Haider Mahdi, MD, MPH[1]; Navid Hafez, MD[2]; Deborah Doroshow, MD[2]; Davendra Sohal, MD[1]; Vickie Keedy, MD[3]; Khanh T. Do, MD[4]; Patricia LoRusso, MD[2]; Juliane Jürgensmeier, MD[2]; Manuel Avedissian, MD[2]; Jeffrey Sklar, MD[2]; Colin Glover, MD[5]; Brunella Felicetti, MD[5]; Emma Dean, PhD[5]; Peter Mortimer, MD[5]; Geoffrey I. Shapiro, MD, PhD[4]; and Joseph Paul Eder, MD[2]

abstract

PURPOSE Poly (ADP-ribose) polymerase (PARP) inhibitors have emerged as promising therapy in cancers with homologous recombination repair deficiency. However, efficacy is limited by both intrinsic and acquired resistance. The Olaparib Combinations basket trial explored olaparib alone and in combination with other homologous recombination–directed targeted therapies. Here, we report the results of the arm in which olaparib was combined with the orally bioavailable ataxia telangiectasia and RAD3-related inhibitor ceralasertib in patients with relapsed or refractory cancers harboring DNA damage response and repair alterations, including patients with *BRCA*-mutated PARP inhibitor–resistant high-grade serous ovarian cancer (HGSOC).

PATIENTS AND METHODS Germline and somatic mutations had to be deleterious by COSMIC or ClinVar for eligibility. Olaparib was administered at 300 mg twice daily and ceralasertib at 160 mg daily on days 1-7 in 28-day cycles until progression or unacceptable toxicities. Primary end points were confirmed complete response (CR) or partial response (PR) rates and clinical benefit rate (CBR; CR + PR + stable disease [SD] at 16 weeks).

RESULTS Twenty-five patients were enrolled, with median four prior therapies. Five patients required dose reductions for myelosuppression. Overall response rate was 8.3% and CBR was 62.5% among the entire cohort. Two of five patients with tumor harboring *ATM* mutation achieved CR or SD ongoing at 24+ months, respectively (CBR 40%). Of seven patients with PARP inhibitor–resistant HGSOC, one achieved PR (–90%) and five had SD ranging 16-72 weeks (CBR 86%).

CONCLUSION Olaparib with ceralasertib demonstrated preliminary activity in *ATM*-mutated tumors and in PARP inhibitor–resistant *BRCA1/2*–mutated HGSOC. These data warrant additional studies to further confirm activity in these settings.

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INTRODUCTION

DNA damage response and repair (DDR) is an essential function to maintain viability in all cells. Homologous recombination (HR) repair is a high-fidelity process used to repair DNA double-strand breaks (DSB), during the S and G_2 phases.[1] Germline mutations in HR genes and other genes involved in DDR and the genes giving rise to Fanconi anemia significantly increase the lifetime risk of certain cancers. HR and DDR pathway genes mutations with accompanying loss of heterozygosity (LOH) have been observed in 17%-21% of patients in large, pancancer data sets.[2,3]

HR deficiency has been shown to be synthetically lethal with inhibition of poly (ADP-ribose) polymerase (PARP) leading to the advent of PARP inhibitors in HR-deficient tumors, particularly in *BRCA*-mutated high-grade serous ovarian cancer (HGSOC), as well as breast, pancreatic, and prostate cancers.[4-6] TCGA data sets have shown that biallelic HR gene inactivation may be present in other cancer types as well and is associated with genomic features of HR deficiency.[7] In HGSOC, approximately 50% are characterized by genetic and epigenetic alterations of the HR pathway genes, particularly *BRCA* genes.[8-10] HR deficiency has been an important therapeutic target in ovarian cancer. In patients with recurrent disease, PARP inhibitors have been used in patients with tumors harboring *BRCA* alterations with response rates typically exceeding 30%.[11-14] Additionally, these agents are now commonly considered irrespective of *BRCA* mutation

ASSOCIATED CONTENT

Protocol

Author affiliations and support information (if applicable) appear at the end of this article.

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CONTEXT

Key Objective

Despite initial benefit from platinum and poly (ADP-ribose) polymerase (PARP) inhibitor–based therapy in high-grade serous ovarian cancer and in other cancers harboring *BRCA* alterations or other homologous recombination repair defects, most tumors develop resistance resulting in treatment failure. Reversing resistance and enhancing response to PARP inhibitors is an area of unmet need.

Knowledge Generated

Inhibition of the ATR-CHK1 pathway reverses restored homologous recombination and compromises replication fork stability in *BRCA*-mutated tumors with acquired PARP inhibitor resistance. Ceralasertib, an ataxia telangiectasia and RAD3-related inhibitor, combined with olaparib in heavily pretreated ovarian cancer patients with *BRCA*-mutated cancers resistant to PARP inhibitors produced an encouraging early clinical benefit signal.

Relevance

Ceralasertib combined with olaparib was well-tolerated and effective in a subset of *BRCA*-mutated patients with PARP inhibitor–resistant high-grade serous ovarian cancer in this exploratory trial and should be further studied in this population.

in the second-line maintenance setting, after a response to a platinum-based chemotherapy.[8-10] Recent work has also examined PARP inhibition in the first-line maintenance setting.[15,16] More common PARP inhibitors use has highlighted the importance of both acquired and de novo resistance. The outlook for patients with HGSOC with acquired or de novo HR proficiency is poor so that reversal of resistance is a pressing clinical problem.

Ataxia telangiectasia and RAD3-related (ATR) is a member of the phosphoinositide 3-kinase-related kinase family. ATR governs checkpoints that serve to ensure cell survival after replication stress or DNA damage. ATR is recruited to stalled replication forks, where it mediates CHK1 activation resulting in cell cycle arrest in S phase. ATR and CHK1 phosphorylate PALB2 and RAD51, respectively, facilitating HR repair.[17,18] ATR also initiates the cascade of events culminating in G2 arrest following DNA damage. Preclinical data have supported the synergism of ATR and PARP inhibition in *BRCA*-mutated PARP inhibitor–sensitive ovarian and breast cancer models.[19] Mechanistically, PARP inhibition leads to G2 accumulation; the addition of ATR inhibition promotes release from G2 with premature mitotic entry with increased chromosomal aberrations and apoptosis.[20]

ATR inhibition is also a promising strategy to overcome PARP inhibitor resistance in *BRCA*-mutated cancers.[20] PARP inhibitor–resistant BRCA-deficient cells are increasingly dependent on ATR for genomic stability and survival. Preclinical data suggest that ATR inhibition targets PARP inhibitor resistance through two potential mechanisms, including disruption of BRCA1-independent RAD51 loading to sites of DSB and reversal of BRCA1-independent replication fork protection.[20] Combined ATR-CHK1 axis and PARP inhibition has been shown to be cooperative in the PARP inhibitor–resistant setting, with synergistic increases in replication fork stalling, DSB, and apoptosis, coupled with compromised HR repair, translating to improved survival in preclinical ovarian cancer models.[20,21]

Like ATR, ataxia telangiectasia-mutated (ATM) has both DNA damage–induced checkpoint and repair functions. ATM deficiency is expected to sensitize malignant cells to ATR inhibition, which has been demonstrated both in preclinical models and in clinical trials.[22-25] Additionally, in preclinical pancreatic and lung cancer models, ATM deficiency also sensitizes to PARP inhibition, suggesting that combined ATR and PARP inhibition may be useful.[26,27] In contrast, other studies showed low sensitivity of ATM-deficient prostate cancers to PARP inhibition.[28,29] Although the role of PARP inhibitor monotherapy in ATM-deficient tumors is not fully clarified, combined ATR and PARP inhibition may still provide clinical benefit.

The Olaparib Combinations (OLAPCO) trial is a basket trial that has explored olaparib alone and in combination with other HR-directed targeted therapies. The objective of this arm of the OLAPCO trial was to assess the efficacy of the combined regimen of olaparib and the ATR inhibitor ceralasertib in previously treated patients with DDR-deficient solid tumors.

PATIENTS AND METHODS

Patient Selection

For the ceralasertib-olaparib arm of OLAPCO (NCT02576444), patients with tumor mutations in HR and other DDR genes were identified by tests performed in a Clinical Laboratory Improvement Amendments–certified laboratory, either locally at one of the participating sites or at a commercial testing facility, before participation in the trial. These platforms included standard local (including Oncomine[30] or Oncopanel[31]) or commercial (including Myriad, FoundationOne, or Tempus) platforms. Patients with tumors harboring deleterious mutations in HR genes (including *BRCA1*, *BRCA2*, *PALB2*, and other genes) and other DNA repair pathway genes (including

ATM and *CHEK2*), as well as mutations in TCA cycle genes implicated in HR defects,[32] were enrolled. Patients with HGSOC harboring germline or somatic mutations in *BRCA1/2* genes and who had prior progression on PARP inhibitors were also permitted to enroll. Germline and somatic mutations had to be deleterious by COSMIC or ClinVar for eligibility.

Eligibility

Eligible patients had to have received standard first-line therapy for metastatic cancer (except for tumors for which no first-line therapy exists) with progressive disease at the time of study entry. Other eligibility criteria include measurable disease by RECIST v1.1 and age ≥ 18 years with life expectancy ≥ 16 weeks. Enrolled patients had Eastern Cooperative Oncology Group performance score of 0-1, adequate hematologic function with no features suggestive of myelodysplastic syndrome/acute myelomonocytic leukemia, and adequate hepatic and renal function. Prior treatment with a PARP inhibitor was required in patients with *BRCA*-mutated HGSOC during the second stage of the study, but there was no specification regarding the duration or response to prior PARP inhibition. The number of prior lines of therapy was not restricted on this study.

Protocol Treatment

Patients received olaparib 300 mg orally twice a day continuously and ceralasertib 160 mg orally on days 1-7 in 28-day cycles until disease progression or unacceptable toxicities. This regimen was based on the recommended phase II dose of ceralasertib that could be combined with full-dose olaparib that was established during a prior phase I study. Toxicities were evaluated using the National Cancer Institute Common Terminology Criteria for Adverse Events version 4.0. Dose reductions for olaparib were 250 mg (dose level –1) and 200 mg (dose level –2) daily, whereas for ceralasertib, dose reductions were 160 mg for day 1-4 for hematologic toxicities or 120 mg oral daily for day 1-7 for nonhematologic toxicities (dose level –1) and 120 mg oral day 1-4 for hematologic or nonhematologic toxicities (dose level –2). Dose reescalation was not permitted.

Ceralasertib and olaparib could be reduced in stepwise fashion for anemia, where dose reduction for olaparib was recommended first, followed by a dose reduction for ceralasertib if the adverse event (AE) recurred. Simultaneous reduction was also allowed depending on the severity and duration of anemia. If a dose reduction was required for neutropenia, leukopenia, or thrombocytopenia, olaparib and ceralasertib were reduced simultaneously because of the greater frequency of these events associated with ceralasertib.

End Points

The primary objectives were to determine overall response rate (ORR) by RECIST v1.1 and clinical benefit rate (CBR), defined as ORR and stable disease (SD) after 16 weeks of treatment.[33] Additional objectives were to determine progression-free survival, duration of ORR and SD, and AEs.

Statistical Design, Sample Size Justification, and Decision Rules

A two-stage accrual design was used.[34,35] A 30% ORR was considered worthy of further study. Initially, 16 eligible patients were to be treated. If there were < 2 responses, accrual would be terminated on the basis of the likelihood of an ORR of ≤ 10%. If ≥ 2 responded in the first stage, the study would continue until 25 patients were treated. This design provided 90% power with a significance level of < .10 (type I error). The second stage was activated with the goal of evaluating more patients with *BRCA*-mutated, PARP inhibitor–resistant HGSOC on the basis of an initial signal observed among the first 16 patients.

Additionally, an early stopping rule for safety was incorporated. If among the first 16 patients, ≥ 4 experienced unacceptable toxicity, enrollment would be terminated early. Unacceptable toxicity was defined as grade 4 hematologic and grade 3 nonhematologic toxicities that failed to resolve to grade 1 despite appropriate supportive care, as defined by Common Terminology Criteria for Adverse Events version 4.0. With this design, the probability of terminating the arm early was .07 if the true but unknown unacceptable toxicity rate was 10% and 0.75 if the true toxicity rate was 30%.

RESULTS

Patient Characteristics

Twenty-five patients were enrolled over 14 months. The median age was 59 years (39-79), including 18 females and seven males. The median number of prior therapies was 4 (0-10). Patient characteristics are presented in Table 1.

Safety and Tolerability

The combined regimen of olaparib and ceralasertib was well-tolerated. Probable or definite treatment-related AEs as judged by the treating investigator are listed in Table 2. All patients had grade 1 AEs that were deemed to be mild and required no dose alterations. These events occurred after many cycles on treatment. Only the most severe grade for each individual patient toxicity is entered. Hematologic toxicity was the most common event as expected from previous clinical trials of each agent. One patient, a woman with germline *BRCA1*-mutated HGSOC, who had received eight prior regimens, experienced grade 3 anemia, grade 4 neutropenia, and thrombocytopenia and required two dose reductions for olaparib and one reduction for ceralasertib. A second patient with grade 4 neutropenia also required a dose reduction for olaparib alone. Three more patients were dose-reduced because of anemia. All adverse hematologic toxicities were reversible. Nonhematologic toxicities were rare and all were ≤ grade 2. No treatment-related deaths occurred. Among the two patients who achieved objective

TABLE 1. Patients' Characteristics (N = 25)

Characteristic	N (range)
Age, years	
Median (range)	59 (39-78)
Sex	
Male	7
Female	18
Prior therapies	
Median (range)	4 (0-10)
Cancer diagnosis	
High-grade serous ovarian cancer	7
Pancreatic ductal adenocarcinoma	6
Castration-resistant prostate cancer	3
ACC	2
Breast cancer	2
Sarcoma	1
Cholangiocarcinoma	1
Colorectal cancer	1
Primary peritoneal	1
Paraganglioma	1

Abbreviation: ACC, adenoid cystic carcinoma.

response, one patient with *BRCA1*-mutated PARP inhibitor–resistant HGSOC required dose reduction but continued to have response to the combination therapy.

Efficacy

The individual germline and somatic mutations of enrolled patients are listed in Table 3, along with diagnosis and clinical outcome. One patient was not evaluable because she withdrew consent after being enrolled. Among the 24 evaluable patients, there was one complete response (CR) and one partial response (ORR 8.3%; Fig 1). These were durable, confirmed responses that occurred among the first 16 patients, allowing the study to progress to the second stage. Thirteen patients (54.2%) had SD for at least 16 weeks with a CBR of 62.5% (Fig 1). The median duration of response was 22 months (18-26+ months), and the median duration of clinical benefit was 5 months (4-26+ months).

Five patients with *ATM* mutations were included. The ORR was 20% (1 of 5) and CBR was 40% (2 of 5, including one patient with durable CR and one patient with durable SD; Fig 2). The CR occurred in a patient with estrogen receptor–positive metastatic breast cancer and a germline *ATM* mutation with LOH. CR was initially achieved at 4 months and has been ongoing for 26+ months. A second patient with primary adenoid cystic carcinoma of minor salivary gland and germline *ATM* mutation with LOH has had an ongoing 22% reduction in target lesions and has also remained on treatment for 26+ months. This patient had previously been treated with surgery for a primary tumor of the sella turcica, proton beam radiation, and cisplatin and has multiple lytic bone metastases. Neither patient has had significant toxicity or required a dose reduction. Two patients with PDAC and *ATM* mutation (unknown LOH status) progressed rapidly. A patient with colon cancer had a 29% reduction in target lesions at 2 months but progressed at 4 months with new lesions.

Seven patients with HGSOC with *BRCA1/2* mutations represented the largest group (Table 4). All patients were heavily pretreated; the median number of prior regimens was 5 (range 2-11). Furthermore, all patients had received 1-3 prior PARP inhibitor–based regimens and had progressed during their most recent PARP inhibitor exposure. The ORR was 14% (1 of 7) and the SD rate was 71% (5 of

TABLE 2. Summary of Treatment-Related AEs

Event	Grade 1, No. (%)	Grade 2, No. (%)	Grade 3, No. (%)	Grade 4, No. (%)
Any AEs	25 (100)	15 (60)	5 (20)	3 (12)
Anemia	4 (16)	6 (24)	3 (12)	0
Neutropenia	1 (4)	2 (8)	1 (4)	2 (8)
Thrombocytopenia	1 (4)	0	1 (4)	1 (4)
Fatigue	4 (16)	3 (12)	0	0
Fever	1 (4)	0	0	0
Nausea or vomiting	3 (12)	1 (4)	0	0
Diarrhea	3 (12)	0	0	0
Dyspepsia	3 (12)	0	0	0
Increased ALT/AST	0	2 (8)	0	0
Increased ALP	1 (4)	1 (4)	0	0
Urticaria/skin rash	1 (4)	0	0	0
Pruritus	1 (4)	0	0	0
Dysgeusia	2 (8)	0	0	0

Abbreviations: AE, adverse event; ALP, alkaline phosphatase.

TABLE 3. Cancer Sites, Mutational Status, and Extent and Duration of Response of Patients Who Received Olaparib and Ceralasertib/AZD6738

Cancer Type	Gene	Mutation	Response	Serious AEs	Duration (months)	Reason Off Study
ACC	*ATM*	p.1069fs, germline LOH	SD	No	26+	
Breast	*ATM*	Exon 61 3′UTRdel germline	CR	Yes	26+	
Pancreatic	*ATM*	Splice 8585-2A>C, germline	PD	No	3	PD
Pancreatic	*ATM*	p.E518fs; p.A2602fs, somatic	PD	Yes, anemia	1	Clinical PD
Colon	*ATM*	p.R2849*, somatic	PD	No	4	PD
Prostate	*BRCA2*	c4638delT, germline	SD	Yes	4	PD
Prostate	*BRCA2*	13q13.1, biallelic del, germline	SD	Yes, anemia	5	PD
Prostate	*BRCA2*	c.2558insA, germline	SD	No	4	PD
Pancreatic	*BRCA1*	p.E23fs*17, germline	PD	No	1	PD
Peritoneal	*BRCA2*	C.4516_4525del11, germline	PD	No	2	PD
Peritoneal	*PALB2*	p.W1038*, germline	PD	Yes, anemia	2	PD
Pancreatic	*PALB2*	c2288_2291del, germline	SD	No	4	Consent withdrawn
Breast	*PALB2*	c20092.delC, germline	PD	No	2	PD
ACC	*PALB2*	F440fs*12, germline	SD	Yes, anemia	14+	
Pancreatic	*MUS81*	c403_404delCA, germline	SD	Yes, anemia	4	Clinical PD
Cholangiocarcinoma	*CHECK2*	P1157T, germline	PD	Yes	1	PD
Myxoid	*IDH1*	R132C, somatic	SD	No	8	PD
Paraganglioma	*SDHD*	c.242T>C, somatic	NE	No	2	Consent withdrawn
Ovarian	*BRCA1*	c3600 del11, germline	SD	No	4	Clinical PD
Ovarian	*BRCA1*	c.2681_2682del, germline	PR	No	18	PD
Ovarian	*BRCA2*	del exons 19-24, germline	PD	Yes	2	PD
Ovarian	*BRCA1*	E23fs*17, germline	SD	No	11	Clinical PD
Ovarian	*BRCA2*	c.1913T>G, somatic	SD	No	8	PD
Ovarian	*BRCA1*	c.3875del4, germline	SD	Yes, anemia	10	PD
Ovarian	*BRCA1*	Loss exon1, germline	SD	No	8	PD

Abbreviations: ACC, adenoid cystic carcinoma; AE, adverse event; CR, complete response; LOH, loss of heterozygosity; NE, non-evaluable; PD, progressive disease; PR, partial response; SD, stable disease.

7), leading to a CBR of 85.7% (6 of 7; Fig 3). The median duration of clinical benefit among the seven patients was 8 months (2-18 months). The duration of benefit from olaparib and ceralasertib exceeded the initial duration of response to the PARP inhibitor in these patients (median 8 months, range 2-18 months *v* 4 months, range 2-12 months; Table 4). Among the five patients with germline *BRCA1* mutations, one had a partial response (–90% tumor reduction for 18 months) and two others had minor responses (–13% for 11 months; –27% for 8 months).

In other solid tumors with pathogenic *BRCA* mutation, three patients had brief periods of SD. Among four patients with *PALB2* mutations, one with metastatic adenoid cystic carcinoma of salivary gland has had ongoing disease stability at 14+ months. Notably, this patient had a 23% increase in pulmonary target lesions in the year preceding study initiation and < 1% increase in these target lesions while receiving olaparib and ceralasertib. Two patients with *MUS81* mutation (presumed to be germline from the family history) and *CHEK2* mutation, respectively, did not derive benefit.

Two patients who had received olaparib monotherapy as part of another cohort in OLAPCO received olaparib and ceralasertib on disease progression. A patient with PDAC harboring *PALB2* mutation received olaparib alone for 11 months previously with SD (23% reduction by RECIST 1.1) but progressed without response on olaparib and ceralasertib. In contrast, a second patient with an *IDH1*-mutated chondrosarcoma had SD for 7 months on olaparib monotherapy and subsequently experienced SD for an additional 7 months on combined olaparib and ceralasertib.

DISCUSSION

The OLAPCO trial is an exploratory basket study that includes several olaparib combinations in genomically targeted patient subsets. In this arm, patients with tumors harboring DDR alterations were treated with combined ceralasertib and olaparib. The regimen used full-dose olaparib along with ceralasertib on the basis of prior phase I data. The ORR was 8.3%, making it a negative trial, although the CBR of 62.5% was promising in this heavily pretreated population. Responses and instances of clinical

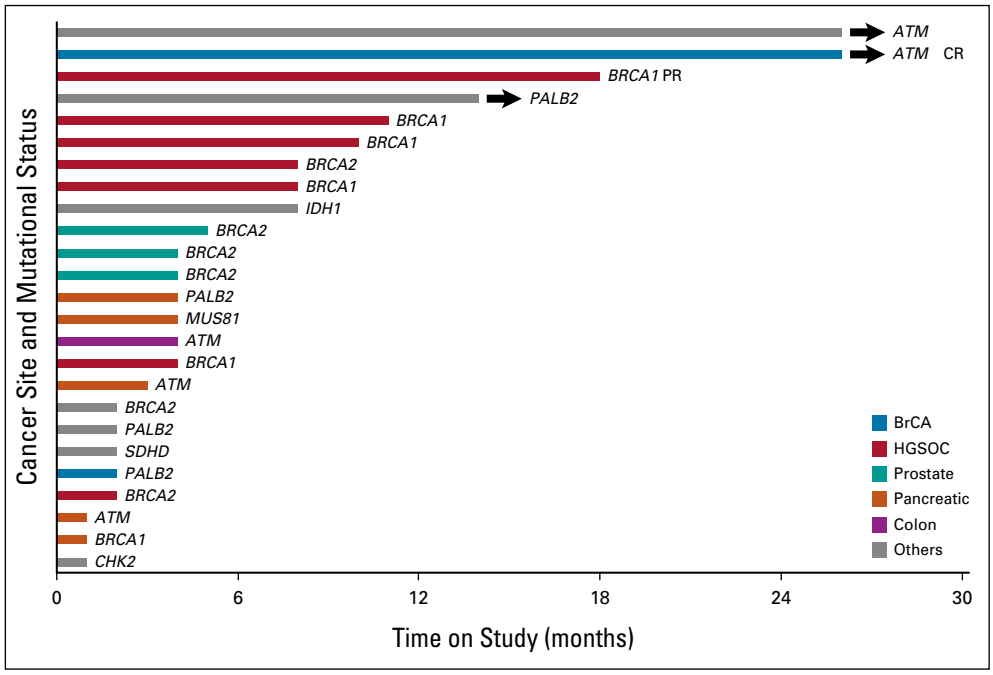
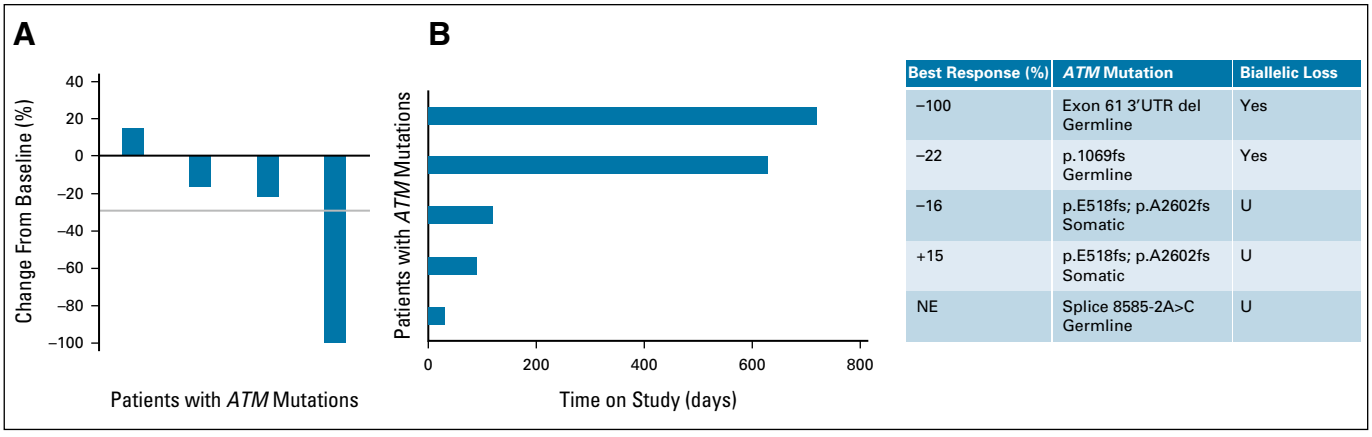


FIG 1. Efficacy of ceralasertib and olaparib in all patients enrolled in the study. A swimmer plot demonstrating time to response and duration of study treatment. CR, complete response; HGSOC, high-grade serous ovarian cancer; PR, partial response.

benefit were observed in subsets of patients with tumors harboring *ATM* mutation and in patients with *BRCA*-mutated HGSOC with acquired PARP inhibitor resistance. These findings warrant further investigation in larger groups of patients.

ATR inhibition has been shown to be synthetically lethal with ATM deficiency in preclinical models, translating to responses to monotherapies that have been reported with agents such as BAY1895344 and M6620.[25,36] The complete loss of ATM protein is ideally confirmed by protein immunohistochemistry, but this is not yet a standard laboratory test. Genetic testing is not as certain, although LOH can sometimes be confirmed. In this trial, two patients with germline *ATM* mutation and evidence of biallelic loss in tumor had durable clinical benefit, with one patient achieving a CR. Loss of the wild-type *ATM* allele could be identified in these two patients as determined by variant allelic frequency in the tumor when compared with germline control. The activity of PARP inhibition in the ATM-deficient setting has primarily been studied in prostate cancer and is less clear, although responses have been reported.[29] Ultimately, randomized trials in ATM-deficient cancers will be required to determine whether the activity is driven by ATR inhibition alone or whether the combination contributed to the benefit observed.

PARP inhibition is now part of the standard armamentarium for HGSOC so that it is critical to develop strategies addressing acquired resistance. This exploratory experience



Best Response (%)	*ATM* Mutation	Biallelic Loss
−100	Exon 61 3′UTR del Germline	Yes
−22	p.1069fs Germline	Yes
−16	p.E518fs; p.A2602fs Somatic	U
+15	p.E518fs; p.A2602fs Somatic	U
NE	Splice 8585-2A>C Germline	U

FIG 2. Efficacy of ceralasertib and olaparib in a subset of patients with *ATM* mutation. (A) Waterfall plot of the best objective response measured as the maximum change from baseline in the sum of the longest diameter of each target lesion. (B) Swimmer plot demonstrating time to response and duration of study treatment. The table on the right defines the cohort, extent of response, ATM mutation details, and biallelic status. NE, non-evaluable; U, unknown.

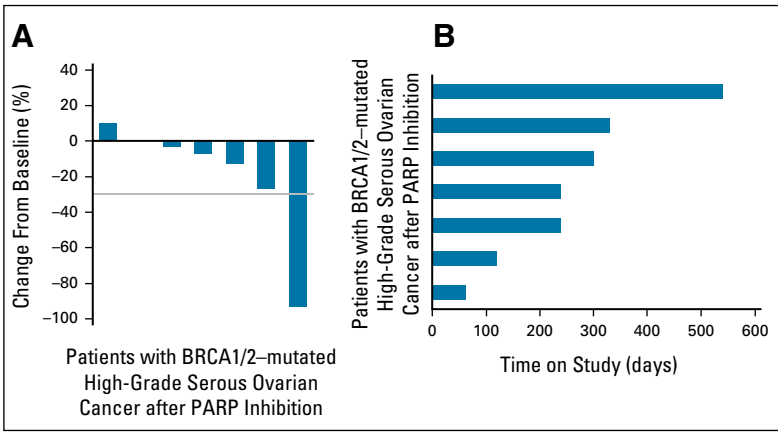
TABLE 4. Prior Therapy, Extent and Duration of Response of Patients With BRCA1/2–Mutated High-Grade Serous Ovarian Cancer Post-PARP Inhibition Received Olaparib and Ceralasertib/AZD6738

Patient No.	BRCA Status	No. of Prior Chemotherapies	Prior PARPi	Line of Therapy	Status of PARPi Before OLAPCO	Duration (months)	OLAPCO Response	AEs, Grade	Duration From Treatment to Disease Progression (months)
1	*gBRCA1*	5	Olaparib Rucaparib Niraparib	Second Fourth Fifth	SD PD PD	6 2 2	SD	N/V, G1 Anemia, G1	4
2	*gBRCA2*	11	Veliparib Rucaparib	Fourth Eighth	PD PD	2	PD	N/V, G3 Diarrhea, G1 Anemia, G3 Neutropenia, G4 Thrombocytopenia, G4 Dose reduced	2
3	*gBRCA1*	9	Olaparib Rucaparib	Third Sixth	SD PD	6 2	SD	N/V, G1 Thrombocytopenia, G2 Anemia, G3 Leukopenia, G2 ALT increase, G2 Dose reduced	11
4	*gBRCA1*	5	Olaparib	Second	PD		SD	Anemia, G3 Thrombocytopenia, G1 Fatigue, G1 Dose delay	8
5	*gBRCA1*	5	Olaparib Niraparib Niraparib plus nivolumab Irinotecan plus veliparib	Third Third Fourth Fifth	Toxicity PD PD Toxicity	12 3 6 1	PR (–90%)	Nausea, G2 Vomiting, G1 Diarrhea, G2 Urticaria, G1 Thrombocytopenia, G1 Leukopenia, G1 Anemia, G1 Pruritus, G1	18
6	*sBRCA2*	2	Niraparib	Second	PD	11	SD	Nausea, G1 Fatigue, G2	8
7	*gBRCA1*	5	Olaparib Rucaparib	Second Third	Toxicity PD	0.1 12	SD	Anemia, G3	10

Abbreviations: AE, adverse event; G1, grade 1; G2, grade 2; G3, grade 3; OLAPCO, olaparib combinations; PARP, poly (ADP-ribose) polymerase inhibitor; PD, progressive disease; PR, partial response; SD, stable disease.

suggests promise for combined ATR and PARP inhibition in this setting. There are several postulated mechanisms of PARP inhibitor resistance including expression of drug efflux pumps or loss of PARP1 protein expression. Restoration of HR pathway function is also a major resistance mechanism that may occur by somatic reversion or restoration of an open reading frame, epigenetic reversion of BRCA1 promoter hypermethylation, express of a hypomorphic protein with residual BRCA function, or by loss of end resection regulation.[37] Additionally, stabilization of replication forks represents another major mechanism of PARP inhibitor resistance.[20,37]

Preclinical evidence supports the importance of the ATR-CHK1 pathway in *BRCA*-mutated cancers, where it is used to maintain genomic stability. PARP inhibitor–resistant BRCA1-deficient cells become increasingly dependent on ATR for survival.[20,21] Despite the lack of BRCA1, PARP inhibitor–resistant cells regain RAD51 loading to DNA double-stranded breaks and stalled replication forks, enabling both restored HR and replication fork stabilization as resistance mechanisms. ATR inhibition will compromise HR and de-stabilize replication forks to overcome both resistance mechanisms. ATR inhibition also overcomes PARP inhibitor resistance in *BRCA2*-mutated ovarian cancer models. In general, responses in PARP inhibitor–resistant preclinical models are superior with combined ATR and PARP inhibition compared with ATR inhibition alone.[21] Our results suggest that the ceralasertib-olaparib combination has potential clinical



Best Response (%)	*BRCA* Mutation	Months on Prior PARPi	Prior Lines of Chemotherapy
−90	*gBRCA1*	6	5
−13	*gBRCA1*	2	9
0	*gBRCA1*	12	5
−27	*gBRCA1*	U	5
−7	*sBRCA2*	11	2
10	*gBRCA1*	2	5
−3	*gBRCA2*	2	11

FIG 3. Efficacy of ceralasertib and olaparib in a subset of patients with *BRCA1/2*–mutated high-grade serous ovarian cancer after PARP inhibition. (A) Waterfall plot of the best objective response measured as the maximum change from baseline in the sum of the longest diameter of each target lesion. (B) Swimmer plot demonstrating time to response and duration of study treatment. The table on the right defines the cohort, *BRCA* mutation details, extent of response and number of prior lines of therapy as well as the duration of therapy on prior PARP inhibition. PARPi, poly (ADP-ribose) polymerase inhibitor. U, unknown.

activity with manageable toxicity in *BRCA*-mutated PARP inhibitor–resistant HGSOC, with a duration of benefit that exceeded the duration achieved on prior PARP inhibitor monotherapy.

The current schedule uses full-dose olaparib with attenuated ceralasertib, which may be most appropriate in less heavily treated, PARP inhibitor–naïve patients. In both the ATM-deficient and PARP inhibitor–resistant settings, a schedule maximizing ceralasertib may be preferable. Dose-finding efforts are underway in other clinical trials with olaparib at 100-150 mg twice daily, which may afford substantially higher doses of ceralasertib that may ultimately be critical for maximizing clinical activity.

Further work will be required for insights into non-responding patients with tumors harboring DDR alterations. It is possible that *BRCA*, *PALB2*, and other mutations have different functional relevance in certain cancer types[38] or that routine assessment of LOH will carry high importance. Additionally, assessment of HR function and replication fork stability at baseline will also be important in future studies to better understand clinical outcomes. Such assessments could include an IHC-based RAD51 assay or DNA fiber assays in organoid cultures derived from patient biopsies.[39] Future studies should include planned translational analyses of paired tissue biopsies and serial circulating tumor DNA samples to assess mechanisms of PARP inhibitor resistance and to identify predictors of response and determinants of resistance to the combination regimen.

This study has several limitations. Although we were able to determine biallelic loss in a few of the tumors, we lacked information about the activity of the nonmutated alleles in most cases. In many cases, germline testing was used for eligibility, rather than analysis of tumor DNA. Furthermore, the clinical outcomes were likely limited because the majority of patients were heavily pretreated and were also resistant to prior therapy, including platinum-based chemotherapy and PARP inhibitors. Finally, higher ceralasertib doses with attenuated olaparib may ultimately be the optimal dosing schedule for this combination.

Despite these limitations and the limited ORR of 8.3%, we were able to confirm safety of ceralasertib combined with full-dose olaparib and generate signals of promising clinical benefit in both ATM-deficient and *BRCA*-mutated PARP inhibitor–resistant ovarian cancer. Further schedule optimization and testing of larger populations in appropriately powered single-arm and randomized trials are warranted.

AFFILIATIONS

[1]Cleveland Clinic, Cleveland, OH
[2]Yale Cancer Center, New Haven, CT
[3]Vanderbilt-Ingram Cancer Center, Nashville, TN
[4]Dana-Farber Cancer Institute, Boston, MA
[5]Oncology Early Clinical Projects, Oncology R&D, AstraZeneca, Cambridge, United Kingdom

CORRESPONDING AUTHOR

Haider Mahdi, MD, MPH, Magee Womens Research Institute, University of Pittsburgh, Division of Gynecologic Oncology, Magee Womens Hospital, University of Pittsburgh Medical Center, 300 Halket St, Suite 2130, Pittsburgh, PA 15213; e-mail: mahdihs@upmc.edu.

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AUTHOR CONTRIBUTIONS

Conception and design: Juliane Jürgensmeier, Brunella Felicetti, Peter Mortimer, Geoffrey I. Shapiro, Joseph Paul Eder
Administrative support: Patricia LoRusso
Provision of study materials or patients: Haider Mahdi, Navid Hafez, Davendra Sohal, Khanh T. Do, Patricia LoRusso, Geoffrey I. Shapiro
Collection and assembly of data: Haider Mahdi, Navid Hafez, Deborah Doroshow, Davendra Sohal, Vickie Keedy, Khanh T. Do, Patricia LoRusso, Manuel Avedissian, Jeffrey Sklar, Geoffrey I. Shapiro, Joseph Paul Eder
Data analysis and interpretation: Haider Mahdi, Navid Hafez, Deborah Doroshow, Davendra Sohal, Vickie Keedy, Patricia LoRusso, Juliane Jürgensmeier, Jeffrey Sklar, Colin Glover, Brunella Felicetti, Emma Dean, Geoffrey I. Shapiro, Joseph Paul Eder
Manuscript writing: All authors
Final approval of manuscript: All authors
Accountable for all aspects of the work: All authors

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Haider Mahdi
Research Funding: Puma Biotechnology

Deborah Doroshow
Consulting or Advisory Role: Ipsen, Atheneum, Boston Healthcare Associates, Dedham Group, Guidepoint Global
Travel, Accommodations, Expenses: Ipsen

Davendra Sohal
Honoraria: Foundation Medicine
Consulting or Advisory Role: Perthera, Ability Pharma
Speakers' Bureau: Incyte
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Vicki Keedy
Consulting or Advisory Role: Karyopharm Therapeutics, Daiichi Sankyo/Lilly
Research Funding: Medpacto, Plexxikon, Daiichi Sankyo, Lilly, Immune Design, GlaxoSmithKline, TRACON Pharma, Advenchen Laboratories, Adaptimmune, Deciphera, SpringWorks Therapeutics

Khanh T. Do
Employment: Moderna Therapeutics
Stock and Other Ownership Interests: Moderna Therapeutics
Consulting or Advisory Role: QED Therapeutics, The Jackson Laboratory

Patricia LoRusso
Honoraria: Five Prime Therapeutics
Consulting or Advisory Role: Genentech, CytomX Therapeutics, Roche/Genentech, Halozyme, Five Prime Therapeutics, Agenus, Agios, Cybrexa Therapeutics, Sotio, AbbVie, Genmab, Takeda, TYME, IQvia, Trial to Reduce IDDM in the Genetically at Risk (TRIGR), Pfizer, ImmunoMet, Black Diamond Therapeutics, GlaxoSmithKline, QED Therapeutics, AstraZeneca, EMD Serono, Shattuck Labs, Astellas Pharma, Salarius Pharmaceuticals, Silverback Therapeutics, Macrogenics, Kyowa Kirin International, Kineta, Zentalis, Molecular Templates, ABL Bio, SK Life Sciences, ST Cube, Bayer, I-Mab
Research Funding: Genentech
Travel, Accommodations, Expenses: Genentech

Juliane Jürgensmeier
Employment: Gilead Sciences
Stock and Other Ownership Interests: Gilead Sciences
Travel, Accommodations, Expenses: Gilead Sciences

Jeffrey Sklar
Stock and Other Ownership Interests: Precipio
Research Funding: Jilin Zixin

Colin Glover
Employment: AstraZeneca
Stock and Other Ownership Interests: AstraZeneca

Brunella Felicetti
Employment: AstraZeneca
Stock and Other Ownership Interests: AstraZeneca

Emma Dean
Employment: AstraZeneca/MedImmune
Stock and Other Ownership Interests: AstraZeneca/MedImmune, AstraZeneca
Travel, Accommodations, Expenses: AstraZeneca

Peter Mortimer
Employment: AstraZeneca
Stock and Other Ownership Interests: AstraZeneca
Travel, Accommodations, Expenses: AstraZeneca

Geoffrey I. Shapiro
Consulting or Advisory Role: G1 Therapeutics, Lilly, Pfizer, Roche, Merck Serono, Sierra Oncology, Cybrexa Therapeutics, Ipsen, Bayer, Fusion Pharmaceuticals, Bicycle Therapeutics, Almac Diagnostics, Astex Pharmaceuticals, Daiichi Sankyo, Angiex, Seattle Genetics, Artios, Boehringer Ingelheim, Concarlo, Atrin Pharmaceuticals, Syros Pharmaceuticals, Zentalis, CytomX Therapeutics
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Travel, Accommodations, Expenses: Lilly, Pfizer, Bicycle Therapeutics, G1 Therapeutics, Sierra Oncology, Bayer

Joseph Eder
Honoraria: Roche Molecular Diagnostics
Consulting or Advisory Role: Roche/Genentech

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H.M. is now at the University of Pittsburgh. D.D. is now at the Mount Sinai School of Medicine. D.S. is now at the University of Cincinnati. J.J. is now at Gilead Sciences Inc.

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