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A Mixed Methods Feasibility Trial of PKC α Inhibition with Auranofin in Asymptomatic Patients with Ovarian Cancer

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Abstract

Purpose—This trial was undertaken to 1) determine the feasibility of enrolling asymptomatic ovarian cancer patients with Ca-125 elevation to a trial with the PKC α inhibitor, auranofin, and 2) understand patients' perceptions of Ca-125 monitoring.

Methods—Asymptomatic ovarian cancer patients with Ca-125 elevation received auranofin 3 mg orally twice/day and were evaluated. Patients participated in interviews about Ca-125 monitoring.

Results—Ten patients were enrolled in slightly over 6 months, exceeding our anticipated rate. Four manifested stable Ca-125 levels for 1 month or longer. The median progression-free survival was 2.8 months (95% confidence interval: 1.3, 3.8 months); auranofin was well tolerated. One patient had baseline and monthly Ca-125 levels of 5570, 6085, 3511, and 2230 units/mL, respectively, stopped auranofin because of radiographic progression at 3 months, and manifested an increase in Ca-125 to 7168 units/mL approximately 3 months later. Patient interviews revealed: 1) the important role of Ca-125 in cancer monitoring; 2) ardent advocacy for Ca-125 testing; and 3) evolution toward the Ca-125 assuming a life of its own.

Conclusions—This study showed feasibility; and patients favored Ca-125 monitoring. One patient had a decline in Ca-125, suggesting that PKC α inhibition merits further study in ovarian cancer.

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Protein kinase C α (PKC α) is a human oncogene that plays a key role in ovarian cancer carcinogenesis and tumor viability [1]. Zhang and others were among the first to demonstrate that the PKC α gene is up-regulated in ovarian cancer, that increased PKC α expression correlates with tumor stage and grade, and that overexpression of PKC α contributes to murine ovarian surface epithelium transformation [2]. Subsequent studies from our group demonstrate that PKC α is required for maintenance of a tumor-initiating cell, stem-like phenotype in human ovarian cancer cells and that PKC α is necessary for ovarian tumorigenesis *in vivo* [3]. Our group further demonstrated that auranofin, a potent and selective inhibitor of oncogenic PKC α signaling, inhibits the tumor-initiating behavior of ovarian tumor cells [3]. These observations -- coupled with the findings that gold compounds, such as auranofin, have been demonstrated *in vitro* to inhibit PKC α and have even shown promise in a recent phase I study -- underscore the need to test the hypothesis that PKC α inhibition leads to antineoplastic effects in patients with ovarian cancer [1,4]. Testing this hypothesis is particularly alluring in view of the fact that ovarian cancer is the most lethal gynecological malignancy and that, in the past 20 years, no new drugs that have been demonstrated to prolong overall survival in patients with this malignancy [5].

In this context, we undertook a pilot trial to test auranofin in patients with epithelial ovarian cancer. Three aspects of our study design merit specific mention. First, because auranofin was to be administered as a single agent, we believe this gold compound was more likely to show proof-of-concept in the setting of a low tumor burden [1]. Consequently, this pilot trial targeted asymptomatic ovarian cancer patients with Ca-125 elevation, as these patients were more likely to have a low tumor burden. Second, previous trials that had defined patient eligibility based on biochemical recurrence alone appeared slow to accrue [6]. Rustin and others went so far as to call into question Ca-125 monitoring, demonstrating that such monitoring does not improve clinical outcomes and further calling into question whether this or future trials would ever be able to accrue with Ca-125 elevation as part of the eligibility criteria [7]. Launching a pilot study and assigning feasibility as the primary endpoint appeared appropriate. Finally, the growing controversy surrounding Ca-125, as alluded to above, offered an opportunity to learn directly from patients how they view and understand the role of this tumor marker. For this reason, qualitative interviews were included in the study design.

For the reasons above, this 10-patient pilot trial focused on feasibility as its primary endpoint and incorporated patient-centered qualitative methodology into the study design. Nonetheless, this trial served as an important platform from which to explore the antineoplastic effects of PKC α inhibition with auranofin in ovarian cancer patients. In essence, this pilot study was designed to lay the groundwork for future clinical trials that focused on PKC α inhibition in ovarian cancer patients.

METHODS

Overview

This study was conducted at the Mayo Clinic in Rochester, Minnesota. The Mayo Clinic Institutional Review Board approved the study protocol, and all patients provided written

informed consent prior to enrollment. This trial was listed on www.clinicaltrials.gov and assigned study number NCT01747798.

Eligibility and Exclusion Criteria

To be eligible, patients had to meet the following criteria: 1) age ≥ 18 years; 2) histologic or cytologic evidence of epithelial ovarian, primary peritoneal, or fallopian tube cancer; 3) completion of initial cancer therapy, which included potentially surgery and/or postoperative chemotherapy; 4) Eastern Cooperative Oncology Group (ECOG) performance score (PS) of 0-2; 5) for patients with a first-time ovarian cancer recurrence, an increase in the Ca-125 as defined as follows: normalization of the Ca-125 during first-line chemotherapy followed by an increase of ≥ 100 units/mL or a nadir of the Ca-125 beyond the upper limit of normal with a confirmatory measurement within 4 weeks or less of the same or higher Ca-125 value; 6) for patients who had received subsequent treatment for progressive cancer, Ca-125 elevation beyond the institutional upper limit of normal; 7) the following laboratory values within 2 weeks of registration: absolute neutrophil count $\geq 1.5 \times 10^9$ cells/liter; platelet count $\geq 100 \times 10^9$ cells/liter; total bilirubin ≤ 1.5 times the institutional upper limit of normal; creatinine within the institutional limit of normal; 8) for women of child-bearing potential, a negative pregnancy test within one week of registration; and 9) willingness to participate in a qualitative telephone interview about Ca-125 monitoring.

Patients were excluded in the event of the following: 1) concurrent illness that, in the opinion of the treating oncologist, would make the patient inappropriate for study participation; 2) symptoms (other than anxiety, depression, and other psychological symptoms) that, in the opinion of the treating oncologist, were a direct result of cancer recurrence; 3) receiving other, medically prescribed, cancer treatment; or 4) pregnant or nursing.

Pretreatment Assessment and Monitoring for Auranofin

All patients underwent a history and physical examination, assignment of ECOG performance score, and adverse event assessment within 14 days of trial registration. Other testing performed within this time frame consisted of a hemogram, chemistry profile (total bilirubin, creatinine, and aspartate aminotransferase), Ca-125, urinalysis, and, in the setting of documented measurable disease, radiographic imaging with the imaging modality left to the discretion of the treating healthcare provider.

Repeat history and physical examination; weight; adverse event assessment; and laboratory testing (including Ca-125), as noted above, were undertaken on a monthly basis while patients remained on auranofin. Every other month, patients underwent testing for tumor measurements, as appropriate, and a urinalysis assessed proteinuria. RECIST criteria, version 1.1, were used to evaluate tumor response in the setting of measurable disease. Further monitoring after drug initiation included a weekly hemogram.

Qualitative Interviews

Patients participated in a 20-30 minute semi-structured interview designed to capture their understanding and impressions of Ca-125 monitoring. These telephone interviews occurred

shortly after study enrollment (interval not specified in the study protocol) and were audio recorded and transcribed. A trained female interviewer queried patients about the first time they had heard of Ca-125, the positive and negative aspects of Ca-125 monitoring, and the subjective value of this test for the patient and for others with ovarian cancer.

Cancer Treatment

Auranofin 3 mg twice a day (6 mg total dose per day) was prescribed orally on days 1-28 of a 28-day treatment cycle. This dose was derived from other indications [8]. Auranofin dose reductions were initiated based on adverse events; the National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE), version 4.0, were used to assess the latter. Grade 4 interim neutropenia or grade 3 or 4 thrombocytopenia was to prompt a 50% dose reduction at the time of starting the next treatment cycle. Similarly, grade 3 or worse skin toxicity, gastrointestinal toxicity, or other toxicity was to prompt a 50% dose reduction. If at retreatment patients manifested grade 2 or worse symptoms, the auranofin was to be held until symptoms abated to grade 1 or less in most circumstances. The exception was grade 2 proteinuria, which permitted restarting the auranofin at the same dose. In the event toxicity did not lessen to an acceptable threshold after 2 weeks, the patient could no longer receive auranofin on study.

Analyses

The primary purpose of this trial was to show that asymptomatic ovarian cancer patients with Ca-125 elevation can be enrolled to a cancer therapeutic trial within a reasonable time frame. We estimated an enrollment rate of 5 patients per year with termination of the trial after one year if three or fewer patients were enrolled. A key secondary endpoint was to assess the rate of Ca-125 stabilization. A 10-patient cohort provided 83% power to detect an improvement in the rate of Ca-125 stabilization from 30% to 60% at one month after study enrollment, assuming a 1-sided significance level of 0.20. Progression-free survival was defined as the time from registration to either cancer progression or death, whichever occurred first. Unless otherwise specified, all adverse event data are reported regardless of attribution.

For the patient interview data, thematic analysis was used [9]. Two investigators (CRB and AJ) independently reviewed the interview transcripts and identified initial codes within the data. Initial codes were subsequently discussed, refined, and collated. Patterns, or themes, were identified with an inductive, data-driven approach that preserved internal homogeneity (similarity within a theme) and external homogeneity (distinction between themes) [10].

RESULTS

Demographics

Ten patients were recruited from March 2013 through October 2013. Patient characteristics are listed in Table 1, which shows the median age of the cohort as 65 years (range 54, 81). Eight patients had an ECOG PS was 0, and two of 1. Four had baseline radiographic evidence of disease.

Auranofin Administration

Of the 10 patients enrolled, the median (range) treatment duration was 2 cycles (1-4 cycles). Reasons for stopping auranofin included cancer progression (n=8 patients), non-specific symptoms that started within only a few day of starting the agent (n=1), and decline of further treatment per the patient (n=1).

Tumor Response Parameters

Only 9 patients were evaluable for tumor response because one stopped therapy prior to the end of the first cycle. Four patients manifested stable disease, as defined as neither a 50%+ increase in Ca-125 (progression) nor a 50% or more drop in Ca-125 (tumor response) within one month (one cycle) or more of starting auranofin (Figure 1).

Of note, one patient had baseline and monthly Ca-125 levels of 5570 units/mL, 6085 units/mL, 3511 units/mL, and 2230 units/mL, respectively, over 3 months only to stop therapy because of radiographic evidence of cancer progression. Her Ca-125 increased to 7168 units/mL approximately 3 months after stopping the auranofin.

At the time of this report, only one patient had died. The median progression-free survival for the cohort was 2.8 months (95% confidence interval: 1.3, 3.8 months) (Figure 2). Among these 10 patients, eight manifested progressive disease with either Ca-125 elevation and/or new radiographic lesions, and one patient died without evidence of disease progression.

Adverse Events

Auranofin was well tolerated, and only grade 1 and 2 adverse events were reported. The most common included diarrhea (n=5 patients), nausea (n=5), and fatigue (n=5), all of which were grade 1 in severity (Table 2).

Qualitative Data on Ca-125 Monitoring

Nine patients completed qualitative interviews; one was not reachable by phone, despite multiple attempts. Three themes were identified from these interviews. The first was termed, “the role of Ca-125 in cancer surveillance and monitoring.” This theme captured perceptions about the test itself as well as the utility and reassurance associated with monitoring Ca-125 levels. Many patients appeared well-informed about the Ca-125 test with a knowledge and understanding that appeared to have been achieved gradually. These patients favored its use as a surveillance tool, but were also able to describe some of this test's limitations. The second theme was termed “ardent advocacy for Ca-125 testing.” This theme conveyed patients’ desire to use the test not only for cancer surveillance but for ovarian cancer screening (for which it is not indicated). This theme captured patients’ zeal for proactively recommending the test to others, irrespective of cancer history, type or degree of cancer risk, and cost of testing. The final theme was termed, “evolution toward the Ca-125 assuming a life of its own.” This theme captured patients’ description of what seemed to be an evolution in their own thinking about Ca-125 elevation. This evolution spanned the emotionally charged nature of Ca-125 elevation/decline to patients’ grappling with a disconnect between Ca-125 elevation and lack of cancer symptoms to an apparent “shift” in therapeutic goals from a focus on the cancer to a focus on treating (lowering) the Ca-125. Indeed, patients

cited their desire to receive cancer treatment primarily to bring down the Ca-125 with little reference to treating the actual cancer. Representative quotes appear in Table 3.

DISCUSSION

This pilot trial succeeded in demonstrating that, despite shifting data on the value of Ca-125 as a monitoring tool for cancer recurrence, asymptomatic ovarian cancer patients who require Ca-125 elevation as part of a trial's eligibility criteria are able to be successfully identified and enrolled. In fact, we demonstrated an ability to exceed our target recruitment rate. Despite the landmark paper from Rustin and others and despite what appeared to be marginal accrual to other trials that relied on Ca-125 in their eligibility criteria, our findings suggest that trials, such as this one, are still able to be completed in a timely manner [6,7].

More importantly, this study showed that PKC α inhibition with auranofin exhibits therapeutic value and merits further investigation. Four patients manifested stable disease with auranofin, albeit over a short interval. Remarkably, one patient manifested a notable drop in Ca-125 which subsequently increased upon stopping the auranofin. Although these data are too preliminary to allow for firm conclusions on therapeutic efficacy, they do suggest that further study of PKC α inhibition, either with auranofin alone or in combination with other therapeutic agents, is indicated. Importantly, auranofin was well-tolerated and appeared to demonstrate antineoplastic effects even within a group of pre-treated patients, thus providing additional justification for further study in ovarian cancer patients.

With respect to feasibility, this trial provided a wealth of information on how patients perceive Ca-125 monitoring. As a result of the data from Rustin and others, we were concerned that patients would voice mixed feelings about Ca-125 monitoring, as described by other groups prior to the publication of the Rustin paper [11]. Admittedly, the Ca-125 is associated with a roller coaster of emotions when the marker fluctuates; and, by interviewing patients who had already enrolled in this auranofin trial, we likely captured the views of only patients who were perhaps more accepting of Ca-125 monitoring in general. However, none of these nine patients described any reluctance to undergo the test, all appeared to value the availability of Ca-125 monitoring – some even to the point of being ardent advocates – and, in aggregate, these patients' comments provided further feasibility data to enable us to conclude future studies that rely on Ca-125 elevation to identify and recruit patients can be accomplished. It should be noted, however, that in accordance with our last two themes, Ca-125 monitoring is fraught with some misunderstanding (it is not a screening tool for ovarian cancer or for other cancers), and the Ca-125 is a highly emotionally-charged test. These last two themes are important and should invite healthcare providers to further instruct patients on the Ca-125 marker and its role in monitoring patients for recurrent or progressive ovarian cancer.

In summary, this trial shows that it is feasible to conduct studies that rely on Ca-125 monitoring to identify and recruit patients. More importantly, the fact that one patient manifested a drop in Ca-125 with auranofin suggests a role for studying auranofin either as monotherapy or combined with other agents. Finally, this study shows a clear need to further educate patients on the role of Ca-125 monitoring, to identify and correct patients'

misunderstandings of this tumor marker, and to help patients cope with the emotionally-charged nature of Ca-125 fluctuations over time.

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Figure 1.

The percent change in Ca-125 levels from baseline appears for all 9 evaluable patients. One patient manifested a notable and sustained drop in Ca-125 of greater than 50%. Cycle length was one month.

Figure 2.

The median progression-free survival for the cohort was 2.8 months (95% confidence interval: 1.3, 3.8 months).

Table 1

Baseline Demographics

DEMOGRAPHIC	NUMBER OF PATIENTS
Age, median, in years (range)	65 (54, 81)
ECOG PS	
0	8
1	2
2	0
Number of Prior Chemotherapy Regimens	
1	4
2	4
3 or more	2
Radiographic Evidence of Cancer at Study Entry	
Yes	4
No	6

Table 2

Maximum Grade Adverse Events Regardless of Attribution

ADVERSE EVENT	GRADE 2 [*]	GRADE 1
Diarrhea	0	5
Nausea	0	5
Vomiting	0	3
Fatigue	2	5
Neutropenia	0	0
Thrombocytopenia	0	4
Thromboembolism	1	0

* Numbers refer to number of patients who experienced this grade of adverse event.

Table 3**Three Qualitative Themes with Supporting Patient Quotes**

The Role of Ca-125 in Cancer Surveillance and Monitoring	Ardent Advocacy for Ca-125 Testing	Evolution Toward the Ca-125 Assuming a Life of Its Own
<p>“... at the moment, it [Ca-125] seems to be the only thing we can check to see if there is possible activity. What I am understanding about the Ca-125 is that it is really not very reliable.”</p> <p>“The doctors tell me well, it's not very accurate, and we really go by the CT scans or the PET scans. I have to believe they focus on those instead of the Ca-125.”</p> <p>“I didn't know the Ca-125 had to be elevated to show it was cancer...”</p> <p>“I am usually happy to hear that the CT is clear and we talk about it [the Ca-125] being spiked... and she [healthcare provider] will say, ‘It is still ok.’ We talk about it and stuff.”</p> <p>“Someone can have a low Ca-125 and someone can have more disease than the other.”</p> <p>“It's my understanding that the number itself [Ca-125] is not indicative of the magnitude of the disease. The number might jump 200 points but that doesn't mean suddenly that you are filled with cancer.”</p> <p>“Oh, and my husband liked the idea that they don't always just look at the Ca-125 and determine treatment as to the numbers. He liked the idea that maybe we need to consider my condition.”</p>	<p>“And right after I had it the first time [Ca-125 check], I told my sisters about it. I told them if they had a chance to get it done...”</p> <p>“My daughter I don't think has tested yet for it [CA-125], but she works in the medical field so is pretty on to things....”</p> <p>“...but I want to make sure my daughter doesn't have to go through this. She could ask the doctor if he could do the blood test [Ca-125]. I wouldn't trust the BRCA.”</p> <p>“It is on my calendar for every month, and then I have a little note card with the numbers [Ca-125 levels] listed.”</p> <p>“... when it started to go up, my doctor...said you know, we'll test you again, do you want to be tested in 2 weeks or 3 weeks or one week? We went with one week at my choice.”</p> <p>“... whether insurance pays for it or not, do it.”</p> <p>“To me, I think it would be good if it was advertised more so that other women know about the test.”</p> <p>“I would talk about it [Ca-125] to anybody of my age who is a female that will listen. I am so glad to have it...it is a great tool.”</p>	<p>“Well each time it went down, I was ecstatic, but I didn't feel any different.”</p> <p>“If you could see it go down, it would give me more hope.”</p> <p>“...I was in absolute fear that it wasn't going down and that it wasn't going to normalize. And then when it did normalize, then my fear and anxiety were that it was going to go up...”</p> <p>“It always feels like the shoe is going to drop.”</p> <p>“My husband is bringing me down from the ceiling every once in a while. It's usually just a matter of when I've got the number....”</p> <p>“I don't feel any differently than I felt when my Ca-125 was normal. It's just mentally that I know it is rising and it's made me, not fatalistic, but close to it.”</p> <p>“...and if it was down from the previous month, I was overjoyed thinking that ‘wow, we're going to kill all of this and be done with it.’”</p> <p>“He said my goal was to get it down to under 34.”</p> <p>“...it [Ca-125] went down 600 points, and then it went down another 2 or 300 So it was making progress going down.”</p> <p>“... the only real, I guess in my mind, the only thing that I was looking at was that Ca-125....”</p> <p>“You want it [Ca-125] to be as close to zero as possible.”</p> <p>“You feel like a walking time bomb.”</p>