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PARAGON: A Phase II study of anastrozole in patients with estrogen receptor-positive recurrent/metastatic low-grade ovarian cancers and serous borderline ovarian tumors

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HIGHLIGHTS

- Anastrozole is well-tolerated in low-grade ovarian cancers (LGOC) and serous borderline ovarian tumors (SBOT)
- Most patients with hormone receptor positive LGOC and SBOT treated with anastrozole have clinical benefit at 3 months
- Median duration of clinical benefit was 9.5 months

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ABSTRACT

Objective. Treatment options are limited for patients with recurrent/metastatic low-grade ovarian cancers (LGOCs) and serous borderline ovarian tumors (SBOTs) as response rates to chemotherapy are low. A subset of patients appears to derive clinical benefit from antiestrogens, but most studies have been retrospective and clinical benefit rates (CBR) remain uncertain. The primary aim of PARAGON was to prospectively investigate the CBR of anastrozole, an aromatase inhibitor, in patients with estrogen receptor (ER) and/or progesterone receptor (PR) positive LGOC and SBOT.

Methods. Post-menopausal women with ER-positive and/or PR-positive recurrent/metastatic LGOCs and

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Low-grade serous carcinoma
Borderline ovarian tumors
Hormonal therapy
Aromatase inhibitors

SBOTs and evaluable disease by RECIST v1.1 or GCIG CA125 criteria were treated with anastrozole 1 mg daily until progression or unacceptable toxicity.

Results. Thirty-six patients were enrolled. Clinical benefit at 3 months (primary endpoint) was observed in 23 patients (64%, 95% CI 48%–78%) and was similar at 6 months (61%, 95% CI 43%–75%). The median duration of clinical benefit was 9.5 months (95% CI 8.3–25.8). Best study response was partial response by RECIST in 5 patients (14%), stable disease in 18 patients (50%) with progressive disease in 13 patients (36%). Median PFS was 11.1 months (95% CI 3.2–11.9). Anastrozole was well-tolerated. Patients with evidence of clinical benefit at 3 months reported less pain, fatigue, and improved physical and role functioning as early as 1 month of commencing treatment.

Conclusions. Anastrozole was associated with a CBR of 61% of patients with recurrent ER-positive and/or PR-positive LGOC or SBOT for at least 6 months with acceptable toxicity.

1. Introduction

Kurman and Shih proposed a dualistic model over 10 years ago to categorize ovarian cancers into two distinct groups designated Type I and Type II, based on evidence for different molecular genetic pathways involved in the pathogenesis of epithelial ovarian carcinomas and the variable clinical behaviour of the different subtypes [1,2]. Type II, predominantly high-grade serous ovarian cancers (HGSOC), are characterized by high response rates to platinum-based chemotherapy compared with Type I tumors, which include low-grade serous, endometrioid, clear cell and mucinous carcinomas. Low-grade serous cancers (LGSCs) comprise 5% to 10% of serous ovarian cancers and 6% to 8% of all ovarian cancers [3–6]. They are typically diagnosed at a younger age than HGSOC, with a median age of 47–54 years at diagnosis, and are characterized by relative resistance to cytotoxic chemotherapy [3,6–13]. In contrast to HGSOC, they lack *TP53* mutations, but may have *KRAS*, *NRAS* or *BRAF* mutations and activation of the *RAS*/*RAF*/*MEK*/*ERK* signaling pathway [14–20]. Low-grade endometrioid carcinomas have aberrations in the Wnt signaling pathway involving somatic mutations of *CTNNB1*, *PTEN* and *PIK3CA* [21]. Estrogen receptor (ER) expression has been reported in the majority of low-grade serous and endometrioid Type I tumors as well as in serous borderline ovarian tumors (SBOTs) and is a potential therapeutic target [22–24].

Despite low response rates to chemotherapy, many patients with low-grade ovarian cancers (LGOCs) receive adjuvant chemotherapy following debulking surgery with uncertain benefit. Neoadjuvant platinum-based chemotherapy for advanced stage LGSC of the ovary or peritoneum was associated with a response rate of 4–37%, which is much lower than response rates of up to 80% in patients with HGSOC [9,10,13]. Similarly, response rates to chemotherapy have been reported to be very low (2–4%) in patients with recurrent LGSC [11]. A recent retrospective, exploratory case-control analysis of over 5000 patients treated with adjuvant chemotherapy in clinical trials included 145 patients (2.8%) with LGSC, of whom 37 had suboptimal debulking at surgery and were evaluable for response. The response rate was higher than other studies at 23.1%, albeit still modest compared with 90.1% in patients with HGSOC [12].

Borderline ovarian tumors, previously known as tumors of low malignant potential, are neoplasms of epithelial origin characterized by nuclear atypia that is less than high grade atypia and increased cellular proliferation but no stromal invasion [25]. SBOTs share molecular and genetic alterations with LGSC and at relapse may have features of LGSC, suggesting that there is a pathway of progression from borderline tumor to low-grade carcinoma in some patients [25–27]. Similar to LGSC, SBOTs are resistant to chemotherapy, and these patients are usually not offered chemotherapy unless there is evidence of LGSC at relapse [26,28,29].

Treatment options are relatively limited for patients with recurrent or metastatic LGSC and SBOTs. Only a minority of patients (2–33%) have *BRAF* mutations and may possibly benefit from a BRAF inhibitor [15,20,30–32]. In view of data indicating mutations within the MAPK pathway, there have been a number of studies exploring MEK inhibitors (MEKi) in patients with LGSC. These include a Phase II trial of the MEK1/2 inhibitor, selumetinib, in 52 patients with LGSC. Eight (15%) patients had an objective response to treatment and 34 (65%) patients had stable disease. The median progression-free survival (PFS) was 11 months and >6 months in 63% of patients. The MILO trial was an open-label Phase III protocol that randomized patients with recurrent LGSC to either chemotherapy or MEK162 (binimetinib) [33,34]. The results of this trial are eagerly awaited as it was stopped after a planned interim analysis showed that the hazard ratio for PFS crossed the predefined futility boundary.

Hence, there remains a need to investigate alternative treatment strategies for patients with recurrent or metastatic LGOC and SBOTs. In particular, there is a strong rationale to investigate endocrine therapy. ER expression has been reported in up to 94% of LGOC and SBOTs, and several retrospective studies suggest that a subset of these patients derive significant benefit from hormonal therapy with antiestrogens [22–24]. The potential benefit of hormonal therapy in recurrent and metastatic SBOTs as well as LGSC has been documented in case reports and retrospective studies [35,36]. Hormonal therapy was associated with overall response rate of 9% in a retrospective analysis of 64 patients with recurrent LGSC [37]. A recent retrospective analysis reported on 203 patients with LGSC of the ovary or peritoneum who received either maintenance hormonal treatment or observation, based on physician discretion, following primary cytoreductive surgery and platinum-based chemotherapy [38]. Patients who received maintenance hormonal therapy had significantly longer median PFS (64.9 vs 26.4 months) compared to the patients in the observation group, without significant prolongation of median overall survival (OS, 115.7 vs 102.7 months). Patients with ER-positive LGSC appeared to derive the most benefit from maintenance hormonal therapy compared to observation, both in terms of median PFS (73.3 vs 29.9 months) and median OS (191 vs 107 months).

Based on the retrospective data suggesting a role for hormonal therapy in LGOC, particularly LGSC, as well as SBOTs which recur as LGSC, prospective studies to determine the proportion of patients who derive clinical benefit are needed but are lacking. The PARAGON trial was designed to investigate the activity of anastrozole in postmenopausal patients with a wide range of ER-positive and/or progesterone receptor (PR) positive recurrent or metastatic gynaecological tumors. It included 7 separate Phase II studies embedded within a single basket protocol. We report on a prospective Phase II trial that was undertaken to determine clinical benefit, quality of life and adverse events with an aromatase inhibitor, anastrozole, in patients with recurrent/metastatic SBOT and LGOC, with the secondary objective being to explore the relationship of ER histoscore with clinical benefit.

2. Materials and methods

2.1. Study design

PARAGON is an investigator-initiated, prospective, multi-centre single-arm Phase II study of anastrozole, 1 mg daily in women with recurrent SBOTs and LGOC. Patients received treatment until disease

progression based on CA125 or RECIST v1.1 criteria or treatment cessation for adverse events.

2.2. Eligibility

All patients had ER-positive and/or PR-positive tumors by immunohistochemical staining, based on assessment by local pathologists with at least 10% of cells staining positive for ER and/or PR. Additional eligibility criteria included a histological diagnosis of recurrent or metastatic SBOTs or LGOC; post-menopausal status; age ≥ 18; life expectancy > 3 months; Eastern Cooperative Oncology Group (ECOG) performance status 0–2; and disease evaluable for response by RECIST v1.1 or GCIG CA125 criteria. Baseline evaluation included history, physical examination, abdominal and pelvic computed tomography (CT) scan, full blood count, blood chemistry, and serum CA125 level.

2.3. Study objectives

The primary objective was to assess clinical benefit rate (CBR), defined as the proportion of patients experiencing either complete or partial response or stable disease at 12 weeks after commencing treatment. Response was determined by RECIST v1.1 in patients with measurable disease and GCIG CA125 response criteria in patients without measurable disease [39,40]. Secondary objectives included PFS, duration of response, quality of life, and toxicity. The European Organisation for Research and Treatment of Cancer (EORTC) QLQ-C30 and the Functional Assessment of Cancer Therapy-Endocrine Symptoms (FACT-ES) subscale were used to assess quality of life and tolerability. An exploratory translational endpoint was the association between ER histoscores and clinical benefit.

2.4. Study conduct

The study was coordinated by the National Health and Medical Research Council (NHMRC) Clinical Trials Centre, University of Sydney in collaboration with Australian New Zealand Gynaecological Oncology Group (ANZGOG). The study was performed in accordance with the NHMRC Statement on Ethical Conduct in Research Involving Humans and the Declaration of Helsinki. Ethical approval was obtained at all participating sites and all participants provided signed, written, informed consent. The study was prospectively registered: ACTRN1261000796088.

2.5. Response and toxicity criteria

Measurable disease was based on documentation at registration and determined whether the patient would be evaluable by RECIST v1.1 or CA125. Clinical deterioration without proven progression was determined by the treating physician. CA125 measurements were recorded at baseline and monthly for the first 3 months of treatment and then 3-monthly thereafter. CT scans were performed at baseline and every 3 months for the first 12 months. For patients responding to treatment at one year, CT scans were then performed as required by the treating physician, including for confirmation of disease progression.

Adverse events were recorded monthly for the first 3 months and then 3-monthly, and toxicity was graded by National Cancer Institute Common Terminology Criteria for Adverse Events v4.0. Quality of life was measured at registration before treatment, monthly for the first 3 months, and then 3-monthly thereafter until progression.

2.6. Tissue microarray and immunohistochemical staining for ER and PR

Twenty-nine of the cases had tumor blocks available for ER and PR confirmation in a central laboratory. Twenty-nine had paraffin-embedded tumor tissue blocks available for construction of a tissue microarray. The study pathologist selected representative areas for coring from hematoxylin- and eosin-stained sections of diagnostic tumor blocks. Duplicate 1 mm cores were taken from each donor block and assembled in a recipient paraffin block by using a manual precision instrument (MTA-1, Beecher Instrument, Sun Prairie, Wisconsin, USA). Endometrium (positive control) and spleen (negative control) were included in each array. Microarray sections (4 μm) were mounted on Superfrost Plus microscope slides and dried at 37 °C for 72 h before staining. Immunohistochemical staining using primary antibodies to ER (Ventana SP1, Ref. 790–4324) and PR (Ventana 1E2, Ref. 790–2223) was performed using a Ventana BenchMark ULTRA IHC staining module according to the manufacturer's instructions (Ventana Medical Systems Inc., Tucson, Arizona, USA), visualized with diaminobenzidine and counterstained with hematoxylin. Staining was evaluated by two observers blinded to treatment response. Nuclear staining intensity (0 = negative, 1 = weak, 2 = moderate, 3 = strong) and a histoscore were determined for each core. The histoscore was the product of the percentage of the tumor within the core that had positive nuclear staining and the staining intensity (range 0–300). The mean histoscore and staining intensity were determined for duplicate cores when present. Tumor histoscore was analysed to determine an association with clinical benefit.

2.7. Ki67, TP53 and BRAF V600E staining

TMA sections were also stained for TP53 (Cat No. NCL-L-p53-DO7, Leica Biosystems), Ki67 (Cat No. 275R-16, Cell Marque) and BRAF V600E (Ref. 790–4855, Ventana) using the Ventana BenchMark ULTRA IHC staining module.

For BRAF V600E, tumors were scored as positive when >90% cells showed moderate to strong cytoplasmic staining and scored negative when there was no staining or only nuclear dot staining or weak staining of single interspersed cells.

TP53 staining was scored as “wild type pattern” (focal nuclear expression i.e. low but variable staining), “overexpression pattern” (intense staining in almost all cells indicates TP53 missense mutation) or “complete absence” (no staining of cancer cell nuclei with retained internal control indicates *TP53* null mutation).

Ki67 score was the average proportion of positive tumor cells in both cores.

2.8. Statistical considerations

The primary endpoint was the CBR at 3 months. The study had a stopping rule to allow for early termination if there was lack of efficacy. There was a pre-planned interim analysis after 25 evaluable subjects had been on study for at least 3 months and received at least 2 weeks of treatment, with the data reviewed by the Independent Data Monitoring and Safety Committee, who recommended continued recruitment to 50 patients.

The expected CBR at 12 weeks in patients with recurrent LGOCs was estimated to be 25%, based on a literature review. There were limited data on the response rates/clinical benefit rates in LGOC and the 25% CBR was selected as we felt that anastrozole would be of clinical interest if the CBR was at least 25%. A sample size of up to 50 patients was selected providing that at least 3 patients in the first 25 patients treated experienced a clinical benefit (i.e., minimum number of responses required to be consistent with the expected CBR).

Analysis of efficacy (overall response and 3-month clinical benefit) used the proportion of patients who had clinical benefit, with a 95% confidence interval (CI) for the estimates. These rates were based on all patients receiving anastrozole for the first 2 weeks (intention-to-treat population) as well as patients who were on study for at least 4 weeks (evaluable for response/clinical benefit) as predefined in the protocol. Comparisons were 2-tailed, with a nominal significance level of 0.05. Analyses of PFS and duration of clinical benefit used time-to-event methods, with Kaplan-Meier survival curves constructed for graphical

display, and unadjusted log-rank tests where appropriate. Death from any cause was considered an event. Clinical benefit rates at 6 and 12 months were estimated using the Kaplan-Meier method to allow for censored patients and 95% CIs calculated using the complimentary log-log transformation. 95% CIs for proportions were constructed by using the modified Wilson method. The conditional binomial exact test was used to test for association between binary variables and the Cochran-Armitage test used to test for trend amongst binomial proportions across levels of an ordinal explanatory variable. For QoL, to compare baseline and on-study scores, changes from baseline to each time-point were calculated and paired *t*-tests were performed. In addition, change scores between baseline and on-study averaged scores were also computed and assessed using one-sample *t*-tests. Linear regression was used to compare changes in quality-of-life scores between patients achieving a 3-month clinical benefit and those who progressed, with adjustment for the baseline score.

3. Results

3.1. Patient characteristics

Between May 2012 and June 2016, 36 patients with mean age 57 years (range 22–77) were recruited from 1 Belgian ($n = 7$), 1 New Zealand ($n = 2$) and 10 Australian centres ($n = 27$). The enrolment target of 50 patients was not reached due to slower than expected recruitment. Relevant baseline characteristics are listed in Table 1. The majority of patients were diagnosed with LGSC and two had SBOTs which recurred as LGSC. At the time of analysis, six patients were still on treatment; at their last clinic visit they had been on treatment for an average time of 31.1 months (range: 14.6–52.4). One of these patients had progressed based on RECIST at 6 months but remained on treatment.

Table 1
Patient characteristics at baseline ($n = 36$).

Characteristic	*n* (%)
Age [years, mean (range)]	57 (22–77)[a]
ECOG performance status	
0	23 (64)
1	13 (36)
Hormone receptor status	
ER+/PR-	16 (44)
ER+/PR+	20 (56)
Histology	
Low-grade serous carcinoma	34 (94)[b,c,d]
Low-grade endometrioid carcinoma	2 (6)
Lines of prior chemotherapy	
0	1 (3)
1	22 (61)
≥2	13 (36)
Prior chemotherapy	35 (97)
Prior radiotherapy	3 (8)
Treatment-free interval	
<6 months	11 (31)
6–12 months	8 (22)
>12 months	16 (44)
No prior chemotherapy	1 (3)
Method of response measurement	
RECIST V1.1	32 (89)
CA125 by GCIG criteria	4 (11)

ER, estrogen receptor; PR, progesterone receptor; GCIG, Gynecologic Cancer InterGroup;
[a] Numbers are n (%), except where shown otherwise.
[b] One case had tissue collected post neo-adjuvant chemotherapy and there was a difference in opinion between initial pathology report and later review. On review, the TP53 staining was considered to be consistent with high grade serous ovarian cancer.
[c] One case was mixed mucinous and serous carcinoma.
[d] Two cases were serous borderline ovarian tumors that recurred as low-grade serous carcinomas.

3.2. Antitumor activity

Amongst 36 enrolled patients, 23 patients (64%, 95% CI 48%–78%) had clinical benefit at 3 months, comprising 22 patients with stable disease and 1 patient with a RECIST partial response (Table 2). Of the 13 patients who progressed, 11 patients had documented progressive disease at 3 months and 2 patients had clinical progression. One additional patient progressed at the 6-month assessment giving a similar CBR at 6 months (61%, 95% CI 43%–75%) to the 3-month CBR. Eleven patients maintained clinical benefit at 12 months, yielding a 12-month CBR of 34% (95% CI 19%–50%). For 32 patients with measurable disease at baseline, CBR at 3 months was 66% (48%–80%) and CBR at 6 months was 62% (95% CI 43–76%).

Best study response amongst 36 evaluable patients included partial response by RECIST v1.1 in 5 patients (14%), stable disease in 18 patients (50%) and progressive disease in the remaining 13 patients (36%). Median PFS was 11.1 months (95% CI 3.2–11.9) (Fig. 1). Of the 23 patients with clinical benefit at 3 months, the median duration of clinical benefit was 9.5 months (95% CI, 8.3 to 25.8 months) (Fig. 2). The two patients with SBOT who recurred as LGSC both had clinical benefit at 3 months and had best study responses of partial response by RECIST v1.1.

There was no significant association between PFS and prior treatment-free interval (TFI), which was defined as the time period between date of last treatment and date of enrolment in the study. Median PFS was 11.1 months amongst patients with TFI < 6 months, 14.0 months in patients with TFI 6–12 months, and 8.4 months in patients with TFI > 12 months (p-value 0.49) (Fig. 3).

3.3. ER staining and clinical benefit

ER-positivity was confirmed in 27 of 29 tumor samples that were available for central review. Most had high ER histoscores, in the range 201–300 ($n = 22$). Twenty of 29 tumor samples were also PR-positive. Of the 7 tumors that were not available for central review, 5 were ER-positive/PR-positive and 2 were ER-positive/PR-negative by local testing.

All 29 patients with tumor samples represented on the tissue microarray were evaluable for clinical benefit at 12 weeks. CBR were not significantly different between patients with ER histoscores of 0–100 (40%, $n = 5$), 101–200 (50%, n = 2), and 201–300 (64%, n = 22) ($p = 0.3$).

3.4. Staining for Ki-67, TP53 and BRAF V600E

Amongst 28 samples with suitable tumor material, median Ki-67 index (percentage of cancer cells with positive nuclear staining) was 10% (range < 1%–20%). One of 29 available tumor samples was collected from a patient who had received neoadjuvant chemotherapy and not suitable for Ki-67 analysis. All but one tumor sample had “wild-type”

Table 2
Objective clinical benefit (complete response, partial response, or stable disease) at 3 and 6 months based on RECIST/CA125 (n = 36).

Endpoint	*Response*	*n (%)*	*95% CI*
Clinical benefit at 3 months	Clinical benefit	23 (63.9%)	(47.6%–77.5%)
	Progressive disease	11 (30.6%)	
	Clinical progression	2 (5.6%)	
Response at 3 months	Partial response[c]	1 (2.8%)	(0.5%–14.2%)
	Stable disease	22 (61.1%)	
	Progressive disease	13 (36.1%)	
Clinical benefit at 6 months	Clinical benefit	21 (60.8%)[a,b]	(42.9%–74.6%)
	Progressive disease	12	
	Clinical progression	2	
	Censored	1	

[a] Actuarial rate (6-month scan took place at 6.3 months for the additional patient that progressed).
[b] 3 partial responses were observed by 6 months.
[c] Partial response was based on RECIST v1.1.

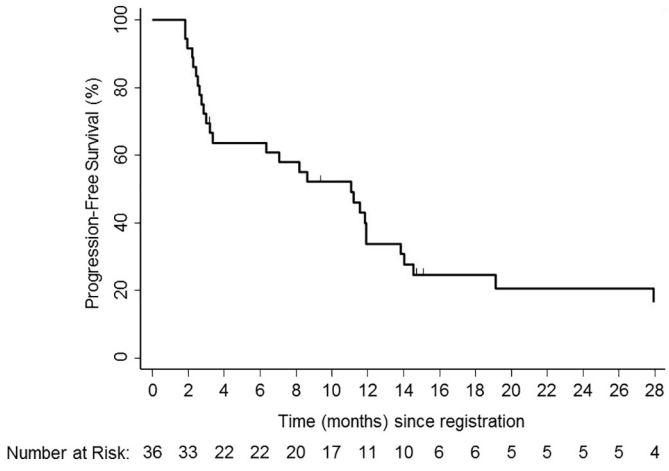

Fig. 1. Progression-free survival.

staining pattern for TP53; the one sample with "overexpression" staining pattern for TP53 was from the patient who was pre-treated with neoadjuvant chemotherapy. Pathology review suggested that this case may be HGSOC because of TP53 staining results and chemotherapy effect may have compromised histo-pathological grading, but this patient was included as she was reported to have LGSC at the site with more tissue available and because the clinical behavior was consistent with a LGSC. Only 1 low grade serous carcinoma had positive staining for BRAF V600E mutation.

3.5. Safety

Treatment with anastrozole was generally well-tolerated (Table 3). The most common side effects were fatigue, hot flushes and arthralgias, the majority of which were grade 1 in severity. Grade 3 hot flushes, anorexia, nausea and vomiting were each reported in <6% of patients. Two patients stopped treatment for adverse events, including arthralgias, hot flushes and decreasing QOL.

3.6. Quality of life

Belgium did not participate in the QoL component of PARAGON. QoL data were available for 28 patients, including 1 patient who had follow-up data only. Compliance with completing QoL questionnaires was high: 93% completed questionnaires at baseline (27/29), 86% at 1 month (25/29), 79% at 2 months (22/28) and 96% at 3 months (23/24).

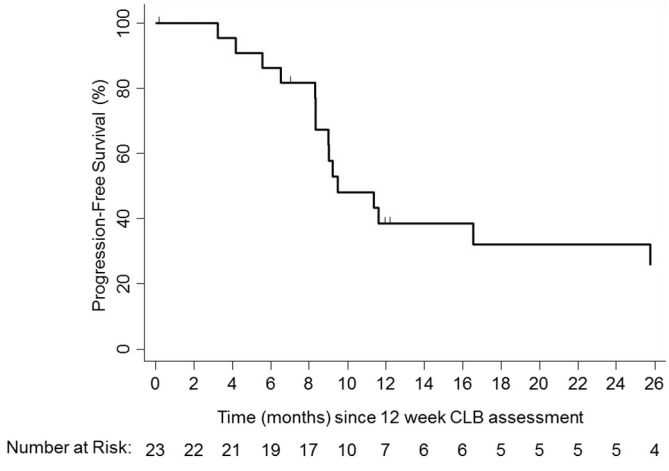

Fig. 2. Duration of clinical benefit for patients with clinical benefit at 3 months ($n = 23$).

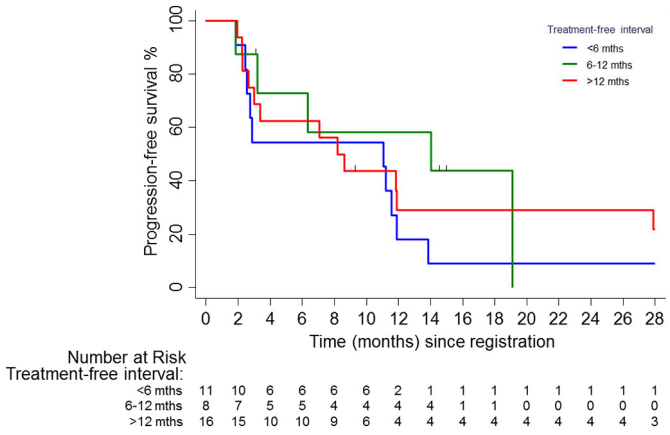

Fig. 3. Progression-free survival by treatment-free interval.

There were no clinically meaningful or statistically significant changes from baseline in the QLQ-C30 subscales (i.e., functioning, global QoL and symptom) averaged over the total time on-study. By time point there was a trend towards improved social functioning at 1 month (mean 10.4 point increase, $n = 24$, 95% CI -0.1 to 21.0, $p = 0.053$), a change defined as small (subtle but clinically relevant) by Cocks et al. [41]

At 1 month after commencing anastrozole, patients with clinical benefit at 3 months ($n = 16$) reported significantly better physical functioning (adjusted difference 10.3 points, 95% CI 1.8 to 18.8, $p = 0.02$), role functioning (adjusted difference 16.9 points, 95% CI 3.4 to 30.5, $p = 0.017$), fatigue (adjusted difference 17.1 points, 95% CI -29.7 to −4.5, p = 0.01) and pain (adjusted difference 12.6 points, 95% CI -25.0 to −0.3, $p = 0.046$) compared to patients who had progressed by 3 months ($n = 9$). Patients with clinical benefit at 3 months ($n = 18$) also reported better global quality of life at 3 months compared to

Table 3
On-study toxicity, worst grade (n = 36).

Event and grade	*n* (%)
Anorexia	
1	2 (6)
2	2 (6)
3	1 (3)
Headaches	
1	9 (25)
Nausea	
1	7 (19)
2	1 (3)
3	2 (6)
Fatigue	
1	19 (53)
2	2 (6)
Vomiting	
1	3 (8)
2	1 (3)
3	2 (6)
Alopecia	
1	2 (6)
Hot flashes	
1	17 (47)
2	3 (8)
3	1 (3)
Rash	
1	4 (11)
Arthralgia	
1	20 (56)
2	3 (8)
Vaginal dryness	
1	5 (14)
2	3 (8)

progressors ($n = 3$) (adjusted difference 27.6 points, 95% CI 3.5 to 51.8, $p = 0.027$).

For the endocrine-therapy specific quality-of-life data from the FACT-ES questionnaire averaged on-study scores declined significantly from baseline for the physical wellbeing subscale (mean 1.8 point decrease, $n = 27$, 95% CI -3.0 to −0.6, $p = 0.006$) and the total FACT-ES score (mean 5.3 point decrease, $n = 26$, 95% CI -9.5 to −1.0, p = 0.017). There was a trend towards worse endocrine subscale scores at 3 months and a significant decrease in endocrine subscale scores by 6 months (mean 3.4 point decrease, $n = 16$, 95% CI -6.5 to −0.3, $p = 0.033$). There was no significant change from baseline in any other individual domain scores for both averaged on-study scores and by individual time-points.

4. Discussion

To the best of our knowledge, this is the first prospective trial of an aromatase inhibitor in women with recurrent or metastatic SBOTs and LGOC. The majority of patients entered had LGSC (32/36), two had low-grade endometrioid cancers and two had SBOTs which had recurred as LGSC. At the 12-week assessment there was 1 patient with a partial response by RECIST v1.1 and 22 patients with stable disease, representing an overall CBR at 3 months of 64%. However, an additional four patients with stable disease at 3 months went on to achieve a partial response, resulting in a best study response rate of 14%. The 6-month CBR was 61%, which may be a more appropriate time point to assess clinical benefit in these patients rather than 12 weeks. This study confirms that anastrozole is generally a well-tolerated treatment, with patients reporting predominantly Grade 1–2 toxicities including fatigue, hot flashes and arthralgias and only 2 patients stopping treatment for adverse effects.

The best tumor response rate of 14% is comparable with previous reports of an objective response rate of 9% in a previous retrospective analysis of patients with recurrent LGSC of the ovary who were treated with a variety of endocrine therapies at MD Anderson [37]. This study reported 6 complete responses amongst 64 patients, whereas in PARAGON study no patients achieved a complete response. This difference may be possibly due to the rigid criteria for RECIST response in a prospective clinical trial and the variety of hormonal treatments patients received in the MD Anderson series, including selective estrogen receptor modulators, gonadotrophin releasing hormone agonists, different classes of aromatase inhibitors and combinations of the above.

A high proportion of patients had stable disease with a CBR (comprising stable disease, partial response and complete response) at 6 months of 61%, which is similar to 6 month progression-free survival of 61% in the MD Anderson series [37]. Furthermore, patients with clinical benefit maintained this benefit for a median of 9.5 months from the 3-month assessment. Given how indolent these tumors can be, it is difficult to know whether the stable disease was related to treatment with anastrozole or is simply a reflection of the biology of these tumors.

When comparing patients with clinical benefit at 3 months and patients who progressed by 3 months, the former reported significantly better physical functioning, role functioning, fatigue and pain scores on the QLQ-C30 questionnaire at 1 month compared to baseline. This suggests that patients who benefit from anastrozole are likely to experience improvements in patient-reported outcomes soon after commencing treatment supporting the notion of “clinical benefit”. The small decreases in the total FACT-ES, endocrine subscale and physical wellbeing scores in the FACT-ES questionnaire are consistent with the known toxicity of anastrozole.

Somewhat surprisingly, the majority of patients with LGOC or SBOTs which relapsed as LGSC in PARAGON (97%) had received prior chemotherapy. We carried out an exploratory analysis of the relationship between prior treatment-free interval and clinical benefit but did not find a significant difference in progression-free survival by treatment-free interval ($p = 0.49$).

There are many challenges to carrying out clinical trials in women with SBOTs and LGOC due to their relative rarity and their generally indolent clinical course, which raises important questions regarding trial endpoints and interpreting stable disease as well as defining clinical benefit. The design of future studies of SBOTs and LGOC should take their natural history into account by investigating efficacy at later time points such as clinical benefit at 6 months and incorporate patient reported outcomes. SBOTs are diagnosed at earlier stages and associated with a much better prognosis than invasive ovarian carcinomas and most patients with Stage I tumors do not relapse after surgery [25,26]. In contrast, LGSC, which account for approximately 10% of all serous ovarian cancers, tend to present in younger patients than HGSOC but are commonly Stage III at initial diagnosis. Despite the limited benefit of chemotherapy, LGSC patients debulked to no residual disease have a much longer survival than HGSOC [3,6,7].

The majority of tumor samples (76%) available for translational research had high ER histoscores in the range 201–300. The high ER histoscores and confirmed ER-positivity in nearly all available samples is consistent with high rates of ER expression previously reported in LGSC and SBOTs [22–24]. Nearly all tumor samples in our study were ER-positive and had high ER histoscore, but not all patients had clinical benefit at 3 months. Furthermore, the rate of clinical benefit did not differ significantly between patients with high, low and intermediate ER histoscores. This suggests that ER-positivity alone is not predictive of benefit from hormone therapy, and other biomarkers should be sought.

In our study, all analysed tumor specimens had low Ki-67 indices (≤20%), as would be expected for low-grade tumors. TP53 overexpression is rarely detected in LGSC, which supports the argument that LGSC and SBOT evolve along a different carcinogenic pathway to HGSOC [42]. The one tumor sample with an “overexpression” pattern of staining for TP53 was from a patient after receiving neoadjuvant chemotherapy. A BRAF V600E mutation was detected in only one of 29 tumor samples (3%) [20]. BRAF mutations have been reported in higher rates in SBOTs in other studies [15,20]; however, SBOTs only made up a small proportion of our study sample (6%).

A potential limitation is the relatively small number of patients treated. The sample size of 36 patients provides 72% of the information available of the planned 50 patients but is still sufficiently large enough to allow for clinical interpretation of the results of this study, which are in keeping with the findings of retrospective studies. The study did not meet its accrual target due to the rarity of these tumors and possibly also because of off label use of aromatase inhibitors. We also excluded patients who had prior hormone therapy. This highlights the difficulties of conducting prospective trials in this population of patients, but nevertheless our results do provide prospective estimates of clinical benefit and response rates to anastrozole (and probably all aromatase inhibitors).

In this prospective trial in women with recurrent or metastatic SBOTs and LGOC, anastrozole was well-tolerated and associated with clinical benefit in just over 60% of patients at both 3 and 6 months and supports the place of aromatase inhibitors in patients with these rare tumors. Combining aromatase inhibitors with other agents may be associated with greater benefit and will be explored in future studies.

Conflicts of interest

MT reports personal fees from Roche, outside the submitted work. PB reports personal fees from Astra Zeneca, outside the submitted work. KMS reports other from AMGEN, personal fees from MERCK, and personal fees from Pfizer, outside the submitted work. PG reports grants from NHMRC Clinical Trials Centre, during the conduct of the study. AdF reports grants from Cancer Institute New South Wales, grants from Cancer Australia, grants from Cancer Council NSW, during the conduct of the study; and grants from AstraZeneca, outside the submitted work. YA reports personal fees from Astra Zeneca, outside the submitted work. JG reports personal fees from AstraZeneca and MSD,

outside the submitted work. MF reports personal fees from ASTRA ZENECA, personal fees from MSD, and non-financial support from ASTRA ZENECA, outside the submitted work.

RLO, FA, ON, AD, PS, LM, TM, CJK, KT, TB and JS have nothing to disclose.

Author contributions

MF conceived of and designed the study. RLO was responsible for statistical analysis. MF, FA, PB, OM, PG, AD, PS, YA, JG and TB were involved in data acquisition. TM, CJK, AdF, KT and JS conducted the translational analysis. MF, RLO and MT analysed and interpreted the data. MF, MT, AdF, TM and RLO prepared the manuscript. All authors contributed to manuscript editing and review and approved the final version.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ygyno.2019.06.011.

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