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Nivolumab and ipilimumab for metastatic castration-resistant prostate cancer with an immunogenic signature: the multicentre, two-cohort, phase 2 NEPTUNES study.

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Running head: Nivo plus Ipi for selected metastatic prostate cancer patients

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1 **Context**

2 **Key objective:** To determine whether selection of metastatic castration-resistant prostate
3 cancer patients according to an immunogenic signature, which included tumour immune
4 infiltrate, enriches for responses to a combination of immune checkpoint inhibitors compared
5 to previous trials in unselected populations.

6 **Knowledge generated:** NEPTUNES reports higher response rates for selected patients
7 treated with nivolumab and ipilimumab compared to previous trials, with some patients
8 achieving long durable responses. This is the first trial that selects prostate cancer patients
9 based on the presence of high immune infiltrate, which as a standalone marker results in a
10 promising response rate of 43%.

1 **Abstract**

2 **Purpose**

3 Efficacy of immune checkpoint inhibitors in unselected patients with metastatic
4 castration-resistant prostate cancer (mCRPC) is limited. The NEPTUNES study evaluated
5 combination nivolumab and ipilimumab in mCRPC patients with an immunogenic
6 signature (ImS+).

7 **Methods**

8 This open-label 2-cohort phase II trial enrolled ImS+ mCRPC patients progressing on ≥ 1
9 previous line of treatment. ImS+ was defined by: 1) mismatch repair deficiency
10 (MMRD); 2) DNA damage repair gene loss; and/or 3) high inflammatory infiltrate (HII).
11 Patients received four doses of nivolumab 1mg/kg+ipilimumab 3mg/kg (C1) or
12 nivolumab 3mg/kg+ipilimumab 1mg/kg (C2) followed by nivolumab 480mg every 4
13 weeks up to 10 cycles. The primary endpoint was composite response rate (CRR)
14 assessed radiologically, biochemically, and by reduction of circulating tumour cells.
15 Secondary endpoints included toxicity, progression-free survival, overall survival, and
16 duration of response.

17 **Results**

18 Between May 2018 and June 2022, 35 (C1) and 36 (C2) patients commenced treatment.
19 The CRR in C1 was 14/35 (40%, 90%CI 26-55%) and in C2 was 9/36 (25%, 90%CI 14-
20 40%). The overall CRR was 23/71 (32%, 90%CI 23-43%). Response rates were higher
21 in patients with MMRD (7/10), BRCA2 loss (4/8), and HII +/- other ImS+ features
22 (13/30). Duration of response for patients with HII without other ImS+ features, DNA
23 repair gene loss without MMRD, and MMRD was 2.6, 17.3 and 10 months, respectively.
24 Grade 3-4 treatment-related adverse events occurred in 22/35 (63%) in C1 and 12/36
25 (33%) patients in C2. There were no treatment-related deaths.

26 **Conclusion**

27 Nivolumab 1mg/kg+ipilimumab 3mg/kg is an active treatment in ImS+ pre-treated
28 mCRPC. Nivolumab 3mg/kg+ipilimumab 1mg/kg has less toxicity but may have lower

- 1 efficacy. HII is a promising prospectively tested predictive biomarker in prostate cancer
- 2 that could be integrated into future trials.

1 Introduction

2 Prostate cancer is the most common cancer in men and, despite new treatments, mortality
3 rates remain high.¹ Checkpoint inhibitor (CPI) therapy has changed the management of many
4 tumours but response rates in metastatic prostate cancer are low. In unselected patients with
5 metastatic castration-resistant prostate cancer (mCRPC), CPI monotherapy has led to
6 radiological or biochemical responses in less than 10% of patients,^{2,3} whereas combination of
7 nivolumab and ipilimumab has led to a 10-25% response rate.^{4,5} Given the well-recognised
8 toxicities of combination CPI therapy, this level of activity has been deemed insufficient for
9 clinical benefit. Despite these underwhelming results, exceptional responders to CPI have
10 been reported.³

11 Prostate cancer has an immunosuppressive tumour microenvironment characterised by low
12 inflammatory infiltrate, low tumour mutational burden (TMB), low neoantigen burden and
13 low checkpoint molecule expression.^{6,7} Enrichment of T-cells within the tumour
14 microenvironment has been correlated with CPI response in a range of tumours.⁸⁻¹¹ In
15 patients with mCRPC and biallelic *CDK12* loss, 2/4 patients responded, specifically those
16 with high inflammatory infiltrate (HII).¹² We and others have shown that a subgroup of
17 patients with prostate cancer have a prominent inflammatory infiltrate,^{7,13} but whether this
18 enriches for response to CPI is unknown. Additionally, genomic aberrations in DNA repair
19 genes¹⁴ have been linked to increased neoantigen burden, higher immune infiltrate, and
20 higher response rates to CPI.^{12,15-17}

21 We hypothesized that patients with mCRPC would be more likely to respond to combination
22 CPI if they had immunogenic molecular or pathological features. We present the results of the
23 NEPTUNES phase 2 study, where we tested two schedules of nivolumab and ipilimumab in
24 molecularly-selected patients with mCRPC.

1 **Methods and materials**

2 *Study design and patients*

3 This multi-centre, open label, phase 2 study recruited patients in 2 sequential dosing cohorts
4 testing the response, survival and safety of the combination of ipilimumab and nivolumab in
5 patients deemed to have a putative immunogenic molecular profile. Cohort 2 (C2) was
6 opened following the successful recruitment to cohort 1 (C1) to test efficacy and safety of
7 lower dose of ipilimumab and higher dose of nivolumab. The study was conducted in 9
8 hospitals across the UK.

9 Metastatic prostate cancer patients who were ≥ 18 years old, had histologically confirmed
10 prostate adenocarcinoma, and either had archival prostate cancer tissue available or were
11 willing to undergo a new biopsy were eligible for pre-screening (see Data Supplement, Trial
12 protocol).

13 Patients were deemed to be immunogenic signature positive (ImS+) if they had at least one of
14 the following: i. loss of expression of mismatch repair proteins (MLH1, MSH2, MSH6 and/or
15 PMS2) by immunohistochemistry (mismatch repair deficiency - MMRD), ii. a deleterious
16 biallelic aberration in one or more genes involved in the DNA repair machinery as assessed
17 by next-generation sequencing (DNA damage repair deficiency - DDRD) and, iii. HII on a
18 multiplexed immunohistochemistry assay defined as T-cells (CD8+/CD4+/FOXP3+)
19 representing more than 20% of the total nucleated cells in the region of interest. (Data
20 Supplement, Figure S1).

21 ImS+ patients could be considered for enrolment into the main study. Patients had to have
22 WHO performance status 0-1, life expectancy ≥ 6 months and confirmed progression to
23 mCRPC, while on continuous androgen deprivation therapy with a serum testosterone < 1.73
24 nmol/L. Patients were also required to have progressed on at least one life-prolonging

systemic therapy for prostate cancer other than androgen deprivation therapy (i.e., androgen receptor pathway inhibitor, taxane-based chemotherapy or Radium223).

The trial protocol was approved by the National Research Ethics Committee (17/SC/0369).

This study adhered to the principles of the Declaration of Helsinki, Good Clinical Practice guidelines, and local regulations. The authors guarantee the accuracy and completeness of the data and analysis and affirm adherence to the protocol throughout the trial. This trial is registered with ClinicalTrials.gov, NCT03061539 (Feb 15, 2017).

Procedures

Patients participating in the pre-screening phase had their tumour tissue analysed (see appendix). Multiplexed immunohistochemistry for CD8, CD4 and FoxP3 was carried out on the tumour samples as described previously⁷. Four slides were stained by immunohistochemistry for MLH1, MSH2, MSH6 and PMS2 using the FDA-approved VENTANA MMR RxDx Panel (Roche) as per manufacturer instructions. UW-OncoPlex comprehensive genomic profiling, that included microsatellite instability and TMB analysis, was performed at the University of Washington.^{18,19}

In C1, patients received 1mg/kg nivolumab and 3mg/kg ipilimumab (Nivo1/Ipi3) on a 3-weekly cycle for up to 4-cycles followed by a 6-week delay and then a 480mg flat dose of nivolumab every 4 weeks for up to 10 cycles. In C2, patients received 3mg/kg nivolumab and 1mg/kg ipilimumab (Nivo3/Ipi1) and had a 3-week delay prior to flat dose nivolumab.

Patients received treatment until unequivocal progression, unacceptable toxicity or withdrawal of consent. Early discontinuation of ipilimumab and start of monotherapy was allowed for patients who could not tolerate combination treatment. Dose reductions were not permitted. If treatment was delayed by >6weeks, the patient had to permanently discontinue trial treatment. Patients could continue treatment, irrespective of PSA and imaging results, providing they were deriving clinical benefit as assessed by the treating physician.

Patients underwent a CT chest abdomen and pelvis and bone scan at baseline, weeks 9, 18, 27, then 12-weekly up to 2 years, and every six months afterwards up to 5 years. Baseline disease features at the time of registration were collected, including presence of measurable disease and burden of disease as defined in the CHAARTED trial.²⁰ Radiological response was assessed using modified RECIST1.1 and PCWG3 criteria for CT and bone scan respectively. A blood sample for circulating tumour cells (CTC) response assessment was collected at the same timepoints together with additional blood samples for exploratory biomarker analyses. CTC were enumerated using the CellSearch Platform. Adverse events (AEs) were recorded based on patients' reports, physical examination, and laboratory tests and graded using Common Terminology Criteria for AEs, version 4.03.

Outcomes

The primary outcome measure was the composite response rate (CRR). A patient was considered to have achieved a response if any of the following criteria were met: i) radiological response confirmed ≥ 4 weeks later; ii) PSA response $\geq 50\%$ confirmed by a second PSA test ≥ 4 weeks later; or iii) conversion of CTC count from ≥ 5 cells/ 7.5 ml at baseline to < 5 cells/ 7.5 ml at 9 weeks confirmed by a second CTC test ≥ 4 weeks later as previously described.²¹

The secondary endpoints included overall survival, progression-free survival, duration of response and an assessment of the frequency and severity of AEs. Overall survival was measured as date of registration to date of death from any cause. Patients were censored at the date last seen alive if they did not have a reported death at the time of final analysis.

Progression-free survival time was measured from registration to either objective radiological progression (RECIST 1.1), PSA progression (PCWG3), unequivocal evidence of clinical progression or death from any cause, whichever occurred first. Patients without documented

1 progression or death were censored at the date of the last follow up. Duration of response was
2 defined as time from composite response to progression or death.

3 An exploratory analysis was conducted to assess the disease control rate at 6 months (DCR6),
4 defined as absence of a progression event (as per same definition used for progression free
5 survival) beyond 6 months from registration. Subgroup analyses based on baseline
6 characteristics were conducted in the combined cohort population.

7 ***Statistical analysis***

8 The study was designed so that each cohort would be analysed separately. The sample size
9 for both arms was calculated using an A'hern single-stage Phase II design, where the null
10 hypothesis was a CCR of 20% or below, while the alternative hypothesis was a CRR of 40%
11 (with a one-sided α level of 0.05 and a β level of 0.20). Following the A'Hern design, if at
12 least 12 (34%) of 35 evaluable patients had a treatment composite response (PSA, CTC or
13 radiological), then the null hypothesis would be rejected. Comparisons for continuous and
14 categorical variables were performed using Wilcoxon rank sum test and Chi-Square test,
15 respectively. A Cox proportional hazards model was used to compare duration of response
16 and survival across cohorts. Time to event outcomes were reported using standard survival
17 analysis based on Kaplan Meier method. AEs were reported descriptively using frequencies
18 and percentages.

Results

Between February 2018 and April 2022, 380 patients were enrolled into pre-screening. The overall rate of ImS+ samples was 31% (n=119) with 11% (n=41) of the samples failing the test, most commonly due to insufficient tumour DNA (Data Supplement, Figure S2). Overall, 74 ImS+ patients (recruitment target was originally 71) were enrolled into the main study. C1 recruited 36 patients between May 2018 and October 2020 and C2 recruited 38 patients between November 2020 and June 2022. One patient in C1 and 2 patients in C2 withdrew from the study prior to drug dosing. These 3 patients were replaced and not included in efficacy analysis. Patients were deemed ImS+ as per the following features: 10/71 (14%) had MMRD, 48/71 (68%) had DDRD, and 30/71 (42%) had HII, with 14/71 (20%) meeting multiple criteria (Figure 1).

The median age was 67 years (range 50-78) in C1 and 70 (range 53-82) in C2 with a performance status of 0 in 47% and 45% patients respectively. Patients were heavily pre-treated with 50% and 60% patients having had ≥ 3 previous life-prolonging therapies, including 89% and 71% having received docetaxel, in C1 and C2, respectively. Measurable disease was detected at baseline in 53% and 50% of patients, with 67% and 61% having high burden of disease at the time of registration in C1 and C2, respectively (Table 1, Appendix and Data Supplement, Table S1).

The CRR was 14/35 (40%) patients in C1 enabling rejection of the null hypothesis. In C2, the CRR of 9/36 (25%) patients was insufficient to reject the null hypothesis. Overall, CRR in both cohorts was 23/71 (32%, 90% CI 23-43), significantly higher than the pre-specified threshold for an ineffective treatment at $\leq 20\%$ (Table 2). CRR was higher in patients with RECIST-measurable disease [51%, 90% CI, 37-66%] than non-RECIST-measurable disease [12%, 90% CI, 4-25%]. In both cohorts, all patients who had a radiological response also had a PSA response (Data Supplement, Figure S3), hence the difference observed is not due to the

absence of radiological responses in the patients with non-RECIST-measurable disease.

Patients also had a higher CRR if they had received prior docetaxel treatment (38% [90% CI, 27-49%] versus 13% [90% CI, 2-36%], Data Supplement, Table S2).

CTC response could not be assessed for 22/35 (63%) and 21/36 (58%) patients, in C1 and C2, respectively, the main reason being insufficient CTCs (<5 cells/7.5ml) at baseline to observe a conversion at week 9. Of the 7 patients with a CTC response, only 2 patients had a CTC-only response.

Responses were recorded for 7/10 (70%) patients with MMRD, 4/8 (50%) patients with biallelic BRCA2 loss-of-function, and 2/7 (29%) patients with biallelic CDK12 loss-of-function. CRR in patients with exclusively ATM, CHD1 or CHEK2 biallelic alterations was low (2/23, 9%). Of the 20 patients that were enrolled into the main study based on HII alone, seven (35%) responded to treatment. The CRR in all patients that had HII, regardless of genomic features, was 43% (13/30).

TMB was evaluable in 64/71 patients (90%) (responders n=21, non-responders n=43) across both cohorts. TMB was higher among responders versus non-responders (Median responders, 4 mut/Mb [IQR: 1.5-15.5] versus non-responders, 2 mut/Mb [IQR: 2-3], p=0.007, Wilcoxon test). However, the difference was primarily driven by enrichment for MMRD patients among the responders (median MMRD 18.5 mut/Mb [IQR 13.5-23] versus MMR proficient 2 mut/Mb [IQR 1-3]). No difference in the TMB between responders and non-responders was observed after excluding MMRD patients (Data Supplement, Figure S4).

With a follow up time of 50.8 months in C1 and 30.6 months in C2 the median progression-free survival time was 6.6 months (95% CI: 4.9-10.0) in C1 and 4.0 months (95% CI: 3.2-7.6) in C2 (Figure 2A). Median overall survival time was 16.2 months (95% CI: 9.2-22.8) and 16.6 months (95% CI: 10.7-33.4) (Figure 2B).

The median duration of response was 10·2 months (95% CI:1·8-17·4) in C1 and 6·4 months (95% CI: 1·1-NR) in C2. Duration of response was significantly longer in patients with MMRD (10·2 months) or DDRD (17·4 months) compared to patients with only HII (2·6 months - Data Supplement, Figure S5). Median duration of response for HII +/- other features was 6.5 months.

The DCR6 was 57% in C1 and 44% in C2. Overall, 36/71 (51%) patients had DCR6. Of the 23 patients achieving a composite response, 15 also had a duration of response greater than 6 months.

The use of steroid was common in the NEPTUNES trial, with 37/71 (52%) patients receiving steroids during treatment or follow-up period. Specifically, 34/37 patients received high-dose steroids (prednisolone>10mg or equivalent dose) during trial treatment. For one additional patient the dose was not specified. Among the 12 responders who received steroids, only 4 had steroids administered before first evidence of response, 6 started steroids after response had already been confirmed, while for 2 patients the date of initiation was missing.

All treated patients were evaluable for toxicity. Treatment related AEs (TRAEs) of any grade occurred in 34/35 patients in C1 and 28/36 patients in C2. Grade 3-4 TRAEs occurred in 22/35 (63%) patients in C1 and 12/36 (33%) patients in C2 (Table 3 and Data Supplement, Table S3). There were no treatment related deaths. At least one serious AE was recorded for 49/71 patients, with 31/71 having a serious TRAE (Data Supplement, Table S4). In C1 there were 17 (49%) patients that required a dose delay and 12 (34%) that had to permanently discontinue treatment due to toxicities. For C2 this was 13 (36%) and 6 (17%), respectively (Appendix).

Discussion

NEPTUNES met its primary endpoint demonstrating a CRR of 40% in C1 and 32% in the combined cohort. This is the first study in prostate cancer to prospectively enrol patients

based on a signature including inflammatory infiltrate. The CRR of 43% in patients with HII is a remarkable increase compared to what observed in unselected patients.^{2,5} Notably, only 40% of MMRD tumours exhibited HII, indicating that the evaluation of multiple biomarkers is crucial to identify mCRPC patients who could respond to CPI. A small phase 2 trial recently recruited patients with early-stage triple negative breast cancer with high tumour infiltrating lymphocytes to receive neoadjuvant nivolumab and ipilimumab, reporting a pathological complete response rate of 33% (5/15 patients) and larger trials using this patient selection are in accrual.²²

NEPTUNES results corroborate the recently published data from the INSPIRE study. This study recruited patients with mCRPC and MMRD, TMB ≥ 7.1 muts/Mb, presence of biallelic CDK12 inactivation, or BRCA2 mutation to receive Nivo3/Ipi1 followed by nivolumab maintenance, creating some overlap with the NEPTUNES trial. Both trials reported excellent outcome for patients with MMRD.²³ Interestingly, in the retrospective series from Graham et al and Sena et al,^{24,25} a lower proportion of patients with MMRD, 8/15 (53%) and 11/17 (65%), respectively, achieved a biochemical response to CPI monotherapy. In the prospective MMRD pan-tumour Keynote-158 study 2/8 (25%) had a radiological response.²⁶

Collectively, this data suggest that combination CPI should be tested against monotherapy in prospective clinical trials.

Response in the BRCA2-aberrant population was 23% in the INSPIRE trial versus 50% in this study. Multiple factors may have contributed to this difference, including the smaller patient subgroup in NEPTUNES (8 versus 20); the higher dose of ipilimumab administered in C1; and the inclusion of patients with mono-allelic BRCA2 mutations and/or BRCA2 signature in INSPIRE versus the more-strict NEPTUNES criteria which only allowed patients with BRCA2 biallelic loss.

We did not include TMB as a selection biomarker in the NEPTUNES study as a clear threshold for TMB had not been defined in prostate cancer at the time of study design. While in our study TMB did correlate with response, the correlation was lost when MMRD patients were excluded from the analysis. Both the CheckMate 650 and the INSPIRE trials did not show a consistent association between efficacy and TMB.^{23,27}

CTC data was lacking, mainly due to insufficient cells detected at baseline. Only 2 patients had a CTC-only response and the duration of response was short. Therefore, CTC enumeration seems to have limited utility in the response assessment of mCRPC patients treated with CPIs.

Two different dose regimens of nivolumab and ipilimumab were tested. This study was not powered to detect differences in efficacy between the two regimens. With this limitation, the numerically lower CRR in the Nivo3/Ipi1 cohort did not impact OS. However, the Nivo3/Ipi1 dose regimen was considerably better tolerated ($\geq$ G3 tox, Nivo1/Ipi3 63% vs Nivo3/Ipi1 33%) and had a lower discontinuation rate, in keeping with data from other tumour types.^{28,29}

The majority of patients in this study were extensively pre-treated. Unexpectedly, docetaxel-naïve patients had a numerically lower CRR compared to previously-exposed patients. Notably, both the CheckMate 650 study and the INSPIRE study reported increased response rates in docetaxel-naïve patients.^{5,23} CRRs were higher in patients with measurable disease. This is consistent with other trials where patients with bone-predominant disease rarely showed a PSA response after immunotherapy,² possibly due to a hostile immune micro-environment enriched in M2-macrophages, T-helper-17 cells and cytokines such as TGF β and IL-6.^{30,31}

Analyses within this trial were limited by lack of contemporaneous tissue meaning that molecular classifications often pre-dates the CPI treatment by many years. Evaluation of composite response was hindered by missing CTC data and the bone-predominant nature of

1 prostate cancer metastases and was therefore mainly driven by biochemical response. Due to
2 the relatively small number of patients enrolled and different length of follow-up between
3 cohorts, the subgroup analyses were not statistically powered and should be considered
4 hypothesis generating.

5 In summary, nivolumab and ipilimumab demonstrated anti-tumour activity in selected
6 patients with pre-treated prostate cancer. The Nivo3/Ipi1 dose schedule was better tolerated
7 than the Nivo1/Ipi3 dose schedule and may be equally effective in prolonging patients'
8 survival. We prospectively tested a composite biomarker which enriched for responders and
9 has the potential to push the risk-benefit dial in favour of combination CPI in mCRPC. Over
10 40% of patients with high inflammatory infiltrate responded to combination CPIs, regardless
11 of the presence of other biomarkers. Larger randomised clinical studies for biomarker-
12 selected metastatic prostate cancer are warranted.

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8 **Contributions**

9 ML was the chief investigator. ML, CS, YNSW, SQ, GA, TM designed the trial. ML acquired
10 funding. CP, EK carried out and interpreted the DNA sequencing. AF, AH, JL and TM carried
11 out the pre-screening immunohistochemistry and LH and JH performed the CTC analysis.
12 MR, GL, and ML performed formal analysis and prepared figures. ML wrote the first draft of
13 the manuscript with crucial input from MR and GL. RJJ, PS, DHJ, SJC, LH, AJ, BK, MK,
14 YNSW and GL recruited patients and contributed to the Trial Management Group. MR, GL,
15 AT and AH have accessed and verified all the data in the study.
16 All authors approved the final manuscript, had full access to all the data in the study and
17 accept responsibility to submit for publication.

18

1 **Declaration of interests**

2 **AP** reports partial employment by Genesis Care UK and payment or honoraria for lectures,
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14 AstraZeneca, and Sanofi-Aventis. GA reports a Patent on blood methylation markers
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Figure Legend

Figure 1: Patient response by molecular selection criteria.

TMB and gene alterations determined using the University of Washington OncoPlex assay. Different types of gene alterations are defined by colour code. Only genes that were altered in ≥ 2 patients are included. TMB and gene alterations were deemed "N/A" (grey boxes) when the OncoPlex assay was not performed (due to insufficient sample quality/quantity) or results were uncertain due to low tumour content. Stacked bars above feature columns represent proportion of patients that are positive/negative for that feature. Duration of response (right) is represented using a discontinuous x axis to include 4 patients with response >4 years. DDR, DNA Damage Repair; MMR, Mismatch Repair; TMB, Tumour Mutation Burden; PSA, Prostate Specific Antigen; CTC, Circulating Tumour Cells; N/A, Data Not Available.

Figure 2: Patient survival and types of response

A) Progression-free survival; B) Overall survival; C) Swimmer plot showing clinical course of individual patients including time and type of response and progression on trial treatment.

Table 1: Baseline characteristics

Baseline characteristics	Cohort 1 (%) N=36	Cohort 2 (%) N=38
Age, in years		
Median (range)	67 (50 to 78)	70 (53 to 82)
WHO PS		
Fully Active	17 (47%)	17 (45%)
Restricted in Physical activity	19 (53%)	21 (55%)
Baseline PSA (ng/ml)		
Median (range)	88.5 (0.4 to 13610.4)	140.7 (2.7 to 3107.7)
Previous Docetaxel		
No	4 (11%)	11 (29%)
Yes	32 (89%)	27 (71%)
Previous ARPI		
No	5 (14%)	1 (3%)
Yes	31 (86%)	37 (97%)
Number of prior therapies		
1	4 (11%)	5 (13%)
2	14 (39%)	10 (26%)
3	11 (31%)	16 (42%)
≥ 4	7 (19%)	7 (18%)
Biomarker Features		
Mismatch repair deficiency by IHC	5 (14%)	5 (13%)
• Mismatch repair deficiency and homologous recombination deficiency	1 (3%)	0 (0%)
• Mismatch repair deficiency with unknown homologous recombination status	0 (0%)	2 (5%)
• Mismatch repair deficiency and high inflammatory infiltrate	2 (6%)	2 (5%)
• Mismatch repair deficiency only	2 (6%)	1 (3%)
Homologous recombination deficiency	23 (64%)	21 (55%)
• BRCA	3 (8%)	7 (18%)
• Non-BRCA	20 (56%)	14 (37%)
High inflammatory infiltrate	14 (39%)	17 (45%)
• High inflammatory infiltrate and mismatch repair deficiency	2 (6%)	2 (5%)
• High inflammatory infiltrate and homologous recombination deficiency	3 (8%)	3 (8%)
• High inflammatory infiltrate with unknown homologous recombination status	3 (8%)	3 (8%)
• High inflammatory infiltrate only	6 (17%)	9 (24%)

ARPI, Androgen Receptor Pathway Inhibitor; IHC, Immunohistochemistry; PSA, Prostate Specific Antigen; WHO PS, World Health Organisation Performance Status

1

Table 2: Composite response by cohort

Responses	All patients N= 71		Cohort 1 N=35		Cohort 2 N=36	
	N (%)	90% CI	N (%)	90% CI	N (%)	90% CI
Composite response rate	23(32%)	23% to 43%	14(40%)	26% to 55%	9(25%)	14% to 40%
PSA Response	21(30%)	21% to 40%	12(34%)	21% to 50%	9(25%)	14% to 40%
Radiological response	8(11%)	6% to 19%	4(11%)	4% to 24%	4(11%)	4% to 24%
CTC Response	7(10%)	5% to 18%	5(14%)	6% to 28%	2(6%)	1% to 16%

2

CI, Confidence Interval; PSA, Prostate Specific Antigen; CTC, Circulating Tumour Cells

3

1 Table 3: Treatment related adverse event by cohorts

Organ Class and AE names (CTCAE v4.03)*	Cohort 1		Cohort 2		Combined cohort	
	Any grade (%)	Grade 3-4 (%)	Any grade (%)	Grade 3-4 (%)	Any grade (%)	Grade 3-4 (%)
Blood and lymphatic system disorders						
Anaemia	3 (8.6%)	2 (5.7%)	1 (2.8%)	-	4 (5.6%)	2 (2.8%)
Endocrine disorders						
Adrenal insufficiency	3 (8.6%)	2 (5.7%)	3 (8.3%)	1 (2.8%)	6 (8.5%)	3 (4.2%)
Hyperthyroidism	7 (20.0%)	-	-	-	10 (14.1%)	-
Hypothyroidism	4 (11.4%)	-	8 (22.2%)	1 (2.8%)	12 (16.9%)	1 (1.4%)
Eye disorders						
Retinal Vascular Disorder	1 (2.9%)	1 (2.9%)	-	-	1 (1.4%)	1 (1.4%)
Uveitis	1 (2.9%)	1 (2.9%)	-	-	1 (1.4%)	1 (1.4%)
Gastrointestinal disorders						
Colitis	8 (22.9%)	5 (14.3%)	2 (5.6%)	1 (2.8%)	10 (14.1%)	6 (8.5%)
Diarrhoea	23 (65.7%)	7 (20.0%)	15 (41.7%)	3 (8.3%)	38 (53.5%)	10 (14.1%)
Nausea	13 (37.1%)	1 (2.9%)	6 (16.7%)	1 (2.8%)	19 (26.8%)	2 (2.8%)
Vomiting	8 (22.9%)	1 (2.9%)	4 (11.1%)	1 (2.8%)	12 (16.9%)	2 (2.8%)
General disorders and administration site conditions						
Fatigue	14 (40.0%)	5 (14.3%)	10 (27.8%)	-	24 (33.8%)	5 (7.0%)
Fever	7 (20.0%)	-	-	-	10 (14.1%)	-
Infusion related reaction	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Immune system disorders						
Other: Hepatitis	1 (2.9%)	-	1 (2.8%)	1 (2.8%)	2 (2.8%)	1 (1.4%)
Other: Hypophysitis	2 (5.7%)	1 (2.9%)	-	-	2 (2.8%)	1 (1.4%)
Other: Myasthenia Gravis	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Infections and infestations						
Anorectal Infection	1 (2.9%)	1 (2.9%)	-	-	1 (1.4%)	1 (1.4%)
Investigations						
Alanine aminotransferase increased	5 (14.3%)	1 (2.9%)	3 (8.3%)	2 (5.6%)	8 (11.3%)	3 (4.2%)
Alkaline phosphatase increased	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Aspartate aminotransferase increased	4 (11.4%)	1 (2.9%)	4 (11.1%)	2 (5.6%)	8 (11.3%)	3 (4.2%)
Serum amylase increased	4 (11.4%)	1 (2.9%)	1 (2.8%)	1 (2.8%)	5 (7.0%)	2 (2.8%)
Weight loss	4 (11.4%)	-	4 (11.1%)	-	8 (11.3%)	-
Metabolism and nutrition disorders						
Acidosis	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Anorexia	6 (17.1%)	-	4 (11.1%)	1 (2.8%)	10 (14.1%)	1 (1.4%)
Hyperglycaemia	2 (5.7%)	1 (2.9%)	3 (8.3%)	1 (2.8%)	5 (7.0%)	2 (2.8%)
Hypokalaemia	2 (5.7%)	1 (2.9%)	-	-	2 (2.8%)	1 (1.4%)
Hyponatremia	2 (5.7%)	1 (2.9%)	1 (2.8%)	1 (2.8%)	3 (4.2%)	2 (2.8%)
Musculoskeletal and connective tissue disorders						
Arthralgia	5 (14.3%)	1 (2.9%)	2 (5.6%)	1 (2.8%)	7 (9.9%)	2 (2.8%)
Back pain	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Myalgia	5 (14.3%)	-	-	-	-	-
Nervous system disorders						
Headache	5 (14.3%)	-	-	-	-	-
Lethargy	5 (14.3%)	-	4 (11.1%)	-	9 (12.7%)	-

Organ Class and AE names (CTCAE v4.03)*	Cohort 1		Cohort 2		Combined cohort	
	Any grade (%)	Grade 3-4 (%)	Any grade (%)	Grade 3-4 (%)	Any grade (%)	Grade 3-4 (%)
Renal and urinary disorders						
Acute kidney injury	4 (11.4%)	-	-	-	-	-
Other: Nephritis	-	-	1 (2.8%)	1 (2.8%)	1 (1.4%)	1 (1.4%)
Respiratory, thoracic and mediastinal disorders						
Dyspnoea	7 (20.0%)	-	3 (8.3%)	1 (2.8%)	10 (14.1%)	1 (1.4%)
Pneumonitis	7 (20.0%)	2 (5.7%)	3 (8.3%)	-	10 (14.1%)	2 (2.8%)
Skin and subcutaneous tissue disorders						
Erythema	1 (2.9%)	1 (2.9%)	-	-	1 (1.4%)	1 (1.4%)
Other: Rash Unspecified	11 (31.4%)	2 (5.7%)	2 (5.6%)	-	13 (18.3%)	2 (2.8%)
Pruritus	9 (25.7%)	-	6 (16.7%)	-	15 (21.1%)	-
Rash maculo-papular	12 (34.3%)	3 (8.6%)	2 (5.6%)	-	14 (19.7%)	3 (4.2%)
Vascular disorders						
Vasculitis	1 (2.9%)	1 (2.9%)	-	-	1 (1.4%)	1 (1.4%)

*Adverse events by organ system included in the table if at least one grade 3-4 event was recorded or incidence of any grade more than 10%