Effects of ADXS–PSA With or Without Pembrolizumab on Survival and Antigen Spreading in Metastatic, Castration–Resistant Prostate Cancer Patients (Results from KEYNOTE–046)

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INTRODUCTION

ADXS–PSA

- ADXS-PSA is a live attenuated *Listeria monocytogenes* (*Lm*)-listeriolysin O (LLO) immunotherapy bioengineered to secrete an antigen adjuvant fusion protein (tLLO-PSA) consisting of a truncated fragment of the listeriolysin (tLLO) fused to human prostate-specific antigen (PSA)[1,2]
  - ADXS-PSA functions as an immunomodulator and T-cell vaccine vector, and is rapidly taken up by antigen-presenting cells where expression of full-length human PSA stimulates anti-tumor immune responses
  - Anti-tumor immunity occurs via several mechanisms, primarily including generation of cytotoxic T lymphocytes targeting PSA and through antigen spreading
  - *Lm*-based immunotherapies such as ADXS-PSA reprogram the tumor microenvironment (TME): the number and function of immunosuppressive regulatory T cells and myeloid-derived suppressor cells within the TME are reduced and expression of programmed cell death protein-1 ligand (PD-L1) is upregulated

PEMBROLIZUMAB

- Pembrolizumab (Keytruda®), a checkpoint inhibitor, is a high-affinity, IgG4/kappa isotype humanized anti-PD-1 monoclonal antibody that blocks the binding of PD-1 receptor to its ligands PD-L1 and PD-L2
- In unselected patients with metastatic castration-resistant prostate cancer (mCRPC), objective response rates with pembrolizumab alone were seen in 3%-5% of patients.[3] In bone-predominant disease the disease control rate was 22% per Response Evaluation Criteria in Solid Tumors (RECIST) v1.1 and median overall survival (95% confidence interval [CI]) was 14.1 (10.8-17.6) months[3]
- Correlative immune analyses suggest a broader immune stimulation with ADXS-PSA + pembrolizumab, including T-cell responses against PSA and antigen spreading in most patients

RATIONALE FOR COMBINATION THERAPY WITH ADXS–PSA AND PEMBROLIZUMAB

- Synergistic activity of the combination of ADXS *Lm*-based immunotherapies with PD-1 blocking antibodies has been shown in animal models[4]
- The magnitude of the PSA-specific T-cell response in ADXS-PSA-treated mCRPC patients is associated with anti-tumor effects, as evidenced by increased T-cell reactivity to PSA and other prostate cancer antigens[5]
- Initial results of Part A and Part B of this study were presented at ASCO 2018.[6] This poster focuses on Part B data

OBJECTIVES

- This is a phase 1/2, open-label, multicenter, dose-determining safety and tolerability study with a phase 2 expansion cohort

PRIMARY

- **Part A:** to evaluate safety and tolerability of ADXS-PSA monotherapy and select the recommended phase 2 dose (RP2D) in patients with mCRPC for use in Part B
- **Part B:** to evaluate safety and tolerability of ADXS-PSA in combination with pembrolizumab and to establish the RP2D for this combination in patients with mCRPC

SECONDARY

- To evaluate anti-tumor activity and progression-free survival (PFS) of ADXS-PSA monotherapy and ADXS-PSA + pembrolizumab combination therapy

EXPLORATORY

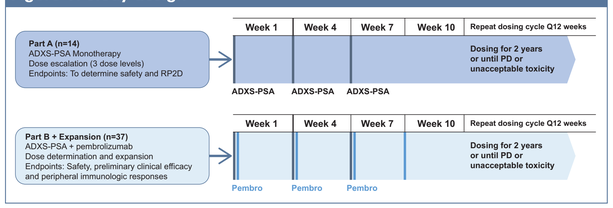
- Exploratory objectives included evaluating correlative immunologic and gene expression profiles of PBMCs for ADXS-PSA monotherapy and ADXS-PSA + pembrolizumab combination therapy as well as direct ELISpot assessment of T-cell responses to PSA and antigen spreading to other prostate cancer–associated antigens and autoantibody detection

METHODS

KEY ELIGIBILITY CRITERIA

- Progressive mCRPC on androgen deprivation therapy
- <3 prior systemic treatment regimens in the metastatic setting
  - Hormonal therapy was not considered prior therapy
- This is a phase 1/2, open-label, multicenter, dose-determining safety and tolerability study with a phase 2 expansion cohort (Figure 1)

Figure 1. Study design



DOSE DETERMINATION

- In the Part A monotherapy phase of the study (completed), the starting dose level (DL1) of ADXS-PSA was 1×10^9 CFU
  - Patients were administered ADSX-PSA intravenously (IV) every 3 weeks × 3 doses in a 12-week cycle for up to 24 months or until disease progression or discontinuation
  - The dose was escalated, remained the same, or de-escalated based on an interim safety data review according to predefined criteria for dose-limiting toxicity
  - DL1 (1×10^9 CFU): n = 7; DL2 (5×10^9 CFU), n = 3; DL3 (1×10^{10} CFU): n = 4
  - The RP2D was 1×10^9 CFU
- In the Part B combination therapy phase of the study (ongoing), patients were administered pembrolizumab 200 mg IV, followed by ADXS-PSA 1×10^9 CFU IV, every 3 weeks as above for up to 24 months or until disease progression or discontinuation

EFFICACY ASSESSMENTS

- Anti-tumor activity and PFS are evaluated based on RECIST v1.1, immune-related RECIST (irRECIST), and Prostate Cancer Working Group 2 (PCWG2) criteria

SAFETY ASSESSMENTS

- Safety is assessed by reported adverse experiences using Common Terminology Criteria for Adverse Events v4.03

BIOMARKER RESEARCH

- T cells are assessed for their specific response to PSA and other prostate cancer antigens, which may include prostate membrane antigen, prostatic acid phosphatase, and prostate stem cell antigen. T-cell responses are determined by enzyme-linked immunosorbent and/or enzyme-linked immunospot (ELISpot) assay
- TCR beta chain sequencing was performed to evaluate changes in clonality and diversity of T cells
- Flow cytometry evaluated changes in PBMC populations during treatment
- Serum cytokine and chemokine changes are determined to assess immune stimulation as a result of treatment
- Nanostring gene expression profiling evaluated changes in gene expression of PBMCs
- MSI status was evaluated via PlasmaSELECT™ R64 by PGDx along with identification of potential somatic and genomic mutations
- Screening for autoantibodies was conducted using HuProt™ Human proteome microarray (V 3.0)

RESULTS as of February 1, 2019

PATIENTS

- A combined total of 50 patients were treated in Part A (ADXS-PSA monotherapy) and Part B (ADXS-PSA + pembrolizumab combination therapy). Tables 1a and 1b present information on patient baseline characteristics and prior patient therapies since diagnosis. 36 patients from Part B were negative for microsatellite instability high (MSI-High) status (1 patient was not tested)

Table 1a. Baseline characteristics

Characteristic	Part A (N = 14[a])	Part B (N = 37)
Dose, n (%)	DL1, 1×10^9 CFU ADXS-PSA: 10 (71)	RP2D, 200 mg pembrolizumab + 1×10^9 CFU ADXS-PSA: 37 (100)
	DL2, 5×10^9 CFU ADXS-PSA: 1 (7)	0
	DL3, 1×10^{10} CFU ADXS-PSA: 2 (14)	0
Median age, years (range)	70.0 (57.0, 80.0)	68.0 (45.0, 92.0)
Time from diagnosis to treatment initiation, years, range	1.4, 20.6	0.9, 17.9
Median Gleason score (range)	8.0 (7.0, 10.0)[b]	9.0 (6.0, 10.0)[c]
Median PSA, ng/mL (range)	19.0 (4.2, 2456.0)	41.5 (0.1, 426.3)
ECOG PS, n (%)		
0	7 (50)	16 (43)
1	7 (50)	21 (57)
Presence of visceral metastases, n (%)		
Yes	4 (29)	11 (30)
No	10 (71)	26 (70)

[a]One patient was withdrawn prior to receiving treatment with ADXS-PSA. [b]Score available for 11 patients. [c]Score available for 22 patients.
DL, dose level; ECOG PS, Eastern Cooperative Oncology Group performance status; PSA, prostate-specific antigen

Table 1b. Prior therapies

Characteristic	Part A (N = 14[a])	Part B (N = 37)
Number of prior therapies since diagnosis, n (%)		
1	1 (7)	5 (14)
2	3 (21)	2 (5)
≥3	9 (64)	30 (81)
Prior therapies, n (%)		
Chemotherapy	5 (36)	21 (57)
Hormonal therapy	12 (86)	33 (89)
Next-generation hormonal agents		
Abiraterone only	3 (21)	7 (19)
Enzalutamide only	1 (7)	12 (32)
Abiraterone + enzalutamide	4 (29)	11 (30)
Immunotherapy[b]	6 (43)	7 (19)

[a]One patient was withdrawn prior to receiving treatment with ADXS-PSA.
[b]Immunotherapy included sipuleucel-T and PCaDCVAC, but no checkpoint inhibitors.

SAFETY

- Overall, of 50 treated patients, 49 (98%) experienced any grade treatment-related adverse events (TRAEs); Part A: 13 (100%); Part B: 36 (97%)
- The majority of TRAEs consisted of Grade 1-2 chills/rigors, fever, hypotension, nausea, and fatigue
- The combination of ADXS-PSA and pembrolizumab appeared safe and tolerable, with no additive toxicity observed

Table 2. Treatment-related adverse event (TRAE) summary*

TRAE, n (%)	Grade 1-2		Grade ≥3	
	Part A (N = 13)	Part B (N = 37)	Part A (N = 13)	Part B (N = 37)
Patients with ≥1 TRAE	13 (100)	36 (97)	5 (38)	12 (32)
Chills	9 (69)	31 (84)	1 (8)	0
Pyrexia	8 (62)	19 (51)	0	1 (3)
Nausea	5 (38)	15 (41)	0	0
Fatigue	4 (31)	10 (27)	1 (8)	3 (8)
Hypotension	4 (31)	7 (19)	2 (15)	1 (3)
Hypertension	0	4 (11)	2 (15)	6 (16)
Anemia	0	4 (11)	1 (8)	3 (8)
Decreased appetite	0	6 (16)	1 (8)	0
Hypothyroidism	0	7 (19)	0	0
Tachycardia	2 (15)	4 (11)	1 (8)	0
Vomiting	2 (15)	5 (14)	0	0
Headache	1 (8)	5 (14)	0	0
Diarrhea	1 (8)	3 (8)	0	1 (3)
Pain	1 (8)	4 (11)	0	0

*TRAEs occurring in ≥10% of patients (Part A + Part B)

EFFICACY

- At the cutoff date of February 1, 2019, median overall survival observed for patients in Part B was 21.1 months (range, 16.0-not reached) (Figure 2)
- Duration of follow-up in months, median (range): Part A, 4.44 (0.76-23.10); Part B, 14.26 (1.94-26.09)
- Prolonged survival can be observed in patients in the combination arm regardless of prior therapies, microsatellite stable (MSS) status, presence of visceral metastasis, or PSA change <50% or ≥50%

Figure 2. Overall survival: Part A (ADXS-PSA monotherapy) and Part B (ADXS-PSA + pembrolizumab combination)

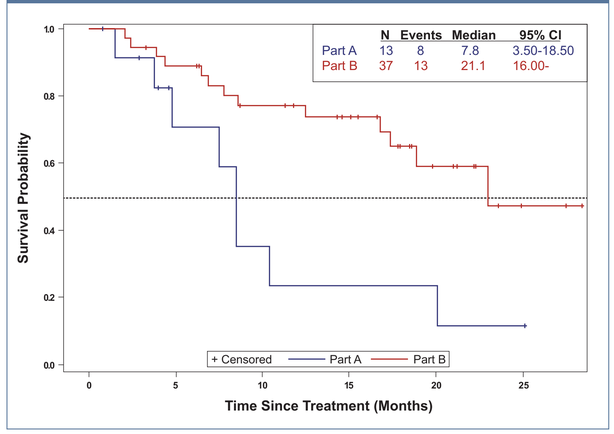
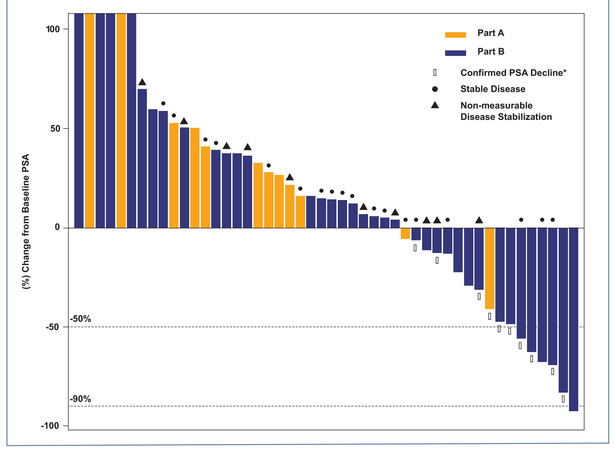


Figure 3. Maximal change in PSA at any time since treatment initiation



- 6 patients in Part B had ≥50% PSA declines from baseline; all 6 were still alive at the time of this analysis (Figure 4)
- Even when selecting for patients in Part B who did not have a ≥50% PSA decline (n = 30), median OS was still 21.1 months at the time of this analysis
- For patients in Part B, stable disease + disease stabilization rate observed with combination treatment was 59% (Table 3)

Table 3. Best overall response per RECIST v1.1

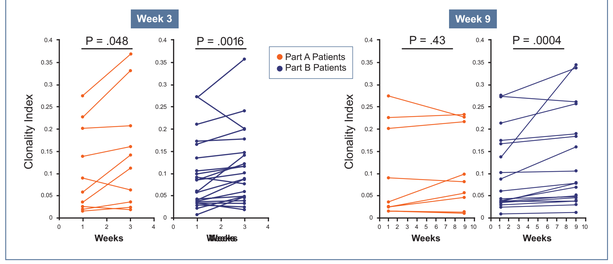
	Part A	Part B
Patients treated, n	13	37
Not evaluable, n (%)	1 (8)	0 (0)
Number with RECIST 1.1 measurable disease at baseline, n (%)	7 (54)	22 (59)
Complete response, n (%)	0 (0)	0 (0)
Partial response, n (%)	0 (0)	0 (0)
Stable disease, n (%)	5 (38)	13 (35)
Progressive disease, n (%)	3 (23)	13 (35)
Disease stabilization*, n (%)	1 (8)	9 (24)
Stable disease + disease stabilization	6 (46)	22 (59)

*Designation for patients who have bone lesions but no measurable soft tissue disease at baseline. Patients had bone scintigraphy and CT/MRI scans every 10 weeks per protocol, and were assessed by the investigator for disease progression/stabilization based on PCWG2 criteria.
CT, computed tomography; MRI, magnetic resonance imaging; RECIST, Response Evaluation Criteria in Solid Tumors; PCWG2, Prostate Cancer Working Group 2

EXPLORATORY ENDPOINTS: RESULTS OF CORRELATIVE/BIOMARKER ANALYSES

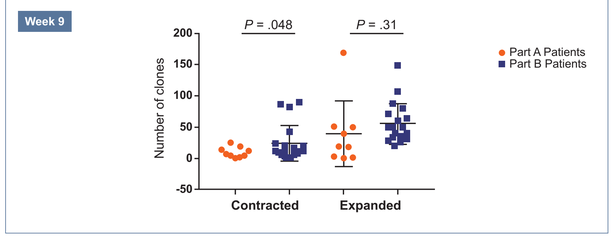
- T-cell receptor beta chain sequencing in peripheral blood mononuclear cells
  - ADXS-PSA combination with pembrolizumab extended T-cell expansion, suggesting a broader immune stimulation (Figure 4a)

Figure 4a. Significant clonal expansions at Weeks 3 and 9 in Part B patients and primarily at Week 3 in Part A patients



  - Increased contraction of T-cell clones in combination suggests that lower-avidity T-cell clones were reduced in favor of high-avidity T cells under PD-1 blockade (Figure 4b)

Figure 4b. No differences in the numbers of contracted and expanded clones between Part B SD and PD patients, Part B PSA responders and PSA nonresponders, and SD patients in Parts A and B. Numbers in each group were small



- NanoString PanCancer Immune Profiling Panel
  - Baseline levels of CCL5, and possibly HLA-C, may help predict patients for whom combination therapy will slow PSA kinetics
  - Part B patients with stable disease have higher expression levels of genes indicative of B-cell activation; Part A patients with stable disease exhibited higher expression levels of genes indicative of T cells
- ELISpot: T cells vs. PSA increased in 5 of 9 Part A patients and 11 of 17 Part B patients
- Antigen spreading was seen in most patients: 100% (9/9) in Part A and 85% (14/17) in Part B
- Microsatellite status has been evaluated in 36 of 37 Part B patients and all 36 were determined to be MSI-High negative

CONCLUSIONS

- The combination of ADXS-PSA and pembrolizumab appeared safe and tolerable in this heavily pretreated, unselected population of patients with bone-predominant and MSI-High-negative mCRPC
  - Treatment-related adverse events were mostly Grade 1-2 chills/rigors, fever, hypotension, nausea, and fatigue
  - No additive toxicity was observed with the combination therapy
- The combination of ADXS-PSA and pembrolizumab appears to show activity in an unselected patient population with MSI-High-negative mCRPC and might be associated with prolonged OS in this population
  - Median OS (95% CI): 21.1 months (16.0–not reached) in this study population including patients having failed chemotherapy for mCRPC and chemotherapy-naive patients
    - Survival benefit was seen regardless of PSA decline and microsatellite stable disease, which is known to be insensitive to immunotherapy
  - 59% (22/37) of patients had stable disease/disease stabilization
  - 40.5% (15/37) of patients had PSA declines; 16% (6/37) had ≥50% PSA declines from baseline; all 6 patients with ≥50% PSA declines were still alive at data cutoff
- There is a broader immune stimulation in the combination arm (which includes B-cell activation) than in ADXS-PSA monotherapy
  - Correlative immune analyses show T-cell responses against PSA (75%) and antigen spreading (85%) in most patients in the combination arm

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