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Combining Immune Checkpoint Blockade and Tumor-Specific Vaccine for Patients With Incurable Human Papillomavirus 16–Related Cancer
A Phase 2 Clinical Trial

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[+] Supplemental content

IMPORTANCE In recurrent human papilloma virus (HPV)–driven cancer, immune checkpoint blockade with anti–programmed cell death 1 (PD-1) antibodies produces tumor regression in only a minority of patients. Therapeutic HPV vaccines have produced strong immune responses to HPV-16, but vaccination alone has been ineffective for invasive cancer.

OBJECTIVE To determine whether the efficacy of nivolumab, an anti–PD-1 immune checkpoint antibody, is amplified through treatment with ISA 101, a synthetic long-peptide HPV-16 vaccine inducing HPV-specific T cells, in patients with incurable HPV-16–positive cancer.

DESIGN, SETTING, AND PARTICIPANTS In this single-arm, single-center phase 2 clinical trial, 24 patients with incurable HPV-16–positive cancer were enrolled from December 23, 2015, to December 12, 2016. Duration of follow-up for censored patients was 12.2 months through August 31, 2017.

INTERVENTIONS The vaccine ISA101, 100 μg/peptide, was given subcutaneously on days 1, 22, and 50. Nivolumab, 3 mg/kg, was given intravenously every 2 weeks beginning day 8 for up to 1 year.

MAIN OUTCOMES AND MEASURES Assessment of efficacy reflected in the overall response rate (per Response Evaluation Criteria in Solid Tumors, version 1.1).

RESULTS Of the 24 patients (4 women and 20 men; 22 with oropharyngeal cancer; median age, 60 years [range, 36-73 years]), the overall response rate was 33% (8 patients; 90% CI, 19%-50%). Median duration of response was 10.3 months (95% CI, 10.3 months to inestimable). Five of 8 patients remain in response. Median progression-free survival was 2.7 months (95% CI, 2.5-9.4 months). Median overall survival was 17.5 months (95% CI, 17.5 months to inestimable). Grades 3 to 4 toxicity occurred in 2 patients (asymptomatic grade 3 transaminase level elevation in 1 patient and grade 4 lipase elevation in 1 patient), requiring discontinuation of nivolumab therapy.

CONCLUSIONS AND RELEVANCE The overall response rate of 33% and median overall survival of 17.5 months is promising compared with PD-1 inhibition alone in similar patients. A randomized clinical trial to confirm the contribution of HPV-16 vaccination to tumoricidal effects of PD-1 inhibition is warranted for further study.

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Human papillomavirus (HPV) is the cause of nearly all cervical cancers and most oropharyngeal, anal, penile, vulvar, and vaginal cancers. Although many cancers are cured with initial treatment, recurrent cancer is frequently incurable and associated with relatively short survival. The E6 and E7 viral proteins, critical in driving HPV oncogenesis and foreign to the human immune system, represent ideal targets for therapeutic cancer vaccination. Recent data indicate that, at initial diagnosis, most patients with HPV-positive oropharyngeal cancer (OPC) exhibit a strong spontaneous immune response to HPV antigens that is associated with substantial infiltration of the cancer with HPV-specific T cells and an excellent prognosis[1]. However, in recurrent HPV-positive OPC, immune checkpoint blockade with anti–programmed cell death 1 (PD-1) antibodies pembrolizumab and nivolumab produces tumor regression in only a minority of patients.[2-5] Thus, we hypothesized that augmentation of the HPV-specific T-cell population by a therapeutic vaccine could increase the proportion of patients benefiting from anti–PD-1 therapy.

The vaccine ISA101, which is among the most promising vaccines targeted to E6 and E7, consists of 9 overlapping long E6 peptides (five 32-mer E6 peptides and four 25-mer E6 peptides) and 4 overlapping 35-mer E7 peptides (synthetic long peptide HPV-16 vaccine), covering the complete sequence of the HPV-16 E6 and E7 oncoproteins.[6] These long peptides effectively deliver antigens to dendritic cells, which then induce CD4$^+$ and CD8$^+$ T-cell responses by HLA classes I and II presentation of the HPV-16 E6 and E7 processed epitope peptides.[7,8]

In a landmark clinical trial, ISA101 demonstrated notable activity in high-grade vulvar intraepithelial neoplasia, with durable and complete remission in 9 of 19 patients at 2 years.[6] Furthermore, clinical responses were directly correlated with vaccine-activated T-cell immune responses against HPV-16. Recently, these results were confirmed with the additional observation that patients with a complete histologic response had also cleared the virus from these sites.[9] However, by itself the vaccine did not affect regression of advanced cervical cancer, suggesting that vaccine-activated T cells are held in check by a tumor-induced immunosuppressive environment.[10,11] Thus, there should be potential to enable the cytotoxic effects of vaccine-activated T cells by inhibiting mechanisms of immunosuppression.[12,13]

Human papillomavirus DNA vaccines targeting E6 and E7, such as VGX-3100 and GX-188E, have also been shown to induce potent HPV-specific CD4$^+$ and CD8$^+$ T-cell responses and regression of high-grade premalignant cervical lesions.[14-16] As with ISA101, the activity of VGX-3100 and GX-188E is limited to premalignant lesions. However, ISA101 is distinguished by the ability to induce HPV-specific T-cell responses in patients with end-stage cervical cancer.[10,17]

We report the results of a single-arm phase 2 clinical trial designed to evaluate the efficacy of ISA101 combined with PD-1 immune checkpoint blockade in patients with incurable HPV-16–positive malignant neoplasms.

Methods

Patients

Patients must have had histologically or cytologically documented diagnosis of incurable HPV-16–positive solid tumors

Key Points

Question Is the efficacy of programmed cell death 1 immune checkpoint inhibition increased by a tumor-specific vaccine in patients with incurable human papillomavirus 16–positive cancer?

Findings In this phase 2 clinical trial of nivolumab and human papillomavirus 16 vaccine ISA101, the primary end point was met, with a 33% overall response rate (8 of 24 patients), compared with response rates of 16% to 22% with programmed cell death 1 inhibitors alone in similar patients. Survival data were also encouraging, with a median survival of 17.5 months.

Meaning These data indicate that HPV-16 vaccination may augment the efficacy of programmed cell death 1 checkpoint inhibition and merit confirmation in a randomized trial.

from oropharyngeal, cervical, vulvar, vaginal, penile, or anal primaries with 0 or 1 line of treatment for recurrent HPV-16–positive cancer. Patients were required to be age 18 years or older, to have an Eastern Cooperative Oncology Group performance status of 0 or 1, and to have measurable disease per Response Evaluation Criteria in Solid Tumors, version 1.1 (RECIST, version 1.1).[18] Consent for a baseline biopsy was required. Major exclusion criteria included active central nervous system metastases and active autoimmune disease, except vitiligo, type 1 diabetes, and hypothyroidism. The study was conducted in accordance with the Declaration of Helsinki[19] and the International Conference on Harmonization Good Clinical Practice Guidelines.[20] The study protocol was approved by the University of Texas MD Anderson Cancer Center Institutional Review Board (trial protocol in Supplement 1). All patients provided informed written consent.

Study Design

This was a single-arm nonrandomized phase 2 clinical trial. The primary objective was assessment of efficacy as reflected in the overall response rate (ORR) per RECIST, version 1.1.[18] Secondary objectives were assessment of safety, tolerability, HPV-specific immune response, and estimation of progression- free survival (PFS) and overall survival (OS). Exploratory biomarker analyses included the correlation of programmed death-ligand 1 (PD-L1) expression with response and survival. Patients were treated with ISA101, 100 μg/peptide in Montanide adjuvant (Seppic) subcutaneously, for a total of 3 doses on days 1, 22, and 50 and nivolumab, 3 mg/kg intravenously, starting on day 8 adminstered every 2 weeks for a total of 12 months or progression of disease, toxic effects, or withdrawal of consent. Imaging was performed at baseline, prior to cycle 6 of nivolumab, and then every 6 weeks thereafter. Tumor biopsies were mandatory before treatment and planned at the time of first restaging. Blood samples were drawn at baseline, before the second and third dose of vaccine, prior to cycle 5 and 6 of nivolumab, and then every 3 months.

Statistical Analysis

Efficacy and safety were evaluated in all patients who received at least 1 dose of ISA101 and nivolumab. The 2-stage MiniMax

design by Simon[21] was used, targeting an alternative hypothesis response rate of 0.3 vs a null hypothesis response rate of 0.10 with 80% power and a 1-sided .03 significance level. A response was defined as ORR (complete response plus partial response), per RECIST, version 1.1.[18] This design required 2 or more responses in 15 patients in the first stage to accrue 10 additional patients in the second stage. A total of 6 or more responses in 25 patients was required to reject the null hypothesis. The ORR was based on investigators' assessment. Independent blinded radiology review was not performed.

Toxic effects were monitored continuously in cohorts of 5 patients and graded according to National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.03.[22] Duration of response, PFS, and OS were estimated with Kaplan-Meier statistics. Duration of response was calculated from time of first response to progression or death, whichever occurred first. Progression-free survival was calculated from treatment start date to progression date or death date, whichever was earlier. Overall survival was calculated from treatment start date to death date or to the last follow-up date. Patients who had not reached a time-to-event end point were censored at the last follow-up date.

The nonparametric Wilcoxon rank sum test was used to evaluate correlation between PD-L1 tumor, immune, and combined scores with ORR. The log-rank test was used to evaluate correlations of PD-L1 scores with PFS and OS. $P < .05$ was considered statistically significant. The Welch 2-sample t test was used to evaluate enzyme-linked immunospot (ELISPOT) values at baseline and after vaccination. The analyses were performed using R, version 3.3.2, a publicly available statistical tool (https://www.r-project.org/).

HPV Genotype
Archival tumors were required to harbor HPV-16 in order to be eligible for this clinical trial. The Cervista HPV-16/18 assay was conducted according to previously published methods.[23] Its use has been validated in formalin-fixed tissue and in HPV-related oropharyngeal cancer.[24]

Immunohistochemistry
Tumor sections from formalin-fixed, paraffin-embedded core needle biopsies were used for immunohistochemical analysis as previously described.[25] Hematoxylin-eosin–stained sections were first assessed visually for evaluable viable tumor cells. An automated staining system (BOND-MAX; Leica Microsystems) with antibodies against PD-L1 (Clone 28-8, dilution 1:400; Abcam, cat#ab205921) was used. Programmed death-ligand 1 was detected using a Novocastra Bond Polymer Refine Detection kit (Leica Microsystems). Immunohistochemistry slides were digitally scanned at ×200 using a ScanScope Aperio AT Turbo slide scanner (Leica Microsystems) and visualized using the ImageScope software program (Leica Microsystems). Visual scoring was also performed. Each individual core needle biopsy was divided by a pathologist (J.R.C.) into tumor cell and tumor stroma compartments using the Aperio tool box (Leica Microsystems). The tumor cell compartment was defined as groups or nests of malignant cells and the tumor stroma compartment was represented by the fibrous

tissue present between tumor cells. Programmed death-ligand 1 expression was scored considering a partial or complete membranous staining at any intensity, on a scale from 0% to 100%, of malignant cells or tumor-associated immune cells, respectively.

Interferon-γ ELISPOT Assay
Interferon-γ ELISPOT assay was performed as described previously with modifications.[9] Peripheral blood mononuclear cells were thawed, plated at 0.5 to 1 × 10⁶ cells/well, and stimulated for 4 days with a pool of 9 HPV-16 E6 synthetic long peptides and 4 HPV-16 E7 synthetic long peptides. After 4 days, cells were harvested, counted, and plated at a concentration of 50 000 or 100 000 cells/well in 200 μL of complete tumor-infiltrating lymphocyte culture media in anti-IFN-γ (5 μg/mL; Mabtech, catalog 3420-3)–coated ELISPOT plates (Millipore; catalog MAHAS4510). After overnight incubation at 37°C, plates were washed with phosphate buffered saline with 0.05% of polysorbate 20 (Invitrogen) and incubated for 1 hour at 37°C with 1 μg/mL of biotin-labeled anti-IFN-γ antibody (Mabtech, catalog 3420-6). Plates were then incubated with diluted extravidin-alkaline phosphatase (1:5000 dilution), for 1 hour at room temperature. Spots were immediately developed by 5-bromo-4-chloro-3-indolyl-phosphate in conjunction with nitro blue tetrazolium and counted on an ImmunoSpot ELISPOT reader (CTL Immunospot Reader, software version 6.0.0.0). The assay was conducted in triplicates. Phorbol 12-myristate 13-acetate and ionomycin were used as positive controls and media alone as negative controls. Values were normalized for reactivity in the negative control. ISA101 was supplied by ISA Pharmaceuticals and nivolumab by Bristol-Myers Squibb.

Results

Patients and Treatment
From December 23, 2015, to December 12, 2016, 34 patients were screened, and 24 patients (22 with oropharyngeal cancer, 1 with anal cancer, and 1 with cervical cancer) were enrolled. Enrollment closed prior to accrual of the 25th patient owing to impending expiration of the ISA101 vaccine lot. Patient characteristics are summarized in **Table 1**. A flowchart of the patients is provided in **Figure 1**.
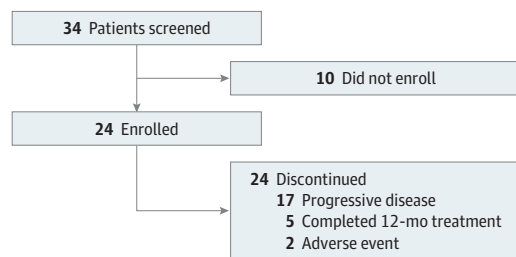
Efficacy
Response data are detailed in **Table 2**. Best percentage change in target lesions (per RECIST, version 1.1) from baseline and duration of response are depicted in **Figure 2** and eFigure 1 in Supplement 2. There were 4 responders in the first 15 patients, directing accrual to the second stage. Of the total 24 patients accrued, 8 patients, all with OPC, achieved a response: 2 complete responses and 6 partial responses, for an ORR of 33% (90% CI, 19%-50%).[26] Three patients achieved their best overall response of partial response subsequent to the first restaging at 11 weeks. To provide a perspective in comparison with monotherapy effects of PD-1 inhibition, response data for patients with OPC and subsets of that group are presented in Table 2, including patients refractory to platins and cetuximab (progression

Table 1. Characteristics of the 24 Study Patients

Characteristic	Patients, No. (%)
Age, median (range), y	60 (36-73)
Male sex	20 (83)
Race/ethnicity	
White	21 (88)
Black	2 (8)
Hispanic	1 (4)
ECOG performance status	
0	12 (50)
1	12 (50)
Primary site	
Oropharynx	22 (92)
Cervix	1 (4)
Anus	1 (4)
Platin status	
Exposed	23 (96)
Resistant, 6 mo	19 (79)
Treatment setting	
First line	10 (42)
Second line	14 (58)
Baseline PD-L1 expression	
Evaluability	
Tumor	18 (75)
Immune	20 (83)
≥1%, No./total No.	
Tumor	7/18 (39)
Tumor and immune	13/18 (72)

Abbreviations: ECOG, Eastern Cooperative Oncology Group;
PD-L1, programmed death-ligand 1.

Figure 1. Patient Flowchart

within 6 months of treatment) and for patients treated second-line for recurrence. The ORR in these admittedly small subsets confirms efficacy similar to the less heavily treated overall population. Median duration of response was 10.3 months (95% CI, 10.3 months to inestimable), with 5 of 8 responses ongoing at the time of analysis (August 25, 2017).

Median PFS was 2.7 months (95% CI, 2.5-9.4 months) and median OS was 17.5 months (95% CI, 17.5 to inestimable), with median follow-up time among censored patients of 12.2 months (eFigures 2 and 3 in Supplement 2). The rate of PFS at 6 months was 37% (95% CI, 22%-63%) and at 12 months was 25% (95% CI, 12%-50%). The rate of OS at 6 months was 75% (95% CI, 59%-94%) and at 12 months was 70% (95% CI, 54%-91%). Estimates of PFS and OS in the 22 patients with OPC were identical to those in the overall population.

Safety

Treatment-related adverse events are listed in eTable 1 in Supplement 2. The toxic effects profile was additive for expected reactions to ISA101, namely, injection site reactions and fever, and those predicted from nivolumab, such as fatigue, diarrhea, and hepatoxicity. Two patients discontinued treatment owing to asymptomatic grade 3 immune adverse events (asymptomatic grade 3 transaminase level elevation in 1 patient and grade 4 lipase and amylase elevation level in 1 patient). There were no other dose-limiting toxic effects.

Efficacy by PD-L1 Status

Data on the evaluability of baseline core needle biopsies for PD-L1 expression and on the incidence of PD-L1 expression of 1% or more are provided in Table 1. The correlation of clinical response to baseline PD-L1 expression score in tumor, immune, and combined compartments is shown in eFigure 4 in Supplement 2. Distribution of PD-L1 expression, medians, and interquartile ranges are in eTable 2 in Supplement 2. In view of the bimodal, nonnormal distribution observed in both tumor and immune cells, data were subjected to the Wilcoxon rank-sum test, using a score of 1% or more as threshold for positivity. A significant correlation of PD-L1 expression with response was demonstrated for tumor score, with an ORR of 43% (3 of 7) compared with 18% (2 of 11) in PD-L1–negative tumors ($P = .04$). Neither the immune scores nor the combined scores were correlated with response. Furthermore, tumor score, immune score, and their combined scores for PD-L1 less than 1% vs 1% or more were not correlated with PFS or OS.

Efficacy by HPV-16–Specific Immune Response

Interferon-γ release data from cultured peripheral blood lymphocytes in response to pooled HPV-16 E6 and E7 peptides, segregated by clinical response, are shown in eFigure 5 in Supplement 2. Of those with baseline data, there was no or minimal reactivity, consistent with what was previously observed in patients with vulvar intraepithelial neoplasia or cervical cancer.[6,9,11] After vaccination, a variable increased number of HPV-specific T cells was observed in both responders and nonresponders. The immune response did not correlate with any efficacy end points, suggesting that local factors in the tumor environment exert preeminent influences on vaccine effect.

Discussion

ISA101, an HPV-16 synthetic long peptide vaccine combined with nivolumab, a PD-1 immune checkpoint inhibitor, exhibited promising efficacy outcomes in patients with recurrent HPV-16–positive cancer. With a total of 24 patients treated instead of the 25 planned, the statistical power was reduced to 77.6%. Despite that, the primary end point was met, with an ORR of 33% (8 of 24 patients) and responses were durable with 63% (5 of 8 patients) ongoing. Furthermore, the 12-month OS rate of 70% and median OS of 17.5 months are encouraging for this population. The combination of ISA101 and nivolumab was very well tolerated, with only additive effects from each agent apparent without increased immune adverse events, relative

Table 2. Response Overall, in Oropharyngeal Cancer, and by Treatment History[a]

| | No. (%) | | | | | |
| | | Patients With Oropharyngeal Cancer | | | | |
Response per RECIST, version 1.1	All Patients (N = 24)	All (n = 22)	Platin-Refractory Disease (n = 17)	Cetuximab-Refractory Disease (n = 8)	Platin- and Cetuximab-Refractory Disease (n = 6)	Received Second-line Treatment (n = 12)
Overall response rate	8 (33)	8 (36)	6 (35)	5 (63)	3 (50)	5 (42)
Complete response	2 (8)	2 (9)	2 (12)	1 (13)	0	2 (17)
Partial response	6 (25)	6 (27)	4 (24)	4 (50)	3 (50)	3 (25)
Stable disease	3 (13)	2 (9)	1 (6)	0	1 (17)	1 (8)
Disease control rate[b]	11 (46)	10 (45)	7 (41)	5 (63)	4 (67)	6 (50)
Progression of disease	13 (54)	12 (55)	10 (59)	3 (38)	2 (33)	6 (50)

Abbreviation: RECIST, Response Evaluation Criteria in Solid Tumors.

[a] Refractory indicates progression of disease within 6 months of treatment.

[b] Disease control rate is calculated by adding the percentages of patients whose tumors exhibited complete response, partial response, and stable disease and indicates lack of progression per RECIST.

Figure 2. Efficacy of ISA101 and Nivolumab
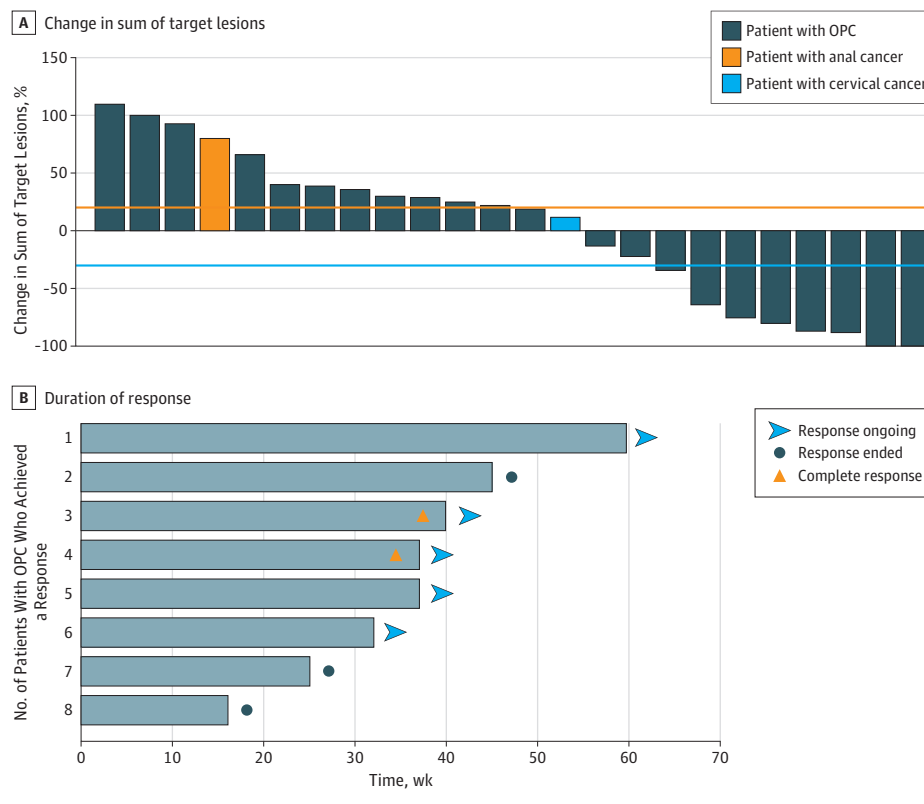
A, Best percentage change in sum of target lesions from baseline and postbaseline assessments per Response Evaluation Criteria in Solid Tumors, version 1.1 (blue horizontal line). The orange horizontal line indicates a 20% increase, considered progressive disease per Response Evaluation Criteria in Solid Tumors, version 1.1. B, Duration of response. OPC indicates oropharyngeal cancer.

to nivolumab monotherapy. The absence of synergistic toxic effects is integral to building rational combination immunotherapy on the anti–PD-1 platform, and combined with the efficacy outcomes, supports further investigation of this approach in a randomized trial. Although combining therapeutic cancer vaccination with immune checkpoint blockade is the focus of much ongoing research, our results are among the first to be reported, to our knowledge.

During the trial's design in 2014, we used preliminary data from Keynote-012, which showed an ORR of 20% (4 of 20 patients) in patients with p16-positive OPC treated with pembrolizumab[27] as a historical reference. Given the accrual of patients with predominantly OPC in our trial, the most appropriate historical references available now are the subsets with p16-positive OPC treated with nivolumab in CheckMate

141 and pembrolizumab in Keynote-012 and Keynote-055 (Table 3).[2-5,28] Eligibility differed among these trials, particularly in regard to prior treatment. Nevertheless, the ORR of 36% among patients in our trial with OPC is numerically higher than that observed in the reference trials, and higher ORRs were confirmed in more comparable subsets of the patients with OPC in this trial, albeit with very small denominators. The median OS of 17.5 months (95% CI, 17.5 months to inestimable) and the 12-month OS rate of 70% are approximately double that observed for the CheckMate 141, Keynote-012, and Keynote-055 trials, from reported rates and inspection of the published survival curves.[2-5] Given the lower bound on the confidence interval of 17.5 months, the survival outcome appears to more clearly distinguish our trial, compared with the ORRs from these previous trials with anti–PD-1 mono-

Table 3. Efficacy of PD-L1 Inhibition in Patients With Recurrent Human Papillomavirus and OPC

Source	Treatment	Eligibility	RR, % (95% CI)	Overall Survival, Median (95% CI), mo
Mehra et al,[28] 2016 (Keynote-012 study)	Pembrolizumab	Treated with mixed platins (n = 28)	22 (12-34)	NR in p16-positive OPC
Bauml et al,[4] 2017 (Keynote-055 study)	Pembrolizumab	Platin-treated and cetuximab-resistant (n = 37)	16 (6-32)	8 (6-11)
Ferris et al,[5] 2016 (CheckMate 141 study)	Nivolumab	Platin-resistant (n = 63)	16 (NR)	9.1 (7.2-10)
Current trial	Nivolumab and ISA101	Up to 1 prior regimen for recurrence (n = 22)	36 (17-59)	17.5 (17.5 to inestimable)

Abbreviations: NR, not reported; OPC, oropharyngeal cancer; PD-L1, programmed death-ligand 1; RR, response rate.

therapy. However, owing to heterogeneity in eligibility and the small number of patients, judgments from this unplanned subset analysis are appropriately limited. Clear demonstration of the contribution of therapeutic HPV vaccine to anti–PD-1 therapy awaits testing in a larger randomized trial with more homogenous eligibility.

Similar to data from CheckMate 141 with nivolumab, we found that PD-L1 expression of 1% or more on tumor cells increased the chance of tumor response to 43% (3 of 7); however, response was also observed in 18% (2 of 11) of patients with PD-L1 expression less than 1%. Although these rates are higher than the 17% (PD-L1 expression ≥1%) and 12% (PD-L1 expression <1%) rates reported for the HPV-positive patients with OPC treated with nivolumab alone,[5] small patient numbers and heterogeneity limit firm conclusions. It can be speculated, however, that the outcome of vaccination on tumor regression in the setting of PD-1 inhibition is predominantly in the PD-L1–positive subset for whom ORR was more than doubled compared with nivolumab alone (43% vs 18%). Neither PD-L1 expression in stromal inflammatory cells nor the combined score with tumor and immune cell expression was associated with response, discordant with what has been reported with both nivolumab and pembrolizumab, for which the combined tumor plus immune cell scores provided an increased association with response vs tumor score alone.[3,29] The method we used differed from those analyses in many ways, including that immune cells were scored only in peritumoral stroma and not in the intratumoral region in our analysis. Scoring of the immune cells, whether intratumoral or stromal, for PD-L1 expression is highly discordant among observers, and has not been validated.[30] The bimodal distribution of PD-L1 expression we observed is also notable. Relevant previous trials with PD-1 inhibition alone have not displayed PD-L1 expression data graphically, although tabular data suggest that PD-L1 expression represents a continuous, more normally distributed variable.[4,5] The bimodal distribution and low number of evaluable biopsies in this trial mandate that this analysis must be viewed as exploratory. More comprehensive immunophenotyping and gene expression profiling are ongoing and may provide further insight as to biomarkers associated with benefit from combined HPV vaccination and PD-1 inhibition.

Limitations

This study has some limitations. This is a small single-arm trial including patients with heterogeneous treatment backgrounds. A larger randomized trial is necessary to confirm the benefit of vaccination added to PD-1 checkpoint inhibition.

Conclusions

Because only a subset of patients could be evaluated for HPV-16–specific immune responses with IFN-γ release, these results do not allow firm conclusions. Although the immune response is encouraging and consistent with earlier studies that demonstrated increased HPV-16–specific T cells after vaccination, it could be that these vaccine-induced T-cell populations are necessary, but not sufficient, for increased ORR in combination with nivolumab, perhaps owing to additional immunosuppressive pathways.[13,31]

The results of our trial are among the first clinical data to support the general concept of combining cancer vaccination with immune checkpoint blockade to enhance efficacy of vaccine-activated T cells in the immunosuppressive tumor environment. A randomized clinical trial testing the contribution of ISA101 to PD-1 inhibition, in patients with platin-resistant HPV-16–positive recurrent OPC is planned.

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