Study protocol: phase II study to evaluate the effect of cetuximab monotherapy after immunotherapy with PD-1 inhibitors in patients with head and neck squamous cell cancer

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Abstract

Background: Immunotherapy with programmed death receptor-1 (PD-1) inhibitors, as a single agent or in combination with chemotherapy, is the standard first-line treatment for recurrent or metastatic head and neck squamous cell cancer (R/M HNSCC). Unfortunately, there is no established second-line treatment for the many patients who fail immunotherapy. Cetuximab is the only targeted therapy approved in HNSCC but historically has a low response rate of 13%.

Objectives: We hypothesize that cetuximab monotherapy following an immune checkpoint inhibitor (ICI) will lead to increased efficacy due to a potential synergistic effect on the antitumor immune response, as a result of activation effects of both treatments on innate and adaptative immune responses. To the authors' knowledge, this is the only ongoing prospective clinical study that evaluates the combination of cetuximab and ICIs administered sequentially.

Methods and analysis: In this non-randomized, open-label, phase II trial, 30 patients with R/M HNSCC who have previously failed or could not tolerate a PD-1 inhibitor as a single agent or in combination with chemotherapy will subsequently be treated with cetuximab monotherapy. Outcomes of interest include overall response rate, duration of response, progression-free survival, overall survival, and treatment toxicity, as well as treatment outcome measured by a patient-reported outcome questionnaire. Saliva and blood will be collected for correlative studies to investigate the immune response status at the end of therapy with an ICI and the effect of cetuximab on the antitumor immune response. The results will be correlated with the response to cetuximab and the time window between the last administration of an ICI and the loading dose of cetuximab. The clinical study is actively recruiting.

Ethics: This study was approved by the Wake Forest Comprehensive Cancer Center Institutional Review Board: IRB00065239.

Clinical trial registration: This study is registered on ClinicalTrials.gov: NCT04375384.

Keywords: cetuximab, Erbitux, head and neck squamous cell cancer, immunotherapy, immunotherapy resistance, recurrent metastatic head and neck squamous cell cancer

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Introduction

In 2020, head and neck cancers were the seventh most common form of malignancy worldwide, with more than 660,000 new cases and 325,000 deaths yearly.[1,2] More than half of all patients with locoregionally advanced head and neck squamous cell cancers (HNSCC) will have local recurrence or distant metastases within 3 years of diagnosis.[3,4]

The advent of immunotherapy has dramatically improved survival in recurrent and metastatic HNSCC (R/M HNSCC). KEYNOTE 048 is the landmark study that established an immune checkpoint inhibitor (ICI), pembrolizumab, alone or combined with chemotherapy, as the standard of care for first-line treatment of patients with R/M HNSCC.[5,6] A recent update of the 5-year overall survival (OS) rate reported 14.4% *versus* 6.5% for pembrolizumab monotherapy *versus* EXTREME chemotherapy and 16.0% *versus* 5.2% for the pembrolizumab combined with chemotherapy *versus* EXTREME chemotherapy in the total populations with further improvement in the combined positive score positive subsets.[7] Unfortunately, <20% of patients with HNSCC respond to treatment with ICI monotherapy and <38% to ICI combined with chemotherapy.[6,8–12] Most patients with R/M HNSCC who fail ICI therapy will require salvage therapy, for which there are no current standard recommendations. Only half of the patients treated in recent, large randomized studies received second-line treatment.[7,13] In the Keynote 048 study for example, only 49.8% of the patients treated with pembrolizumab alone, 42.3% of the patients treated with pembrolizumab-chemotherapy, and up to 53.7% of patients treated with the EXTREME chemotherapy continued with a subsequent anticancer therapy. This is likely due to a variety of factors, including the lack of standardized second-line treatments, the lack of subsequent treatments with limited toxicities more appropriate for patients with declining performance status, and also the possible location of tumor progression with severe complications leading to decisions to stop active anticancer treatment and pursue comfort care. Significant research efforts are focused on other classes of immunotherapy and targeted drug therapies to address this critical need for effective R/M HNSCC salvage agents. Cetuximab's anticancer effect combines the immune and targeted therapeutic mechanisms and should therefore be more closely scrutinized as a potential post-ICI salvage agent.

Cetuximab is the only targeted therapy approved for HNSCC. It has been considered a pivotal agent for treating HNSCC in some scenarios, for example in combination with chemotherapy in the EXTREME regimen or as a radiosensitizer. However, older prospective studies of cetuximab monotherapy in R/M HNSCC reported a disappointingly low response rate of 13%, with a median time to progression of only 70 days.[14] ICIs as first-line therapy have changed the management of R/M HNSCC, and there is some evidence that cetuximab may enhance immune-mediated anticancer pathways primed by previous treatment with ICIs. The low toxicity profile of cetuximab monotherapy further supports the rationale for its investigation as a salvage therapy in R/M HNSCC patients who have failed therapy with ICIs.

This study protocol addresses this clinical need by assessing the effectiveness and tolerability of cetuximab monotherapy as a potential low-toxicity salvage therapy after first-line single-agent immunotherapy with pembrolizumab or the combination of pembrolizumab and chemotherapy for the management of R/M HNSCC.

Background and rationale

Mechanism of ICIs

Pembrolizumab and nivolumab are ICIs that comprise the backbone of R/M HNSCC treatment. These ICIs are humanized monoclonal IgG4 kappa antibodies that affect the programmed death receptor-1 (PD-1) on the surface of human lymphocytes. When bound to the programmed death ligand-1 (PD-L1), the PD-1 receptor plays an essential immunoregulatory role in preventing the immune system from attacking healthy cells by inhibiting T-cell function.[15] Many tumor types have capitalized on this regulatory function by expressing a high amount of the PD-L1, thus creating an immunosuppressed microenvironment and allowing malignant cells to evade the immune response. ICIs block the formation of the PD-1:PD-L1 complex, improving T-cell function and re-instating the role of adaptive immunity in the antitumor response.[16,17]

Mechanism of cetuximab

Cetuximab is a chimeric human/mouse IgG1 monoclonal antibody that targets the extracellular domain of the endothelial growth factor receptor

(EGFR). EGFR surface receptor overexpression is present in up to 90% of HNSCC and is often accompanied by overexpression of activating ligands.[18] EGFR's overexpression and dysregulation is an adverse prognostic factor, making targeting EGFR a rational approach to controlling HNSCC.[19,20] The binding of EGFR by cetuximab prevents further cell proliferation through competitive inhibition of EGF and TGF-α *via* cessation of intracellular signaling cascades[19,21–24] and leads to EGFR receptor internalization promoting downregulation of EGFR cell surface expression and EGFR-dependent transcriptional processes.[23,25]

In addition to its actions on EGFR, pre-clinical data suggest that cetuximab also induces complex immunostimulatory and immunoinhibitory effects. The Fc constant region of cetuximab binds to natural killer (NK) cells, causing the NK cells to target tumor cells for destruction *via* antibody-dependent, cell-mediated cytotoxicity (ADCC).[26–28] The released tumor-specific antigens are then cross-presented by dendritic cells, priming the CD8+ T cells for further adaptive tumor immune attack and increasing the immune cell infiltration in the tumor microenvironment (TME).[26,28] In addition, cytokines such as interferon γ released by the activated NK cells will recruit and activate dendritic cells and macrophages.[27,28] These phagocytic cells are further boosted by the cetuximab activation of the complement cascade, with C3 fragments known to have synergistic interactions with FcγR-mediated phagocytosis and ADCC.[29,30] Other cytokines released from NK cells may have direct antitumor effects by promoting FasL/Fas-mediated tumor cell apoptosis. Cetuximab also increases CD3+/CD8+ T cells and NK cells in the peripheral blood. Ultimately, by increasing CD137 expression on activated T cells, cetuximab enhances the activation and survival of tumor-specific CD8+ cells.[31] The increase in the TME immune cell infiltrate by cetuximab has been noted in the clinical setting where patients with metastatic colorectal cancer treated with cetuximab had higher markers of immune infiltration (CD3+, CD8+, CD56+ cells, etc.) than their counterparts who were not treated with cetuximab containing regimens.[32–34]

Despite the preponderance of immunostimulatory effects, several cetuximab-activated pathways may ultimately lead to an immunosuppressive outcome. For example, the direct EGFR signaling and the interferon γ released by NK cells can upregulate the PD-L1 expression in tumor cells *via* Janus kinase two signaling.[35] Increased infiltration of TME with regulatory T cells (Tregs) is another counterregulatory immunosuppressive mechanism activated by the cetuximab effects, which may ultimately lead to treatment resistance and cancer progression.[26]

Treatment with cetuximab and ICIs – mechanism of synergistic action. The scientific foundation of combining PD-1 inhibitors and cetuximab is built upon the complementary actions of these drugs on the innate and adaptive immune systems. As mentioned above, the efficacy of cetuximab monotherapy might be attenuated by the development of therapeutic resistance secondary to cetuximab-induced activation of several immunoinhibitory pathways. These pathways, such as the increased expression of PD-L1 in tumor tissue or increased PD-L1 expression on T regulatory cells in the TME, are targeted explicitly by PD-1/PD-L1 inhibitors. Furthermore, the pathways affected by cetuximab and ICIs are interconnected at several other sites. These sites include the EGFR pathway's influence on the major histocompatibility complex (MHC) expression (which plays a critical role in T-cell activation), the Phosphatidylinositol 3-kinase (PI3K)-AKT pathway activation (which lies downstream of EGFR and likewise plays a role in MHC expression), EGFR's regulation of interferon γ and activator of transcription 1 (STAT1), and modulation of cytokines including interleukin 6, interleukin 10, interleukin 8, and VEGF.[36] Therefore, there is increasing interest in evaluating cetuximab given concurrently with or sequentially after ICIs to improve tumor control in R/M HNSCC.

Few retrospective and prospective studies have sought to assess the safety and efficacy of cetuximab used concurrently with an ICI for R/M HNSCC. Sacco *et al.* reported results from an open-label, multi-arm, non-randomized, phase II trial studying the safety profile and response rate of R/M HNSCC to cetuximab and pembrolizumab used in combination. The overall response rate (ORR) was 45% (95% confidence interval 28–62%) at 6 months, and the median OS was 18.4 months (95% confidence interval 11.0 months–not reached) in their cohort of 33 patients. Treatment was given as a first line for R/M HNSCC in 88% of the patients. The response rate of the combination was significantly higher than the published results for each agent. A

total of 15% of participants suffered severe treatment-related adverse events, with mucositis being the most common. Furthermore, Sacco *et al.*[37] reported that responses were frequently seen early in the treatment course. Similarly, Glazar *et al.*[38] reported that early response dynamics could predict treatment failure in patients with R/M HNSCC treated with cetuximab and nivolumab. Chung *et al.* conducted a phase II multi-institutional clinical trial to determine OS in R/M HNSCC patients treated with a combination of cetuximab and nivolumab given every 2 weeks. Their study had two cohorts of patients: cohort A enrolled 45 patients with prior therapy for R/M HNSCC (including an ICI) and cohort B enrolled 43 patients with no prior treatment. The 6-month ORR was 23% in cohort A and 36% in cohort B. With a median follow-up of 15.9 months, the median OS was 11.4 months for patients with prior therapy and 20.2 months for those treated without prior treatment.[39,40] Results for patients in cohort B are similar to the Keynote 048 results (first-line treatment with pembrolizumab and chemotherapy) with an objective response rate of 36% and a median OS of 16 months.[6] With an excellent toxicity profile, these response and survival findings justify further investigation of therapies combining ICI and cetuximab *via* a larger randomized study. One such large population study was written by Chung *et al.* This meta-analysis studied 802 patients with R/M HNSCC who received ICI as monotherapy ($n=684$) or the combination therapy of cetuximab plus an ICI ($n=118$) demonstrated that the addition of cetuximab benefited only patients with human papillomavirus (HPV)-negative disease.[41] Other studies have suggested that cytokine profiles and T-cell repertoire in peripheral blood may distinguish the patients who will respond to combination treatment with ICI and cetuximab from those who will not.[42,43]

Evaluation of cetuximab after ICIs should be performed to address the critical need for a salvage therapy option after the standard ICI-based therapy first line in R/M HNSCC. With an extensive literature search, only one retrospective study by Pestana *et al.* was identified that reported outcomes for R/M HNSCC patients who progressed after ICIs and received cetuximab monotherapy. This review of 16 cases demonstrated an ORR of 37.5%, a progression-free response of 4.24 months, and a median OS of 8.41 months.[44] Cabezas-Camarero *et al.* presented data from a retrospective study of 23 patients with HNSCC treated with cetuximab combined with chemotherapy after ICIs and reported an ORR of 56.5% and a median OS of 12 months. However, 65% of patients developed grade 3 or 4 adverse events.[45] Yang *et al.* reported a retrospective review comparing the use of cetuximab followed by ICI to the use of ICI followed by cetuximab for the treatment of R/M HNSCC. This study demonstrated that both groups benefited from therapy and observed no significant survival difference between the two cohorts.[46]

Study rationale. The complex multilevel actions of cetuximab and ICIs on innate and adaptative immune response raised the hypothesis of synergistic antitumor effects. This synergism is further supported by several retrospective and prospective small clinical reports and is currently under investigation in large prospective studies. Given the long half-life of ICIs (approximately 27 days), we reason that the hypothesized synergism is not limited to concurrent therapy with both agents but will also occur with sequential administration of cetuximab following ICI.[47] The critical need for salvage therapy in the many R/M HNSCC patients who fail standard first-line ICI-based treatment highlights the need for prospective evaluation of cetuximab as monotherapy after treatment with a PD-1 inhibitor in these patients.

The excellent toxicity profile of cetuximab monotherapy further supports its use in the palliative setting. However, reports of a higher incidence and severity of the known skin toxicity of cetuximab when administered with ICIs underline the importance of close monitoring and reporting of the toxicity events. In the Chung *et al.* study of cetuximab and concurrent immunotherapy with nivolumab, the incidence of skin and nail toxicity was 85% for all grades and 9% and 14% for grade 3 in cohorts A and B, respectively,[38] while the incidence of skin toxicities reported by Vermorken *et al.*[48] on cetuximab monotherapy before the ICIs era was 49% for all grades and 1% for grades 3 and 4.

Phase II study

Herein, we describe the design and methods of a single-institution phase II clinical trial aimed at evaluating the efficacy of cetuximab monotherapy on R/M HNSCC after the failure or intolerance of ICI-based therapy. At this time, this is the only prospective clinical trial that evaluates the combination of cetuximab and ICIs administered sequentially. The clinical hypothesis is that the synergism between the two treatments will lead to

improved response and survival compared to previously reported data on cetuximab monotherapy. This study will be conducted at the Wake Forest Baptist Comprehensive Cancer Center and is registered on ClinicalTrials.gov (NCT04375384).

Study design

This is a single-arm, non-randomized, open-label study to evaluate the efficacy and tolerability of single-agent cetuximab for patients with R/M HNSCC who have progressed on or have been unable to tolerate treatment with ICI. In all, 30 eligible patients will be enrolled and treated with the standard dose of cetuximab weekly. Although a specific time window from previous treatment with ICI is not mandated, starting treatment with cetuximab as soon as possible after the last administration of pembrolizumab or nivolumab is encouraged. The correlation of this time window with response to treatment with cetuximab will be analyzed. The initial staging will be done using CT or MRI, performed no more than 28 days before treatment. Follow-up imaging to evaluate treatment response will be done with the same imaging modalities every 6–8 weeks throughout the treatment.

The demographic characteristics of patients, including the staging, smoking status, treatment history, PD-L1 expression level, tumor mutational burden (TMB) in tumor and/or blood, and HPV status, will be collected in all patients. Next-generation sequencing is encouraged and will be done from the blood and tumor tissue if available. The patients' status will also be evaluated and monitored with the patient-reported outcomes (PRO) questionnaire.

Saliva and blood will be collected for correlative studies to investigate the immune response status at the end of therapy with an ICI and the effect of cetuximab on the antitumor immune response. The results will be correlated with the response to cetuximab and the time window between the last administration of an ICI and the loading dose of cetuximab (Figure 1).

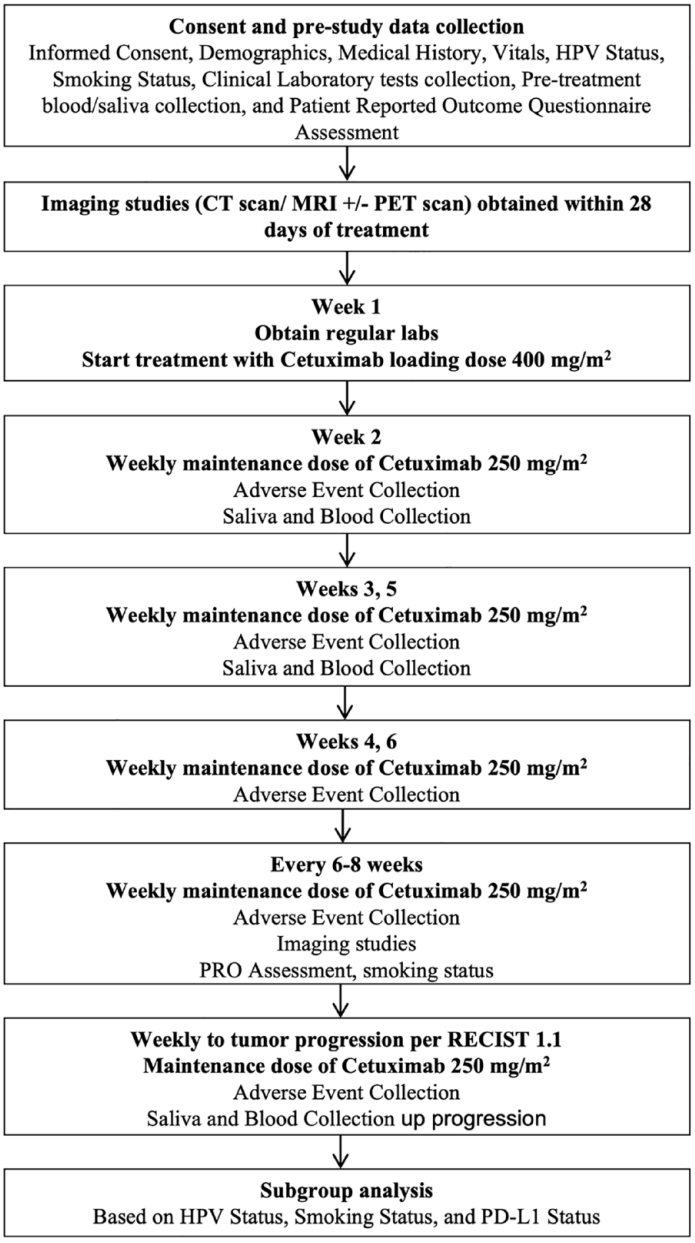

Figure 1. Study schema for the study protocol of cetuximab monotherapy after immune checkpoint inhibitor for recurrent/metastatic head and neck squamous cell cancer.

Eligibility criteria

Eligibility criteria are summarized in Table 1. Patients 18 years of age or older with biopsy-proven R/M HNSCC who have received and progressed on or failed to tolerate treatment with a PD-1 inhibitor (pembrolizumab or nivolumab) alone or in combination with chemotherapy are eligible for the study. The Eastern Cooperative Oncology Group (ECOG) performance status must be two or less. Patients will be excluded if they have ever received previous treatment with cetuximab, have a history of interstitial lung disease, require treatment with corticosteroids or other immunosuppressive agents, or have any relative or absolute contraindications to therapy with cetuximab.

Table 1. Inclusion and exclusion criteria for the study protocol of cetuximab monotherapy after immune checkpoint inhibitor for recurrent/metastatic head and neck squamous cell cancer.

Inclusion criteria
• Patients must have histologically or cytologically confirmed HNSCC. • Patients must have measurable disease (at least one measurable lesion) by CT or MR imaging, as defined by RECIST 1.1. • Patients must have received previous treatment with immunotherapy with a PD-1 inhibitor alone or in combination with chemotherapy. There will be no time window requirement between cetuximab and the previous immunotherapy. • Patients must have a performance status of 0–2 as measured by the ECOG Status Scale. • Patients must be ⩾18 years old. • Patients must be willing to consent to collect blood and saliva samples as scheduled throughout the treatment. • Patients must be willing and able to comply with the protocol for the duration of the study. • Patients must be willing to donate two tablespoons of blood and one teaspoon of saliva for research before treatment, three more times during the first 5 weeks of treatment, and then at cancer progression. • Patients must be able to understand and willing to sign an IRB-approved informed consent document.
Exclusion criteria

• Patients who have received prior treatment with cetuximab or therapy that specifically and directly targets the EGFR pathway will be excluded from the study.
• Patients who have had a prior allergic reaction to cetuximab will be excluded from the study.
• Patients with a history of allergic reactions attributed to compounds of chemical or biological composition similar to cetuximab will be excluded from the study.
• Patients receiving any other investigational agents will be excluded from the study.
• Patients using medications that need to be continued and might interact with cetuximab will be excluded from the study.
• Patients with any uncontrolled condition that, in the investigator's opinion, would interfere with the safe and timely completion of study treatment and procedures will be excluded from the study.
• Patients with a history of interstitial lung disease (e.g. pneumonitis or pulmonary fibrosis) or evidence of interstitial lung disease on screening chest imaging will be excluded from the study.
• Any persons who are pregnant, breastfeeding, or expecting to conceive or father children within the projected duration of the trial will be excluded from the study.
• In addition, patients with any of the following conditions will be excluded:
  ○ Patients with a severe or non-healing wound, ulcer, or bone fracture at the discretion of the treating physician.
  ○ Patients with a history of abdominal fistula, gastrointestinal perforation, or intraabdominal abscess within 28 days of study enrollment.
  ○ Patients with a history of cerebrovascular accident or transient ischemic attack within 12 months before study enrollment.
  ○ Patients with a history of myocardial infarction, ventricular arrhythmia, stable/unstable angina, symptomatic congestive heart failure, coronary/peripheral artery bypass graft or stenting, or another significant cardiac disease within 6 months before study enrollment.
  ○ Patients with a history of arterial or venous thrombosis/thromboembolic event, including pulmonary embolism, within 6 months of study enrollment.
  ○ Patients with any condition requiring the use of immunosuppression, excluding rheumatologic conditions treated with stable doses of corticosteroids.

ECOG, Eastern Cooperative Oncology Group; HNSCC, head and neck squamous cell cancer; RECIST, Response Evaluation Criteria in Solid Tumors.

Study objectives

The primary objective of this study is to evaluate the ORR of treatment with cetuximab monotherapy following treatment with PD-1 inhibitor immunotherapy. The secondary objectives include the duration of response (DoR), progression-free survival (PFS), OS, and the evaluation of treatment toxicity with cetuximab monotherapy after ICIs. The exploratory objectives include the assessment of the overall tolerability of treatment based on PRO data, as well as the assessment of cetuximab-induced antitumor immune response effects measured in blood and saliva.

Study endpoints and procedures

The primary endpoint, the ORR, will be measured according to Response Evaluation Criteria in Solid Tumors 1.1. The percentage of patients who develop a complete response (CR) or partial response (PR) to treatment, measured by imaging studies, will be determined. Follow-up imaging

with CT or MRI to assess response will be obtained before starting treatment and every 6–8 weeks through the treatment until progression is identified. The DoR will be measured in days and defined as the interval from tumor response documentation (when either CR or PR is first determined) to tumor progression or death, whichever occurs first; patients lost to follow-up will be censored. PFS will be defined as the interval from the start of treatment to tumor progression or death, whichever occurs first. OS is defined as the time from the beginning of treatment to death from any cause.

The study will also evaluate the tolerability of the cetuximab monotherapy administered after a PD-1 inhibitor. Treatment toxicity will be measured by the rate of adverse effects assessed at each clinic visit. Adverse events will be graded according to the revised National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) version 5.0. Treatment tolerability will be evaluated with patient self-reported quality of life assessments. Patient-reported outcomes will be obtained every 6–8 weeks using a targeted checklist of head and neck cancer-specific PRO-CTCAE version 1.0 items.

Saliva and blood will be collected before starting treatment, before treatments 2, 3, and 5, and then upon progression. These samples will be processed and frozen, and biomarkers will be evaluated retrospectively to address mechanistic questions regarding the immune response status at the time of failure of PD-1 inhibitors and then the effect of treatment with cetuximab on the immune response. Nonspecific markers of immune activation, such as immune-regulatory miRNAs, will be tested in blood and saliva. Profiling of circulating immune cells including but not limited to, CD3, CD8, CD16, CD69, CD107a, CD 137, NKG2D, NKG2A, NKp46, as well as interferon, perforin, granzyme B, and other immune cytokines and interleukins, will be tested in blood. Multi-omics profiling of saliva and blood will be performed to address specific mechanistic questions.

Biomarkers and demographic data will be evaluated and collected for correlation with response. HPV status will be determined by polymerase chain reaction when tumor tissue is available or by p16 immunohistochemistry as an established surrogate marker. PD-L1 level will be tested using the 22C3 PharmDx assay, and results will be reported as a Combined Positive Score (CPS). Smoking status will be collected and defined as follows: smoker – actively smoking or has more than a 10-pack-year history of smoking; non-smoker – not actively smoking and has less than a 10-pack-year history of smoking.

Treatment with cetuximab

Cetuximab will be administered at the standard loading dose of 400 mg/m^2, followed by weekly maintenance doses of 250 mg/m^2. The maintenance dose of cetuximab will be given every 7 days ($\pm$2 days). In any case of treatment delay, there will be no re-loading infusion, and all subsequent treatments will be at the assigned dose level. Cetuximab will not be dose-reduced or held for hematologic adverse events such as neutropenia, neutropenic fever, or thrombocytopenia. Cetuximab will be held for any non-hematologic toxicity equal to or higher than grade 3 until less than or equal to grade 2. There will be no dose adjustment for infusion reactions. Cetuximab dose modifications for dermatologic toxicities are presented in Table 2. The infusion rate will be permanently reduced by 50% for grade 1 or 2 reactions, and treatment will be permanently discontinued for grade 3 or 4 reactions. If cetuximab is omitted for more than four consecutive infusions for adverse events or an intercurrent illness (e.g. infection) requiring interruption of therapy, the patient will be discontinued from further cetuximab therapy.

Discontinuation from the study treatment and follow-up

Treatment on the study protocol will continue until there is tumor progression or severe adverse events. The treatment segment will also be discontinued if the patients develop conditions that are incompatible with the continuation of treatment with cetuximab, could affect the integrity of the study, or meet the exclusion criteria. A last set of biologic samples for correlative studies will be collected at the time of tumor progression. The follow-up will continue for at least 1 week after the last administration of cetuximab and weekly until all treatment-related adverse events become less than or equal to grade 1. During this time, adverse events will be recorded weekly. Patients will continue to be followed every 4–6 weeks for survival. If patients cannot come or decline the follow-up visits, the information can be obtained by phone or outside medical records with the patient's permission. The patients who withdraw

Table 2. Cetuximab dose modifications for dermatologic changes (⩾grade 3).

Cetuximab	Outcome	Cetuximab dose modification
First occurrence		
Delay infusion 1–2 weeks	Improvement to ⩽grade 2	Continue at 250 mg/m^2
	No improvement; remains ⩾grade 3	Discontinue cetuximab
Second occurrence		
Delay infusion 1–2 weeks	Improvement to ⩽grade 2	Reduce dose to 200 mg/m^2[a]
	No improvement; remains grade ⩾3	Discontinue cetuximab
Third occurrence		
Delay infusion 1–2 weeks	Improvement to ⩽grade 2	Reduce dose to 150 mg/m^2[a]
	No improvement; remains grade ⩾3	Discontinue cetuximab
Fourth occurrence		
Discontinue cetuximab		

[a]Cetuximab dose can be restored to the previous dose level if the skin rash resolves to grade 1 or less.

voluntarily from the study or are lost to follow-up will be discontinued from the entire study. The patients who do not have at least the first set of follow-up imaging studies and the corresponding scheduled collection of the correlative blood and saliva samples will be replaced.

Statistics

Planned sample size. The planned sample size for this single-arm phase II study is 30 patients. The ORR will be compared to treatment with cetuximab monotherapy following treatment with immunotherapy with PD-1 inhibitors and will be assessed using Simon's (1989) two-stage design.[49] The null hypothesis that the confirmed ORR is less than 13% will be tested against a one-sided alternative of a response rate of 30%. An interim analysis will be performed after the first 16 patients are accrued. The study will be stopped if there are two or fewer responses from these 16 patients. Otherwise, the 14 additional patients will be accrued for a total of 30 patients. The null hypothesis will be rejected if 7 or more responses are observed in 30 patients. This design yields a type 1 error rate of 0.077 and a power of 80% when the true response rate is 30%.

Statistical considerations. The primary objective of this study is to compare the ORR to treatment with cetuximab monotherapy. With a total sample size of 30 patients, this design yields a type I error rate of 0.077 and a power of 80% when the actual response rate is 30%. In addition to performing this test comparing the ORR, we will estimate the ORR and the corresponding 95% Exact Clopper–Pearson confidence interval for the ORR. To examine the relationship between treatment response and HPV, PD-L1, and smoking status, Fisher's exact tests will be performed with response status (yes/no) against the mentioned variables (positive/negative) or smoking status (never/ever) as the variables in each 2×2 table.

For time-to-event measures such as DoR, PFS, and OS, we will generate Kaplan–Meier survival curves and estimate the median days-to-event and the percent of patients who have experienced PFS and survived at 6 months and 1 year post-treatment. Finally, for tolerability, we will create frequency tables that count the number and severity of toxicities, as well as the items collected *via* the PRO-CTCAE and calculate corresponding 95% Clopper–Pearson exact confidence intervals for these toxicities.

A subset analysis will estimate the correlation between the time window from previous ICI treatment and the beginning of treatment with cetuximab and ORR.

Conclusion

There are no standard recommendations for salvage therapy for R/M HNSCC after the failure of ICI-based regimens. This study will assess response, survival, toxicity, and the mechanistic immune effects associated with single-agent cetuximab after progression or inability to tolerate ICI. Correlation with demographic information and biological markers (such as smoking status, PD-L1 level, TMB, and HPV), as well as exploratory results (such as immune miRNA, circulating immune cells, and cytokines), are expected to better inform clinicians as to which patients are most likely to benefit from this therapy. To the authors' knowledge, this is the only ongoing prospective clinical study that evaluates the combination of cetuximab and ICIs administered sequentially. Data obtained in this study may later inform confirmatory clinical trials investigating salvage therapies for R/M HNSCC.

Declarations

Ethics approval and consent to participate

This study was approved by the Wake Forest Comprehensive Cancer Center Institutional Review Board (IRB00065239). Written and verbal consent has been obtained from all current participants and will be obtained from all future participants.

Consent for publication

Not applicable. No identifying information has been included in this publication.

Author contributions

Kimberly M. Burcher: Conceptualization; Visualization; Writing – original draft; Writing – review & editing.

Chance H. Bloomer: Methodology; Visualization; Writing – original draft; Writing – review & editing.

Elena Gavrila: Visualization; Writing – original draft; Writing – review & editing.

John M. Kalada: Visualization; Writing – review & editing.

Mark J. Chang: Writing – original draft; Writing – review & editing.

Rediet R. Gebeyehu: Methodology; Writing – review & editing.

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Lara M. Khoury: Visualization; Writing – review & editing.

Thomas W. Lycan: Conceptualization; Methodology; Supervision.

Rebecca Kinney: Visualization; Writing – review & editing.

Ralph D'Agostino Jr.: Conceptualization; Methodology.

Paul M. Bunch: Conceptualization; Methodology; Visualization; Writing – original draft.

Kirtikar Shukla: Methodology; Writing – review & editing.

Pierre Triozzi: Conceptualization; Methodology; Supervision.

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