Published in final edited form as:

Curr Treat Options Oncol. ; 19(12): 68. doi:10.1007/s11864-018-0587-0.

Window Studies in Squamous Cell Carcinoma of the Head and Neck: Values and Limits

Dan P. Zandberg, MD* and Robert L. Ferris, MD, PhD

UPMC Hillman Cancer Center, 5150 Centre Avenue, 5th floor, Room 552, Pittsburgh, PA, 15232, USA

Opinion statement

In head and neck cancer, we continue to work towards a more personalized approach to treatment of patients, where analysis of a patient's tumor guides targeting of molecular or immunologic pathways. Critically important to this pursuit is a better understanding of the direct biologic effect of a drug or combination on the tumor microenvironment in humans, as well as biomarker discovery. These goals are consistent with the primary purpose of a "window of opportunity" trial and while conduct of these trials requires a careful balance of benefits and potential risks, to date these trials have been both feasible and safe in HNSCC in the curative intent setting. In the era of immunotherapy, with countless possible combinations and ongoing clinical trials, window trials are even more important for informing clinical trial design and appropriate combination therapy, and ultimately a more personalized approach to our patients that leads to improvement in outcomes.

Keywords

Window trial; HNSCC; Neoadjuvant; Anti-PD-1; Immunotherapy; Targeted therapy; Cetuximab

Introduction

Squamous cell carcinoma of the head and neck (HNSCC) is the sixth most common cancer worldwide and includes cancers that involve the larynx, hypopharynx, oropharynx, and oral cavity [1]. While smoking and alcohol intake are still traditional risk factors for HNSCC, in recent decades, it has become apparent that the human papillomavirus (HPV) also has an etiological role in squamous cell carcinoma of the oropharynx, and these tumors are associated with an improved prognosis compared to HPV-negative HNSCC [2–5]. This has led to de-escalation strategies in locally advanced HPV-positive oropharyngeal cancer to improve functional outcomes and escalation of therapy intensity in HPV-negative HNSCC,

* zandbergdp@upmc.edu.

This article is part of the Topical Collection on *Head and Neck Cancer*

Conflict of Interest

Dan P. Zandberg has received research support for role as PI for clinical trials conducted by Merck, Bristol-Myers Squibb, and AstraZeneca.

Human and Animal Rights and Informed Consent

This article does not contain any studies with human or animal subjects performed by any of the authors.

where there is a clear need for improvement in oncologic outcomes. Additionally, prognosis remains poor in the recurrent/metastatic setting for all HNSCC patients, with a median survival with systemic therapy alone of 10–12 months [6].

Across tumor types, including in HNSCC, the field has focused on more personalized approaches to treatment of patients. This includes the use of immunotherapy and targeting of molecular pathways, and the only approved drugs for HNSCC are anti-PD-1 mAbs nivolumab and pembrolizumab as well as IgG1 mAb cetuximab, respectively. These approvals, especially more recently with anti-PD-1 mAbs, have led to a great expansion of research and clinical trials evaluating new drug combinations and predictive biomarkers for efficacy. Important to this pursuit is a better understanding of the effect of immunotherapeutic or targeted agents on the tumor microenvironment, and window of opportunity trials can help to achieve these goals.

Definition of a “window of opportunity trial”

A “window trial” in the broadest sense is a trial in which treatment-naïve patients receive a systemic agent or a combination of agents prior to initiation of standard of care therapy, with the primary goal of understanding the mechanisms of action or biomarkers of response of the agent or combination [7–9]. This is accomplished most often with a paired pre-treatment biopsy as well as post-treatment tissue, the latter usually obtained as part of surgical resection. In addition to pathologic evaluation, window trials can also include pre- and post-treatment imaging for evaluation of response.

In this review, we will summarize ongoing and reported window of opportunity trials in HNSCC and focus on the values and limitations of these trials. While the purest definition of a window trial would exclude a neoadjuvant therapeutic trial where the goal of the trial is to improve efficacy, important mechanistic information can be additionally gained when these trials include methodology typically used in window trials. As such, we have included some of these trials in this review.

Targeted therapy window trials: review of the literature

A number of window of opportunity trials have focused on targeted agents against the epidermal growth factor receptor (EGFR) including monoclonal antibodies such as cetuximab and tyrosine kinase inhibitors erlotinib and afatinib. The epidermal growth factor receptor is a transmembrane receptor that is part of the ERBB family, and stimulation of EGFR leads to activation of downstream pathways including phosphatidylinositol-3-kinase/v-akt murine thymoma viral oncogene homolog, phospholipase-C-γ/protein kinase C, and ras/raf/mitogen-activated protein kinase, resulting in increased proliferation and survival of cells [10–12]. Cetuximab is an IgG1 human-murine monoclonal antibody which blocks EGFR signaling by binding irreversibly to the extra-cellular domain of the EGFR receptor [13]. Cetuximab remains the only FDA-approved targeted agent for HNSCC, in combination with platinum and 5-fluorouracil in the first-line recurrent/metastatic setting and in combination with radiation as part of definitive therapy in locally advanced disease [14, 15]. While overall survival has been increased with the addition of cetuximab in these settings,

the magnitude of benefit is small, and this has led to evaluation for predictive biomarkers to select those that may benefit the most. Schmitz et al. conducted a neoadjuvant window trial with cetuximab given prior to surgical resection in patients with T1–T4 SCC of the oral cavity, oropharynx, larynx, or hypopharynx. Cetuximab was given weekly starting day – 15 prior to planned surgical resection and shorter intervals between the last dosage of cetuximab and surgery were evaluated. Therapy was determined to be safe given at an interval of 24 h prior to surgical resection. After treatment with cetuximab, pathologic response was evaluated and 47% (15/32) of patients had < 50% tumor cellularity with 5 having < 25% tumor cellularity. PET was also done preand post-cetuximab and decrease in SUV after treatment was associated with decreased residual tumor cellularity on post-operative pathology. While comparison of pre- and post-tissue revealed significantly decreased expression of pEGFR and pERK, neither of these biomarkers at baseline correlated with change in PET avidity after treatment with cetuximab [16].

Erlotinib either alone or in combination has been evaluated in the window of opportunity setting. Erlotinib alone was compared to erlotinib plus non-selective COX inhibitor sulindac and placebo with the primary objective of evaluating change in Ki-67 proliferation index comparing pre- and post-paired tumor samples. Ki-67 was significantly reduced with erlotinib or erlotinib plus sulindac compared to placebo, with a trend towards combination therapy having the greatest effect. No clinical correlation was evaluated [17]. Baumen et al. conducted a randomized four-arm window trial with erlotinib, dasatinib, erlotinib plus dasatinib, or placebo in stage I–IV HNSCC prior to resection. All treatment arms were safe without unexpected toxicity. Erlotinib alone or with dasatinib led to a significantly higher response compared with dasatinib alone or placebo ($P = 0.0006$). Treatment with erlotinib was associated with reduction in tumor size in 29% of patients with no additive or synergistic effects when combined with dasatinib. None of the intervention arms altered any of the 16 selected potential biomarkers in pre- and post-tissue compared to placebo. Furthermore, while baseline pMAPK expression had some association with response to erlotinib, none of the other pre-selected biomarkers (pSrc, pMet, serum IL-6, change in KI-67) correlated with response [18]. Afatinib was evaluated in a multicenter phase II randomized window trial comparing afatinib given for 14 days prior to surgery to placebo. The primary objective was PET response and 70% had a partial metabolic response. Twenty-two percent had a partial response by RECIST. While there was no G4 or G5 toxicity experienced and a pre-specified safety stop rule was not crossed, one patient developed G3 diarrhea and renal failure as well as G3 acute pancreatitis after 11 days on afatinib, and a delay in surgery of 24 days occurred for this patient. Two other patients had delays in surgery of 2 and 4 days. No treatment-related surgical complications were observed. No pre-treatment biomarkers examined (PTEN, HER3, EGFR) were predictive of response. Wild-type p53 status and high cluster3-hypoxia score were both significantly associated with partial metabolic response but not response by RECIST [19]. Additional targeted therapy window trials are shown in Table 1.

Immunotherapy window trials: review of the literature

Several drugs targeting the immune system have been recently evaluated in the window of opportunity setting (Table 1). As part of a multicohort study, 29 patients received nivolumab

240 mg days 1 and 15 and then underwent surgical resection of their HNSCC. Importantly, no delays in surgery were seen due to treatment-related adverse events, and 48% of patients had reduction in the size of their tumor on repeat imaging done prior to surgery, with 3 patients having a greater than 40% reduction. Analyses of changes in the immune microenvironment are ongoing at this time [20]. In another study, patients with locally advanced (stage III/IV) HPV-negative resectable HNSCC underwent treatment with pembrolizumab X 1 dose followed by surgical resection. Similar to the nivolumab study, no serious drug-related AEs were observed and there were no unexpected delays in surgery or postop complications. Treatment effect on post-operative pathology was seen in 42% (10/24) of patients with downstaging observed. For example, one patient with T2N2b SCC of the buccal mucosa responded and on resection was pathologic stage T1N0. A significant correlation was seen between pre-treatment tumor PD-L1 expression, CD8 and CD4 T cell infiltration, and pathologic response. For example, 7/8 evaluable pathologic responders were PD-L1 positive (> 1% TC expression) on pre-treatment biopsy [21]. Bell and colleagues conducted a neoadjuvant window trial with MEDI6469, an agonist antibody to OX40 in resectable stage II–IVa SCCHN patients. Patients received three doses of MEDI6469 followed by surgical resection either 2 days, 1 week, or 2 weeks after treatment. Seventeen patients were treated and similar to the studies with anti-PD-1 mAbs, there were no G3 or 4 AEs, delays in surgery, or unexpected surgical complications. They evaluated activation markers Ki-67 and CD38 by flow cytometry in the peripheral blood and found that there was increased activation of CD4 T cells and effector and memory CD8 T cells after receipt of MEDI6469, which peaked approximately 14 days after treatment. Twenty-four percent (4/17) of patients treated experienced proliferation of tumor reactive CD8 T cells double positive for CD39 and CD103 and these four patients remain recurrence free with a median of 24 months of follow-up [22••]. Colevas et al. reported preliminary results of an ongoing study evaluating the ability of PET tracer [23] F-AraG to predict immunologic response after anti-PD-1 mAb given prior to surgical resection. Activated T cells can overexpress dGK and this PET tracer is taken up in cells with high levels of dGK. In 3 patients reported, 1 patient exhibited increased tracer uptake which correlated with an increase in the proportion of activated CD8 T cells in the tumor micro-environment. Pathologic response was not reported [23].

IRX-2 is composed of cytokines that have been obtained from stimulated peripheral blood mononuclear cells. It was evaluated in a phase IIa neoadjuvant trial in SCCHN which included 27 patients [24]. Patients received 10 days of IRX-2 injections with low-dose cyclophosphamide prior to resection. Higher lymphocyte density on resected tumors was associated with increased response and overall survival. Seven patients had matched pre- and post-resection tumor specimens for analysis. By multiplex IHC analysis, 5/7 had increase in CD68 cells, 4/7 had increase in PD-L1, and 4/7 had increase in density of T cells post-IRX-2 treatment, and there was no increase in regulatory T cell subsets in any patients. Although there was variability among individual patients, gene expression analysis by nanostring showed mean increase in genes associated with CD8, CD4, NK cell, and B cell activity as well as increases in gene expression associated with interferon receptors and chemokine pathways, including CXCL12, CCL2, CCL14, CCL19, and CCL21, after treatment with IRX-2 [25].

In addition to targeting the EGFR pathway, cetuximab can induce the innate immune system through NK cell-mediated antibody-dependent cellular cytotoxicity (ADCC) and additionally stimulate adaptive immunity, specifically the development of EGFR-specific CD8 T cells via NK cell/dendritic cell crosstalk [26, 27]. As such, there is potential for synergy with other drugs targeting the immune system. After safety was established in a phase I trial in the R/M setting, a neoadjuvant window trial combining cetuximab with small molecule TLR8 agonist motolimod was conducted to evaluate quantitative changes in immune cell populations and immune biomarkers [28••]. Therapy was given for 3–4 weeks with surgery conducted 2 days after the last dosage. Evaluation of pre- and post-treatment paired tumor samples showed that combination therapy led to increased infiltration of myeloid antigen-presenting cells, and a decrease in expression of markers of suppression on Tregs, including CTLA4, CD73, and membrane-bound TGF-B. The latter was an important finding given that cetuximab monotherapy led to increased Treg infiltration of the tumor microenvironment with increased expression of CTLA-4 and TGF-B [29]. In addition, combination treatment led to increased CD8 T cell tumor infiltration and these cells had a more activated phenotype, with decreased expression of inhibitory co-signaling molecules TIGIT, PD-1, and CTLA4 [28••].

Advantages and disadvantages of window of opportunity trials

The window trial is a careful balance of benefits and risks in maximizing information gained while ensuring patient safety. This balance is especially critical as window trials are most often conducted in a curative intent patient population. Potential risks include unexpected toxicity delaying or preventing curative surgical resection, or causing post-operative complications precluding or affecting the tolerance of standard of care adjuvant therapy. Additionally, there is risk of progression during trial treatment which could hamper the ability to provide curative intervention. To reduce these risks, there are a number of important considerations in designing and conducting a window trial. First, patient safety is paramount. As such, a drug or combination therapy should have an established tolerated dosage from a phase I trial prior to inclusion in a window trial. Additionally, adequate toxicity stopping rules should be included and/or frequent discussions about toxicity during trial should be conducted by the research team. As discussed above, importantly, most window trials in HNSCC have not had unexpected toxicity. Timing and the length of the intervention must be considered, including the additional lag time for screening and enrollment to the trial. Most window trials in HNSCC have had a therapeutic intervention of 2–4 weeks (range 1–6 weeks) and there were no delays in the planned surgical resection in the majority of reported trials (Table 1). Still, window trials should be conducted at centers experienced in multidisciplinary care and clinical trial monitoring, to reduce these risks as much as possible. While these potential risks without likelihood of direct benefit can deter patients from enrolling, importantly, trials discussed in this review highlight the feasibility of single arm or randomized trials with modest sample sizes (median 31 patients). The primary end points in most window trials are biomarker based or safety. These end points are typically most feasible and in line with the goals of a window trial. If the primary end point includes comparison to control or another intervention arm, it is important that adequate sample size be considered, in order to strengthen the conclusions that can be made from the

data obtained. Also, given the risks of window trials, it is important that designs maximize collection of additional information. Therefore, if appropriate, pre- and post-imaging for response correlation and peripheral blood samples should also be collected.

Given the limitations of in vitro and in vivo experiments in mouse models, a major advantage of a window trial is the ability to examine the effect of the therapeutic intervention, with pre- and post-tissue samples, directly in patients. Paired tumor sample collection is often not feasible or ethical in trials in advanced patients. Since surgery follows intervention in most window trials, adequate tissue for more detailed analysis is typically available. This approach can allow for confirmation or rejection of pre-clinical hypothesis especially important to combination therapies and to guidance of future trial design. For example, pre-clinical HNSCC models showed that activation of Src led to resistance to EGFR blockade [30, 31], providing rationale for combination dasatinib and erlotinib; however, despite dual blockade of Src and EGFR with this combination, no synergy was observed in the window trial [18]. Also, whereas cetuximab monotherapy led to increase in suppressive co-signaling molecule expression on Tregs, combination with motolimod was able to reverse this leading to reduction of these suppressive co-signaling molecules [28•, 29]. Towards the goal of being able to decipher the effect of a therapeutic intervention, having a control or comparator arm is important. If no drug is appropriate for comparison, then placebo can be used or rather as some investigators have done, using a randomized design whereby a portion of the patients go right to surgery, so this pathologic specimen can be used as a control, without risking progression on placebo. We favor this latter design rather than having a placebo arm.

The traditional drug development pathway in oncology has been that a new drug is first tested in a phase I setting in the most advanced refractory disease and then if safe transitioned to a phase II and then phase III trial in the advanced setting. If the drug proves itself in the advanced setting, then it is incorporated in trials in the upfront curative setting. While a phase I trial establishes safety, these patients are resistant to multiple lines of therapy, making the likelihood of an efficacy signal, even when cohorts are expanded, lower with subsequent abandonment of drug development if no preliminary efficacy is seen [9, 32, 33]. An open question is how an efficacy signal in a treatment-naïve window trial can be most helpful to inform the development of a new agent or combination in the advanced setting. The limited duration of treatment during a window trial precludes evaluation of the duration of response, which is where many systemic agents fall short in the advanced setting. Additionally, the response to systemic therapy is often higher in the treatment-naïve setting compared to recurrent treatment refractory setting. For example, afatinib had a response rate of 22% compared to 10% by RECIST 1.1 in the treatment-naïve and second-line R/M setting respectively [19•, 34]. Therefore, a low response rate in an unselected treatment-naïve patient population could serve as a better signal that this agent should not be moved forward in any setting. Additionally, a biomarker-focused window trial done in a timely fashion after phase I dosing is established could serve to inform subsequent selection criteria in phase II/III trials in the advanced setting.

The window trial is especially important in the era of immunotherapy. Currently, both pembrolizumab and nivolumab are approved in R/M HNSCC after failure of platinum-based

chemotherapy. Nivolumab, for example, significantly improved overall survival compared to investigator choice chemotherapy in a randomized phase III trial [35]. While this is the first drug in a randomized trial to ever prolong overall survival in R/M disease after platinum failure, response rate was only 13% with an additional 20% achieving SD. Therefore, the majority of patients will progress and not benefit from single-agent anti-PD-1 mAb therapy. With a seemingly infinite number of possible IO combinations, validation of proposed mechanisms of action and synergy and biomarker selection of patients will be critical to determining which combinations to move forward into a phase III trial, and ultimately to be able to select a combination most likely to benefit each individual patient. There are a number of immunotherapy-based window of opportunity trials in HNSCC currently enrolling (NCT02919683, NCT02002182, NCT03618654).

Conclusion

The design and conduct of a window of opportunity trial require adequate maximization of the potential benefits and reduction in the potential risks. Importantly, to date, these trials have been feasible and conducted safely in HNSCC in the curative intent setting. As the oncology field continues to work towards a more personalized approach to treatment, both with immunotherapy and targeted therapy, window of opportunity trials will continue to become even more important for guiding appropriate combinations and biomarker-driven clinical trial design.

Acknowledgments

Robert L. Ferris has received research funding for clinical trials conducted by AstraZeneca/MedImmune, Bristol-Myers Squibb, Merck, Tesaro, and VentiRx Pharmaceuticals, and has served on advisory boards for Amgen, AstraZeneca/MedImmune, Bristol-Myers Squibb, EMD Serono, PPD, Lilly, Merck, Pfizer, and Tesaro.

References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:

- Of importance
- Of major importance

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. *CA Cancer J Clin.* 2011;61(2):69–90. [PubMed: 21296855]
2. Ang KK, Harris J, Wheeler R, Weber R, Rosenthal DI, Nguyen-Tan PF, et al. Human papillomavirus and survival of patients with oropharyngeal cancer. *N Engl J Med.* 2010;363(1):24–35. [PubMed: 20530316]
3. Zandberg DP, Bhargava R, Badin S, Cullen KJ. The role of human papillomavirus in nongenital cancers. *CA Cancer J Clin.* 2013;63(1):57–81. [PubMed: 23258613]
4. Rischin D, Young RJ, Fisher R, Fox SB, Le QT, Peters LJ, et al. Prognostic significance of p16INK4A and human papillomavirus in patients with oropharyngeal cancer treated on TROG 02.02 phase III trial. *J Clin Oncol.* 2010;28(27):4142–8. [PubMed: 20697079]
5. Posner MR, Lorch JH, Golubeva O, Tan M, Schumaker LM, Sarlis NJ, et al. Survival and human papillomavirus in oropharynx cancer in TAX 324: a subset analysis from an international phase III trial. *Ann Oncol.* 2011;22(5):1071–7. [PubMed: 21317223]
6. Vermorken JB, Specenier P. Optimal treatment for recurrent/metastatic head and neck cancer. *Ann Oncol.* 2010;21 Suppl 7:vii252–61. [PubMed: 20943624]

7. Graham PJ, Brar MS, Foster T, McCall M, Bouchard-Fortier A, Temple W, et al. Neoadjuvant chemotherapy for breast cancer, is practice changing? A population-based review of current surgical trends. *Ann Surg Oncol*. 2015;22(10):3376–82. [PubMed: 26202561]
8. Tsao AS. Current readings: window-of-opportunity trials for thoracic malignancies. *Semin Thorac Cardiovasc Surg*. 2014;26(4):323–30. [PubMed: 25837547]
9. Schmitz S, Duhoux F, Machiels JP. Window of opportunity studies: do they fulfil our expectations? *Cancer Treat Rev*. 2016;43:50–7. [PubMed: 26827692]
10. Sacco AG, Worden FP. Molecularly targeted therapy for the treatment of head and neck cancer: a review of the ErbB family inhibitors. *Onco Targets Ther*. 2016;9:1927–43. [PubMed: 27110122]
11. Egloff AM, Grandis JR. Targeting epidermal growth factor receptor and SRC pathways in head and neck cancer. *Semin Oncol*. 2008;35(3):286–97. [PubMed: 18544443]
12. Moreira J, Tobias A, O'Brien MP, Agulnik M. Targeted therapy in head and neck cancer: an update on current clinical developments in epidermal growth factor receptor-targeted therapy and immunotherapies. *Drugs*. 2017;77(8):843–57. [PubMed: 28382569]
13. Goldstein NI, Prewett M, Zuklys K, Rockwell P, Mendelsohn J. Biological efficacy of a chimeric antibody to the epidermal growth factor receptor in a human tumor xenograft model. *Clin Cancer Res*. 1995;1(11):1311–8. [PubMed: 9815926]
14. Vermorken JB, Mesia R, Rivera F, Remenar E, Kawecki A, Rottey S, et al. Platinum-based chemotherapy plus cetuximab in head and neck cancer. *N Engl J Med*. 2008;359(11):1116–27. [PubMed: 18784101]
15. Bonner JA, Harari PM, Giralt J, Azarnia N, Shin DM, Cohen RB, et al. Radiotherapy plus cetuximab for squamous-cell carcinoma of the head and neck. *N Engl J Med*. 2006;354(6):567–78. [PubMed: 16467544]
16. Schmitz S, Hamoir M, Reychler H, Magremanne M, Weynand B, Lhommel R, et al. Tumour response and safety of cetuximab in a window pre-operative study in patients with squamous cell carcinoma of the head and neck. *Ann Oncol*. 2013;24(9):2261–6. [PubMed: 23704200]
17. Gross ND, Bauman JE, Gooding WE, Denq W, Thomas SM, Wang L, et al. Erlotinib, erlotinib-sulindac versus placebo: a randomized, double-blind, placebo-controlled window trial in operable head and neck cancer. *Clin Cancer Res*. 2014;20(12):3289–98. [PubMed: 24727329]
18. Bauman JE, Duvvuri U, Gooding WE, Rath TJ, Gross ND, Song J, et al. Randomized, placebo-controlled window trial of EGFR, Src, or combined blockade in head and neck cancer. *JCI Insight*. 2017;2(6):e90449. [PubMed: 28352657]
19. Machiels JP, Bossi P, Menis J, Lia M, Fortpied C, Liu Y, et al. Activity and safety of afatinib in a window preoperative EORTC study in patients with squamous cell carcinoma of the head and neck (SCCHN). *Ann Oncol*. 2018;29(4):985–91. [PubMed: 29346507] Important, well-conducted window trial.
20. Ferris RL, Goncalves A, Baxi S, UM Martins, Gauthier H, Langenberg M, et al. LBA46—an open-label, multicohort, phase 1/2 study in patients with virus-associated cancers (CheckMate 358): safety and efficacy of neoadjuvant nivolumab in squamous cell carcinoma of the head and neck. *Ann Oncol*. 2017;28(suppl_5):ESMO 2017 Congress.
21. Uppaluri R, Zolkind P, Lin T, Nussenbaum B, Jackson R, Rich J, et al. Neoadjuvant pembrolizumab in surgically resectable, HPV negative, locally advanced head and neck squamous cell carcinoma (HNSCC). *J Clin Oncol* 35, 2017 (Suppl; abstr 6012) ASCO Annual Meeting 2017. 2017.
22. Bell R, Duhon R, Leidner R, Curti B, Ballesteros-Merino C, Piening B, et al. Neoadjuvant anti-OX40 (MEDI6469) prior to surgery in head and neck squamous cell carcinoma. *J Clin Oncol* 36, 2018 (suppl; abstr 6011). Oral abstract presentation at ASCO 2018 Annual Meeting. 2018. Important thorough window trial with a newer checkpoint inhibitor.
23. Colevas AD, Bedi K, Chang S, Nieves U, Chatterjee S, Davidzon A, et al. A study to evaluate immunological response to PD-1 inhibition in squamous cell carcinoma of the head and neck (SCCHN) using novel PET imaging with [18F]F-AraG. *J Clin Oncol* 36, 2018 (suppl; abstr 6050) ASCO 2018 Annual Meeting Poster Presentation. 2018.

24. Wolf GT, Fee WE Jr, Dolan RW, Moyer JS, Kaplan MJ, Spring PM, et al. Novel neoadjuvant immunotherapy regimen safety and survival in head and neck squamous cell cancer. *Head Neck*. 2011;33(12):1666–74. [PubMed: 21284052]
25. Berinstein NL, McNamara M, Nguyen A, Egan J, Wolf GT. Increased immune infiltration and chemokine receptor expression in head and neck epithelial tumors after neoadjuvant immunotherapy with the IRX-2 regimen. *Oncoimmunology*. 2018;7(5):e1423173. [PubMed: 29721379]
26. Ferris RL, Lenz HJ, Trotta AM, Garcia-Foncillas J, Schulten J, Audhuy F, et al. Rationale for combination of therapeutic antibodies targeting tumor cells and immune checkpoint receptors: harnessing innate and adaptive immunity through IgG1 isotype immune effector stimulation. *Cancer Treat Rev*. 2017;63:48–60. [PubMed: 29223828]
27. Srivastava RM, Lee SC, Andrade Filho PA, Lord CA, Jie HB, Davidson HC, et al. Cetuximab-activated natural killer and dendritic cells collaborate to trigger tumor antigen-specific T cell immunity in head and neck cancer patients. *Clin Cancer Res*. 2013;19(7):1858–72. [PubMed: 23444227]
- 28••. Shayan G, Kansy BA, Gibson SP, Srivastava RM, Bryan JK, Bauman JE, et al. Phase Ib study of immune bio-marker modulation with neoadjuvant cetuximab and TLR8 stimulation in head and neck cancer to overcome suppressive myeloid signals. *Clin Cancer Res*. 2018;24(1):62–72. [PubMed: 29061643] Important combination immunotherapy window trial.
29. Jie HB, Schuler PJ, Lee SC, Srivastava RM, Argiris A, Ferrone S, et al. CTLA-4(+) regulatory T cells increased in cetuximab-treated head and neck cancer patients suppress NK cell cytotoxicity and correlate with poor prognosis. *Cancer Res*. 2015;75(11):2200–10. [PubMed: 25832655]
30. Stabile LP, He G, Lui VW, Thomas S, Henry C, Gubish CT, et al. c-Src activation mediates erlotinib resistance in head and neck cancer by stimulating c-Met. *Clin Cancer Res*. 2013;19(2):380–92. [PubMed: 23213056]
31. Brand TM, Iida M, Wheeler DL. Molecular mechanisms of resistance to the EGFR monoclonal antibody cetuximab. *Cancer Biol Ther*. 2011;11(9):777–92. [PubMed: 21293176]
32. Postel-Vinay S, Collette L, Paoletti X, Rizzo E, Massard C, Olmos D, et al. Towards new methods for the determination of dose limiting toxicities and the assessment of the recommended dose for further studies of molecularly targeted agents—Dose-Limiting Toxicity and Toxicity Assessment Recommendation Group for Early Trials of Targeted Therapies, an European Organization for Research and Treatment of Cancer-led study. *Eur J Cancer*. 2014;50(12):2040–9. [PubMed: 24880774]
33. Eisenhauer EA, O'Dwyer PJ, Christian M, Humphrey JS. Phase I clinical trial design in cancer drug development. *J Clin Oncol*. 2000;18(3):684–92. [PubMed: 10653884]
34. Machiels JP, Haddad RI, Fayette J, Licitra LF, Tahara M, Vermorken JB, et al. Afatinib versus methotrexate as second-line treatment in patients with recurrent or metastatic squamous-cell carcinoma of the head and neck progressing on or after platinum-based therapy (LUX-Head & Neck 1): an open-label, randomised phase 3 trial. *Lancet Oncol*. 2015;16(5):583–94. [PubMed: 25892145]
35. Ferris RL, Blumenschein G Jr, Fayette J, Guigay J, Colevas AD, Licitra L, et al. Nivolumab for recurrent squamous-cell carcinoma of the head and neck. *N Engl J Med*. 2016;375(19):1856–67. [PubMed: 27718784]

Table 1.

Window of opportunity trials in HNSCC

Trial	N	Agent	Control arm	Timing	Duration	Primary end point	Significant toxicity related to drug	Delay in surgery
Baumen	55	1 Erlotinib	Yes	Neoadj	7–21 days	Biomarker	No	NR
		2 Desatinib						
		3 Combination						
Schmitz	33	Cetuximab	Yes	Neoadj	21 days	Safety	No	No
Gross	47	1 Erlotinib	Yes	Neoadj	7–14 days	Biomarker	No	NR
		2 Erlotinib pLus sul.indac						
MachielS	30	Afatinib	Yes	Neoadj	14 days	FDG-PET response	1 patient with G3 diarrhea and ARF	3 patients
Curry	39	Metformin	No	Neoadj	9–24 days	Biomarker	No	NR
Thomas	35	Erlotinib	No	Neoadj	8–29 days	Biomarker	6 patients discontinued because of G3 pruritis and G2 rash	No
Campo	107	Lapatinib	Yes	Prior to CRT	2–6 weeks	Biomarker	No	NA
Berinstein	27	IRX-2	No	Neoadj	10 days	Biomarker Safety	1 post-op wound complication possibly reLated	No
Ferris	29	Nivolumab	No	Neoadj	29 days	Safety	No	No
Uppaluri	21	PembroLizumab	No	Neoadj	14–21 days	LRR	No	No
BeLL	17	MEDI6469	No	Neoadj	8–14 days	Safety	No	No
Shayan	14	Cetuximab pLus motoLimod	No	Neoadj	21–28 days	Biomarker	No	No

LRR locoregional recurrence

Neoadj neoadjuvant prior to surgical resection